BASF
The Chemical Company

Alpha-Cypermethrin

Document M-CA, Section 1

IDENTITY OF THE ACTIVE SUBSTANCE

Compiled by:

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

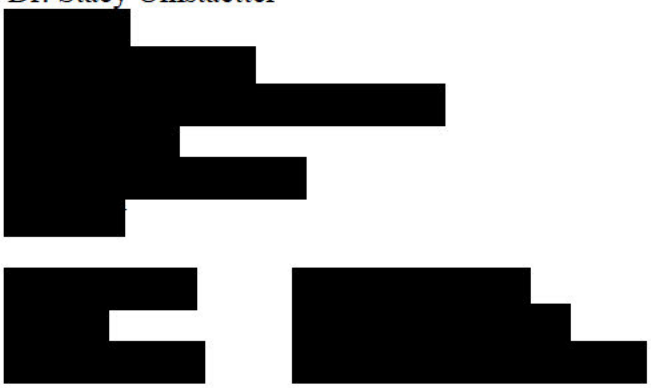
CA 1 IDENTITY OF THE ACTIVE SUBSTANCE

CA 1.1 Applicant

BASF Agro B.V. Arnhem (NL) – Zürich Branch
8036 Zürich-Wiedikon
Switzerland

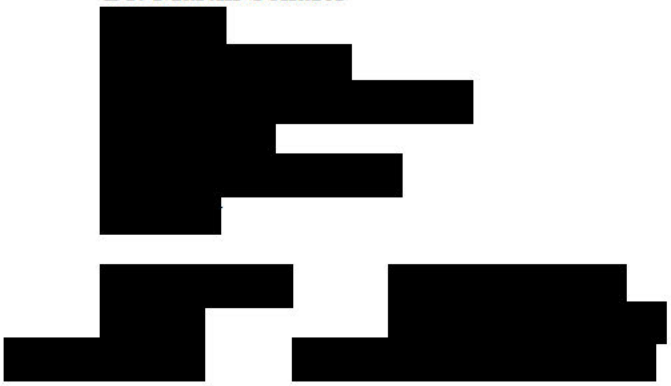
(a) Contact:

Dr. Stacy Umstaetter



(b) Alternative:

Dr. Martin Schäfer



CA 1.2 Producer

Producer of alpha-cypermethrin (legal entity):

BASF Agro B.V. Arnhem (NL) – Zuerich Branch
8036 Zuerich-Wiedikon
Switzerland

Contact person:

Alternative contact:
Dr. Martin Schaefer

Information on the production sites is contained in the confidential part (Doc JCA) of this dossier.

CA 1.3 Common Name Proposed or ISO-accepted and synonyms

Alpha-Cypermethrin

CA 1.4 Chemical Name (IUPAC and CA nomenclature)

IUPAC: Racemate of:
(S)-α-cyano-3-phenoxybenzyl-(1R)-cis-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylate and

(R)-α-cyano-3-phenoxybenzyl-(1S)-cis-3- (2,2-dichlorovinyl)- 2,2-dimethylcyclopropane carboxylate

(= cis-2 isomeric pair of Cypermethrin)

CA: Racemate of:
[1S[1α (R*), 3α]]-cyano-(3-phenoxymethyl-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylate and

[1R[1α (S*), 3α]]-cyano-(3-phenoxymethyl-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylate

(= cis-2 isomeric pair of Cypermethrin)

CA 1.5 Producer's Development Code Numbers

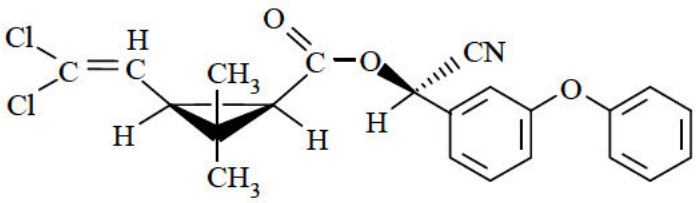
BASF Number:	BAS 310 I
BASF Registry Number:	Reg.No. 4078193
BASF CL Number (old):	CL 900049

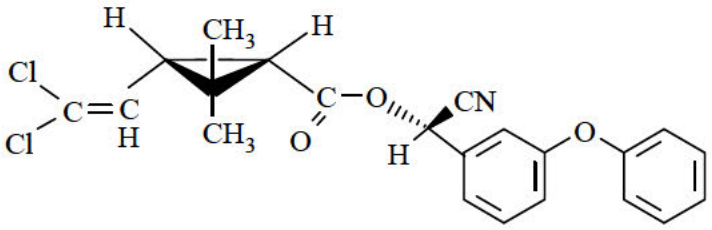
CA 1.6 CAS, EC and CIPAC Numbers

CAS:	67375-30-8
CIPAC:	454
EINECS:	Not allocated

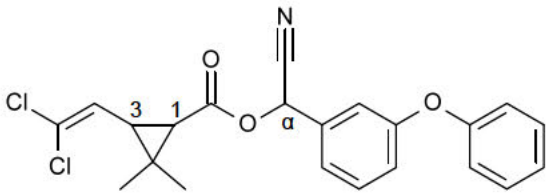
CA 1.7 Molecular and Structural Formula, Molar Mass

Structural formula:





or



Configuration at C Atom		
1	3	α
S	S	R
R	R	S

Molecular formula: $C_{22}H_{19}Cl_2NO_3$

Molecular mass: 416.3 g/mol

CA 1.8 Method of Manufacture (synthesis pathway) of the active substance

CONFIDENTIAL information - data provided separately (Document J)

CA 1.9 Specification of Purity of the Active Substance in g/kg

CONFIDENTIAL information - data provided separately (Document J)

CA 1.10 Identity and Content of Additives (such as Stabilisers) and impurities

CA 1.10.1 Additives

CONFIDENTIAL information - data provided separately (Document J)

CA 1.10.2 Significant impurities

CONFIDENTIAL information - data provided separately (Document J)

CA 1.10.3 Relevant impurities

CONFIDENTIAL information - data provided separately (Document J)

CA 1.11 Analytical Profile of Batches

CONFIDENTIAL information - data provided separately (Document J)

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Alpha-Cypermethrin

Document M-CA, Section 2

PHYSICAL AND CHEMICAL PROPERTIES OF THE ACTIVE SUBSTANCE

Compiled by:

[redacted]

[redacted]

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
July 10, 2017	CA 2.6 Solubility in organic solvents: values for acetonitrile were changed (values were incorrect) 2 formerly submitted studies have been added, which contain data on the valiation of the analytical methods.	BASF DocID 2017/1134411
	CA 2.14: additional studies (solubility in water of metabolites) were added (DocID 2001/1009912 and DocID 2001/1009851)	
	CA 2.5: additional data (+footnote)	

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Table of Contents

CA 2 PHYSICAL AND CHEMICAL PROPERTIES OF THE ACTIVE SUBSTANCE

<table>
<tr><th>Test or Study & Data Point</th><th>Guideline and method</th><th>Test material purity and specification</th><th>Findings</th><th>GLP Y/N</th><th>Reference</th></tr>
<tr><td>CA 2.1 Melting point and boiling point</td><td>OECD 102 (DSC/TG)</td><td>PMAM000630: 99.3%</td><td>Melting point (onset): 82.1 °C
Decomposition temp. (onset): approx.. 248 °C</td><td>Y</td><td>[see 2014/1097996 Kroehl T. 2014 a]</td></tr>
<tr><td>CA 2.2 Vapour pressure, volatility</td><td>OECD 104,
EEC A.4
(thermo-gravimetry)</td><td>PMAM000630: 99.3%</td><td><table><tr><th>Temperature</th><th colspan="2">Vapour Presure</th></tr><tr><th>[°C]</th><th>[hPa, mbar]</th><th>[Pa]</th></tr><tr><td>20</td><td>3.8 x 10^{-9}</td><td>3.8 x 10^{-7}</td></tr><tr><td>25</td><td>8.5 x 10^{-7}</td><td>8.5 x 10^{-7}</td></tr></table></td><td>Y</td><td>[see 2014/1097996 Kroehl T. 2014 a]</td></tr>
<tr><td></td><td>Calculation</td><td></td><td>Henry’s Law Constant:
H = 5.3 * 10^{-2} Pa m^3 / mol</td><td>N</td><td>[see 2014/1161030 Kroehl T. 2014 i]</td></tr>
<tr><td>CA 2.3 Appearance (Physical state, colour)</td><td>OPPTS 830.6302
OPPTS 830.6303
(visual examination)</td><td>PMAM000630: 99.3%</td><td>The material is a fine powder of white colour.</td><td>Y</td><td>[see 2014/1097996 Kroehl T. 2014 a]</td></tr>
</table>

Test or Study & Data Point	Guideline and method	Test material purity and specification	Findings	GLP Y/N	Reference
CA 2.4 Spectra (UV/VIS, IR, NMR, MS), molar extinction at relevant wavelengths, optical purity			Information already reported and peer-reviewed previously (see Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004) at λ_{max} (276 nm) : ε = 2073 L mol^{-1} cm^{-1} at 300 nm : ε = 38.64 L mol^{-1} cm^{-1}		Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004
CA 2.5 Solubility in water	OECD 105 (Column elution method)		The solubility of alpha-cypermethrin in water at 20 °C was determined to be 3 (± 1.1) µg/L. No colloidal alpha-cypermethrin in the aqueous samples (see note below).	N	[see 2014/1159447 Bacher R. 2014a]
CA 2.6 Solubility in organic solvents	CIPAC MT 181	PMAM000630: 99.3%	Solubility of alpha-cypermethrin in: Toluene > 250 g/L solvent Dichloromethane > 250 g/L solvent Acetone > 250 g/L solvent Ethyl acetate > 250 g/L solvent n-Heptane 10 – 14 g/L solvent Methanol 25 – 29 g/L solvent ~~Acetonitrile 200 – 500 g/L solvent~~ Acetonitrile 200 – 250 g/L solvent	Y	[see 2014/1159449 Bacher R. 2014c]
	FIFRA subdivision D, §63-8 (cfr. OECD 105) (flask method + capillary GCFID - see available data in M-CA 4.1.2 (g))	ST91/118: 97.3 %	Solubility of alpha-cypermethrin at 21 °C in: n-Hexane: 6.5 g/L Toluene: 596 g/L Dichloromethane: miscible (> 10^3 g/L) Methanol: 21.3 g/L 2-Propanol: 9.6 g/L Acetone: miscible (> 10^3 g/L) Ethyl acetate: 584 g/L	Y N	Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004 [see AL-312-002 Bohle J.F. 1991a] [see AL-210-004 Bohle J.F. 1991b]

Test or Study & Data Point	Guideline and method	Test material purity and specification	Findings	GLP Y/N	Reference
CA 2.7 Partition co-efficient n-octanol/water	OECD 117 EC A.8 (HPLC method)	PMAM000630: 99.3%	Partition coefficient of alpha-cypermethrin in n-octanol/water: Pow = 689005 log Pow = 5.8	N	[see 2014/1159448 Bacher R. 2014b]
CA 2.8 Dissociation in water - dissociation constant(s) (pKa values) - identity of dissociated species - dissociation constant(s) (pKa values) of the active principle			Information already reported and peer-reviewed previously (see Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004) No dissociation of the a.s.		Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004
CA 2.9 Flammability and self-heating			Information already reported and peer-reviewed previously (see Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004) Not highly flammable Not auto-flammable		Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004
CA 2.10 Flash point			Not applicable, as the melting point > 40 °C.		
CA 2.11 Explosive properties			Information already reported and peer-reviewed previously (see Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004) Not explosive		Review Report for alpha-cypermethrin, SANCO/4335/2000 final, 13 February 2004

Test or Study & Data Point	Guideline and method	Test material purity and specification	Findings	GLP Y/N	Reference
CA 2.12 Surface Tension			Not applicable, as alpha-cypermethrin technical is solid at ambient temperature.		
CA 2.13 Oxidising properties			Information already reported and peer-reviewed previously (see Addendum to EU registration monograph, Annex B, B.2 Physical and chemical properties, December 2002) Not oxidizing		Addendum to EU Registration Mono-graph, Annex B, B.2 Physical and chemical properties, December 2002
CA 2.14 Other studies			~~not required~~		
	EC A.6 OECD 105	Reg.No. 4080830 (CL# 912554) AC12717-65: 99.0 %	Solubility in water of the metabolite Reg.No. 4080830: Deionized water 129.0 (± 0.2) mg/L, pH 4.0 NaOH 240.0 g/L, pH 9.1 HCl (0.1 Mol/L) 109.4 (± 0.9) mg/L, pH 0.7	Y	[see 2001/1009912 Daum A. 2001a]
	EC A.6 OECD	Reg.No. 130213 (CL# 206128) AC12251-34 99.0 %	Solubility in water of the metabolite Reg.No. 130213: Deionized water 24.68 (± 0.08) mg/L, pH 4.2 NaOH 221 g/L, pH 9.1 HCl (0.1 Mol/L) 9.2 (± 0.74) mg/L, pH 0.9		[see 2001/1009851 Daum A. 2001b]

Note: Absence of colloidal matter checked (please see below the statement of the contract institute):

There are no observations and related remarks in the lab journal regarding the presence of any colloidal matter in the aqueous extracts derived from the column elution experiments. In the case of the test item alpha-cypermethrin, a presence of colloidal matter in the aqueous fractions is very improbable:

- The water solubility of this analyte is extremely low (approx. 3 µg/L water, as experimentally determined within the study). Colloidal alpha-cypermethrin resulting in a Tyndall effect and generating a visible turbidity of the aqueous fractions would require much higher concentrations of alpha-cypermethrin. Thus, there is no evidence for presence of colloidal alpha-cypermethrin in the aqueous samples.
- If colloidal alpha-cypermethrin would be present in the aqueous fractions, it will be significantly dissolved in the extracts from the column elution experiments after 1:1 dilution with acetonitrile (see sample preparation in section 2.4.3). However, all concentrations of alpha-cypermethrin observed in these fractions were in the low µg/L concentration range excluding the presence of undissolved alpha-cypermethrin in the samples.
- In addition, alpha-cypermethrin present as colloidal material would be significantly eliminated from the aqueous fractions by filtering through the polyethylene frits present in the SPE reservoirs used in the experiments (see section 2.4.2 of the report).

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Alpha-Cypermethrin

Document M-CA, Section 3

FURTHER INFORMATION ON THE ACTIVE SUBSTANCE

Compiled by:

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[redacted]
[redacted]
[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
10/Jul/2017	additional study - MSDS (DocID 2016/1236516) in CA 3.8, CA 3.9 and CA 3.10	BASF DocID 2017/1135054

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Table of Contents

CA 3 FURTHER INFORMATION ON THE ACTIVE SUBSTANCE

CA 3.1 Use of the Active Substance

Alpha-cypermethrin exhibits high levels of efficacy on a broad range of crop-relevant noxious insects at dose rates between 10 to 30 g ai/ha. The proposed use of the active is accordingly as a foliar-applied agricultural / horticultural insecticide for the control of a range of sucking and chewing pests in a variety of crops.

CA 3.2 Function

Alpha-cypermethrin is used as an insecticide.

CA 3.3 Effects on Harmful Organisms

Alpha-cypermethrin acts as a broad-spectrum insecticide with efficacy against chewing/biting and piercing/sucking insect pests in many agricultural and horticultural crops. Independent from the crop growth-stage or specific cultivation systems, applications are made as foliar sprays in order to control relevant phyto-phagous insect pests which have exceeded economical threshold.

CA 3.4 Field of Use Envisaged

Alpha-cypermethrin is used in horticulture and agriculture as insecticide in order to protect the relevant crop against the attack of plant-destructive insects and so to prevent quantitative and qualitative losses.

CA 3.5 Harmful Organisms Controlled and Crops or Products Protected or Treated

Alpa-cypermethrin is used for the control of most important crop-infesting insect species of the out of the groups of Homoptera, Heteroptera, Lepidoptera, Coleoptera, Diptera and Thysanoptera.

The area of use covers a broad range of crops including:

Cereals (Barley, Wheat, Oat, Rye), Maize, Rice, Millet, Linseed, Cotton, Tobacco,
Oilseed Crops (Oilseed Rape, Mustard, Soybeans,)
Beet roots (Sugarbeet, Cichory roots),
Legume Crops used as dry pulses/dry harvest such as beans (field beans), peas (chickpeas, field peas) or used as legume vegetables/fresh harvest such as beans with & without pods (green beans, french beans, snap beans,), peas with & without pods, Alfalfa,
Fruiting vegetables - Solanacea (Tomato, Eggplant, Peppers), Artichoks,
Fruit.Vegetables – Cucurbitacea (Cucumber, Cucurbits, Melon, Pumpkin, Zucchini/Courgette),
Brassica Vegetables (Broccoli, Brussel Sprouts, Cauliflower, Head cabbage, Leafy cabbage, Flowering cabbage, Kohlrabi).
Bulb Vegetables (Onion, Welsh onions, Shallot, Garlic, Chives, Salad onions).
Root & Tuber Vegetables (Carrots, Potato, Horse radish, Turnips, Swedes, Parsnip, Celeriac, Rocket, Scarole),
Stem Vegetables (Leek, Celery, Asparagus, Fennel, Spinach)
Leafy Vegetables & Herbs (Lettuce, Cichory, Dill), Poppy
Biannual / perennial crops:
Berries & Small Fruits (Currants, Raspberries, Strawberries),
Pomefruits (apple, pear, Quince)
Stonefruits (cherry, plums, peaches, apricots),
Citrus, , Olives, Table- and vine grapes.

Following insect pests are controlled with Alpha-cypermethrin in relevant major crops (table 1):

Crops	Key targets	
Apple	*Cydia pomonella*	codling moth
	Aphis pomi	green apple aphid
	Dysaphis plantaginea	rosy apple aphid
	Dysaphis devecta	*leaf-curling aphid*
	Anthonomus pomorum	*apple blossom weevil*
	Hoplocampa testudinea	*apple sawfly*
Citrus	Ceratitis capitata	Mediterranean fruit fly
	Toxoptera aurantiae	black citrus aphid
	Aphis spiraecola	green citrus aphid
Peach	*Grapholita molesta*	oriental peach moth
	Myzus persicae	peach aphid
	Ceratitis capitata	Mediterranean fruit fly
Olive	*Bactrocera oleae*	olive fly
	Preys oleae	olive moth
Grapes	*Eupoecilia ambiguella*	grape berry moth
	Lobesia botrana	vine moth
	Scaphoideus titanus	golden flavescence cicadella
	Empoasca vitis	green leafhopper
Tomatoes	*Heliothis armigera*	bollworm
	Spodoptera sp (armyworms)	armyworms
	Plusia sp	*Plusia sp*
Pepper	*Aphis gossypii*	*cotton aphid*
	Myzus persicae	*green peach aphid*
	Tuta absoluta	*Tuta absoluta* (tomato leafminer)
Leafy veg.	*Plutella maculipennis*	diamond back moth
	Pieris brassicae	cabbage white
	Delia radicum	*cabbage root fly*
	Nasonovia ribisnigri	*lettuce aphid*
	Myzus persicae	*peach aphid*
	Brevicoryne brassicae	*cabbage aphid*
Potatoes	Leptinotarsa decemlineata	Colorado potato beetle
	Macrosiphum euphorbiae, A. nasturtii, Aulacorthum solani,	Several aphids
	Phtorimaea operculella	potato moth

Crops	Key targets	
Cereals	*Rhopalosiphum padi*	*apple grain aphid*
	Metopolophium dirhodum	*rose grain aphid*
	Sitobion avenae	*grain aphid*
	Oscinella frit	frit fly
	Delia coarctata	wheat bulb fly
	Oulema melanopus	cereal leaf beetle
	Agromyza sp	miner flies
	Psammottetix alienus	leafhopper – virus transmission
	Cnephasia pumicana	cereal tortrix moth
	Sitodiplosis mosellana & Contarinia tritici	midges
Cotton	*Heliothis armigera*	tobacco budworm
	Pectinophora gossypiella	pink bollworm
	Aphis gossypii	cotton aphid
OSR	*Ceutorhynchus napi*	cabbage stem weevil
	Ceutorhynchus assimilis	*pod weevil*
	Ceutorhynchus quadridens	*small stem weevil*
	Meligethes aeneus	pollen beetle
	Phyllotreta sp.	cabbage flea beetle
	Psylliodes chrysocephala	cabbage stem flea beetle
	Brevicoryne brassicae	cabbage aphid
	Myzus persicae	green peach aphid
	Athalia rosae	*turnip saw fly*
	Dasineura brassicae	brassica pod midge
Peas	*Acyrtosiphum pisum*	Pea aphid
Beans	*Aphis fabae*	Black bean aphid
Corn	*Diabrotica virgifera*	western corn root worm
	Ostrinia nubilalis	European Corn borer
	Rhopalosiphum padi	apple grain aphid
	Metopolophium dirhodum	rose grain aphid
	Sitobion avenae	grain aphid

CA 3.6 Mode of Action

Alpha-cypermethrin, a synthetic pyrethroid, acts like naturally occurring pyrethrum on the nervous system of insects. The nervous system serves to transmit information in the organism. Stimuli act on sensory receptors which trigger a response. The nerves transmit this response e.g. to muscles where a reaction takes place. Changes in electrical charge to the membranes of the nerve fibres play a decisive role in producing and transmitting responses. In an idle state (resting potential) the nerve fibre membranes are charged with a concentration of Na+ ions, positive on the exterior and negative on the interior. The stimuli cause specific Na+ channels to open through which Na+ ions flow into the interior of the nerve fibres and cause a reversal in the state of charge. A so-called action potential is created along the length of the membrane. In order to be able to receive new stimuli the stimulated areas have to be returned to their idle state. This is achieved by actively transmitting ions and closing the specific Na+ channels. If the Na+ channels opened by the creation of action potentials are kept open by the effect of Alpha-cypermethrin, this leads to a succession of sustained action potentials which destroys the impulse sequence and transmission. The affected insect is thus subject to uncoordinated movements and finally death.

Although Alpha-cypermethrin is highly effective against insects it is not toxic for warm-blooded animals because of its high degree introduction of selectivity in the relevant application quantities. Alpha-cypermethrin is active against both piercing-sucking and chewing insects. The effect is triggered by the insects contact with, or ingestion of the active ingredient. In addition to its effect on mature and immature insects Alpha-cypermethrin has also demonstrated a significant ovicidal effect. The excellent knock-down and residual control of Alpha-cypermethrin products are complemented by a strong repellent effect on pests. Because the effect of Alpha-cypermethrin is increased at lower temperatures, Alpha-cypermethrin products can also ideally be used under cool climatic conditions. Due to low solubility in water and excellent rain-fastness the product is not washed off plant surfaces by rain. This means that a high degree of control can be achieved in unfavorable climatic conditions.

CA 3.7 Information on Occurrence or Possible Occurrence of the Development of Resistance and Appropriate Management Strategies

Preliminary remarks

Structure and headlines of the following text are in accordance with the chapter 'Registration Requirements' from the EPPO-Guideline PP1/213/3.

The term 'resistant insect' can be defined in different ways. The agricultural field definition requires that the resistant population must survive the recommended use rate of the insecticide under normal field conditions. However, the recommended field use rate of an insecticide is subjective, and may vary from region to region depending on the crop, cultural practices, or economics. In addition, the environment (i.e. weather, soil type, growing conditions, stress, etc.) plays a significant role toward the insecticide's efficacy. For marginal cases of resistance, a population that typically survives a recommended field use rate may be controlled by the same rate under greenhouse conditions. From a scientific point of view resistance can be defined as a genetically inherited statistical difference in insecticide responses between two populations of the same species. Even though there is a statistical difference among populations, this does not necessarily mean the most resistant population will survive normal field use rates.

IRAC-Classification of Alpha-cypermethrin

The Insecticide Resistance Action Committee (IRAC) developed a system of classifying insecticides according to their mode of action. This classification system groups insecticides into various categories designated by groups. One of the purposes of this system is to make it easier for farmers and farm advisors to understand which insecticides share the same mode of action without having to know the actual biochemical basis.

The classification system is not based on resistance risk assessment; therefore, different insecticides of the same IRAC-class may have different degrees of risk for developing resistance.

According to the IRAC-classification, Alpha-cypermethrin is in the Group 3 (Sodium channel modulators).

Mechanism of Resistance of Alpha-cypermethrin

Insecticide resistance to synthetic pyrethroids occurs via two main basic mechanisms. The first is modification of the target site, reducing its sensitivity to the insecticide (e.g., the *kdr* gene). The second mechanism involves metabolism or sequestration of the insecticide molecule by various enzymes. This is achieved by increased activity levels of specific esterases mixed function oxidases, glutathione S-transferases or GSTs), or a combination of these enzyme systems. A third mechanism occurs when insects absorb toxins slower than susceptible insects (penetration resistance). And the last recognized resistance mechanism occurs when insects detect or recognize the presence of the insecticide and avoid the toxin (behavioral resistance). Target site resistance and metabolic resistance are overwhelmingly the most important forms of resistance in key economic insect pests. Penetration and behavioral resistance have not been reported in Europe with regards to pyrethroids (and specifically not to alphacypermethrin).

Target site resistance

The common target site resistance to pyrethroids is the knock-down resistance associated with amino acid mutations in the voltage-gated sodium channel (the *kdr* and super-*kdr* genes) conferring nerve insensitivity. Multiple knock-down mutation alleles have been identified in multiple agriculturally important species, such as *Myzus persicae*, *Thrips tabaci*, *Plutella xylostella*, and *Tuta absoluta*, etc. The *kdr* L1014F allele is associated to moderate resistance to pyrethroids and DDT and is the most common *kdr* allele encountered in insects. The *super-kdr* allele is consisted of L1014F and M918T mutations and is associated to higher levels of pyrethroid resistance Target resistance of pyrethroids alpha-subunit sodium channel mutants ($para^{ts1}$ and $para^{ts2}$) in *Drosophila melanogaster* have been shown to have reduced sodium channel numbers on neural membranes, neurophysiological defects, and are 10-30 times more resistant to pyrethroids. Target site resistance to pyrethroids has been associated with deficits in reproductive success suffered by the resistant populations in *Heliothis virescens*.

Metabolic resistance

Metabolic resistance to pyrethroids is achieved by increased activity levels of specific esterases, mixed function oxidases, glutathione S-transferases or GSTs, or a combination of these enzyme systems. For example, metabolic resistance to pyrethroids and carbamates in *Myzus persicae* is due to the overproduction of E4 and/or FE4 carboxylesterases. Resistance to pyrethroids in *Meligethes aeneus* is due to increased cytochrome P450. Studies have shown that a typical characteristic of pyrethroid metabolic resistance is overexpression of detoxification genes at transcription level, resulting in increased protein amounts and enzyme activities that lead to higher level of detoxification. Up-regulated detoxification genes conferring pyrethroid resistance have been identified in different species. Overexpressed CYP9A12 and CYP9A14 from pyrethroid-resistant *Helicoverpa armigera* were confirmed to metabolize pyrethroids; CYP6AY3v2 and CYP6FU1 were significantly overexpressed (16 and 24 folds) in a *Laodelphax striatellus* resistant strain. Metabolic resistance to pyrethroids has been suggested to cause low fecundity in *H. virescens*.

In addition to the mechanisms of resistance described above, it is important to note that additional factors could precipitate a field failure without invoking resistance mechanisms, even when alphacypermethrin is applied correctly in the field. One factor influencing the activity of a pyrethroid is the developmental status of an insect. Certain life stages are more susceptible to pyrethroids. For example, the fifth instar (L5) larvae of *Heliothis virescens* are 130 times less sensitive than the younger second instar (L2) larvae as shown in laboratory tests. Similarly, Soderlund et al. (1983) found *Laphygma frugiperda* L5 stage larvae are 25 times less sensitive than the L2 stage. Controlling insects at the correct life stage is essential. Temperature effects also contribute to the efficacy of certain pyrethroids.

Evidence of Resistance to Alpha-cypermethrin

Inherent risk

Alpha-cypermethrin is a chemically resolved product consisting of two isomers of the eight, which make up cypermethrin (*cis*-isomer of cypermethrin). Alpha-cypermethrin is a fast-acting insecticide that works by either contact or ingestion providing control of insect pests in a wide range of crops including cereals (wheat, barley), potatoes, vegetables and oilseed rape.
Based on factors including genetics, pest life cycle, number of pest generation/year, frequency of insecticide application, etc., there is a risk for resistance development to synthetic insecticides in the Alpha-cypermethrin targeted crops.

Resistance Risk per Crop

Cereals
Aphids are the most serious pests of cereals. These include grain aphid, *Sitobion avenae*, rose-grain aphid, *Metopolophium dirhodum*, oat bird-cherry aphid, *Rhopalosiphum padi*, and corn leaf aphid, *R. maidis*. Aphids can also be important vectors of plant viruses in cereals including Barley Yellow Dwarf Virus, BYDV.
Major insect pests of cereals are controlled by Alpha-cypermethrin. Insecticide resistance risk in aphids may generally be considered high because of their short generation time, the occurrence of multiple generations per year, abundant populations, and documented ability to adapt to insecticides. However, there are reports indicating that in aphids, adverse selection is imposed through poorer winter survival, maladaptive behavior, reduced reproductive fitness, and increased predation and parasitism. There is no resistance reported for those major aphid species listed above.
Therefore, based on more than two decades of pyrethroids use on cereals in Europe with virtually no recorded resistance, it can be concluded that the inherent risk for pyrethroid resistance is low in cereals. Modifiers are therefore not required to reduce the risk of resistance development to an acceptable level.

Potatoes

Alpha-cypermethrin is recommended for controlling of the Colorado potato beetle (CPB), *Leptinotarsa decemlineata*, and aphids in this crop.

In Europe some level of resistance against pyrethroids including Alpha-cypermethrin has been reported for CPB. In Germany an annually increasing tolerance to pyrethroids was observed in areas with extensive insecticide use. Complete control could only be achieved in CPB instars L1-L2 at these locations.

The peach-potato aphid, *Myzus persicae*, the melon aphid *Aphis frangulae*, the buckthorn aphid *Aphis nasturtii*, and potato aphid, *Macrosiphum euphorbiae*, are the most important European aphid species of potatoes.
On a global scale, insecticide resistance is widespread in *M. persicae*. Several forms of resistance to insecticides have been reported for this insect. Products used for control of other pests, occurring simultaneously with *M. persicae*, may further complicate resistance development in this aphid. Fortunately, resistant aphids tend to be less fit and have a reduced ability to survive the winter. As a result, individuals with lower levels of resistance are more common the following spring and these are relatively well controlled with established aphicides (IRAC).

Pyrethroids remain highly effective in the field in many areas, even against populations that show pyrethroid resistance in laboratory bioassays. This might be explained by the initial, transient effects of the compounds via hyperactivity and ataxia causing exposed aphids to fall from the foliage.
In Europe, there is no documented strong and wide spread CPB and peach-potato aphid resistance to Alpha-cypermethrin. Regardless, the inherent risk and abundant documented resistance of these two species to pyrethroids remains high. BASF therefore proposes use of modifiers to reduce the risk to an acceptable level.

Fruiting and leafy vegetables, lettuces

Table 1 shows major and minor insect pests of vegetables in Europe. Aphids are some of the most serious pests of vegetables. Melon-cotton aphid, *Aphis gossypii*, and peach-potato aphid, *M. persicae*, are two of the most economically important aphid pests of vegetables in Europe.

A. gossypii is largely confined to glasshouse vegetable crops in Europe. Resistance to organophosphoates by *A. gossypii* was first reported in 1964 and subsequently to carbamates, pyrethroids, neonicotinoids, and others. A *kdr*-type mechanism of resistance to pyrethroids is known to occur in this species, but its geographical range is unclear. Although resistance of many insecticides was reported in Europe, Alpha-cypermethrin resistance has been reported only from Pakistan.
The currant-lettuce aphid, *Nasonovia ribisnigri*, is a pest of lettuce. Control failures have been reported in Spain and France. Bioassays with nine strains of *N. ribisnigri* collected from lettuce in the UK showed widespread, varied, but overall low levels of resistance to pyrethroids.
Resistance to etofenprox, a non-ester pyrethroid molecule, in cabbage aphid, *Brevicoryne brassicae*, has also been reported in Spain. This species has not been reported as being resistant to Alpha-cypermethrin (IRAC), but it is nonetheless assumed as a result of pyrethroid cross-resistance.

Whiteflies are also important insect pests, especially in greenhouse crop production. The glasshouse whitefly, *Trialeurodes vaporariorum*, has developed resistance to all major insecticide classes, but no report on Alpha-cypermethrin resistance yet. The sweetpotato whitefly, *Bemisia tabaci*, is another whitefly species especially in southern European glasshouses. *B. tabaci* has developed high levels of resistance to pyrethroids and other established modes of action. Resistance in *B. tabaci* to Alpha-cypermethrin has been reported from Greece and China.
Alpha-cypermethrin controls piercing-sucking (e.g., aphids, whiteflies) and chewing Lepidoptera (e.g., armyworms, diamondback moth) insect pests in vegetable crops. The inherent resistance risk associated with *M. persicae*, *A. gossypii*, *T. vaporariorum*, *B. tabaci*, and *Spodoptera* spp. is considered by BASF as being “high” because of short generation times, the occurrence of multiple generations per year, abundant populations, wide range of potential host plants, and documented ability to develop insecticide resistance (IRAC Groups in European countries).

The diamondback moth, *Plutella xylostella*, is an important pest of cole crops in many regions of the world. It is however relatively rare in Europe. This species has a long history of developing resistance to a wide variety of insecticides. It is reported to be resistant to 15 pyrethoid insecticides, however, resistance to Alpha-cypermethrin has not been reported in the EU.

Therefore, in cole crops and kale the inherent risk of pyrethroid resistance for field pest populations is considered as being moderate, and BASF recommends that modifiers should be used to reduce the risk to an acceptable level. For *B. tabaci* however, BASF considers the risk of pyrethroid resistance as high. BASF recommends that efficacy of Alpha-cypermethrin applications against this pest is assessed carefully, and alternative products with different mode of action are used as soon as failures in *B. tabaci* control occur or pyrethroid resistance is considered problematic.

Oilseed Rape

The major and minor insect pests of oilseed rape (OSR) are shown in Table 1. It should be noted that several rape pests are also pests of other *Brassica* crops, therefore, there is always possibility of movement between crops. Information presented in the EPPO guidelines for vegetable brassicas may also have relevance for OSR.

An increasing number of failures in controlling pollen beetle (*Meligethes aeneus*) with pyrethroids has been reported from Denmark, Poland, France and Germany. A reduction in sensitivity in *M. aeneus* has also been observed for Alpha-cypermethrin.
Insecticide Resistance Action Committee (IRAC) pollen beetle resistance monitoring efforts from 2007-2009 show pyrethroid resistant populations dominating in many countries in Europe (Belgium, Czech Republic, France, Germany, Netherlands, Sweden, Switzerland, and Poland). Additional surveys indicate presence of resistant populations in the UK, Finland, Hungary, Russia, Austria, Denmark, Norway, and Lithuania..

Alpha-cypermethrin is recommended for controlling several Coleoptera species including flea beetles, pollen beetles, and stem weevils, all of which are important pests of OSR. Only pollen beetles have been recorded to have Alpha-cypermethrin resistance. Pollen beetle populations should be monitored carefully for resistance issues and only where control levels were reported as sufficient, application of Alpha-cypermethrin should be considered.

Based on the extensive use of Alpha-cypermethrin and other pyrethroids on oilseed rape and the occurrence of control failures in pollen beetles, the inherent risk of pyrethroid resistance development to oilseed rape pests is considered by BASF to be “high”.

Cross Resistance to Alpha-cypermethrin

Cross resistance among insecticides of a same mode MoA is a common phenomenon. A given species found to be resistant to one pyrethroid is often resistant to other pyrethroids. The *kdr* or "knockdown" gene confers resistance to DDT and synthetic pyrethroids. Esterases and mixed function oxidases are known to detoxify a number of organophosphate and carbamate insecticides as well as pyrethroids. For example, cytochrome P450 and esterase confer resistance to organophosphates, organochlorine, carbamates and pyrethroids in *H. virescens*, and *M. persicae*. Esterases conferred fenitrothoin and deltamethrin cross resistance in *Anophelus albimanus*.
Alphacypermethrin and other pyrethroids can therefore be affected by a number of resistance mechanisms, or combinations of these mechanisms.

Sensitivity Data

A database of baseline sensitivity was not developed at the time that Alpha-cypermethrin and other synthetic pyrethroids were developed. However, results of numerous in-house and outside contract field studies with various Alpha-cypermethrin formulations that show high levels of pest control are available. The target species in these studies include many of the high-risk insect pests for resistance development such as *M. persicae*, *R. padi*, *R. maidis*, *M. dirhodum*, other aphids, *L. decemlineata*, and *P. xylostella*. These efficacy trials serve as the best available indication of the susceptibility of target pests in Europe.
Most recently, BASF sponsored a study testing Alpha-cypermethrin against various aphid populations in Italy and Spain. The results showed Alpha-cypermethrin susceptibility variation among species and populations, but most of populations maintained full susceptibility. No major shifts in pest susceptibility or field failures were detected or reported.
The low and acceptable level of resistance risk with field use of Alpha-cypermethrin is best evidenced by nearly two decades of commercial field performance.

Use Pattern (unrestricted use)

The major recommended risk modifiers to minimize the likelihood of the development of pest resistance to Alpha-cypermethrin are:

Cereals:
- maximum two applications/year/pest
- if multiple applications are required, alternate with compounds having different modes of action and no cross-resistance
- do not alternate with products of the same mode of action or those showing cross-resistance

Potato:
- maximum two applications/year/pest
- alternate with compounds with different modes of action and no cross-resistance
- do not alternate with products of the same mode of action or those showing cross-resistance

Cole crops/kale:
- maximum two applications/year/pest
- alternate with compounds having different modes of action and no cross-resistance
- do not alternate with products of the same mode of action or those showing cross-resistance

Oilseed rape:
- maximum two applications/year/pest
- alternate with compounds having different modes of action and no cross-resistance
- do not alternate with products of the same mode of action or those showing cross-resistance

Resistance Risk Assessment of Unrestricted Use Pattern and Acceptability of Resistance Risk

For the last 20 years, Alpha-cypermethrin applications have permitted growers to use the product without restriction for resistance management reasons. Furthermore, other members of the pyrethroid family have been similarly applied with minimal or no restriction of use for resistance management reasons, over the last 30 years. This creates an opportunity to observe the effects of an "unrestricted" use pattern based on historical evidence. The supported "safe use" GAPs for Annex I inclusion are cereals, oilseed rape, cabbage and cucumber/courgettes. Resistance to pyrethroids in these crops in the EU has been reported (based on field failures, laboratory work, or rumors) for *A. gossypii* on vegetables (Greece, UK), *M. persicae* on vegetables (Greece, UK), pollen beetle, *meligethes aeneus* in oilseed rape and *Spodoptera exigua* on vegetables (Spain).

High inherent risk profiles include single-site mode of action, monogenetic resistance, ease of metabolism, short life cycle, high fecundity, widespread distribution, existence of a mechanism that the pest metabolizes a range of pesticides, as well as the existence of cross resistance. Despite these risk factors, resistance development in insect pests found in tropical zones using similar application rates have been much more prone to the development of resistance when compared to temperate zones like Europe. Fewer life cycles and harsher winters appear to select against resistant genotypes, which may contribute to a slow development of insecticide resistance in Europe. Two conclusions are clear. First, reported cases of Alpha-cypermethrin resistance in arthropod species on the intended crops even after three decades of using pyrethroids in Europe are still restricted to four pest species. Secondly, Europe climate and geography appears to contribute to a reduced rate of resistance development when compared to tropical zones.

Implementation of Management Strategy

BASF is founder member of the IRAC Working Group and has participated in forming the guidelines for management strategy.

The basic principles of resistance management are similar in both the prevention of resistance in a given population, as well as in the limitation of resistance after its first occurrence. Once problems have been detected, management strategies have to be adapted to the particular situation. As a rule, the following methods can be recommended:

- Sequences of insecticides with different modes of action
- Crop rotation
- Cultivation practices

BASF promotes further awareness of general aspects of insecticide resistance management in product and technical leaflets, training sessions to sales personnel, distributors and growers' associations. Additionally, specific aspects on the product use are provided for achieving best control results.

Following an example on key elements of the resistance management strategy for FASTAC including general and product specific aspects:

- Always follow IRAC guidelines for preventing and managing insecticide resistant insects.
- Maximise the use of cultural control measures wherever possible
- Use sequences of effective insecticides with different modes of action within individual crops, or successive crops.
- Monitor fields regularly and investigate the reasons for any poor control

Monitoring, Reporting, and Reaction to Changes in Performance

In the case that BASF obtains information about a loss of field performance after application of Alpha-cypermethrin, and that resistance to Alpa-cypermethrin is identified as the cause, BASF will inform IRAC and the registration authorities. Furthermore, BASF will advise the respective farmer to apply the appropriate resistance management strategies as developed and recommended by IRAC. Once resistant strains have been detected, management strategies must be customized to the particular situation under consideration of points of view published by IRAC.

CA 3.8 Methods and Precautions Concerning Handling, Storage, Transport or Fire

Report:	CA 3.8/1 Anonymous, 2016a Safety data sheet – Alpha-Cypermethrin tech-biocide use 2016/1236516
Guidelines:	EEC 1907/2006
GLP:	no

Personal protection: Control parameters

Components with workplace control parameters: No occupational exposure limits known.

Exposure controls

Personal protective equipment

Respiratory protection:
Suitable respiratory protection for lower concentrations or short-term effect: Particle filter with high efficiency for solid and liquid particles (e.g. EN 143 or 149, Type P3 or FFP3).

Hand protection:
Suitable chemical resistant safety gloves (EN 374) also with prolonged, direct contact (Recommended: Protective index 6, corresponding > 480 minutes of permeation time according to EN 374): E.g. nitrile rubber (0.4 mm), chloroprene rubber (0.5 mm), butyl rubber (0.7 mm) and other.

Eye protection:
Safety glasses with side-shields (frame goggles) (e.g. EN 166)

Body protection:
Body protection must be chosen depending on activity and possible exposure, e.g. apron, protecting boots, chemical-protection suit (according to EN 14605 in case of splashes or EN ISO 13982 in case of dust).

General safety and hygiene measures
Handle in accordance with good industrial hygiene and safety practice. Wearing of closed work clothing is recommended. Store work-clothing separately. Keep away from food, drink and animal feeding stuffs.

Precautions for safe handling

No special measures necessary if stored and handled correctly. Ensure thorough ventilation of stores and work areas. When using do not eat, drink or smoke. Hands and/or face should be washed before breaks and at the end of the shift. Remove contaminated clothing and protective equipment before entering eating areas.

Protection against fire and explosion:
Avoid dust formation. Dust can form an explosive mixture with air.
Prevent electrostatic charge - sources of ignition should be kept well clear - fire extinguishers should be kept handy.

Conditions for safe storage, including any incompatibilities

Segregate from foods and animal feeds.
Further information on storage conditions: Keep away from heat. Protect from direct sunlight. Protect against moisture.

Storage stability:
Storage duration: 48 Months

Transport

Land transport

ADR

UN number	UN3349
UN proper shipping name:	PYRETHROID PESTICIDE, SOLID, TOXIC (contains ALPHA-CYPERMETHRIN 93%)
Transport hazard class(es):	6.1, EHSM
Packing group:	III
Environmental hazards:	yes
Special precautions for user:	Tunnel code: E

RID

UN number	UN3349
UN proper shipping name:	PYRETHROID PESTICIDE, SOLID, TOXIC (contains ALPHA-CYPERMETHRIN 93%)
Transport hazard class(es):	6.1, EHSM
Packing group:	III
Environmental hazards:	yes
Special precautions for user:	None known

Inland waterway transport

ADN

UN number	UN3349
UN proper shipping name:	PYRETHROID PESTICIDE, SOLID, TOXIC (contains ALPHA-CYPERMETHRIN 93%)
Transport hazard class(es):	6.1, EHSM
Packing group:	III
Environmental hazards:	yes
Special precautions for user:	None known
Transport in inland waterway vessel:	Not evaluated

Sea transport

IMDG

UN number:	UN 3349
UN proper shipping name:	PYRETHROID PESTICIDE, SOLID, TOXIC (contains ALPHA-CYPERMETHRIN 93%)
Transport hazard class(es):	6.1, EHSM
Packing group:	III
Environmental hazards:	yes
Marine pollutant:	YES
Special precautions for user:	None known

Air transport

IATA/ICAO

UN number:	UN 3349
UN proper shipping name:	PYRETHROID PESTICIDE, SOLID, TOXIC (contains ALPHA-CYPERMETHRIN 93%)
Transport hazard class(es):	6.1
Packing group:	III
Environmental hazards:	No Mark as dangerous for the environment is needed
Special precautions for user:	None known

Fire-fighting measures

Extinguishing media

Suitable extinguishing media: water spray, foam, dry powder
Unsuitable extinguishing media for safety reasons: carbon dioxide, water jet

Special hazards arising from the substance or mixture
hydrogen chloride, carbon monoxide, Carbon dioxide, nitrogen oxides, organochloric compounds.
The substances/groups of substances mentioned can be released in case of fire.

Advice for fire-fighters
Special protective equipment:
Wear self-contained breathing apparatus and chemical-protective clothing.

Further information: Collect contaminated extinguishing water separately, do not allow to reach sewage or effluent systems. Dispose of fire debris and contaminated extinguishing water in accordance with official regulations. In case of fire and/or explosion do not breathe fumes. Keep containers cool by spraying with water if exposed to fire.

CA 3.9 Procedures for Destruction or Decontamination

Report:	CA 3.9/1 Anonymous, 2016a Safety data sheet – Alpha-Cypermethrin tech-biocide use 2016/1236516
Guidelines:	EEC 1907/2006
GLP:	no

Waste treatment methods

Must be sent to a suitable incineration plant, observing local regulations.

Contaminated packaging:
Contaminated packaging should be emptied as far as possible and disposed of in the same manner as the substance/product.

Methods and material for containment and cleaning up

For small amounts: Contain with dust binding material and dispose of.

For large amounts: Sweep/shovel up.

Avoid raising dust. Cleaning operations should be carried out only while wearing breathing apparatus. Collect waste in suitable containers, which can be labeled and sealed. Clean contaminated floors and objects thoroughly with water and detergents, observing environmental regulations. Dispose of absorbed material in accordance with regulations.

CA 3.10 Emergency Measures in Case of an Accident

Report:	CA 3.10/1 Anonymous, 2016a Safety data sheet – Alpha-Cypermethrin tech-biocide use 2016/1236516
Guidelines:	EEC 1907/2006
GLP:	no

First-aid measures

Description of first aid measures

First aid personnel should pay attention to their own safety. If the patient is likely to become unconscious, place and transport in stable sideways position (recovery position). Immediately remove contaminated clothing.

If inhaled:
Keep patient calm, remove to fresh air, seek medical attention.

On skin contact:
Immediately wash thoroughly with soap and water, seek medical attention.

On contact with eyes:
Wash affected eyes for at least 15 minutes under running water with eyelids held open.

On ingestion:
Immediately rinse mouth and then drink 200-300 ml of water, seek medical attention.

Most important symptoms and effects, both acute and delayed
Toxic if swallowed; harmful if inhaled (causes temporary irritation after single exposure); repeated oral exposure may affect certain organs (damages the peripheral nerve system).
Please refer to MSDS for further details.

Indication of any immediate medical attention and special treatment needed
Treatment: Treat according to symptoms (decontamination, vital functions), no known specific antidote.

Personal precautions, protective equipment and emergency procedures
Use personal protective clothing. Avoid contact with the skin, eyes and clothing. Avoid dust formation.

Environmental precautions
Do not discharge into the subsoil/soil. Do not discharge into drains/surface waters/groundwater.

Alpha-Cypermethrin

Document M-CA, Section 4

ANALYTICAL METHODS

Compiled by:

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
10/Jul/2017	Summaries of already peer-reviewed analytical methods used in support of analysis of plant, animal, e-fate matrices. Also updated are the summaries of analytical methods used in support of ecotoxicological studies. Soil AL-230-001 AL-242-001 AL-242-006 AL-210-012 2015/1000961 Water AL-243-001 AL-243-002 AL-243-003 AL-243-006 AL-210-012 Air AL-210-002 AL-241-001 AL-241-002 Tox AL-245-005 2016/1232572 AL-210-001 Plant 1983/7001664 1977/7000275 1979/7000414 AL-714-002 AL-123-088 AL-244-006 AL-244-012 AL-240-001 AL-244-010 AL-244-001 1989/5000067 AL-230-001 AL-244-011 AL-244-007 2016/1232571 AL-244-008 AL-244-009 2004/1010543	BASF DocID 2017/1134413

	2003/1001290 2006/1029533 Animal AL-245-001 AL-245-003 AL-245-006 AL-245-007 AL-245-008 AL-210-012 AL-440-018 Ecotox 2009/1114317 2009/1031203 2007/1016502 2009/1109081 2009/1109080 2009/1109079 2009/1108874 2009/1011443 2009/1085205 2009/1102214 2011/1124187 2013/1250848 2016/1232573 2012/1205915 2008/1010445 2008/1009839 2008/1051526 2009/1050236 2009/1050237 2014/1000203 2015/1000382 2007/1013272 2016/1232650 2014/1162697 2014/1246534 2016/1232658 2012/1000062 2013/1404157 AL-511-001 AL-512-002 AL-523-001 AL-523-002 AL-249-002 AL-243-004 2002/1004851 AL-123-029 AL-560-023 AL-243-007 AL-243-008 AL-534-003 2002/1004682 2002/1004683 2001/1014673	

	2001/1017462 2002/1004139 2002/1004140 2014/1125983 2002/1004857 2001/1015123 CA 4.1.2/103: Additional study AL-210-004 CA 4.2/10: Additional study: Air monitoring method with a lower LOQ of 0.06 µg/m^3 2015/1225669	

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 4 ANALYTICAL METHODS

CA 4.1 Methods used for the generation of pre-approval data

CA 4.1.1 Methods for the analysis of the active substance as manufactured

Report: CA 4.1.1/1
Anonymous, 1997a
Determination of Alpha-Cypermethrin in TGAI and WP-, EC-, SC- and UL-formulations
1997/1005661
Guidelines: none
GLP: no

Principle of the method

The active ingredient content in the test samples was determined by means of gas chromatography with a flame ionisation detector and internal calibration according to CIPAC method "ALPHA-CYPERMETHRIN 454". This method contains experimental details regarding sample preparation, chromatographic conditions, evaluation, and calculation.

Applicability of CIPAC Methods

This method corresponds to CIPAC method 454 [Alpha-cypermethrin].

Identity

The identity of the analyte is confirmed by comparison of the retention time with that of authentic reference material and by infrared (IR) spectrometry.

Description of the methods for validation

The validity of the method was confirmed by an international collaborative CIPAC trial. The inter-lab repeatability and inter-lab reproducibility was determined according to ISO 5725.

Data Origin: CIPAC Method 454
Repeatability r: 17 g/kg at 954 g/kg active ingredient content
Reproducibility R: 20 g/kg at 954 g/kg active ingredient content.

The applicability has further been shown in the Analysis Reports of Five Representative Batches of alpha-cypermethrin (see chapter 1.11, confidential part, Document JCA).

For further information on significant impurities, please refer to confidential Document JCA.

CA 4.1.2 Methods for risk assessment

A concordance list of structures and designations of reference compounds mentioned in CA 4.1.2 and 4.2 as well as the respective sections of Doc N is given below.

(a) Methods in soil, water, sediment, air and any additional matrices used in support of environmental fate studies

The following methods cover the compounds that have to be considered for the discussion on environmental risk assessment as summarized in Document N, chapter 8.5 and discussed in CA 7.4:

Soil:	BAS 311 I (cypermethrin, Reg. No. 127266; Cis I, Cis II [alpha-cypermethrin], Trans III, and Trans IV), 3-phenoxybenzoic acid (3-PBA, Reg. No. 130213), 2,2-dimethyl-3-(2,2'-dichlorovinyl)cyclo-propane carboxylic acid (DCVA, cis- and trans-isomers, Reg. No. 180011)
Sediment:	As no aquatic field studies were conducted, no separate stand-alone method validation for the determination of unlabelled alpha-cypermethrin or any of its metabolites was required. Due to similarity to soil matrix, determination of such, if required, can be accomplished by applying the fully validated soil method.
Water:	BAS 311 I (cypermethrin, Reg. No. 127266; Cis I, Cis II [alpha-cypermethrin], Trans III, and Trans IV), 3-phenoxybenzoic acid (3-PBA, Reg. No. 130213), 2,2-dimethyl-3-(2,2'-dichlorovinyl)cyclo-propane carboxylic acid (DCVA, cis- and trans-isomers, Reg. No. 180011), 3-phenoxybenzaldehyde (Reg. No. 4080665)
Air:	BAS 310 I (alpha-cypermethrin, Reg. No. 4078193)

A summary of the methods can be found in the respective section of Doc N, chapter 5.1.3.

Soil

Analytical methods for the determination of alpha-cypermethrin residues in soil were evaluated in the context of the inclusion in Annex I of Directive 91/414/EEC. Methods evaluated previously are listed in Table 4.1.2-1 for the reviewer´s convenience and summarized shortly below.

Table 4.1.2-1: Summary of peer-reviewed analytical methods for determination of alpha-cypermethrin residues in soil

Method No.	Matrix	Method principle	Target analytes	LOQ mg/kg	year	DocID	EU reviewed
SAMS 349-1	Soil	HPLC/UV or GC/ECD after derivatization; TLC/GC or GC/MS for confirmation	DVA	0.05	Issued 1982	AL-230-001 (1983)	Yes
SAMS 348-1	Soil	HPLC/UV; HPLC/UV or GC/MS after derivatization	3-PBA	0.05	Issued 1982	AL-230-001 (1983)	Yes
SAMS 354-2	Soil	GC/ECD; GC/MS for confirmation	Alpha-cypermethrin	0.01	1990	AL-242-001	Yes
SAMS 354-2	Soil	GC/ECD; GC/MS for confirmation	Alpha-cypermethrin	0.05	1999	AL-242-006	Yes
M 3499	soil	sample prep. acc. to SAMS 354-2; GC/MS (confirmatory method)	Alpha-cypermethrin	0.05	2001	AL-210-012	Yes

DVA: 2,2-dimethyl-3-(2,2-dichlorovinyl)cyclopropane carboxylic acid (WL 44776)
3-PBA: 3-phenoxybenzoic acid (WL 44607)

For completeness of information, summaries of already EU-peer reviewed analytical methods are presented for completeness of information.

Report:	CA 4.1.2/1 Hitchings E. et al., 1983 The development of methods for the determination of Ripcord - WL 44776 and WL 44607 in crops and soil AL-230-001
Guidelines:	none
GLP:	No

Principle of the methods SAMS 349-1 and SAMS 349-2:
The methods for both acid metabolites (WL 44776 = 2,2-dimethyl-3-(2,2-dichlorovinyl)cyclopropane carboxylic acid; WL 44607 = 3-phenoxybenzoic acid) are essentially the same.

Soil samples are extracted by tumbling end-over-end with a mixture of acetic acid/water/acetonitrile (1:30:70 v/v), after which the extract is filtered. An aliquot of the filtrate is diluted with water and partitioned under acidic conditions with a diethyl ether/petroleum spirit mixture (1:1 v/v). The organic fraction is concentrated and subsequently cleaned up by normal phase HPLC (LiChrosorb Diol (10 µm); isocratic elution) with UV detection at 225 nm, after which WL 44776 (resp. WL 44607) is determined by reversed phase HPLC (Spherisorb S5 ODS (5 µm); isocratic elution) with UV detection at 216 nm; quantification by external standardization.

If the extracts are not sufficiently clean for satisfactory analysis to be carried out, the acid WL 44776 (as obtained after normal phase HPLC) may be converted into the parent WL 43467 (cypermethrin) by reaction with α-cyano-3-phenoxybenzyl bromide, after which the derivative is cleaned up using a Porasil column. Analysis is then carried out using GC (GE-XE 60 1.2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD).
The acid WL 44607 (as obtained after normal phase HPLC) may be converted into its methyl ester, after which the derivative is cleaned up by partition with petroleum spirit. Analysis is then carried out by either reversed phase HPLC (ODS-Hypersil (5 µm); isocratic elution) with UV detection at 212 nm or GC-MS (SE-30 2% (m/m) on Gas Chrom Q (100-120 mesh)) with multiple ion monitoring (MS operated in EI mode, ions monitored: m/e 196 and 228). The latter procedure may also be adopted to confirm the identity of any residues found.
Residues of WL 44776 determined by HPLC may be confirmed by preparing the parent compound WL 43467 as described above and using either TLC-GC (GE-XE 60 1.2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD) or GC-MS (SE-30 1% (m/m) on Gas Chrom Q (100-120 mesh)) as the determination step, MS operated in EI mode (ions monitored: m/e 207 and 209).

Specificity/Interferences: Typical HPLC chromatograms are shown and from these chromatograms it is clear that control samples show no major interferences at the retention time of WL 44776 or WL 44607; blank values are stated to be < 0.05 mg/kg.
Chromatograms of recovery extracts fortified with WL 44776 and WL 44607 at the same time were not shown, but based on the retention times that were stated, both metabolites should be well resolved under the test conditions used.

Recovery/Precision: see Table **4.1.2-2**

Limit of quantitation*:* The limit of quantitation (LOQ) of the method for both metabolites, defined by the lowest fortification level is 0.10 mg/kg.

Table 4.1.2-2: Validation of methods SAMS 349-01 and SAMS 348-01 (1983/7001072)

		Fortification level (mg/kg commodity)	Number of samples	Range (%)	Mean (%)	RSD (%)
Loam, sandy clay loam	WL 44776 free	0.10	2	75 - 95	85	(16.6)
	WL 44607 free	0.10	2	75 - 80	77.5	(4.6)

Conclusion **The method was developed and validated before SANCO/3029/99 rev. came into force. A newly developed and fully validated method (R0034/01) allows the quantification of the analytes down to 0.001 mg/kg.**

Report:	CA 4.1.2/2 Anonymous, 1990 Determination of residues of Alphacypermethrin in soils - Gas chromatographic method AL-242-001
Guidelines:	none
GLP:	No

Report:	CA 4.1.2/3 Anonymous, 1990 Determination of residues of Alphacypermethrin in soils - Gas chromatographic method AL-242-006
Guidelines:	EEC 91/414,EEC 96/46,BBA Guideline Residue Analytical Methods for Post-Registration Control Purposes of July 21 1998,Guidance Document of Residue Analytical Methods 8064/VI/97 rev. 4 15.12.1998
GLP:	Yes (certified by Umweltministerium Baden-Wuerttemberg, Stuttgart)

Principle of the method:

SAMS 354-02: Soil samples are mixed with anhydrous sodium sulphate and extracted by tumbling end-over-end with acetone/hexane (1:1 v/v). Extracts are washed with water to remove the acetone and cleaned up by liquid-solid chromatography using Florisil.
Alpha-cypermethrin is determined by packed column GC (GE-XE 60 2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD); quantification by external standardization. Confirmation of the residues is carried out by capillary GC-ECD (SE-54; 25m × 0.32 mm, 0.5 µm) and GC-MS (SE-30 1% (m/m) on Gas Chrom Q (100-120 mesh)), MS operated in negative ion chemical ionisation mode (ions monitored: m/e 207 and 209).
It should be noted that a slightly adapted method SAMS 354-02 (incl. i.e. capillary column: DB5, 30m × 0.25 mm, 0.25 µm, temperature program, GC-MS conditions: Optima 5, 25m × 0.25 mm, 0.25 µm) has been validated in the following study by H. Werle (1999) – *Alpha-cypermethrin (CL 900049): Validation of Method SAMS 354-2 for the Determination of Residues in Soils* (BASF DocID 1999/7001540), but the results were not reported in the initial monograph.

Specificity/Interferences *:*	typical packed column GC-ECD chromatograms are shown. Control samples exhibit no significant interfering peaks at the retention time of alpha-cypermethrin; control and blank data were stated to be < 0.01 mg/kg.
Linearity:	Linearity data were not provided for the validation using the packed GC-ECD method but linearity data were presented for the capillary GC-ECD method. Method was found to be linear in the 0.0126 – 0.25 µg/mL. Linearity was checked before sample analysis (n=4), concurrently to analysis (n=3) and at the end of the analysis (n=4). Correlation coefficient R^2 was in each case ≥ 0.997. The regression equations were provided but not the linear curves

Recovery/Precision : see Table **4.1.2-3**

Limit of quantitation: The limit of quantitation (LOQ) of the method for both metabolites, defined by the lowest fortification level is 0.05 mg/kg.

Table 4.1.2-3: Validation of method SAMS 354-02 (AL-242-001, packed GC-ECD and AL-242-006, capillary GC-ECD)

Matrix	Analyte	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
clay loam soil SAMS 354-2 (packed column)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	100 - 100 90 - 100 105 - 105	100 95 105	(0.0) (7.4) (0.0)
		0.05 - 0.50	6	90 - 105	100	5.5
sandy loam soil SAMS 354-2 (packed column)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	100 - 100 95 - 115 90 - 105	100 105 97.5	(0.0) (13.5) (10.9)
		0.05 - 0.50	6	90 - 115	100.8	8.5
silty clay soil SAMS 264-2 (packed column)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	95 - 115 85 - 90 90 - 115	105 87.5 102.5	(13.5) (4.0) (17.3)
		0.05 - 0.50	6	85 - 115	98.3	13.5
Overall SAMS 354-2 (packed column)	alpha-cypermethrin	0.05 0.20 0.50	6 6 6	95 - 115 85 - 115 90 - 115	101.7 95.8 101.7	6.7 11.2 9.7
		0.05 - 0.50	18	85 - 115	99.7	9.2
Soil 2.3 (LUFA) SAMS 354-2 (capillary column)	alpha-cypermethrin	0.05 0.5	4* 5	86-123* 83-91	90 87	6.1 3.0

* 123 was identified as an outlier and not taken into account to calculate the mean recovery.

Conclusion **Both methods (packed column and capillary column) are suitable for alpha-cypermethrin residue analysis in soil with a LOQ of 0.05 mg/kg. The method was developed and validated before SANCO/3029/99 rev. came into force. A newly developed and fully validated method (R0034/01) allows the quantification of the analytes down to 0.001 mg/kg.**

Report:	CA 4.1.2/4 Xu B., 2001 BAS 310 I (Alpha-Cypermethrin): Validation of method M 3499 for the confirmation of BAS 310 I residues in water soil and blood by GC/MS
	AL-210-012
Guidelines:	Guidance Document on Residue Analytical Methods (SANCO/825/00rev.6)
GLP:	Yes (certified by United States Environmental Protection Agency)

Principle of the method:
Method M3499 is used to confirm the specificity of Method SAMS 354-2 (GC-ECD), which was proposed for enforcement in soil. The procedure for the sample preparation is the same as the one outlined in SAMS 354-2, but an alternative detection method is used: determination by capillary GC (DB-5MS; 5 m x 0.25 mm i.d; 0.25 µm) with MSD in SIM mode (m/z 207, 209 and 211).

Specificity/Interferences:	specificity of method SAMS 354-2 is demonstrated by using GC-MS as an alternative method of detection (confirmation based on retention times and ion ratios: see Table **4.1.2-4**). - untreated control samples exhibit no significant interferences at retention time of alphacypermethrin (control values < 30% of LOQ).
Linearity:	the response of the GC-MS system (= peak area) to alphacypermethrin was demonstrated to be linear over a concentration range from 0.0025 to 0.04 µg/mL (n = 5); r > 0.99 for each ion. Calibration seems to have been performed in solvent without assessment of the matrix effects.

Recovery/Precision: no data provided; only qualitative validation was performed.

Limit of quantitation:	The limit of quantitation (LOQ) of the method for both metabolites, defined by the lowest fortification level is 0.05 mg/kg.

Table 4.1.2-4: Validation of confirmatory method M 3499 for soil

Analyte	Fortification level (mg/kg commodity)	Number of samples	% Retention Time Difference (RTD) (ion m/z 207)	% Ion Ratio Difference (IRD)	
				207/209	207/211
alphacypermethrin	0.05	3	0.0 0.1 0.0	- 0.6 - 1.0 0.0	- 4.6 - 1.4 - 3.0

% RTD = (sample retention time – average standard retention time) x 100/average standard retention time
% IRD = (sample ion ratio – standard average ion ratio) x 100/standard average ion ratio

Conclusions	**Method M 3499 (GC-MS) is suitable as qualitative confirmatory method to SAMS 354-2 (GC-ECD) for determination of alphacypermethrin residues in soil (LOQ 0.05 mg/kg). A newly developed and fully validated method (R0034/01) allows the quantification of the analytes down to 0.001 mg/kg.**

Report:	CA 4.1.2/5 Carter M.L., Perez S., 2014a Validation of BASF analytical method R0034/01: Method for the quantitation of the diastereomeric forms of BAS 311 I (Reg. 127266) and its metabolites 3-Phenoxybenzoic acid (Reg. No. 130213) and DCVA (cis and trans isomers, Reg. No. 180011) in soil by LC-MS/MS 2014/7002375
Guidelines:	EPA 850.7100, EPA 835.6100, SANCO/3029/99 rev. 4 (11/07/00), SANCO/825/00 rev 8.0 (20/06/2000)
GLP:	yes (certified by United States Environmental Protection Agency)

Principle of the method

The BASF analytical method R0034/01 allows the determination of the diastereomeric forms of BAS 311 I (cypermethrin, Reg. No. 127266; Cis I, Cis II [alpha-cypermethrin], Trans III, and Trans IV), and its three metabolites 3-phenoxybenzoic acid (PBA, Reg. No. 130213) and DCVA (cis- and trans-isomers, Reg. No. 180011) in soil.

Soil samples (5 g) are extracted for cypermethrin isomers (Cis I, Cis II, Trans III, and Trans IV) by shaking with 50 mL of 0.1% formic acid in acetonitrile. An aliquot (20%) from the extract is evaporated to dryness and then re-dissolved with acetonitrile/water with 0.1% formic acid (50:50, v/v). The residues are determined using LC-MS/MS.

Soil samples (5 g) are extracted for the metabolites of cypermethrin (c/s-DCVA, trans-DCVA, and 3-PBA) by shaking twice with 25 mL acetonitrile/water (70:30, v/v). An aliquot (10%) from the combined extract is evaporated to dryness and then dissolved in methanol/water (20:80, v/v) with 0.1% formic acid. The residues are determined using LC-MS/MS.

LC-MS/MS conditions were specifically adapted for the determination of the individual analytes:

Primary and confirmatory mass transitions (*m/z*) for the diastereomeric forms of BAS 311 I (Cis I, Cis II, Trans III, and Trans IV) were analyzed using UPLC and HPLC Methods B and C, respectively.

Primary and confirmatory mass transitions for 3-PBA were analyzed using UPLC and HPLC Methods G and E, respectively. The ions monitored for DCVA (Cis and Trans) were analyzed using UPLC and HPLC Methods G and E, respectively. For DCVA (Cis and Trans), Method E was used for primary quantitation in HPLC mode and confirmatory quantitation for Method G. Additionally, Method F was used for confirmatory quantitation both for HPLC mode and for Method E.

The limit of quantification (LOQ) is 0.001 mg/kg for each analyte, which corresponds to a concentration in the extract of 0.5 ng/mL. The limit of detection in soil is 0.0002 mg/kg (corresponds to 0.1 ng/mL).

Remark: Comparison of extraction recoveries of cypermethrin applying the extraction scheme of the aerobic soil metabolism study and the reported data generation method (M-CA 4.1.2/1) was additionally assessed using ^{14}C-labelled soil samples in the respective soil metabolism study. Details are reported in chapter 7.1.2.1.1 in the respective study: Metabolism of BAS 310 I in soil under aerobic conditions (DocID 2014/1159491).

Recovery findings

The analytical method was developed for the determination of the diastereomeric forms of BAS 311 I (Cis I, Cis II [alpha-cypermethrin], Trans III, and Trans IV), and its three metabolites 3-phenoxybenzoic acid (PBA) and DCVA (cis- and trans-isomers) in soil. The method was validated at two fortification levels (0.001 and 0.01 mg/kg (ppm)) for sandy and clay loam soil samples. For each fortification level and matrix, five replicates were analyzed. Additionally, a method blank and at least two replicates of unfortified samples were examined.

Solvent-based and matrix-matched standards were also analyzed within the study to check for possible matrix effects. Clay and sandy loam soil samples were tested for matrix effects for the diastereomers of BAS 311 I. Clay soil samples were tested for matrix effects for the metabolites of BAS 311 I. No significant suppression (greater than 20%) occurred for any of the analytes when analyzed by LC-MS/MS. If significant suppression occurs, matrix-matched standards may be utilized.

It was proven that the method R0034/01 is suitable to determine residues of the diastereomeric forms of BAS 311 I (cypermethrin, Reg. No. 127266), and its metabolites, 3-phenoxybenzoic acid (Reg. No. 130213), and DCVA (cis- and trans-isomers, Reg. No. 180011) in soil. As shown in the following tables, the mean recovery values were found to be within the acceptable range of 70-120% for all methods tested. The overall relative standard deviations (RSD, %) for all fortification levels were below 20%.

Detailed results of recoveries for each mass transition and soil are given in Table **4.1.2-5** to Table **4.1.2-11**.

Table 4.1.2-5: Recoveries for Cis I, Cis II, Trans III, and Trans IV (Reg. No. 127266) in Sandy Loam Soil Using Method B (UPLC Mode): Primary and Confirmatory Quantitation[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Sandy Loam	Cis I: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	70, 74, 68, 70, 73	71	2.4	3.4
	0.01	5	75, 76, 69, 71, 70	72	2.9	4.0
	Overall	**10**	**Range: 68-76**	**72**	**2.7**	**3.8**
	Cis I: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	70, 79, 70, 73, 77	74	4.1	5.5
	0.01	5	77, 79, 71, 76,72	75	3.5	4.6
	Overall	**10**	**Range: 70-79**	**74**	**3.7**	**4.9**
	Cis II: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	67, 79, 71, 71, 77	73	4.7	6.5
	0.01	5	75, 77, 69, 78, 72	74	3.9	5.3
	Overall	**10**	**Range: 67-79**	**74**	**4.2**	**5.7**
	Cis II: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	68, 77, 70, 70, 75	72	3.7	5.1
	0.01	5	75, 75, 69, 76, 71	73	3.1	4.3
	Overall	**10**	**Range: 68-77**	**73**	**3.3**	**4.5**
	Trans III: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	85, 89, 72, 77, 90	83	7.7	9.3
	0.01	5	85, 77, 76, 78, 76	78	3.7	4.7
	Overall	**10**	**Range: 72-90**	**81**	**6.1**	**7.6**
	Trans III: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	85, 94, 71, 71, 88	82	10.3	12.6
	0.01	5	91, 81, 72, 84, 76	81	7.4	9.1
	Overall	**10**	**Range: 71-94**	**81**	**8.5**	**10.4**
	Trans IV: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0					
	0.001	5	77, 86, 70, 72, 78	77	6.0	7.9
	0.01	5	85, 77, 65, 74, 68	74	7.7	10.4
	Overall	**10**	**Range: 65-86**	**75**	**6.7**	**8.9**
	Trans IV: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	79, 83, 68, 72, 79	76	6.1	7.9
	0.01	5	86, 77, 64, 74, 69	74	8.4	11.4
	Overall	**10**	**Range: 64-86**	**75**	**7.0**	**9.4**

[a] All calculations are based on unrounded values.

[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Table 4.1.2-6: Recoveries for Cis I, Cis II, Trans III, and Trans IV (Reg. No. 127266) in Clay Loam Soil Using Method B (UPLC Mode): Primary and Confirmatory Quantitation[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Clay	Cis I: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	81, 82, 82, 80, 81	81	0.6	0.7
	0.01	5	85, 91, 88, 88, 89	88	2.0	2.3
	Overall	**10**	**Range: 80-91**	**85**	**4.0**	**4.7**
	Cis I: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	86, 87, 85, 86, 87	86	0.9	1.0
	0.01	5	87, 92, 88, 88, 89	89	2.0	2.2
	Overall	**10**	**Range: 85-92**	**87**	**2.1**	**2.4**
	Cis II: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	85, 86, 85, 85, 87	86	0.9	1.1
	0.01	5	90, 95, 90, 91, 92	91	2.2	2.4
	Overall	**10**	**Range: 85-95**	**89**	**3.4**	**3.8**
	Cis II: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	88, 88, 86, 87, 90	88	1.6	1.8
	0.01	5	88, 95, 89, 91, 89	90	2.6	2.9
	Overall	**10**	**Range: 86-95**	**89**	**2.4**	**2.7**
	Trans III: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	101, 95, 96, 102, 92	97	4.0	4.1
	0.01	5	86, 94, 91, 89, 90	90	3.1	3.4
	Overall	**10**	**Range: 86-102**	**94**	**5.1**	**5.4**
	Trans III: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	96, 95, 94, 100, 90	95	3.4	3.6
	0.01	5	90, 103, 89, 89, 93	93	6.1	6.5
	Overall	**10**	**Range: 89-103**	**94**	**4.8**	**5.1**
	Trans IV: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	86, 90, 93, 94, 92	91	3.2	3.5
	0.01	5	81, 95, 85, 88, 91	88	5.7	6.4
	Overall	**10**	**Range: 81-95**	**90**	**4.6**	**5.2**
	Trans IV: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	86, 96, 93, 93, 89	91	3.8	4.2
	0.01	5	82, 94, 85, 84, 94	88	5.7	6.5
	Overall	**10**	**Range: 82-96**	**89**	**4.9**	**5.5**

[a] All calculations are based on unrounded values.
[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Table 4.1.2-7: Recoveries for Cis I, Cis II, Trans III, and Trans IV (Reg. No. 127266) in Sandy Loam Soil Using Method C (HPLC Mode): Primary and Confirmatory Quantitation[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Sandy Loam	Cis I: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	85, 86, 93, 105, 96	93	8.0	8.6
	0.01	5	94, 93, 86, 90, 91	91	3.0	3.4
	Overall	**10**	**Range: 85-105**	**92**	**5.8**	**6.4**
	Cis I: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	88, 86, 94, 102, 101	94	7.5	7.9
	0.01	5	100, 93, 92, 85, 93	93	5.4	5.8
	Overall	**10**	**Range: 85-102**	**94**	**6.2**	**6.6**
	Cis II: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	96, 98, 100, 107, 97	100	4.2	4.2
	0.01	5	100, 101, 94, 100, 96	98	3.0	3.1
	Overall	**10**	**Range: 94-107**	**99**	**3.5**	**3.6**
	Cis II: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	86, 98, 100, 111, 96	98	9.0	9.1
	0.01	5	97, 101, 93, 101, 92	97	4.6	4.7
	Overall	**10**	**Range: 86-111**	**97**	**6.7**	**6.9**
	Trans III: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	84, 96, 97, 112, 108	100	10.8	10.9
	0.01	5	105, 97, 98, 118, 99	103	9.0	8.7
	Overall	**10**	**Range: 84-118**	**101**	**9.6**	**9.5**
	Trans III: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	101, 100, 108, 103, 83	99	9.5	9.7
	0.01	5	103, 97, 93, 119, 104	103	9.9	9.6
	Overall	**10**	**Range: 83-119**	**101**	**9.4**	**9.3**
	Trans IV : Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	93, 103, 100, 114, 95	101	8.1	8.1
	0.01	5	108, 107, 98, 119, 94	105	9.6	9.2
	Overall	**10**	**Range: 93-119**	**103**	**8.7**	**8.4**
	Trans IV: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	105, 113, 107, 106, 107	108	3.1	2.9
	0.01	5	108, 102, 93, 116, 97	103	9.0	8.7
	Overall	**10**	**Range: 93-116**	**106**	**6.8**	**6.4**

[a] All calculations are based on unrounded values.

[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Table 4.1.2-8: Recoveries for Cis I, Cis II, Trans III, and Trans IV (Reg. No. 127266) in Clay Loam Soil Using Method C (HPLC Mode): Primary and Confirmatory Quantitation[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Clay Loam	Cis I: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	84, 83, 74, 85, 91	83	6.0	7.2
	0.01	5	103, 87, 77, 83, 109	92	13.6	14.9
	Overall	**10**	**Range: 74-109**	**87**	**10.8**	**12.4**
	Cis I: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	103, 120, 94, 99, 106	104	9.9	9.5
	0.01	5	126, 102, 91, 102, 131	110	17.2	15.6
	Overall	**10**	**Range: 91-131**	**107**	**13.6**	**12.7**
	Cis II: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	98, 98, 84, 92, 95	94	5.8	6.2
	0.01	5	112, 91, 86, 82, 104	95	12.8	13.4
	Overall	**10**	**Range: 82-112**	**94**	**9.4**	**10.0**
	Cis II: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	98, 90, 81, 103, 95	93	8.3	8.9
	0.01	5	108, 91, 83, 87, 106	95	11.3	11.9
	Overall	**10**	**Range: 81-108**	**94**	**9.4**	**10.0**
	Trans III: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	91, 92, 89, 89, 93	91	1.7	1.9
	0.01	5	106, 88, 90, 95, 108	97	9.2	9.5
	Overall	**10**	**Range: 88-108**	**94**	**7.2**	**7.6**
	Trans III: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	83, 97, 73, 79, 91	84	9.5	11.3
	0.01	5	106, 88, 95, 90, 104	96	7.9	8.2
	Overall	**10**	**Range: 73-106**	**90**	**10.4**	**11.5**
	Trans IV: Primary Quantitation (*m/z* 433.0 → *m/z* 191.0)					
	0.001	5	97, 105, 94, 89, 86	94	7.3	7.8
	0.01	5	110, 89, 93, 93, 100	97	8.3	8.5
	Overall	**10**	**Range: 86-110**	**96**	**7.5**	**7.9**
	Trans IV: Confirmatory Quantitation (*m/z* 435.0 → *m/z* 193.0)					
	0.001	5	93, 100, 88, 102, 99	97	5.7	5.9
	0.01	5	109, 90, 97, 89, 106	98	9.3	9.5
	Overall	**10**	**Range: 88-109**	**97**	**7.3**	**7.5**

[a] All calculations are based on unrounded values.
[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Table 4.1.2-9: Recoveries for DCVA (Reg No. 180011, Cis and Trans) and 3-PBA (Reg No. 130213) in Sandy Loam and Clay Loam Soil Using Method G (UPLC Mode): Primary and Confirmatory Quantitation of 3-PBA and Primary Quantitation of DCVA (Cis and Trans)[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Sandy Loam	*cis*-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	101, 98, 92, 90, 88	94	5.8	6.2
	0.01	5	92, 94, 100, 99, 93	96	3.7	3.9
	Overall	**10**	**Range: 88-101**	**95**	**4.7**	**4.9**
	trans-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	106, 112, 100, 105, 103	105	4.5	4.3
	0.01	5	92, 97, 104, 105, 97	99	5.3	5.4
	Overall	**10**	**Range: 92-112**	**102**	**5.6**	**5.5**
	3-PBA: Primary Quantitation (*m/z* 213.0 → *m/z* 93.0)					
	0.001	5	105, 102, 97, 96, 93	99	5.1	5.2
	0.01	5	88, 94, 98, 103, 98	96	5.7	6.0
	Overall	**10**	**Range: 88-105**	**97**	**5.3**	**5.4**
	3-PBA: Confirmatory Quantitation (*m/z* 213.0 → *m/z* 169.0)					
	0.001	5	103, 96, 96, 95, 91	96	4.2	4.4
	0.01	5	89, 97, 100, 104, 98	98	5.3	5.4
	Overall	**10**	**Range: 89-104**	**97**	**4.6**	**4.8**
Clay Loam	*cis*-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	88, 98, 92, 96, 104	96	6.1	6.4
	0.01	5	95, 103, 101, 94, 106	100	4.9	4.9
	Overall	**10**	**Range: 88-106**	**98**	**5.7**	**5.8**
	trans-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	108, 115, 96, 91, 98	102	9.5	9.3
	0.01	5	96, 104, 107, 108, 107	104	4.8	4.6
	Overall	**10**	**Range: 91-115**	**103**	**7.2**	**7.0**
	3-PBA: Primary Quantitation (*m/z* 213.0 → *m/z* 93.0)					
	0.001	5	100, 106, 101, 108, 107	105	3.4	3.3
	0.01	5	98, 107, 107, 104, 110	105	4.6	4.3
	Overall	**10**	**Range: 98-110**	**105**	**3.8**	**3.6**
	3-PBA: Confirmatory Quantitation (*m/z* 213.0 → *m/z* 169.0)					
	0.001	5	92, 110, 96, 99, 111	102	8.1	8.0
	0.01	5	103, 107, 107, 108, 110	107	2.7	2.5
	Overall	**10**	**Range: 92-111**	**104**	**6.4**	**6.1**

[a] All calculations are based on unrounded values.
[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Table 4.1.2-10: Recoveries for DCVA (Reg No. 180011, Cis and Trans) and 3-PBA (Reg No. 130213) in Sandy Loam and Clay Loam Soil Using Method E: Primary and Confirmatory Quantitation of 3-PBA in HPLC mode; Primary Quantitation in HPLC mode of DCVA (Cis and Trans) and Confirmatory Quantitation for Method G[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Sandy Loam	*cis*-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	106, 103, 81, 85, 83	92	11.7	12.8
	0.01	5	74, 92, 83, 85, 77	82	7.2	8.7
	Overall	**10**	**Range: 74-106**	**87**	**10.6**	**12.0**
	trans-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	105, 101, 113, 97, 98	103	6.5	6.4
	0.01	5	77, 97, 94, 93, 89	90	7.9	8.8
	Overall	**10**	**Range: 77-113**	**96**	**9.5**	**9.9**
	3-PBA: Primary Quantitation (*m/z* 213.0 → *m/z* 93.0)					
	0.001	5	97, 88, 110, 101, 88	97	9.4	9.7
	0.01	5	85, 102, 107, 97, 99	98	8.2	8.4
	Overall	**10**	**Range: 85-110**	**97**	**8.3**	**8.6**
	3-PBA: Confirmatory Quantitation (*m/z* 213.0 → *m/z* 169.0)					
	0.001	5	96, 90, 109, 97, 99	98	6.7	6.8
	0.01	5	83, 101, 110, 101, 94	98	9.8	10.0
	Overall	**10**	**Range: 83-110**	**98**	**7.9**	**8.1**
Clay Loam	*cis*-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	92, 97, 90, 88, 100	94	4.9	5.2
	0.01	5	89, 100, 93, 101, 99	**96**	5.2	5.4
	Overall	**10**	**Range: 88-101**	**95**	**4.9**	**5.2**
	trans-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	83, 99, 83, 82, 101	90	9.5	10.6
	0.01	5	94, 104, 89, 103, 99	98	6.4	6.5
	Overall	**10**	**Range: 82-104**	**94**	**8.8**	**9.4**
	3-PBA: Primary Quantitation (*m/z* 213.0 → *m/z* 93.0)					
	0.001	5	89, 114, 89, 93, 115	100	13.4	13.4
	0.01	5	96, 110, 94, 104, 102	101	6.4	6.3
	Overall	**10**	**Range: 89-115**	**100**	**9.9**	**9.9**
	3-PBA: Confirmatory Quantitation (*m/z* 213.0 → *m/z* 169.0)					
	0.001	5	92, 109, 83, 88, 104	95	10.7	11.2
	0.01	5	100, 104, 95, 105, 103	102	4.3	4.2
	Overall	**10**	**Range: 83-109**	**98**	**8.4**	**8.5**

[a] All calculations are based on unrounded values.
[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Table 4.1.2-11: Recoveries for DCVA (Reg No. 180011, Cis and Trans) in Sandy Loam and Clay Loam Soil Using Method F: DCVA (Cis and Trans) Confirmatory Quantitation both for HPLC Mode and for Method E[a]

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	Standard Deviation	%RSD [b]
Sandy Loam	cis-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	118, 111, 98, 76, 102	101	16.2	16.0
	0.01	5	87, 97, 90, 96, 100	**94**	5.1	5.4
	Overall	**10**	**Range: 76-118**	**98**	**11.9**	**12.2**
	trans-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	105, 95, 94, 93, 97	97	4.7	4.9
	0.01	5	95, 98, 93, 106, 99	**98**	4.9	4.9
	Overall	**10**	**Range: 93-106**	**97**	**4.6**	**4.7**
Clay Loam	*cis*-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	85, 90, 91, 75, 93	**87**	7.2	8.3
	0.01	5	93, 96, 94, 101, 103	97	4.4	4.5
	Overall	**10**	**Range: 75-103**	**92**	**7.9**	**8.6**
	trans-DCVA: Primary Quantitation (*m/z* 207.0 → *m/z* 207.0)					
	0.001	5	115, 110, 87, 90, 97	100	12.3	12.3
	0.01	5	93, 98, 99, 102, 94	97	3.8	3.9
	Overall	**10**	**Range: 87-115**	**99**	**8.7**	**8.8**

[a] All calculations are based on unrounded values.
[b] Relative Standard Deviation = (Standard Deviation/Average Recovery) x 100

Linearity

Good linearity (r > 0.99) was observed in the range of 0.05 to 10 ng/mL (nominal) for the two mass transitions of Cis I, Cis II, Trans III, and Trans IV in mixed standard solutions. Good linearity was observed in the range of 0.1 to 10 ng/mL (nominal) for the two mass transitions of 3-PBA and the one mass transition of cis- and trans-DCVA in mixed standard solutions.

Specificity

The method allows the specific determination of the diastereomeric forms of BAS 311 I (Reg. No. 127266, Cis I, Cis II [alpha-cypermethrin], Trans III, and Trans IV), and its three metabolites 3-phenoxybenzoic acid (Reg. No. 130213, 3-PBA) and DCVA (Reg. No. 180011, cis- and trans-isomers) in soil. Interferences may be observed during analysis of the metabolites of BAS 311 I depending on soil type. This can be alleviated by testing the LC-MS/MS gradient before GLP analysis. Retention time shifting was observed for the diastereomeric forms of BAS 311 I and is instrument dependent. However, this does not affect quantitation as long as the retention pattern is observed and compared with the reference standards. The instrument must be sufficiently conditioned with injections of sample matrix prior to analysis.

Limit of Quantitation The limit of quantitation (LOQ) was defined by the lowest fortification level successfully tested. LOQ is 0.001 mg/kg for each analyte, which corresponds to a concentration in the extract of 0.5 ng/mL. The limit of detection in soil is 0.0002 mg/kg (corresponds to 0.1 ng/mL).

Repeatability The overall relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-5** to Table **4.1.2-11**.

Reproducibility Reproducibility of the method was not determined within this validation study.

Extractability The extractability of the diastereomers of BAS 311 I in soil was tested using the residue and metabolism extraction procedures. An extractability set consisted of one control, two fortifications at the LOQ (0.001 mg/kg for each diastereomer), one fortification at 10 x LOQ (0.01 mg/kg) and one sample with incurred residues from the terrestrial field dissipation study number 380198. Extractability data using the residue and metabolism extraction procedure are presented in Table **4.1.2-12** and Table **4.1.2-13**, respectively. Results show that the diastereomers of BAS 311 I in soil are comparable from both residue and metabolism extraction procedures.

Table 4.1.2-12: Extractability Data using the Residue Extraction Procedure

Matrix	Final Reported ppm			
	Cis I	Cis II	Trans III	Trans IV
Soil	0.0015	0.0052	ND	ND

Table 4.1.2-13: Extractability Data using the Metabolism Extraction Procedure

Matrix	Final Reported ppm			
	Cis I	Cis II	Trans III	Trans IV
Soil	0.0024	0.0033	ND	ND

Standard stability During the method validation, it was found that the calibration standard solutions of BAS 3111 were stable (less than 20% decline) for at least 64 days refrigerated and the fortification and concentrated stock solutions were stable for at least 65 days. The metabolites of BAS 311 I were stable (less than 20%) for at least 38 days for calibration and fortification solutions and at least 69 days for the concentrated stock solution.

Conclusion

The method for analysis of the diastereomeric forms of BAS 311 I (Cis I, Cis II [alpha-cypermethrin], Trans III, and Trans IV), and its three metabolites 3-phenoxybenzoic acid (Reg. No. , PBA) and DCVA (Reg. No. , cis- and trans-isomers) in soil uses LC-MS/MS for final determinations, which is a modern and highly specific technique. The limit of quantitation was 0.001 mg/kg for each analyte.

It was demonstrated that method R0034/01 fulfills the requirements with regard to specificity, repeatability, limit of quantitation, and recoveries and is, therefore, applicable to correctly determine residues of the insecticide BAS 311 I (cypermethrin, Reg. No. 127266) and its metabolites 3-phenoxybenzoic acid (Reg. No. 130213) and DCVA (cis- and trans-isomers, Reg. No. 180011) in soil.

~~**Report:** CA 4.1.2/2~~
~~N.N.~~
~~Determination of metabolite M310I017 (Reg. No. 6009306) in soil by LC-MS/MS~~
~~2015/1000961~~
~~**Guidelines:** OPPTS 850.7100, OPPTS 835.6100, PR Notice 2011-3,~~
~~SANCO/3029/99 rev. 4, 2000 (11 July 2000)~~
~~SANCO/825/00 rev. 8.1, 2010 (16 November 2010)~~
~~**GLP:** yes~~

~~The title has slightly changed from the application submitted as the analyte was allocated a different internal BASF Reg. No. This changed from 6002320, which was supposed to contain only the relevant cis II isomer, whereas the provided analyte 6009306 is a mixture of all four cypermethrin isomers, hence it was allocated a different unique identifier number. Apparently, during synthesis the cis II isomer (alpha-cypermethrin) could not be purified for technical reasons.~~

~~The method allows the determination of the metabolite M310I017 (Reg. No. 6009306, (cyano[3-(4-hydroxyphenoxy)phenyl]methyl (1RS,3RS) 3-(2,2-dichloroethenyl) 2,2-dimethylcyclopropanecarboxylate) in soil by LC-MS/MS.~~

~~The study is at the time of submission of the dossier still ongoing as synthesis and certification of required reference compound has been accomplished late in the process by the fourth quarter 2014. Expected finalisation date of the GLP study will be the first quarter of 2015. Upon finalisation of the study, the relevant documentation will be immediately submitted.~~

Report: CA 4.1.2/6
Geschke, S. (2015)
Validation of BASF analytical method L0286/01: Determination of Residues of Reg. No. 6009306 (M310I017cis, hydroxylated cypermethrin) in two different Soil Types
2015/1000961
Guidelines: OPPTS 850.7100, OPPTS 835.6100, PR Notice 2011-3, SANCO/3029/99 rev. 4, 2000 (11 July 2000), SANCO/825/00 rev. 8.1, 2010 (16 November 2010)
GLP: yes
(Landesamt fuer Umwelt, Messungen und Naturschutz Baden-Wuerttemberg, Karlsruhe, Germany)

Principle of the method L0286/01:
The analytical method was used for the determination of a cypermethrin metabolite. The analytical method was based on method R0034/01. The sample extraction was adapted to extract the hydroxylated cypermethrin. A 5.0 g soil sample aliquot was extracted with 0.1% formic acid in acetonitrile. One aliquot of the extract was then centrifuged for 5 min at 4000 rpm. An aliquot was taken and evaporated to dryness under a stream of nitrogen at 50°C and reconstituted in acetonitrile/pure water 1/1 v/v + 0.1% formic acid. The residue in the extract was determined using LC-MS/MS (Waters Acquity UPLC HSS T3, 150 × 2.1 mm, 1.8 µm, quantitation transition: 430 → 207, confirmation transition: 432 → 209). External calibration.

Conditions for gradient elution at a flow rat of 400 µL/min:

Time (min)	Mobile phase A : 4 mM ammonium formate solution in water + 0.1% formic acid	Mobile phase B: 4 mM ammonium formate solution in methanol + 0.1% formic acid
0.0	95	5
1.0	95	5
4.0	2	98
6.9	2	98
7.0	95	5
8.0	95	5

Specificity/Interferences: Method is highly selective and specific as 2 mass transitions were monitored (no further confirmatory method is required). The method was validated for both mass transitions.

No interferences have been observed at the retention time of the analyte from the soil control samples.
Representative chromatograms for matrix-matched calibration solutions, controls and fortified samples at LOQ and 10 × LOQ are provided for each concerned mass transition monitored. Product ion spectrum for the analyte was provided for the 430 and 432 m/z.

Linearity: Determination at 8 concentrations (single injection). Method is linear $r \geq 0.9999$ in the 0.1 - 20 ng/mL range (corresponding to a 0.0002 – 0.04 mg/kg range) for the two mass transitions.
The concentrations of the samples meet the calibration range.

Accuracy: Soil matrices were fortified with the analyte at two fortification levels (0.001 and 0.01 mg/kg). For each fortification levels, 5 replicates were analysed. 2 replicates of unfortified samples were also examined for each soil type. Mean recoveries can be consulted in Table **4.1.2-14**.

Repeatability: RSD remains below 20% at each fortification level for all analytes. RSD are presented in Table **4.1.2-14**.

LOQ: 0.001 mg/kg.

Table 4.1.2-14: Recoveries for hydroxylated cypermethrin in soils using LC-MS/MS method L0286/01

Matrix	Fortification Levels (ppm)	n	Recovery (%)	Average Recovery (%)	%RSD
Soil Lufa 2.2	Primary Quantitation (*m/z* 430.0 → *m/z* 207)				
	0.001	5	98, 96, 106, 102, 101	101	4
	0.01	5	94, 90, 95, 89, 94	72	3
	Confirmatory Quantitation (*m/z* 432.0 → *m/z* 209)				
	0.001	5	98, 95, 101, 97, 92	97	3
	0.01	5	95, 91, 96, 90,99	94	4
Soil Lufa 5M	Primary Quantitation (*m/z* 430.0 → *m/z* 207)				
	0.001	5	97, 95, 95, 91, 92	94	3
	0.01	5	99, 97, 100, 100, 94	98	3
	Confirmatory Quantitation (*m/z* 432.0 → *m/z* 209)				
	0.001	5	97, 101, 103, 95, 98	99	3
	0.01	5	98, 98, 101, 97, 95	98	2

Stability

The calibration solutions were stable for at least 10 days when stored refrigerated. The analyte is stable in the final extracts if stored in the refrigerator for 9 days.

Conclusion

Method L0286/01 (based on method R0034/01 - LC-MS/MS) is fully validated according to SANCO/3029/99 rev.4 and SANCO/825/00 rev. 8.1. The mean recovery at each fortification level is within the 70-110 % range for both mass transitions and the repeatability is always below 20%. The method has been shown to be linear (second order) and specific. No confirmatory method is required since two mass transitions have been monitored and validated.

The method is considered suitable to hydroxylated cypermethrin (metabolite of cypermethrin) in soil with a validated LOQ of 0.001mg/kg. The method can be used as a pre- as well as a post-registration method.

Water

Analytical methods for the determination of alpha-cypermethrin residues in water were evaluated in the context of the inclusion in Annex I of Directive 91/414/EEC. Methods evaluated previously are listed in Table 4.1.2-11 and summarized shortly below.

Table 4.1.2-15: Summary of peer-reviewed analytical methods for determination of alpha-cypermethrin residues in water

Method No.	Matrix	Method principle	Target analytes	LOQ µg/l	year	DocID	EU reviewed
SAMS 469-2	Water	GC/ECD; GC/MS for confirmation	Alpha-cypermethrin	0.01	1990	AL-243-001	Yes
SAMS 470-1	Water	GC/ECD; GC/ECD with alternative capillary column for confirmation	Permethrin	0.01	1988	AL-243-002	Yes
SAMS 468-1	Water	GC/ECD; GC/MS for confirmation	Cypermethrin	0.01	1988	AL-243-002	Yes
SAMS 469-1	Water	GC/ECD; GC/MS for confirmation	Alpha-cypermethrin	0.01	1988	AL-243-002	Yes
SAMS 469-2	Pond water	GC/ECD	Alpha-cypermethrin	0.01	1997	AL-243-003	Yes
SAMS 469-2	Surface water	GC/ECD; GC/MS for confirmation	Alpha-cypermethrin	0.05	1999	AL-243-006	Yes
M 3499	Water	sample prep. acc. to SAMS 469-2; GC/MS (confirmatory method)	Alpha-cypermethrin	0.05	2001	AL-210-012	Yes

For completeness of information, summaries of already EU-peer reviewed analytical methods are presented for completeness of information.

Report:	CA 4.1.2/7 Anonymous, 1990 Determination of residues of Alphacypermethrin in water - Gas chromatographic method
	AL-243-001
Guidelines:	none
GLP:	No

Report:	CA 4.1.2/8 Sherren A.J., 1990 Development of methods for the analysis of water for residues of Permethrin, Cypermethrin and Alphacypermethrin
	AL-243-002
Guidelines:	none
GLP:	No

Report:	CA 4.1.2/9 Pelz S. et al., 1997 Validation of method SAMS 469-2 for the determination of AC 900049 (Alphacypermethrin) applied as a formulation (FastacTM 100 G/L OESC insecticide) in pond water
	AL-243-003
Guidelines:	none
GLP:	Yes (Behoerde fuer Arbeit, Gesundheit und Soziales, Freie und Hansestadt Hamburg, Hamburg)

Report:	CA 4.1.2/10 Werle H., 1999 Alphacypermethrin (CL 900049) validation of method SAMS 469-2 for the determination of residues in surface water
	AL-243-006
Guidelines:	EEC 91/414,EEC 96/46,BBA Guideline Residue Analytical Methods for Post-Registration Control Purposes of July 21 1998,Guidance Document of Residue Analytical Methods 8064/VI/97 rev. 4 15.12.1998
GLP:	Yes (Umweltministerium Baden Wuerttemberg, Stuttgart)

Principle of the method:
Water samples are extracted by solvent partition with hexane and, if required, the extract is cleaned up using a Florisil disposable cartridge.
Alpha-cypermethrin is determined by GC with electron capture detection (ECD), using either *capillary column* (5% phenyl methyl silicone; 15m×0.32mm, 0.25 µm) or *packed column* (GE-XE 60 2% (m/m) on Gas Chrom Q (100-120 mesh)); quantification by external standardization. Analysis by capillary column is the preferred method. If required, residues may be confirmed by GC-MS (CPSil 5CB), MS operated in negative ion chemical ionisation mode (ions monitored: m/e 207 and 209).
The packed column does not allow the distinction between the different isomers of cypermethrin whereas the capillary column does. Under the conditions listed for GC-MS for confirmation purposes, the four isomers of cypermethrin are not resolved and elute as a single peak. Thus the confirmation indicates the presence of cypermethrin but does not distinguish the Cis-2 isomer.
It should be noted that for validation of SAMS 469-2 under the study by Werle 1999, modifications to the GC conditions occurred compared to the initial method (for GC-ECD: DB5 column, 30m×0.25mm, 0.25µm, differences in the injection and oven program and for GC-MSD incl. column Optima 5, 25m×0.25mm, 0.25µm and different carrier gas).
The method development of SAMS 469-2 in pond water was performed in support of aquatic toxicity studies with modified conditions (column FS capillary column 30m × 0.53 mm, 100% methyl silicone, splitless mode in place of FS capillary column 15 m × 0.32 mm, 5 % phenyl methyl silicone, split ratio 10:1) compared to the initial SAMS 469-2 method.

Specificity/Interferences*:*	Typical GC-ECD chromatograms, resp. obtained on packed column (SAMS 469-02) and capillary column (SAMS 469-01), are shown. Control samples exhibit no interfering peaks at the retention time of alpha-cypermethrin; control and blank data were stated to be < 0.01 µg/L.
	The method can be confirmed by GC-MS with method M3499, described below.
Linearity:	Method was found to be linear in the 0.0126 to 0.502 µg/mL (n=5, duplicate injection) with a correlation coefficient R^2 = 0.9926. The calibration curve and the regression equation were provided in the study report (determination with the capillary GC-ECD modified method only). Matrix effects were not demonstrated but can be sensibly reduced by the clean-up step applied.
	In SAMS 469-2 applied in pond water: method was found linear in the 0.00503 – 0.503 µg/mL range (n = 4 - duplicate injections for some concentrations) with a correlation coefficient r = 0.9997. The linear curve and the corresponding equation were provided.
Recovery/Precision:	see Table **4.1.2-16**
Limit of quantitation*:*	The limit of quantitation (LOQ) of the method for both metabolites, defined by the lowest fortification level is 0.05 mg/kg.

Table 4.1.2-16: Validation of methods SAMS 469-01 and SAMS 469-02

Matrix	Analyte	Fortification level (µg/L commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
source water (analysis by packed column) SAMS 469-02	alpha-cypermethrin	0.05 0.20 0.50	5 5 5	90 - 120 90 - 105 80 - 120	107 95 100	10.2 6.5 15.4
		0.05 - 0.50	15	80 - 120	100.7	11.7
source water (analysis by capillary column) SAMS 469-01	alpha-cypermethrin	0.10 0.20	2 2	85 - 90 95 - 95	87.5 95	(4.0) (0.0)
		0.10 - 0.20	4	85 - 95	91.3	5.3
surface water (analysis by capillary column) SAMS 469-02 (modified)	alpha-cypermethrin	0.05 0.5	5 5	80-103 78-107	88 89	8.9 12
		0.05-0.5	10	78-107	89	9.7

Storage stability: Water samples must be stored cold (at 4 °C) and extracted within 3 days of sampling.

Conclusion: **The packed column method is suitable for alpha-cypermethrin residue analysis in source water with a LOQ of 0.05 µg/L. The capillary column method is suitable for alpha-cypermethrin residue analysis in source and surface water with a LOQ of 0.05 µg/L.**

Report:	CA 4.1.2/4 Xu B., 2001 BAS 310 I (Alpha-Cypermethrin): Validation of method M 3499 for the confirmation of BAS 310 I residues in water soil and blood by GC/MS AL-210-012
Guidelines:	Guidance Document on Residue Analytical Methods (SANCO/825/00rev.6)
GLP:	Yes (United States Environmental Protection Agency)

Principle of the method:
Method M 3499 is used to confirm the specificity of method SAMS 469-2 (GC-ECD). The procedure for sample preparations is the same as the one outlined in SAMS 469-2 but an alternative detection method is used (determination by capillary GC (DB-5MS, 5m × 0.25 mm i.d., 0.25 µm) with MSD in SIM mode (m/z 207, 209 and 211). Ground water was used as matrix.

Specificity/Interferences:	Specificity of method SAMS 469-2 is demonstrated by using GC-MS as an alternative method of detection. Untreated control samples exhibit no significant interferences at retention time of alphacypermethrin (control values < 30% of LOQ)
Linearity:	The response of the GC-MS system (= peak area) to alphacypermethrin was demonstrated to be linear over a concentration range from 0.0025 to 0.04 µg/mL (n = 5); r > 0.999 for each ion (207, 209 and 211). Samples are diluted if necessary to meet the calibration range.

Recovery/Precision: See Table **4.1.2-17**.

Limit of quantitation:	The limit of quantitation (LOQ) of the method, defined by the lowest fortification level is 0.05 µg/L.

Table 4.1.2-17: Validation of confirmatory method M 3499 for groundwater

Analyte	Fortification level (µg/L commodity)	Number of samples	% Retention Time Difference (RTD) (ion m/z 207)	% Ion Ratio Difference (IRD)	
				207/209	207/211
alphacypermethrin	0.05	3	0.1 0.0 0.1	- 0.1 - 0.8 - 0.2	- 4.7 - 3.0 - 5.0

% RTD = (sample retention time – average standard retention time) x 100/average standard retention time
% IRD = (sample ion ratio – standard average ion ratio) x 100/standard average ion ratio

Conclusion:	**Method M 3499 (GC-MS) is suitable as qualitative confirmatory method to SAMS 469-2 (GC-ECD) for determination of alphacypermethrin residues in water (LOQ 0.05 µg/L).**

No new methods for water were required for risk assessment in support of environmental fate studies. A newly developed analytical method for post-approval and monitoring purposes is described in detail under CA 4.2/7, respective independent laboratory validation under CA 4.2/8.

Air

Analytical methods for the determination of alpha-cypermethrin residues in air were evaluated in the context of the inclusion in Annex I of Directive 91/414/EEC. Methods evaluated previously are listed in Table **4.1.2-18** and summarized shortly below.

Table 4.1.2-18: Summary of peer-reviewed analytical methods for determination of alpha-cypermethrin residues in air

Method No.	Matrix	Method principle	Target analytes	LOQ µg/m³	year	DocID	EU reviewed
AMS 887-01	Air	HPLC-UV	Alpha-cypermethrin	3 µg/m³ for 15L air, 0.1 µg/m³ for 480L air	Not reported	AL-210-002	Yes
SAMS 534-1	Air	GC/NPD, GC/ECD, GC/MS	Alpha-cypermethrin	0.02	1992	AL-241-001	Yes
RCC project no. 249120	Air	GC/NPD	Alpha-cypermethrin	0.01	1992	AL-241-002	Yes

For completeness of information, summaries of already EU-peer reviewed analytical methods are presented for completeness of information.

Report: CA 4.1.2/11
Anonymous, 1992
Determination of crystalline Fastac in aerosols
AL-210-002
Guidelines: none
GLP: No

Principle of the method:
A known volume of air (15 L or 480 L, dependent on the expected concentration) is drawn through a silver membrane filter with glassfibre prefilter and cellulose pad (at 1 L/min) to trap the crystalline alpha-cypermethrin present, after which the inlet and outlet of the filter holder are sealed. The filters are extracted by sonication with diethylether and the resulting solution is reduced by evaporation. The enriched solution is analyzed by HPLC (Spheri 5 RP 18; isocratic elution) with UV detection at 215 nm.

As the method has already been evaluated as not appropriately validated before, not further data are summarised for this method.

Report:	CA 4.1.2/12 Anonymous, 1992 Determination of Alphacypermethrin in air using an adsorbent trap - Gas chromatographic method AL-241-001
Guidelines:	none
GLP:	No

Principle of the method:
Air to be sampled (120 L) is pumped through a tube packed with Chromosorb 102 at 250 mL/min, after which the tube is capped and stored at 4 °C pending analysis. The alpha-cypermethrin is desorbed from the Chromosorb 102 (front and back section separately) by sonication with acetone, dodecane is added and the acetone is evaporated.
The resulting solution is analyzed by capillary GC (DB-5; 0.25 µm) with nitrogen phosphorous selective detection (NPD); quantification by external standardization. An electron capture detector could also be used, provided the final extracts are sufficiently clean; clean-up of the extract may be achieved by using a Florisil cartridge. If required, residues may be confirmed by GC-MS (CPSil 5CB), MS operated in negative ion chemical ionisation mode (ions monitored : m/e 207 and 209).

Specificity/Interferences: Typical GC-NPD chromatograms are shown. Control sample appears to exhibit some interference at retention time of alpha-cypermethrin, but control and blank data were nevertheless stated to be < 0.02 µg/m³.

Recovery/Precision: see Table **4.1.2-19**

Limit of quantitation: The limit of quantitation (LOQ) of the method, defined by the lowest fortification level is 0.02 µg/m³.

Table 4.1.2-19: Validation of method SAMS 534-01

Matrix		Analyte	Fortification level (µg/m³ commodity)	Recovery			
				Number of samples	Range (%)	Mean (%)	RSD (%)
air (T : 21-30 °C; RH : 40-90%)		alpha-cypermethrin	0.02 0.10 0.20 0.50	5 5 4 5	98 - 115 86 - 105 91 - 94 75 - 94	104.6 92.6 92.5 83	7.8 8.1 1.4 10.3
			0.02 - 0.50	19	75 - 115	93.2	11.2

Conclusion: **The method is suitable for alpha-cypermethrin residue analysis in air with a LOQ of 0.02 µg/m³.**

Report:	CA 4.1.2/13 Mueller-Kallert H., 1992 Development of a method for the determination of Alphacypermethrin in air AL-241-002
Guidelines:	BBA I 1-2
GLP:	Yes (Eidgenoessisches Department des Inneren, Bern, Schweiz)

This study referenced as Method AL-241-002 (1992/7001981) is similar to the peer-reviewed method AL-241-001 / 1992/7001991 (SAMS 534-01 – see above) and is based on SAMS 534-01. Method AL-241-001 has been validated again as a repeat of the method AL-241-002 for a regional submission.

Validation data found in the report are summarized here below:

Specificity/Interferences:	Typical GC-NPD chromatograms are shown. Considerable background was visible with NPD detection. However, this background noise was lower than signals resulting from a concentration equal to the LOQ.
Linearity:	Investigated range 0.01 – 05 µg/mL with a correlation coefficient of 0.9984. Equation and calibration curve were provided.
Recovery/Precision:	see Table **4.1.2-19**
Limit of quantitation:	The limit of quantitation (LOQ) of the method, defined by the lowest fortification level is 0.02 µg/m³.
Conclusion:	**The method is suitable for alpha-cypermethrin residue analysis in air with a LOQ of 0.02 µg/m³.**

No new methods for air are required for risk assessment in support of environmental fate studies. A newly developed analytical method for post-approval and monitoring purposes is described in detail under CA 4.2/9.

(b)Methods in soil, water and any additional matrices used in support of efficacy studies

No stand-alone validation of analytical methods was required in support of efficacy studies.

(c)Methods in feed, body fluids and tissues, air and any additional matrices used in support of toxicological studies

No stand-alone methods for toxicological studies were required. However, a newly developed method for the determination of alpha-cypermethrin in blood and urine was developed and validated for post-approval and monitoring purposes and is described in detail in CA 4.2/10.
A summary table of methods already evaluated is also presented in CA 4.2/10 for the reviewer's convenience.

Detailed summaries of the analytical methods used for data-generation purposes in support of toxicological gavage and feeding studies are given below. Analytical methods used for data generation of stability assessment are also summarised below. The analytical methods were not allocated a unique document identifier, but the summaries below are given under the unique document identifier (DocID) of the respective toxicological study.

Report:	CA 4.1.2/14 Maas X., Bending P., 2015 a Alpha-Cypermethrin: Analysis of alpha-Cypermethrin in rodent samples originating from a mammalian toxicity study 2015/1175543
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Messungen und Naturschutz Baden Wuerttemberg, Karlsruhe, Germany)

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Principle of the method

The analytical procedure/method was developed and validated based on QueChERS extraction with subsequent clean-up and LC-MS/MS and adapted for the determination of alpha-cypermethrin (Cis II isomer).
A homogenized sample (rodent body or rodent brain) was weighed into a 50 mL centrifuge tube and water was added. After shaking manually for 30 sec, acetonitrile was added and the sample was shaken manually for 1 min. Then, the content of a Dispersive SPE (dSPE) citrate extraction tube was added and shaken manually for 1 min. After centrifugation for 5 min at 3000 rpm, 6 mL of the acetonitrile layer was transferred to a Dispersive SPE PSA SPE clean-up tube. The sample was shaken manually for 30 sec and then centrifuged for 5 min at 3000 rpm. An aliquot of 1 mL of the supernatant was transferred to an autosampler vial and 10 µL of acetonitrile/formic acid (50/1, v/v) was added.
Analysis was accomplished by LC-MS/MS. The chromatography was performed on a Waters Acquity UPLC HSS T3 column with a flow rate of 0.4 mL/min and a gradient mixture of water/formic acid/10 mM ammonium formate (1000/1/0.63, v/v/w) and methanol/formic acid/10 mM ammonium formate (1000/1/0.63, v/v/w). Detection was accomplished by electrospray ionization (positive mode) at mass transitions 433→191 for quantification and 435→193 for confirmation.

Recovery findings

The described method is suitable to determine residues of alpha-cypermethrin in samples of rodent body and rodent brain. Samples were spiked with alpha-cypermethrin with 0.01 mg/kg (LOQ) and 0.1 mg/kg (10xLOQ) and rodent body samples additionally with 0.2 mg/kg (20xLOQ). All average recovery values were between 70% and 120%. The detailed results are given inTable 4.1.2-20.

Table 4.1.2-20: Recovery results of alpha-cypermethrin (Cis II isomer) in rodent brain and body samples

Matrix	m/z	Fortification level [mg/kg]	Number of Replicates	Mean Recovery [%]	RSD [%]	Overall Recovery [%]	RSD [%]
Rodent Brain	433 → 191	0.01	3	103	1	105	3
		0.1	3	107	4		
	435 → 193	0.01	3	104	2	105	3
		0.1	3	106	4		
Rodent Body	433 → 191	0.01	3	96	2	95	7
		0.1	3	98	6		
		0.2	1	83	--		
	435 → 193	0.01	3	102	3	98	8
		0.1	3	99	6		
		0.2	1	84	--		

RSD = Relative standard deviation

Linearity

The linearity was tested at concentrations between 0.01 ng/mL and 25 ng/mL for rodent brain and 0.25 ng/mL to 30 ng/mL for rodent body, prepared in acetonitrile/formic acid (1000/1, v/v). Linear correlation with coefficients of $r > 0.99$ were obtained. Seven (rodent brain) and eight (rodent body) calibration standards distributed over the range given above were used, respectively.

Specificity

LC-MS/MS is a highly specific self-confirmatory technique. Under the described conditions, the method is specific for the determination of alpha-cypermethrin in rodent brain and rodent body samples. Analysis is possible at two mass transitions.

Due to the high selectivity and specificity of LC-MS/MS an additional confirmatory technique was not necessary.

Interference

No residues of alpha-cypermethrin were present above 0.002 mg/kg (20% of LOQ) in any of the blank control specimens.

Matrix Effects

Matrix effects were tested by comparing the response of the analyte in calibration solutions in solvent versus response in matrix-matched calibration standards. No significant effects (< 20%) were observed.

Limit of Quantification

The limit of quantification (LOQ) is defined by the lowest fortification level of 0.01 mg/kg for alpha-cypermethrin.

Limit of Detection

The limit of detection (LOD) of alpha-cypermethrin is defined by the lowest calibration level with 0.0002 mg/kg for rodent brain and 0.001 mg/kg for rodent body.

Repeatability The relative standard deviation (RSD, %) for all fortification levels were < 20% for alpha-cypermethrin.

Reproducibility Reproducibility of the method was not determined within the validation study.

Conclusion **The described method for analysis of alpha-cypermethrin (Cis II isomer) in rodent brain and rodent body samples used LC-MS/MS for final determination, which is a highly specific technique.**

It could be demonstrated that the method fulfils the requirements with regard to linearity, specificity, repeatability, limit of quantification, and recoveries and is therefore applicable to correctly determine residues of alpha-cypermethrin (Cis II isomer) in rodent brain and body samples.

Report:	CA 4.1.2/15 Becker M., Kamp H., 2015 a Analytical report - BAS 310 I (Alpha-Cypermethrin) - Stability analysis in corn oil 2014/1170685
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Executive Summary

In the context of toxicological studies, the stability of alpha-cypermethrin (BAS 310 I) in the vehicle corn oil was investigated in one stability study.

Standard solutions were stored at room temperature and analyzed at day 0 and after 4 h, 4 d, and 7 days.

Stability of alpha-cypermethrin at a nominal concentration of 200 mg/L was given over a time period of 7 days of storage at ambient temperature.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Internal code:	BAS 310 I
Common name:	alpha-cypermethrin
Batch No.:	PMAM000622
Test substance No.	01/0265-5
Purity:	99.2%

B. STUDY DESIGN

1. Experimental Conditions

50.0 mg of alpha-cypermethrin were dissolved in 20 mL acetone. An aliquot of 80 µL was taken and evaporated at room temperature. Then, 1 mL of corn oil was added. For each sampling time point a separate sample was prepared. The final nominal concentration was 200 mg/L.

2. Description of analytical procedures

Analysis was accomplished by gas chromatography (GC) equipped with FID (detector temperature 340°C). The chromatography was performed on a DB-1 MS column at a constant flow of 1.4 mL/min using helium as carrier gas. A temperature gradient was run, starting with a temperature of 230°C (hold time 23 min), followed by a temperature increase of 25°C/min to 340°C (hold time 30 min). Run time was approximately 58 min.

Principle of the method:
Samples were diluted completely with acetone using appropriate volumetric flasks to obtain sample solutions with alpha-cypermethrin concentrations that match the actual calibration range of 19.2 mg/L to 192 mg/L. Adequate dilutions with matrix solutions were performed as necessary. It was ensured, that the test substance was completely dissolved in the solutions.

Validation:

Specificity/Interferences:	The study report provided chromatograms for blank matrix, matrix-matched calibration standard and the stability sample after 7 days. No interferences seem to occur at the retention time of the analyte. Chromatograms were also provided for control, calibration and samples in study (DocID 2014/1275120) and did not show any interferences above 30% of the lowest calibration level.
Linearity:	The linearity of the method was investigated at concentrations between 19.2 to 192 mg/L in corn oil matrix (matrix-matched calibration solutions). Linear correlation coefficient and regression equation were not provided but the calibration curve showed the good linearity of the method in the investigated range (six calibration concentrations were used to demonstrate the linearity). The method mentioned that samples are diluted if required to meet the calibration range.
Recovery/Precision:	Not demonstrated as such. The stability of alpha-cypermethrin in corn oil was determined in one stability study over a time period of 7 days at room temperature at a nominal concentration of 200 mg/L. Over the investigated time period of 7 days, all measured concentrations were in the range between 90% to 110% of the nominal concentration. The detailed results are summarized in Table 4.1.2-21.
Limit of quantification:	An LOQ of 200 mg/L is proposed.
Conclusion:	**The method described is suitable for the quantification of alpha-cypermethrin in corn oil with a limit of quantification of 200 mg/L.**

II. RESULTS AND DISCUSSION

The stability of alpha-cypermethrin in corn oil was determined in one stability study over a time period of 7 days at room temperature at a nominal concentration of 200 mg/L.

Over the investigated time period of 7 days, all measured concentrations were in the range between 90% to 110% of the nominal concentration. The detailed results are summarized in Table 4.1.2-21.

Table 4.1.2-21: Results for stability analysis of alpha-cypermethrin in corn oil

Matrix	Time after starting	Target concentration [mg/L]	Concentration found [mg/L]	Nominal concentration [%]
Corn oil	0 h	200	196.87	98.4
	4 h	200	198.43	99.2
	4 d	200	196.65	98.3
	7 d	200	195.54	97.8

4.1.2-22: Results for concentration control of alpha-cypermethrin in corn oil (2014/1275120)

Matrix	Target concentration [mg/L]	N	% Mean recovery (range)	RSD
Corn oil	200	3	97.3 (96.2, 99.7, 99.0)	1.5
	350	3	100.2 (10.7, 99.0, 100.8)	1.0
	660	3	99.6 (100.1, 99.1 99.7)	0.5

III. CONCLUSION

Based on the analytical results it was demonstrated that alpha-cypermethrin was stable in corn oil over a time period of 7 days stored at room temperature at a nominal concentrations of 200 mg/L.

Report:	CA 4.1.2/16 Grauert E.,Kamp H., 2014 a BAS 310 I (Alpha-Cypermethrin) - Stability analysis in 1% Carboxymethylcellulose in drinking water 2014/1275135
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/17 Grauert E.,Kamp H., 2015 a BAS 310 I (Alphacypermethrin) - Stability analysis in 1% Carboxymethylcellulose in drinking water 2015/1093547
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

The data from M-CA 4.1.2/17 and M-CA 4.1.2/18 are summarised together and given below. An overview of the analytical procedures given below is done under the unique document identifier of the original toxicological studies.

Executive Summary

In the context of toxicological studies, the stability of alpha-cypermethrin (BAS 310 I) in the vehicle 1% carboxymethylcellulose (CMC) in drinking water was investigated in two stability studies.
In each stability study, standard solutions were stored at room temperature and analyzed at day 0 and after 4 h, 24 h, 96 h and 7 days.

Stability of alpha-cypermethrin at nominal concentrations of 201.6 mg/L and 104.4 mg/L was given over a time period of 7 days of storage at ambient temperature.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Internal code:	BAS 310 I
Common name:	alpha-cypermethrin
Batch No.:	PMAM000622
Test substance No.	01/0265-5
Purity:	99.2%

B. STUDY DESIGN

1. Experimental Conditions

BASF DocID 2014/1275135
50.4 mg of alpha-cypermethrin were dissolved in 10 mL acetone. An aliquot of 40 µL was taken and evaporated at room temperature. Then, 1 mL of 1% carboxymethylcellulose (CMC) in drinking water was added. For each sampling time point a sample was prepared. The final nominal concentration was 201.6 mg/L.

BASF DocID 2015/1093547
52.2 mg of alpha-cypermethrin were dissolved in 25 mL acetone. An aliquot of 50 µL was taken and evaporated at room temperature. Then, 1 mL of 1% carboxymethylcellulose (CMC) in drinking water was added. For each sampling time point a sample was prepared. The final nominal concentration was 104.4 mg/L.

2. Description of analytical procedures

BASF DocID 2014/1275135
Analysis was accomplished by gas chromatography (GC) equipped with FID (detector temperature 340°C). The chromatography was performed on a DB-1 MS column at a constant flow of 1.4 mL/min using helium as carrier gas. A temperature program was run, starting with a temperature of 230°C (hold time 23 min), followed by a temperature increase of 25°C/min to 340°C (hold time 5 min). Run time was approximately 32 min.

Principle of the method:
Samples were diluted completely with solvent solutions (900 mL acetone + 100 mL citric acid 5% in water, v/v) using appropriate volumetric flasks to obtain sample solutions with alpha-cypermethrin concentrations that match the actual calibration range of 9.48 mg/L to 94.8 mg/L. Adequate dilutions with matrix solutions were performed as necessary. It was ensured, that the test substance was completely dissolved in the solutions.

BASF DocID 2015/1093547
Analysis was accomplished by gas chromatography (GC) equipped with FID (detector temperature 340°C). The chromatography was performed on a DB-1 MS column at a constant flow of 1.4 mL/min using helium as carrier gas. A temperature program was run, starting with an initial temperature of 70°C (hold time 3 min), followed by a temperature increase of 25°C/min to 230°C (hold time 22 min) and further increase by 30°C/min to 340°C (hold time 5 min). Run time was approximately 40 min.

Principle of the method:
Samples were transferred into volumetric flasks with approximately 5 to 10 mL of a mixture of acetone/citric acid 5% in water (1+1, v/v) and filled up to the mark with solvent solution (950 mL acetone + 50 mL citric acid 5% in water, w/v) to obtain sample solutions with alpha-cypermethrin concentrations that match the actual calibration range of 1.002 mg/L to 10.02 mg/L. Adequate dilutions with matrix solutions were performed as necessary. It was ensured, that the test substance was completely dissolved in the solutions.

Validation:

Specificity/Interferences: The study report provides chromatograms for blank matrix, matrix-matched calibration standard and the stability sample after 4 hours for the first study and after 7 days for the second study. No interferences occur at the retention time of the analyte.

Linearity: The linearity of the method was investigated at concentrations between 9.48 to 94.8 mg/L and 1.002 to 10.02 mg/L in 1% CMC in drinking water (matrix-matched calibration solutions), respectively. Linear correlation coefficient and regression equation are not provided but the calibration curve shows the good linearity of the method in the investigated range (six and five calibration concentrations were used to demonstrate the linearity, respectively).

Recovery/Precision: Not demonstrated by fortification at different levels by means of several replicates. Only stability data are available. In the investigated time period of 7 days, all measured concentrations in both stability studies were in the range between 90% to 110% of the nominal concentrations. The detailed results for both studies are summarized in Table 4.1.2-23.

Limit of quantitation: Not stated.

II. RESULTS AND DISCUSSION

The stability of alpha-cypermethrin in 1% carboxymethylcellulose (CMC) in drinking water was determined in two stability studies [*BASF DocID 2014/1275135 and BASF DocID 2015/1093547*] over a time period of 7 days at room temperature at nominal concentrations of 201.6 mg/L and 104.4 mg/L.

Over the investigated time period of 7 days, all measured concentrations in both stability studies were in the range between 90% to 110% of the nominal concentrations. The detailed results for both studies are summarized in Table 4.1.2-23.

Table 4.1.2-23: Results for stability analysis of alpha-cypermethrin in 1% carboxymethylcellulose in drinking water

Matrix	BASF DocID	Time after starting	Target concentration [mg/L]	Concentration found [mg/L]	Nominal concentration [%]
1% CMC in drinking water	2014/1275135	0 h	201.6	203.10	100.7
		4 h	201.6	191.85	95.2
		24 h	201.6	202.50	100.4
		96 h	201.6	205.33	101.9
		7 d	201.6	202.07	100.2
	2015/1093547	0 h	104.4	107.52	103.0
		4 h	104.4	101.73	97.4
		24 h	104.4	103.08	98.7
		96 h	104.4	102.09	97.8
		7 d	104.4	96.88	92.8

CMC = carboxymethylcellulose

III. CONCLUSION

Based on the analytical results it was demonstrated that alpha-cypermethrin was stable in 1% carboxymethylcellulose in drinking water over a period of 7 days stored at room temperature at nominal concentrations of 201.6 mg/L and 104.4 mg/L.

Report:	CA 4.1.2/18 Becker M.,Kamp H., 2015 b Analytical report - 3-Phenoxybenzoic acid (metabolite of BAS 310 I, Alpha-Cypermethrin) - Concentration control analysis in Polyethylene glycol 400 2015/1003984
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Principle of the method

Samples were diluted completely with acetonitrile using appropriate volumetric flasks to obtain sample solutions with 3-phenoxybenzoic acid (metabolite of alpha-cypermethrin) concentrations that match the actual calibration range of 5.21 mg/L to 52.1 mg/L. Adequate dilutions with matrix solutions were performed as necessary. It was ensured, that the test substance was completely dissolved in the solutions.
Analysis was accomplished by HPLC with UV-detection (DAD) at 230 nm with reference wavelength of 360 nm. The chromatography was performed on a Kinetex C18 column with a flow rate of 3.0 mL/min and a gradient mixture of acetonitrile/water/formic acid (950/50/1, v/v/v) and water/acetonitrile/formic acid (950/50/1, v/v/v).

Recovery findings

The described method is suitable to determine residues of 3-phenoxybenzoic acid in polyethylene glycol 400. Samples were spiked with 3-phenoxybenzoic acid with concentrations of 50 mg/g, 100 mg/g and 200 mg/g. The calculated mean values of both sample sets (main and retain samples) were in the range between 85% and 88%. Values that are in the range of ± 15% of the nominal concentrations can be regarded as acceptable for liquid test substance preparations. The detailed results are given in Table 4.1.2-24.

Table 4.1.2-24: Results of concentration control of 3-phenoxybenzoic acid in polyethylene glycol 400

Matrix	Sample set	Amount [mg/g]	Nominal concentration [mg/g]	Nominal concentration [%]	Average nominal concentration [a] [%]	RSD [a] [%]
Polyethylene glycol 400	Sample set a (main samples)	42.071	50	84.1	84.7	0.7
		85.172	100	85.2		
		169.433	200	84.7		
	Sample set b (retain samples)	44.061	50	88.1	87.5	0.6
		87.352	100	87.4		
		174.111	200	87.1		

RSD = Relative standard deviation
[a] Average nominal concentrations and RSD values are based on calculations of the data originating from the study report.

Linearity The linearity was tested at concentrations between 5.21 mg/L and 52.1 mg/L, prepared in matrix solution (preparation procedure according to the description in principle of the methods). Linear correlation with coefficients of $r > 0.995$ were obtained. Five calibration standards distributed over the range given above were used.

Specificity The identification and quantification of 3-phenoxybenzoic acid were based on the selected wavelength at 230 nm and the retention time. Under the described conditions, the method is specific for the determination of 3-phenoxybenzoic acid in polyethylene glycol 400. As the method is only used for dose verification of known substances and known nominal concentrations, no additional confirmatory technique is necessary.

Interference No test substance was detected in the polyethylene glycol 400 control samples with a concentration of $\geq 30\%$ of the lowest calibration solution.

Matrix Effects Matrix-matched calibration solutions were used for quantification of 3-phenoxybenzoic acid, hence any potentially occurring matrix effect has already been accounted for by adjusting the matrix in the standard solutions used for calibration.

Limit of Quantification The limit of quantification (LOQ) is defined by the lowest fortification level of 50 mg/g for 3-phenoxybenzoic acid.

Limit of Detection The limit of detection (LOD) of 3-phenoxybenzoic acid is defined by the lowest calibration level with 5.21 mg/L.

Repeatability	The relative standard deviation (RSD, %) for concentration levels were < 20% for 3-phenoxybenzoic acid.
Reproducibility	The average nominal concentrations in Table 4.1.2-24 are mean values of samples measured at different days (sample set a and b). The data are comparable, so a reproducibility is given.
Conclusion	**The described method is suitable for the determination of 3-phenoxybenzoic acid (metabolite of alpha-cypermethrin) in polyethylene glycol 400.**

Report:	CA 4.1.2/19 Becker M.,Kamp H., 2015 c Analytical report - 3-Phenoxybenzoic acid (metabolite of BAS 310 I, Alpha-Cypermethrin) - Plasma analysis for external studies 2015/1032402
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.
The method was used to verify the mere presence of the test substance 3-phenoxybenzoic acid (metabolite of alpha-cypermethrin) in mouse plasma by using confirmation analysis.

Principle of the method

30 µL of plasma were mixed with 270 µL acetonitrile in a plastic micro centrifuge tube. After vortex mixing and protein precipitation, the samples were centrifuged at 13000 rpm for 5 min. The supernatant was directly analyzed by HPLC-MS. The chromatography was performed on an Ascentis Express C18 column with a flow rate of 0.25 mL/min and a gradient mixture of acetonitrile/water/formic acid (950/50/0.1, v/v/v) and water/acetonitrile/formic acid (950/50/0.1, v/v/v). Detection was accomplished by electrospray ionization (negative mode) at mass transitions 214.1→169.9 as quantifier and 214.1→93.3 as qualifier.

Recovery findings

The described method is suitable to verify residues of 3-phenoxybenzoic acid in mouse plasma by confirmation analysis. Samples spiked with 3-phenoxybenzoic acid with 2000 mg/kg body weight were analyzed 1 h and 4 h after application. The calculated mean values after 1 h and 4 h after application were in the range between 67.2% and 68.9%. Based on the analytical results it was concluded that 3-phenoxybenzoic acid was detected and confirmed in all mouse plasma samples, as the ion ratios of the quantifier and qualifier ion were comparable between the test substance 3-phenoxybenzoic acid at a dose level of 2000 mg/kg body weight and the calibration solution (± 20%). The detailed results are given in Table 4.1.2-25.

Table 4.1.2-25: Results of 3-phenoxybenzoic acid in mouse plasma at 1 hour and 4 hours after application

Matrix	Hours after application	Dose [mg/kg body weight]	Retention time (reference) 4.70 – 5.20	Ion ratio (reference) 53.13% - 79.68%	Average ion ratio [a] (reference) 53.13% - 79.68%	RSD [a] [%]	Results
Mouse plasma	1 h	2000	4.95	69.1	68.9	1.9	Complies
			4.95	70.1			
			4.95	67.5			
	4 h	2000	4.95	67.9	67.2	0.9	Complies
			4.95	66.7			
			4.95	67.0			

RSD = Relative standard deviation
[a] Average ion ratio and RSD values are based on calculations of the data originating from the study report.

Linearity Not applicable and in case of mere positive confirmatory analysis (positive test) not relevant.

Specificity The identification and quantification of 3-phenoxybenzoic acid were based on the analysis by HPLC-MS and by its retention time. Under the described conditions, the method is specific for the confirmation analysis of 3-phenoxybenzoic acid in mouse plasma. As the method is only used for dose verification of known substances and known nominal concentrations, no additional confirmatory technique is necessary.

Interference No test substance was detected in the mouse plasma control samples with a concentration of ≥ 20% of the calibration solution with 27.1 ng/mL.

Matrix Effects Matrix-matched calibration solutions were used for quantification of 3-phenoxybenzoic acid, hence any potential matrix effects were accounted for by adjusting the matrix in the standard solutions used for calibration.

Limit of Quantification Not applicable and in case of mere positive confirmatory analysis (positive test) not relevant.

Limit of Detection Not applicable and in case of mere positive confirmatory analysis (positive test) not relevant.

Repeatability The relative standard deviation (RSD, %) for the tested concentration level was < 20% for 3-phenoxybenzoic acid.

Reproducibility The average ion ratios in Table 4.1.2-25 are mean values of samples measured at different time points. The data are comparable, so a reproducibility is given.

Conclusion	**The described method is suitable for the confirmation analysis of 3-phenoxybenzoic acid (metabolite of alpha-cypermethrin) in mouse plasma.**

Report:	CA 4.1.2/20 Becker M.,Kamp H., 2015 d 3-Phenoxybenzoic acid (metabolite of BAS 310 I, Alpha-Cypermethrin) - Stability analysis in DMSO (4h) 2015/1029517
Guidelines:	none
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Executive Summary

In the context of toxicological studies, the stability of 3-phenoxybenzoic acid (metabolite of alpha-cypermethrin - BAS 310 I) in the vehicle DMSO was investigated in one stability study.

Standard solutions were stored at room temperature and analyzed at day 0 and after 4 h.

Stability of 3-phenoxybenzoic acid at a nominal concentration of 103.2 mg/L was given over a time period of 4 hours of storage at ambient temperature.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Common name:	3-phenoxybenzoic acid
Batch No.:	AC12251-34
Test substance No.	01/0418-1
Purity:	100%

B. STUDY DESIGN

1. Experimental Conditions

51.6 mg of 3-phenoxybenzoic acid were dissolved in 20 mL DMSO. This solution was further diluted 1:25 with DMSO. The final nominal concentration was 103.2 mg/L.

2. Description of analytical procedures

Analysis was accomplished by HPLC with UV-detection (DAD) at 230 nm. The chromatography was performed on a Kinetex C18 column with a flow rate of 3.0 mL/min and an isocratic mixture (50/50) of acetonitrile/formic acid (1000/1, v/v) and water/formic acid (1000/1, v/v).

Principle of the method:
Samples were diluted completely with acetonitrile using appropriate volumetric flasks to obtain sample solutions with 3-phenoxybenzoic acid concentrations that match the calibration range of 2.04 mg/L to 20.4 mg/L. Adequate dilutions with matrix solutions were performed as necessary. It was ensured, that the test substance was completely dissolved in the solutions.

Validation:

Specificity/Interferences:	The study report provided chromatograms for blank matrix, matrix-matched calibration standard and the stability sample after 4 hours. No interferences seem to occur at the retention time of the analyte.
	The identification and quantitation of 3-phenoxybenzoic acid were based on the selected wavelength at 230 nm and the retention time. Under the described conditions, the method is specific for the determination of 3-phenoxybenzoic acid in DMSO as the method is only used for dose verification of known substances and known nominal concentrations. Hence, no additional confirmatory technique is necessary.
Linearity:	The linearity of the method was investigated at concentrations between 2.04 to 20.4 mg/L (matrix-matched calibration solutions), respectively. Linear correlation coefficient and regression equation were not provided but the calibration curve showed the good linearity of the method in the investigated range (five calibration concentrations were used to demonstrate the linearity).
Recovery/Precision:	Not demonstrated by means of fortification at different levels using different replicates. Only stability data are available. Over the investigated time period of 4 hours, all measured concentrations were in the range between 90% to 110% of the nominal concentrations. The detailed results for both studies are summarized in Table 4.1.2-26.

Limit of quantitation: Not stated but the lowest fortified level of 103.2 mg/L is proposed as limit of quantification.

Conclusion: **The method is suitable for the quantification of alpha-cypermethrin in DMSO; this was confirmed by the stability assessment.**

II. RESULTS AND DISCUSSION

The stability of 3-phenoxybenzoic acid in DMSO was determined in one stability study stored over a time period of 4 hours at room temperature at a nominal concentration of 103.2 mg/L.

Over the investigated time period of 4 hours, all measured concentrations were in the range between 90% to 110% of the nominal concentration. The detailed results are summarized in Table 4.1.2-26.

Table 4.1.2-26: Results for stability analysis of 3-phenoxybenzoic acid in DMSO

Matrix	Time after starting	Target concentration [mg/L]	Concentration found [mg/L]	Nominal concentration [%]
DMSO	0 h	103.2	108.9	105.5
	4 h	103.2	105.1	101.8

III. CONCLUSION

Based on the analytical results it was demonstrated that 3-phenoxybenzoic acid (metabolite of alpha-cypermethrin) was stable in DMSO over a time period of 4 hours stored at room temperature at a nominal concentration of 103.2 mg/L.

Report:	CA 4.1.2/21 [redacted] 1995 a WL85871: 52 week oral (dietary) toxicity study in dogs AL-427-001
Guidelines:	EPA 83-1
GLP:	yes (certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Report:	CA 4.1.2/22
	[redacted] (1993) Validation of analytical method no. 5458 for the analysis of WL85871 in dog dietary formulations
	AL-245-005
Guidelines:	none stated
GLP:	not stated

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Principle of the method

10 g of the dog diet (SDS Type A) were weighed into a screw cap jar and the internal standard butylated hydroxytoluene was added. WL85871 (active substance alpha-cypermethrin) was extracted with acetone from dog diet by shaking the mixture for 1 hour at ambient temperature on a mechanical shaker. After filtration (0.45 µm) an aliquot of the supernatant was analyzed.
Analysis was accomplished by HPLC with spectrophotometric detection at 220 nm. The chromatography was performed on a TSK GEL ODS 80 Tm column with a flow rate of 2.0 mL/min and a mixture of methanol/water (80/20, v/v).

Recovery findings

The described method is suitable to determine residues of WL85871 in dog diet. Samples were spiked with WL85871 at concentration levels of 60 ppm, 120 ppm and 240 ppm and analyzed at the respective dose week. The calculated average values of all samples were in the range between 96.2% and 99.4%. The detailed results are given in Table 4.1.2-27.

Table 4.1.2-27: Results of the analysis of the test formulation WL85871 in dog diet

Matrix	Theoretical concentration [ppm]	Analysis of test diets	Mean found concentration [ppm] [a]	Mean found concentration [b] [%]	Average found concentration [b] [%]	RSD [%]
Dog diet	60	Week 1	60.5	100.8	96.2	4.2
		Week 13	59.5, 60.5	99.2, 100.8		
		Week 26	52.0, 55.6	86.7, 92.7		
		Week 28	58.2, 57.3	97.0, 95.5		
		Week 39	59.2, 58.2	98.7, 97.0		
		Week 52	57.0, 56.8	95.0, 94.7		
	120	Week 1	120	100.0	96.9	3.0
		Week 13	112, 113	93.3, 94.2		
		Week 26	116, 116	96.7, 96.7		
		Week 28	-/-	-/-		
		Week 39	119, 120	99.2, 100.0		
		Week 52	111, 119	92.5, 99.2		
	240	Week 1	234	97.5	99.4	1.8
		Week 13	242, 245	100.8, 102.1		
		Week 26	235, 238	97.9, 99.2		
		Week 28	-/-	-/-		
		Week 39	237, 238	98.8, 99.2		
		Week 52	245, 234	102.1, 97.5		

RSD = Relative standard deviation
[a] Mean of 3 measurements.
[b] Conversion of [ppm] to [%] was calculated using the data originating from the study report and by considering the respective nominal concentration; average concentration and RSD values are based on these calculations.

Specificity The identification and quantification of WL85871 were based on the selected wavelength at 220 nm, the retention time and the use of the internal standard butylated hydroxytoluene. Under the described conditions, the method is specific for the determination of WL85871 in dog diet. As the method is only used for dose verification of known substances and known nominal concentrations, no additional confirmatory technique is necessary.

Interference No test substance was detected in the control diet samples.

Matrix Effects Matrix-matched calibration solutions were used for quantification of WL85871, hence any potentially occurring matrix effects were automatically accounted for as calibration standards and unknown samples contained identical matrix load.

Limit of Quantification The limit of quantification (LOQ) is defined by the lowest fortification level of 60 ppm for WL85871.

Limit of Detection	Good detectability is achieved at a signal to noise ratio of 3:1, which is defined as the limit of detection (LOD).
Repeatability	The relative standard deviation (RSD, %) for all concentration levels were < 20% for WL85871.
Reproducibility	The results in Table 4.1.2-27 are mean values of samples measured at different days. The data are comparable, so a reproducibility is given.
Conclusion	**The described method is suitable for the determination of WL85871 (active substance alpha-cypermethrin) in dog diet.**

Report: CA 4.1.2/23
Done J.N., 1984 a
Establishment of methodology for the analysis of WL 85871 in SDS dog diet (A) and the assessment of the dietary mixing procedures and the stability of WL 85871 in SDS dog diet (A)
AL-245-004
Guidelines: none
GLP: no

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Principle of the method

About 20 g of dog diet samples (SDS dog diet (A)) were weighed in a 250 mL glass screw top jar. 2 mL of the internal standard butylated hydroxytoluene (BHT) in acetone was added and then extracted with 50 mL acetone by mechanical shaking for 1 h at ambient temperature. Samples of the clear supernatant were taken for analysis.
Analysis was accomplished by reverse phase HPLC using UV detection at 220 nm. The chromatography was performed on a column packed with 3 µm ODS Hypersil, using a mixture of methanol/distilled water (70/25, v/v) as mobile phase with a flow rate of 2 mL/min.

Recovery findings

The described method is suitable to determine residues of WL85871 (active substance alpha-cypermethrin) in dog diet. 20 g and 100 g samples formulated with WL85871 at a concentration level of 200 ppm were taken from 3 levels (bottom, middle and top) in the mixing drum and analyzed. The results showed a homogeneous distribution and an overview is given in Table 4.1.2-31.

Additionally, the repeatability of the extraction procedure and the repeatability of the chromatography was tested by analyzing 10 samples from the same extract and this analysis was repeated once. The results showed excellent repeatability in both cases and the detailed results are summarized in Table 4.1.2-32.

Table 4.1.2-28: Results of the analysis of WL85871 in dog diet at 200 ppm

Matrix	Concen-tration [ppm]	Sample origin	Observed concentration [ppm]	Mean observed concentration [ppm]	Mean observed concentration [a] [%]	RSD [a] [%]
Dog diet (20 g samples)	200	bottom	184, 186, 191, 180, 178, 186	184	92	2.5
		middle	187, 205, 189, 187, 182, 186	189	95	4.2
		top	179, 194, 182, 184, 184, 184	185	92	2.7
Dog diet (100 g samples)	200	bottom	197, 195, 202	198	99	1.8
		middle	200, 203, 206	203	102	1.5
		top	204, 209, 209	207	104	1.4

RSD = Relative standard deviation
[a] Conversion of [ppm] to [%] was calculated using the data originating from the study report and by considering a nominal concentration of 200 ppm, RSD values are based on these calculations.

Table 4.1.2-29: Results of the analysis and repeat analysis of 10 samples of the extract from one diet

Matrix	Concen-tration [ppm]	Analysis	Observed concentration [ppm]	Mean observed concentration [ppm]	Mean observed concentration [a] [%]	RSD [a] [%]
Dog diet	200	Analysis of 10 samples	184, 182, 182, 189, 185, 186, 181, 180, 186, 185	184	92	1.5
		Repeat analysis of 10 samples	184, 181, 182, 183, 185, 183, 185, 181, 184, 186	183	92	0.9

RSD = Relative standard deviation
[a] Conversion of [ppm] to [%] was calculated using the data originating from the study report and by considering a nominal concentration of 200 ppm, RSD values are based on these calculations.

Specificity

The identification and quantification of WL85871 were based on the retention time, UV detection at 220 nm and the use of an internal standard. Under the described conditions, the method is specific for the determination of WL85871 in dog diet. As the method is only used for dose verification of known substances and known nominal concentrations, no additional confirmatory technique is necessary.

Interference

Samples of blank diet showed no significant peaks at the retention times of WL85871 or the internal standard butylated hydroxytoluene.

Matrix Effects

Matrix-matched calibration solutions were used for quantification of WL85871, hence potentially occurring matrix effects were accounted for. Furthermore, the control sample / blank analysed did not show any eluting matrix at the elution time of interest.

Stability The diet samples were stored in the dark at room temperature and analyzed after 1, 10 and 21 days. The results showed stability over 10 days of storage, however, after 21 days of storage the results indicated a slight loss of WL85871 (Table 4.1.2-30).

Table 4.1.2-30: Results of the storage stability of WL85871 in dog diet

Matrix	Concen-tration [ppm]	Sample origin	Day of analysis	Observed concentration [ppm]	Mean observed concentration [ppm]	Mean observed concentration [a] [%]
Dog diet	200	bottom	Day 0	197, 195, 202	198	99
			Day 10	198, 206, 197	200	100
			Day 21	191, 189, 190	190	95
		middle	Day 0	200, 203, 206	203	102
			Day 10	200, 196, 198	198	99
			Day 21	194, 193, 182	190	95
		top	Day 0	204, 209, 209	207	104
			Day 10	198, 194, 197	196	98
			Day 21	189, 192, 189	190	95

[a] Conversion of [ppm] to [%] was calculated using the data originating from the study report and by considering a nominal concentration of 200 ppm.

Limit of Quantification The limit of quantification (LOQ) is defined by the used fortification level of 200 ppm for WL85871.

Limit of Detection Good detectability is achieved at a signal to noise ratio of 3:1, which is defined as the limit of detection (LOD).

Repeatability The relative standard deviation (RSD, %) for all concentration levels were < 20% for WL85871.

Reproducibility Reproducibility was not tested within the validation study.

Conclusion **The results show that the analytical methods is accurate and precise. Therefore, the described method is suitable for the determination of WL85871 (active substance alpha-cypermethrin) in dog diet.**

Report: CA 4.1.2/24
[redacted] 1984 a
WL 85871: 13 week oral (dietary) toxicity study in dogs
AL-425-005
Guidelines: none
GLP: yes
(certified by Department of Health and Social Security, London, United Kingdom)

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Remark: For concentration control experiments (see Table 4.1.2-34), the validated method of J.N. Done, 1984 [*AL-245-004*] was used. Under the described conditions, the analytical method was accurate and precise and therefore suitable to determine WL85871 (active substance alpha-cypermethrin) in dog diet.

Principle of the method

Homogeneity
The diet samples (18.7 to 20.0 g) were weighed into a cellulose extraction thimble and extracted with a mixture of hexane/acetone (2/1, v/v) in a Soxhlet extractor for 3 hours. After cooling, the extracts were diluted with 100 mL hexane. A portion of 1 mL of each extract was diluted with hexane to 100 mL.
Analysis was accomplished by gas-liquid chromatography equipped with an electron capture detector (ECD). The chromatography was performed on a glass column (3% (m/m) of OV 17 on 100 to 120 mesh Supelcoport) with nitrogen as carrier gas with a flow rate of 54 mL/min. An external standard solution of 96% pure WL85871 with a concentration of 0.0597 µg/mL was used for calibration.

Recovery findings

Dog diet samples formulated with WL85871 at concentration levels of 30 ppm, 90 ppm and 270 ppm were analyzed in the respective dose week (week 1, 6, 12 and additionally in week 7, 8, 9 at concentration level 30 ppm as reserve). The calculated average values of all samples were in the range between 93.3% and 96.5%. The detailed results are given in Table 4.1.2-31.

Furthermore, to test the homogenous distribution of the test substance in the diet samples, samples at a concentration level of 30 ppm WL85871 were analyzed in diet samples formulated for dose weeks 1, 6 and 12. For this purpose, duplicate samples were taken from different positions (bottom, middle and top) of the mix. The results of the analysis demonstrate that each of the diet samples contained WL85871 at a concentration close to the nominal concentration. Results are presented in Table 4.1.2-32.

Table 4.1.2-31: Results of concentration control of WL85871 in dog diet at concentration levels of 30 ppm, 90 ppm and 270 ppm

Matrix	Nominal concentration [ppm]	Gender male/female	Analysis of test diets [a]	Mean found concentration [b] [ppm]	Mean found concentration [c] [%]	Average found concentration [c] [%]	RSD [c] [%]
Dog diet	30	male	Week 1	29.6	98.7	93.3	7.8
			Week 6	24.7	82.3		
			Week 7	30.2	100.7		
			Week 8	31.1	103.7		
			Week 9	29.1	97.0		
			Week 12	26.3, 28.3 [d]	87.7, 94.3 [d]		
		female	Week 1	28.1	93.7		
			Week 6	23.1	77.0		
			Week 7	27.9	93.0		
			Week 8	29.8	99.3		
			Week 9	28.8	96.0		
			Week 12	26.6, 28.1 [d]	88.7, 93.7 [d]		
	90	male	Week 1	88.9	98.8	96.3	3.8
			Week 6	81.8	90.9		
			Week 12	86.9	96.6		
		female	Week 1	91.1	101.2		
			Week 6	87.3	97.0		
			Week 12	84.1	93.4		
	270	male	Week 1	255	94.4	96.5	2.6
			Week 6	259	95.9		
			Week 12	261	96.7		
		female	Week 1	257	95.2		
			Week 6	258	95.6		
			Week 12	274	101.5		

RSD = Relative standard deviation

[a] Weeks 7, 8 and 9 were used as reserve samples.

[b] Mean of 3 measurements.

[c] Conversion of [ppm] to [%] was calculated using the data originating from the study report and by considering the respective nominal concentration, average concentration and RSD values are based on these calculations.

[d] Analysis was repeated using phenanthrene as internal standard.

Table 4.1.2-32: Results of the analysis of WL85871 in dog diet sampled at different positions in the mix

Matrix	Nominal concen-tration [ppm]	Position of diet sample in the mix	Gender male/female	Analysis of test diets	Mean found concentration [a] [%]	Average found concentration [b] [%]	RSD [b] [%]
Dog diet	30	bottom	male	Week 1	100	102	2.6
				Week 6	99		
				Week 12	101		
			female	Week 1	105		
				Week 6	105		
				Week 12	100		
		middle	male	Week 1	103	100	2.0
				Week 6	99		
				Week 12	99		
			female	Week 1	100		
				Week 6	97		
				Week 12	101		
		top	male	Week 1	101	101	1.2
				Week 6	102		
				Week 12	101		
			female	Week 1	100		
				Week 6	99		
				Week 12	102		

RSD = Relative standard deviation
[a] Mean of duplicates.
[b] Average found concentration and RSD values are based on calculations of the data originating from the study report.

Specificity The identification and quantification of WL85871 were based on the retention time and the use of an external standard. Under the described conditions, the method is specific for the determination of WL85871 in dog diet. As the method is only used for dose verification of known substances and known nominal concentrations, no additional confirmatory technique is necessary.

Interference No test substance or interfering signal was detected in the diet control samples at the specific wavelength and elution time of the analyte.

Limit of Quantification The limit of quantification (LOQ) is defined by the lowest fortification level of 30 ppm for WL85871.

Limit of Detection Good detectability is achieved at a signal to noise ratio of 3:1, which is defined as the limit of detection (LOD).

Repeatability	The relative standard deviation (RSD, %) for all concentration levels were < 20% for WL85871.
Reproducibility	The results in Table 4.1.2-31 and Table 4.1.2-32 are mean values of samples measured at different days. The data are comparable, so a reproducibility is given.
Conclusion	**The described method is suitable for the determination of WL85871 (active substance alpha-cypermethrin) in dog diet.**

Report:	CA 4.1.2/25 [redacted] 1993 a WL85871 (Fastac): An acute oral (gavage) neurotoxicity study in the rat AL-451-004
Guidelines:	none
GLP:	yes (certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Report:	CA 4.1.2/26 N.N. Except of raw data supporting analytical methods used in toxicological studies 2016/1232572
Guidelines:	not stated
GLP:	not stated

Report:	CA 4.1.2/27 N.N. Sittingbourne analytical method series determination of Fastac and the ratio of enantiomer pairs in technical material and formulated products liquid chromatographic method AL-210-001
Guidelines:	not stated
GLP:	not stated

An overview of the analytical procedure given below is done under the unique document identifier of the original toxicological study.

Principle of the method

The test substance FASTAC (active substance alpha-cypermethrin) was prepared by weighing a defined amount of the test substance in corn oil and diluting to the required volume. Solutions containing 0 and 0.6% m/v were supplied.
For stability experiments, 250 mL of a 0.6% m/v solution was prepared. The solution was divided into four 50 mL samples for dosing and one separate sample for analysis. In addition, 4 x 50 mL corn oil samples were used for control dosing. The samples were stored at room temperature in der dark.
For concentration control, 250 mL of a 0.6% m/v solution was prepared and divided as described above for stability experiments.
Analysis was accomplished using analytical method SAMS 490 (AL-210-001) by HPLC with ultraviolet absorption detection at 230 nm. HPLC analysis was done using a packed Spherisorb CN column equipped with a guard column and a mobile phase of n-hexane and MTBE (99.5 / 0.5 v/v) at a flow rate 3.0 mL/min at 22°C.

Recovery findings

The described method is suitable as concentration control for the determination of WL85871 (active substance alpha-cypermethrin) in solutions prepared in corn oil. Samples dosed with WL85871 at a concentration level of 6 mg/mL WL85871 were analyzed; two as received and two as pre-warmed samples. The mean of all analyzed samples was 90.2%, which is acceptable within the ±10% tolerance. The detailed results are given in Table 4.1.2-33.

Table 4.1.2-33: Results of concentration control of WL85871 in corn oil

Matrix	Sample set	Concentration [%]	Mean concentration [%]	Average concentration [%]	RSD [a] [%]
Corn oil	received	89.7, 89.7	89.7	90.2	1.0
	warmed	89.8, 91.6	90.7		

RSD = Relative standard deviation
[a] RSD value is based on calculations using data originating from the study report

Linearity Linearity is not stated in the report itself; however, calibration curves from the original raw data assessing the linearity of the detector response versus concentration (n=5 with single injection) confirmed good correlation, although the regression factor was not given in the raw data.

Specificity The identification and quantification of WL85871 were based on the analysis using HPLC with ultraviolet absorption detection (230 nm). Under the described conditions, the method is specific for the determination of WL85871 in corn oil. As the method is only used for dose verification of known substances and known nominal concentrations, no additional confirmatory technique is necessary.

Interference No test substance was detected in the corn oil control samples (< 0.003 mg/mL).

Matrix Effects No interference at the detection wavelength and retention times of interest were reported.

Limit of Quantification As the LOD is defined as at least 30% of the LOQ, the proposed LOQ of this method is 1.7 mg/mL. However, as no solid samples are extracted, but formulations were analyzed after dilution, no fortification experiments were conducted as not required.

Limit of Detection Good detectability is achieved at a signal to noise ratio of 3:1, which is defined as the limit of detection (LOD). The lowest standard was 0.5 mg/mL

Repeatability The relative standard deviation (RSD, %) for the tested concentration level was < 20% for WL85871.

Reproducibility Reproducibility of the method was not determined within the study.

Stability Data from the stability analysis indicated that the formulations were stable for four days and furthermore, that the isomerization ratio (cis I to cis II) in the formulations remained constant over the storage period. Results are presented in Table 4.1.2-34.

Table 4.1.2-34: Results of concentration control of WL85871 in corn oil

Matrix	Date of analysis	alpha-cypermethrin [mg/mL]	Isomer ratio	
			CIS I	CIS II
Corn oil	0 d	6.1	3	97
	4 d	5.9	3	97

Conclusion **The described method is suitable for the determination of WL85871 (active substance alpha-cypermethrin) in corn oil.**

(d)Methods in body fluids, air and any additional matrices used in support of operator, worker, resident and bystander exposure studies

No stand-alone validated analytical methods for the determination of alpha-cypermethrin were required for exposure studies.

(e)Methods in or on plants, plant products, processed food commodities, food of plant and animal origin, feed and any additional matrices used in support of residues studies

The following methods cover the compounds necessary for the discussion on the residue definition for risk assessment for products of plant and animal origin as summarized in Document N, chapter 7.3 and discussed in CA 6.7:

Plant: BAS 310 I [alpha-cypermethrin, Reg. No. 4078193]

Animal:
in milk and kidney: BAS 310 I [alpha-cypermethrin, Reg. No. 4078193]

in other animal matrices: BAS 310 I [alpha-cypermethrin, Reg. No. 4078193]

Plant

Analytical methods for the determination of alpha-cypermethrin residues in plant matrices were evaluated in the context of the inclusion in Annex I of Directive 91/414/EEC. Methods evaluated previously are listed in Table **4.1.2-35** and shortly summarized below. Methods not yet evaluated are summarized in detail in the following chapter.

Table 4.1.2-35: Summary of peer-reviewed analytical methods for determination of alpha-cypermethrin residues in plant matrices

Method No.	Matrix	Method principle	Target analytes	LOQ mg/kg	year	DocID	EU reviewed
SAMS 383-1	Crops	GC-ECD and GC-MS	Cypermethrin	0.01	1983	1983/7001664	Yes
SAMS 233-1 and SAMS 234-1	Crops (high water)	GC-ECD	Cypermethrin	0.01	1977 and 1979	1979/7000414, 1977/7000275	Yes
AGROLABSOP-RAD.M004	Olive oil	GC-ECD	Alpha-cypermethrin	0.02	1998	AL-714-002	Yes
FAMS 045-02	Savoy cabbage, leek, wheat grain	GC-MS	Cypermethrin	0.01	1995	AL-123-08	Yes
FAMS 066-01	Cereal grain and straw	GC-MS	Alpha-cypermethrin	0.01	1996	AL-244-006	Yes
RLA 12513.03.V (extraction) RLA 12644.01.V (determination)	cabbage, carrots, lettuce	GC/MS	cypermethrin isomers	0.01	2002 and 1998	AL-244-012 and AL-240-001	Yes
RLA 12644.00 RLA 126644.01V	cabbage, oilseed rape, barley grain, grapes	GC/MS	Alpha-cypermethrin	0.01	2001	AL-244-010	Yes
SAMS 351-02	Dry, oily, high water-content crops	GC/ECD and GC/MS (confirmation)	Alpha-cypermethrin	0.01	1989	AL-244-001	Yes
SAMS 320-2	Plant matrices (general)	HPLC/UV or GC/ECD after derivatization; GC/MS or TLC/GC for confirmation	DCVA	0.05	Issued 1982	AL-230-001 (1983)	Yes
SAMS 295-2	Plant matrices (general)	HPLC/UV, HPLC/UV or CS/MS after derivatization for confirmation	3-PBA	0.05	Issued 1981	AL-230-001 (1983)	Yes
RLA 12644	Wheat grain and straw	GC/MS	Cypermethrin isomers	0.01	2002	AL-244-011	Yes
RLA 12594.01	Olives and olive oil	GC/ECD; GC-MS for confirmation	Alpha-cypermethrin	0.05	2000	AL-244-007	Yes
DFG-S19	Grapes, wheat grain, cabbage, oilseed rape	GC/ECD	Alpha-cypermethrin	0.01	2000	AL-244-008	Yes
DFG-S19	Cabbage leaves, boiled cabbage leaves, sauerkraut, oilseed rape press cake, refined rapeseed oil	GC/ECD	Alpha-cypermethrin	0.01	2000	AL-244-009	Yes

DCVA: 2,2-dimethyl-3-(2,2-dichlorovinyl)cyclopropane carboxylic acid (WL 44776)
3-PBA: 3-phenoxybenzoic acid (WL 44607)

For completeness of information, summaries of already EU-peer reviewed analytical methods are presented for completeness of information.

Report:	CA 4.1.2/28 Anonymous, 1983 Determination of residues of Ripcord (WL 43467) in crops - Gas chromatographic method 1983/7001664
Guidelines:	not stated
GLP:	not stated

Method SAMS 383-1

Principle of the method

For analytical method SAMS 383-1, samples of homogenized crop are extracted and partitioned at the same time by macerating with a mixture of aqueous acetonitrile (1:1, v/v) and re-distilled petroleum spirit. The mixture is either allowed to stand (straw, fruit) or centrifuged (grain) to obtain phase separation, depending on the matrix. The organic phase is water washed, dried with sodium sulphate and cleaned up using a pre-washed Bond Elut-CN cartridge. Final determination is performed with GC on a glass column (1.4 m × 3.0 mm) with OV 201 2% (m/m) or polar phases such as GEXE60, OV225 and Ultrabond 20 M as stationary phase and nitrogen as mobile phase. An electron capture (EC) detector (^{63}Ni source) is used. GC-MS determination is performed on a glass column (1.5 m × 2.0 mm) with SE 30 1% (m/m) as stationary phase and helium as mobile phase. Mass spectrometric detection is performed in the negative ionization mode with m/z 207 and 209.

Recovery findings	Not reported.
Linearity	Not reported.
Specificity	The signal to noise ratio is 5:1. For confirmation, GC-MS determination can be performed on a glass column (1.5 m × 2.0 mm) with SE 30 1% (m/m) as stationary phase and helium as mobile phase. Mass spectrometric detection is performed after ionization (negative mode) with m/z 207 and 209.
Limit of Quantification	The limit of quantification for cypermethrin (RIPCORD; WL 43467) is 0.01 mg/kg for all investigated plant matrices.
Repeatability	The repeatability of this method was found to be 0.17 for milled grain containing 0.8 mg/kg cypermethrin (22 degrees of freedom) and 0.09 for milled grain containing 1.5 mg/kg alpha-cypermethrin (20 degrees of freedom).
Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.

Standard stability	Solutions of the test substance in hexane can be stored for at least three months at ambient temperature in the dark.
Conclusion	**The information given for the method number SAMS 383-1 indicates its suitability to determine cypermethrin residues in grain and other plant matrices.**

Report:	CA 4.1.2/29 Anonymous, 1977 Determination of residues of WL 43479 in crops 1977/7000275
Guidelines:	not stated
GLP:	not stated

Report:	CA 4.1.2/30 Cole E.R., Woodbridge A.P, 1979 The development of methods for the analysis of crops and soil for residues of WL 43467 (Ripcord) and WL 43479 (Talcord) 1979/7000414
Guidelines:	not stated
GLP:	not stated

Methods SAMS 233-1 and SAMS 234-1

Only data on cypermethrin (WL 43467; RIPCORD) are summarized below.

Principle of the method

Chopped or ground samples of aqueous based and non-oily crops (apples, grapes and maize grain and silage, lemon and kale) are extracted with a mixture of acetone and petroleum spirit (1:1, v/v). Extracts are washed with water to remove the acetone and are cleaned up by column chromatography on Florisil. Final analysis is performed with GC with electron capture (EC) detection. One of the following stationary phases has been used: OV-101, SE-30, Apiezon L, Oronite polybutene, DC-200 (grapes), OV-17, Reoplex 400, OV-210, OV-225 (other crops), Apolar 9CP or OV-275.

Extraction efficiency: Radiolabelled extractability studies and extraction of stored and aged samples carried out with cypermethrin (WL 43467, BAS 311 I) have shown the mixture of acetone and petroleum spirit to be an efficient solvent for the extraction of pyrethroid residues.

Recovery findings

The overall average recovery was 109%.There were no residues detected in the control samples above the limit of quantification (0.01 mg/kg). Individual recoveries ranged from 90 to 120%. Average recoveries for each matrix are summarised in Table **4.1.2-36**.

Table 4.1.2-36: Results of method validation: BAS 311 I (cypermethrin) in apple, maize, grape, lemon and kale specimens

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Apple	BAS 311 I	0.1	2	110	N/A	N/A
Maize grain	BAS 311 I	0.1	1	120	N/A	N/A
Maize silage	BAS 311 I	0.1	1	120	N/A	N/A
Grape	BAS 311 I	0.1	1	100	N/A	N/A
Lemon	BAS 311 I	0.1	1	90	N/A	N/A
Kale	BAS 311 I	0.1	1	110	N/A	N/A
Overall mean			**7**	**109**	**11**	**9.8**

Linearity	No data on linearity are presented in the report.
Specificity	Residues are confirmed by TLC on silica gel F_{254} plates followed by GC or GC-MS operating in the EI mode with multiple ion monitoring; ions m/z 163, 181 and 415 were monitored. Helium was used as carrier gas. No interference from co-extractives is usually observed.
Limit of Quantification	The limit of quantification (LOQ) for cypermethrin is 0.01 mg/kg for all investigated plant matrices.
Repeatability	The overall relative standard deviation (RSD, %) was below 20%. The detailed values are shown in Table **4.1.2-36**.
Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The method SAMS 233-1 is considered to be fit for purpose for the determination of residues of cypermethrin in apple, maize, grape, lemon and kale.**

Report:	CA 4.1.2/31 Klitsinaris A., 2000 Alphacypermethrin (AL 900049) 100 g ai/L OESC: Decline curve residue study on Alphacypermethrin in olives and olive oil - Hellas 1998 AL-714-002
Guidelines:	EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8),EEC 96/68,EEC 7029/VI/95 rev. 5
GLP:	not stated

Method AGROLABSOP-RAD.M004

Principle of the method

Olive oil is cleaned using gel permeation chromatography for the removal of lipids and other macromolecules. The fraction containing alpha-cypermethrin is collected and dried. The residue is diluted in n-hexane and further cleaned up through a Florisil column. Quantification is performed by GC-ECD.

Recovery findings

Average recoveries for all fortification levels were between 70 and 110%. Average recoveries for each matrix are summarised in Table **4.1.2-37**.

Table 4.1.2-37: Results of method validation: BAS 310 I (alpha-cypermethrin) in olive oil specimens

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Olive oil	BAS 310 I	0.020	3	82.1	3.2	3.9
		0.200	3	112.7	13.1	11.6
		1.000	2	97.6	2.8	2.8
Overall mean			**8**	**97.4**	**15.9**	**16.3**

Linearity	Linearity was tested using six calibration standards in the range of 0.002-0.080 mg/kg.
Specificity	A confirmatory method has not been performed; however, no interferences of matrix or labware have been reported. Good validation results have been achieved with the primary method.
Limit of Quantification	The limit of quantification (LOQ) for BAS 310 I is 0.02 mg/kg for olive oil.
Repeatability	The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-37**.

Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The method AGROLABSOP-RAD.M.004 is considered to be fit for purpose for the determination of residues of alpha-cypermethrin in olive oil.**

Report:	CA 4.1.2/32 Memmesheimer H., 1997 Alphacypermethrin (CL 900049): Analytical method for the determination of residues in savoy cabbage, leek and wheat grain AL-123-088
Guidelines:	not stated
GLP:	not stated

Method FAMS 045-02

Principle of the method

Alpha-cypermethrin is extracted from the sample with hexane/acetone (8:2, v/v). This is followed by liquid/liquid partitioning with water. The extract is evaporated to dryness. Further clean-up is achieved alternatively with solid phase extraction (savoy cabbage, leek) or gel permeation chromatography (wheat grain). Final analysis is performed by capillary gas chromatography with either a nitrogen/phosphorous (GC-NPD) or a mass selective detector (GC-MS) on a fused silica capillary column with methyl silicone as liquid phase and helium as carrier gas at a flow rate of 3.5 (NPD) or 0.85 mL/min (MSD). Mass spectrometric detection is performed after electron impact ionization (SIM mode) with m/z 165.

Recovery findings

Average recoveries for all fortification levels were between 70 and 110% using both the primary (GC-NPD) and the confirmatory method (GC-MS) for detection. Average recoveries for each matrix are summarized in Table **4.1.2-38** and Table **4.1.2-39**.

Table 4.1.2-38: Results of method validation: BAS 310 I (alpha-cypermethrin) in plant specimens – GC-NPD

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Savoy cabbage	BAS 310 I	Overall (0.01-0.5)	not reported	92	n.r.	7
Leek	BAS 310 I	Overall (0.01-0.5)	not reported	94	n.r.	6
Wheat grain	BAS 310 I	Overall (0.01-1.0)	not reported	85	n.r.	15

Table 4.1.2-39: Results of method validation: BAS 310 I (alpha-cypermethrin) in plant specimens – GC-MS

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Savoy cabbage	BAS 310 I	Overall (0.01-0.5)	not reported	91	n.r.	7
Leek	BAS 310 I	Overall (0.01-0.5)	not reported	93	n.r.	11
Wheat grain	BAS 310 I	Overall (0.01-1.0)	not reported	85	n.r.	3

Linearity Linearity was tested using calibration standards in the range of 0.05-2.0 µg/mL. Standard solutions are prepared in cyclohexanone.

Specificity For confirmation, either GC-NPD or GC-MS determination can be performed. No significant blank signals or unspecific interferences in untreated control samples were observed. The limit of detection is ≤50% LOQ (0.005 mg/kg).

Limit of Quantification The limit of quantification (LOQ) for alpha-cypermethrin is 0.01 mg/kg for all matrices tested.

Repeatability The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-38** and Table **4.1.2-39**.

Reproducibility Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.

Extract stability Stability of extracts was not determined within this validation study.

Standard stability Stability of standards was not determined within this validation study.

Conclusion **The method FAMS 045-02 is considered to be fit for purpose for the determination of residues of alpha-cypermethrin in cabbage, leek and wheat grain.**

Report:	CA 4.1.2/33 Memmesheimer H., 1996 Alphacypermethrin (CL900049): Validation of method FAMS 066-01 for the determination of CL 900049 residues in cereal grain and straw (Germany, 1996)
	AL-244-006
Guidelines:	IVA-Leitlinie Rueckstandsversuche Teil I (1992),BBA Guideline for the evaluation of pesticides for registration of a pesticide (Ribbesbuettel 2nd edition 1990),DFG Method Series for Pesticide Residue Analysis V and VIII (1991)
GLP:	Yes (Ministerium fuer Arbeit, Soziales, Familie und Gesundheit, Mainz, Germany)

Method FAMS 066-01

Principle of the method

For analytical method FAMS 066-01, samples of homogenized wheat grain and straw are extracted with hexane/acetone (8:2, v/v) and centrifuged. For clean-up the extracts were partitioned with water and the organic phase was reduced almost to dryness. The residue was dissolved in acidic methanol. Further clean-up was carried out by gel permeation chromatography. Final determination is performed with GC-NPD on a fused silica capillary column (12 m × 0.32 mm) with methyl silicone as liquid phase and helium as carrier gas at a flow rate of 3.5 mL/min.

Recovery findings

Average recoveries for all fortification levels were between 70 and 110% using both the primary (GC-NPD) and the confirmatory method (GC-MS) for detection. Average recoveries for each matrix are summarized in Table **4.1.2-40** and Table **4.1.2-41**.

Table 4.1.2-40: Results of method validation: BAS 310 I (alpha-cypermethrin) in wheat grain and straw specimens – GC-NPD

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Wheat grain	BAS 310 I	0.01	3	94	1.2	1.2
		0.10	3	74	2.1	2.8
		1.0	3	80	3.2	4.0
		Overall	9	82	9.1	11.1
Wheat straw	BAS 310 I	0.03	3	96	2.5	2.6
		0.30	3	82	2.0	2.4
		3.0	3	80	2.6	3.3
		Overall	9	86	8.0	9.3

Table 4.1.2-41: Results of method validation: BAS 310 I (alpha-cypermethrin) in wheat grain and straw specimens – GC-MS

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Wheat grain	BAS 310 I	0.01	3	93	1.2	1.2
		0.10	3	79	2.5	3.2
		1.0	3	76	1.0	1.3
		Overall	9	83	8.1	9.8
Wheat straw	BAS 310 I	0.03	3	100	5.7	5.7
		0.30	3	90	4.2	4.6
		3.0	3	77	2.6	3.4
		Overall	9	89	10.5	11.9

Linearity Linearity was tested using seven calibration standards in the range of 0.2-2 µg/mL. Calibration curves gave acceptable correlation coefficients ≥ 0.99. For grain and straw, cubic and linear curve fit were applied, respectively. Standard solutions are prepared in cyclohexanone.

Specificity For confirmation, GC-MS determination can be performed on a fused silica capillary column (12 m × 0.2 mm) with methyl silicone as liquid phase and helium as mobile phase at a flow rate of 0.85 mL/min. Mass spectrometric detection is performed after electron impact ionization (SIM mode) with m/z 165. No significant blank signals or unspecific interferences in untreated control samples were observed. The limit of detection is ≤30% LOQ (0.003 mg/kg for grain and 0.01 mg/kg for straw).

Limit of Quantification The limit of quantification (LOQ) for alpha-cypermethrin is 0.01 mg/kg for wheat grain and 0.03 mg/kg for wheat straw.

Repeatability The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-40** and Table **4.1.2-41**.

Reproducibility Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.

Extract stability Stability of extracts was not determined within this validation study.

Standard stability Stability of standards was not determined within this validation study.

Conclusion **The method FAMS 066-01 is considered to be fit for purpose for the determination of residues of alpha-cypermethrin in wheat grain and straw.**

Report:	CA 4.1.2/34 Atkinson S., 2002 Validation of the analytical procedure carried out for the analysis of Cypermethrin isomers in cabbage, carrots and lettuce carried out in study 83265
	AL-244-012
Guidelines:	none
GLP:	Yes (certified by Dempartment of Health of the Government of the United Kingdom, United Kingdom

Report:	CA 4.1.2/35 Cuthbert M., 1998 Analysis of Alphacypermethrin (AC 900049) in vining peas, field beans, strawberries, cherries and green beans
	AL-240-001
Guidelines:	none
GLP	not stated

Remark: Methods 12513.03.V, RLA 12644.01V are part of the document AL-244-012 which was submitted as part of the Addendum 2002. The data were initially not reported in the section of the analytical methods of the addendum 2002 because part of the residue study 83265 (CEMS-1691). In order to be complete, validation data are therefore presented here below.

The objective of the study was to demonstrate that the analytical procedure used in residue study 83265 may be used for quantitative analysis of cypermethrin isomers and to demonstrate the stability of the cis-2 isomer (alpha-cypermethrin) in crops.

Principle of the method:
The cabbage, carrot and lettuce specimens were extracted according to the analytical procedure described by BASF Agro Reasearch Procedure RLA 12513.03V “Analysis of Alphacypermethrin (AC 900049) in Vining Peas, Field Beans, Strawberries, Cherries and Green Beans”.
A 20g specimen was homogenized in 50/50 heptane/acetone, filtered and transferred to a round bottomed flask and evaporated to about 20mL. The extract was transferred to a separating funnel and water and heptane added. The specimen was partitioned into the heptane by inverting the funnel. The aqueous layer was removed. The heptane layer was portioned twice with water. The heptane was then collected through anhydrous sodium sulphate into a volumetric flask and made up to volume. An aliquot of the heptane was subjected to further clean up using silica solid phase extraction cartridges. All specimen matrices were analysed using conditions in the analytical procedure RLA 12644.01V. Final determination was by gas liquid chromatography with spectrometric detection. Detailed conditions are described in AL-240-001.

Specificity/Interference: Not demonstrated within the study report. However, GC-MS (RLS 12644.01V) is known to be a highly specific method by the possibility to monitor three m/z ions (see AL-244-009). Validation data were provided for the m/z ion 181.
Untreated control samples exhibit no significant interferences at retention time of the analytes (chromatograms were provided for all the analytes for matrix-matched standards, controls and fortified samples at LOQ and 10 × LOQ for the m/z ion 181).

Linearity: Typical results for a matrix matched calibration were provided for each analyte without indicating if it concerned calibration in cabbage, lettuce or carrots. The linearity response during the validation procedure was acceptable with a correlation coefficient > 0.995 for all analysis determination of the provided example. Seven matrix matched batch standard solutions over the range 0.005 – 0.15 µg/mL were injected. Regression equations and calibration curves were provided.

Recovery/Precision: see Table **4.1.2-42** and Table **4.1.2-43**

Limit of Quantification: 0.01mg/kg for each of the 4 isomers of cypermethrin in all matrices.

Table 4.1.2-42: Validation data (Atkinson, 2002)

Fortification Levels mg/kg	Cis I		Cis II		Trans III		Trans IV		Total isomers	
	Mean recovery %	% RSD	Mean recovery %	% RSD	Mean recovery %	% RSD	Mean recovery %	% RSD	Mean recovery %	% RSD
Cabbage										
0.01	77	13.3	82	7.3	81	10.4	75	16.2	79	8.4
0.1	70	10.9	77	10.6	74	9.8	71	10.9	73	10.7
Carrots										
0.01	70	4.4	67	5.9	78	4.9	87	3.3	75	2.6
0.1	91	7.9	99	8.5	96	9.8	89	8.1	94	8.1
Lettuce										
0.01	83	10.5	73	7.7	79	14.8	92	9.9	82	8.5
0.1	78	5.5	80	3.6	79	6.2	82	5.6	80	5.0

Note: fortification performed with mixed isomers.
n = 5 replicates at each fortification level and for each analyte.

Table 4.1.2-43: Data from method check (Grolleau 2002)

Fortification Levels mg/kg	Cis I		Cis II		Trans III		Trans IV	
	Reco-very %**	Reco-very %***	Mean recovery %**	Reco-very %***	Mean recovery %**	Reco-very %***	Mean recovery %**	Reco- very %***
Cabbage								
0.01	82, 73	73	74, 73	72	78, 76	71	70, 70	58*
0.1	110, 103	-	111, 103	-	111, 102	-	108, 103	-
Carrots								
0.01	78, 80	82	72, 73	75	73, 70	81	74, 68	70
0.1	100, 97	-	106, 98	-	91, 84	-	89, 86	-
Lettuce								
0.01	77, 71	75	71, 70	76	76, 80	78	70, 69	65*
0.1	93, 99	-	95, 102	-	96, 102	-	104, 108	-
Wheat whole plant								
0.01	99, 96	96	108, 104	95	97, 100	98	101, 101	101
0.1	108, 113	-	111, 120	-	110, 108	-	113, 120	-
Wheat straw								
0.01	78, 80	102, 109	78, 81	97, 104	84, 83	99, 100	77, 77	106, 97
0.1	94, 89	-	98, 88	-	99, 87	-	96, 90	-
Wheat grain								
0.01	82, 79	69*	89, 80	71	77, 70	72	80, 70	70
0.1	102, 105	-	110, 108	-	110, 100	-	110, 109	-

* A reference item solution of 0.005 µg/mL in matrix (equivalent to 0.005 mg/kg) was analysed with this batch thus demonstrating that this lower level is detectable. Therefore a recovery value of less than 70% has been accepted.
** method check
*** recovery data from procedural recoveries.

Conclusion: **The method is suitable to quantify the cypermethrin isomers at a limit of quantification of 0.01 mg/kg in crop matrices. The method has been used in support of a rotational crop study.**

Report:	CA 4.1.2/36 Young H., 2001 Confirmatory analysis of BAS 310 I (Alphacypermethrin) in cabbage, oilseed rape, barley grain and grapes using RLA 12644
	AL-244-010
Guidelines:	none
GLP:	Yes (certified by Dempartment of Health of the Government of the United Kingdom, United Kingdom

Principle of the method (RLA 12644.00 and RLA 12644.01 V issued method at the completion of the validation (RLA 12644.01V supersedes RLA 12644.00)):
Qualitative method RLA 12644 is used to confirm the specificity of method RLA 12642 (= DFG Method S19 (extended revision) – module D1) which was initially proposed for enforcement (cfr. addendum 1 dd. January 2001). The procedures for extraction and clean-up are the same as those outlined in RLA 12642, but an alternative detection method is used:

- extraction according to module E1 (grapes, cabbage) or E2 (grain) or E7 (oilseed rape)
- cleanup of extract using gel permeation chromatography (module GPC)
- supplemental cleanup using silica gel column chromatography (module C1)
- determination by capillary GC (Phenomenex Zebron ZB1; 30 m x 0.25 mm i.d.; 0.25 µ m) with MSD in SIM mode (monitoring ions m/z 163, 165 and 181)

Specificity/Interferences:	Specificity of method RLA 12642 (GC-ECD) is demonstrated by using GC-MS as an alternative method of detection. Samples were analysed in scan mode to obtain the retention time of the analyte and mass spectrum. The mass spectrum (full scan) was used to select the monitoring ions m/z 163, 165 and 181 and the resulting TIC was integrated. Untreated control samples exhibit no significant interferences at retention time of alphacypermethrin (control values < 20% of LOQ) (chromatograms were provided for matrix-matched standards, controls and fortified samples).
Linearity:	The response of the GC-MS system (= peak area) to alphacypermethrin was demonstrated to be linear over a concentration range from 0.01 to 0.1 µ g/mL (n = 5); r > 0.995 in all cases (matrix-matched calibration standards).
Recovery/Precision:	see Table **4.1.2-44**
Limit of Quantification:	0.01 mg/kg

Table 4.1.2-44: Validation of confirmatory method RLA 12644 (Young, 2001)

Matrix	Analyte	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
cabbage	Alpha-cypermethrin	0.01	3	72 – 103	86	18.4
oilseed rape		0.01	3	78 – 97	85	12.6
barley grain		0.01	3	65 – 80	71	11.2
grapes		0.01	3	90 – 94	92	2.3

Note: fortification with alpha-cypermethrin

Conclusion: **Method RLA 12644 (GC-MS) is suitable as confirmatory method to DFG Multi-residue Method S19 (extended revision) (GC-ECD) for determination of alphacypermethrin residues in plant matrices.**

Report:	CA 4.1.2/37 Author not specified, 1989 Determination of residues of Alphacypermethrin in crops - Gas chromatographic method AL-244-001
Guidelines:	none
GLP:	not stated

Report:	CA 4.1.2/38 Furr H., 1989 Certification of analysis: The determination of Fastac (WL 85871) residues in oilseed rape seed 1989/5000067
Guidelines:	none
GLP:	not stated

Principle of the method:
Crop samples are mixed with anhydrous sodium sulphate and extracted by homogenising with acetone/hexane (1:1 v/v). Extracts are washed with water to remove the acetone and in the case of oily crops (e.g. linseed, soybean, cotton seed and tobacco) the extracts are cleaned up by partitioning between hexane and water/acetonitrile (1:9 v/v). All extracts are cleaned up by liquid-solid chromatography using Florisil. Alpha-cypermethrin is determined by packed column GC (GE-XE 60 2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD); quantification by external standardization. Residues of alpha-cypermethrin are confirmed by capillary GC-ECD (SE-54; 0.5 µm) and GC-MS (SE-30 1% (m/m) on Gas Chrom Q (100-120 mesh)), MS operated in negative ion chemical ionisation mode (ions monitored: m/e 207 and 209).

Remark: SAMS 351-01 was also used in support of residue trials. Methods SAM 351-01 and SAM 351-02 do not differ (extraction procedure and analytical conditions). Description of method SAMS 351-01 is given in **DocID 1989/5000067** (Jones 1989 - *The determination of FASTAC (WL 85871) residues in oilseed rape seed* – Hazleton UK – HUK Project No. 717/1) submitted in the course of the assessment. Only slight differences in the chromatographic conditions (incl. packed column) occur.

Specificity/Interferences:	Typical packed column GC-ECD chromatograms are shown. Most control samples exhibit no major interfering peaks at the retention time of alpha-cypermethrin; control and blank data are < 0.01 mg/kg.
Linearity:	Not reported in this report. However, linearity is reported in DocID 1989/5000067 for analysis in oil seed rape seed (SAMS 351- 01). Calibration occurred solvent in the 0.002 – 0.1 µ g/mL (n = 5) and was linear (r = 0.9998, calibration curve and equation provided).
Recovery/Precision:	see Table **4.1.2-45**
Limit of Quantification:	The lowest fortified level is 0.05 mg/kg.

Table 4.1.2-45: Validation of method SAMS 351-02 (no author specified, 1989)

Matrix	Analyte	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
Apples (high water)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	95 - 110 95 - 105 90 - 110	102.5 100 100	(10.4) (7.1) (14.1)
		0.05 - 0.50	6	90 - 110	100.8	8.5
Wheat (dry – high starch)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	90 - 95 90 - 90 100 - 100	92.5 90 100	(3.8) (0.0) (0.0)
		0.05 - 0.50	6	90 - 100	94.2	5.2
Cabbage (high water)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	95 - 95 95 - 100 110 - 110	95.0 97.5 110	(0.0) (3.6) (0.0)
		0.05 - 0.50	6	95 - 110	100.8	7.3
Soybean (oily)	alpha-cypermethrin	0.05 0.20 0.50	2 2 2	85 - 100 75 - 90 85 - 95	92.5 82.5 90	(11.5) (12.9) (7.9)
		0.05 - 0.50	6	75 - 100	88.3	9.9
overall	alpha-cypermethrin	0.05 0.20 0.50	8 8 8	85 - 110 75 - 105 85 - 110	95.6 92.5 100	7.6 9.6 9.6
		0.05 - 0.50	24	75 - 110	96.0	9.2

Conclusion: **In terms of interfering blanks, accuracy (mean recoveries between 70 and 110%) and precision (RSD's lower than 20%), the method is suitable for alpha-cypermethrin residue analysis in dry, oily and aqueous based crops with a LOQ of 0.05 mg/kg.**

Report:	CA 4.1.2/1 Hitchings E., 1983 The development of methods for the determination of Ripcord - WL 44776 and WL 44607 in crops and soil AL-230-001
Guidelines:	none
GLP:	not stated

Methods SAMS 320-02 and SAMS 295-02

Principle of the method: The methods for both acid metabolites (WL 44776 = 2,2-dimethyl-3-(2,2- dichlorovinyl)cyclopropane carboxylic acid; WL 44607 = 3-phenoxybenzoic acid) are essentially the same.
Aqueous and dry crops are extracted by homogenizing with a mixture of water/acetonitrile to give a final solvent ratio of 30:70 v/v. The extract is filtered, after which an aliquot of the filtrate is diluted with water and partitioned under acidic conditions with a diethylether/petroleum spirit mixture (1:1 v/v), to extract any free WL 44776 (resp. WL 44607) present.
Oily crops are extracted by homogenizing with acetonitrile, after which the extract is filtered. An aliquot of the filtrate is concentrated to low volume, diluted with aqueous sodium carbonate and partitioned with petroleum spirit to remove oil and any parent pyrethroid which may be present. The resulting aqueous extract is diluted further with aqueous sodium chloride solution and is partitioned with diethyl ether/petroleum spirit (1:1 v/v) to remove any remaining traces of oil and other co-extractives. After acidification of the aqueous phase, free WL 44776 (resp. WL 44607) is extracted from the aqueous phase with diethyl ether/petroleum spirit (1:1 v/v).
Conjugated WL 44776 (resp. WL 44607) is hydrolyzed by concentrating the remaining aqueous phase under alkaline conditions and subsequently treating it with HCl. Any WL 44776 (resp. WL 44607) released is partitioned into diethyl ether/petroleum spirit (1:1 v/v). Residues of WL 44776 (resp. WL 44607) in the “free acid” or “hydrolyzed” extracts are cleaned up by normal phase HPLC (LiChrosorb Diol (10 µm); isocratic elution) with UV detection at 225 nm, after which WL 44776 (resp. WL 44607) is determined by reversed phase HPLC (Spherisorb S5 ODS (5 µm); isocratic elution) with UV detection at 216 nm; quantification by external standardization.
If the extracts are not sufficiently clean for satisfactory analysis to be carried out, the acid WL 44776 (as obtained after normal phase HPLC) may be converted into the parent WL 43467 (cypermethrin) by reaction with α-cyano-3-phenoxybenzyl bromide, after which the derivative is cleaned up using a Porasil column. Analysis is then carried out using GC (GE-XE 60 1.2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD).
The acid WL 44607 (as obtained after normal phase HPLC) may be converted into its methyl ester, after which the derivative is cleaned up by partition with petroleum spirit. Analysis is then carried out by either reversed phase

HPLC (ODS-Hypersil (5 µm); isocratic elution) with UV detection at 212 nm or GC-MS (SE-30 2% (m/m) on Gas Chrom Q (100-120 mesh)) with multiple ion monitoring (MS operated in EI mode, ions monitored : m/e 196 and 228). The latter procedure may also be adopted to confirm the identity of any residues found.

Residues of WL 44776 determined by HPLC may be confirmed by preparing the parent compound WL 43467 as described above and using either TLC-GC (GE-XE 60 1.2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD) or GC-MS (SE-30 1% (m/m) on Gas Chrom Q (100-120 mesh)) as the determination step, MS operated in EI mode (ions monitored : m/e 207 and 209).

Specificity/Interference: Typical HPLC chromatograms are shown. Most control samples exhibit no major interfering peaks at the retention time of the metabolites; blank values are stated to be < 0.05 mg/kg. Chromatograms of recovery extracts fortified with WL 44776 and WL 44607 at the same time were not shown, but based on the retention times that were stated, both metabolites should be well resolved under the test conditions used.

Recovery/Precision: see Table **4.1.2-46**

Limit of Quantification: Based on the recovery results that were submitted, a LOQ of 0.10 mg/kg seems appropriate for WL 44607 (free and conjugated), while for WL 44776 (free and conjugated), a LOQ of 0.50 mg/kg appears defensible.

Table 4.1.2-46: Validation of methods SAMS 320-02 and SAMS 295-02 (Hitchings at al., 1983)

Matrix	Analyte	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
Lettuce, potatoes, maize, apples, pears	WL 44776 free	0.20 0.50	1 4	80 70 - 100	- 80	- 16.9
		0.20 - 0.50	5	70 - 100	80	14.7
	WL 44766 conjugated	0.50	5	65 - 85	76	9.8
Cabbage, potatoes, maize, apples, pears	WL 44607 free	0.10 0.20 0.50	2 3 1	100 - 115 100 - 120 90	107.5 113.3 -	(9.9) 10.2 -
		0.10 - 0.50	6	90 - 120	107.5	11.7
	WL 44607 conjugated	0.10 0.50	2 4	85 - 100 60 - 105	92.5 85	(11.5) 22.0
		0.10 - 0.50	6	60 - 105	87.5	17.8

Conclusion: **In terms of interfering blanks, accuracy (overall mean recoveries between 70 and 110%) and precision (overall RSD's lower than 20%), the method appears suitable for WL 44776 (free and conjugated) residue analysis in crops with a LOQ of 0.50 mg/kg. The methods were however not considered by the residue section in the frame of the renewal and are no longer considered as relied upon.**

Report:	CA 4.1.2/39 Smalley R., 2002 Validation of method RLA 12644 for the analysis of Cypermethrin isomers in wheat straw and grain AL-244-011
Guidelines:	none
GLP:	not stated

Principle of the method:
The residues of BAS 310 I are extracted using acetone followed by liquid-liquid partitioning into ethyl acetate/cyclohexane. The extract is then further cleaned up using GPC followed by silica SPE cartridge prior to analysis by GC-MSD (m/z ions 163, 165 and 181, TIC integrated).

Specificity/Interferences:	Not demonstrated within the study report. However, GC-MS is known to be a highly specific method by the possibility to monitor three m/z ions (see Study No. 7 here above). Untreated control samples exhibit no significant interferences at retention time of the analytes (chromatograms were provided for matrix-matched standards, controls and fortified samples at LOQ and 10 × LOQ).
Linearity:	Typical results for matrix matched calibrations were provided for each analyte. The linearity response during the validation procedure was acceptable with a correlation coefficient > 0.995 for all analysis determinations. Five (for wheat straw) and four (for wheat grain) matrix matched batch standard solutions over the range 0.01 – 0.1 µg/mL were injected.
Recovery/Precision:	see Table **4.1.2-47**
Limit of Quantification:	0.01mg/kg (nominal) for each of the 4 isomers of cypermethrin in all matrices. The actual lowest fortification per isomer is lower than 0.01 mg/kg in function of the isomeric ratio.

Table 4.1.2-47: Validation data (Smalley 2002)

Fortification Levels mg/kg	Cis I		Cis II		Trans III		Trans IV		Total isomers	
	Mean recovery %	% RSD	Mean recovery %	% RSD	Mean recovery %	% RSD	Mean recovery %	% RSD	Mean recovery %	% RSD
Wheat straw (no group)										
0.01	84	7.1	75	10.5	82	6.8	91	12.2	83	8.8
0.1	106	7.4	110	9.2	114	9.8	118	6.8	112	6.1
Wheat grain (dry)										
0.01	78	6.0	69	8.6	80	9.3	78	7.0	77	5.7
0.1	92	3.9	98	7.8	90	5.1	97	2.8	94	2.7

* Note: fortification performed with mixed isomers. The fortification level stated corresponds to the sum of the cis or trans-isomers, hence 0.01 and 0.1 mg/kg are the nominal levels. The level per individual isomer needs to be corrected by the isomeric ratio. The isomeric ratio of the Cis-I-Cis-II in the corresponding cypermethrin standard and the isomeric ratio of Trans-III-Trans-IV in the corresponding cypermethrin standard was however not availavle within the study.The actual lowest fortification per isomer is hence lower than 0.01 mg/kg. n = 5 replicates at each fortification level and for each analyte.

Conclusion: **This data generation method has been used in support of a cereal rotational crop study. As the results obtained generally lead to an overestimation of the residues determined, this method is considered to fully support the data generated.**

Report:	CA 4.1.2/40 Smalley R., 2002 Method validation of RLA 12594.01 analysis of Alphacypermethrin (AC 900049) in olives and olive oil AL-244-007
Guidelines:	EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 96/98
GLP:	Yes (Department of Health of the United Kingdom, United Kingdom)

Report:	CA 4.1.2/41 Anonymous, 2016 Excerpt of raw data supporting analytical methods used for the analysis of plant & animal matrices 2016/1232571
Guidelines:	not stated
GLP:	not stated

Method RLA 12594.01

Note: The method RLA 12594.01 (AL-244-007) was used as analytical method in residue study AL-750-038 (addendum 2002)- Alpha-cypermethrin 150 g as/kg WG(RLM 11203): A harvest residue study on alphacypermethrin in oil seed rape, south France, 2000 (KII 6.3/011), Grolleau G. The data were initially not reported in the section of the analytical methods of the addendum 2002 because part of the residue study AL-750-038. In order to be complete, validation data are therefore presented here below.

Method RLA 12594.02V supersedes RLA 12594.01 (method was amended only for calculation due to change in the volume used for extraction). The study reported results obtained for the primary validation but also for independent laboratory validation.

Principle of the method:

A 10 g specimen portion was extracted two times with acetonitrile. The acetonitrile was partitioned three times with hexane. An aliquot of the acetonitrile is evaporated and reconstituted in heptane. Clean-up was achieved by solid phase extraction using silica cartridges. Analysis was then performed by GC-ECD (DB5 column, 15 m × 0.53 mm i.d., 0.5 µm film thickness, oven temperature: 50°C for 0.5 min., then 35°C/min. to 200°C, then 10°C/min. to 280°C, hold 4 min.). External calibration.

Specificity/Interferences:	The report stated that specificity of method RLA 12594.01 (GC-ECD) was demonstrated by using GC-MS as an alternative method of detection. However, no results were provided. Untreated control samples exhibit no significant interferences at retention time of alpha-cypermethrin (chromatograms were provided for standards, controls and fortified samples at LOQ and 10 × LOQ).

Linearity: The linearity response during the validation procedure was acceptable with a correlation coefficient > 0.995. Five standard solutions over the range 0.02 – 0.1 µg/mL (40 – 200% of the normal working standard concentration, corresponding to 0.04 – 2 mg/kg) were injected. Calibrations occurred in solvent whereas matrix effects were not assessed in the study report. *Matrix was removed by the extensive clean-up using liquid-liquid partitioning of the acetonitrile phase with hexane. Untreated specimen confirm the absence of interference (example chromatograms in the study report)*". Additionally, **DocID 2016/1232571** (excerpt of raw data supporting analytical methods used for the analysis of plant & animal matrices) provided during the course of the assessment, that the ion ratios of all ions were comparable in the samples and standards (when confirmation by GC-MS), hence indicating that matrix-effects can be excluded (calibration with MS detection in the 0.02 – 0.1 µg/mL range).

Matrix effects: Matrix was removed by the extensive clean-up using liquid-liquid partitioning of the acetonitrile phase with hexane. Untreated specimen confirm the absence of interference (example chromatograms in the study report)". Additionally, **DocID 2016/1232571** (excerpt of raw data supporting analytical methods used for the analysis of plant & animal matrices) provided during the course of the assessment, that the ion ratios of all ions were comparable in the samples and standards (when confirmation by GC-MS), hence indicating that matrix-effects can be excluded (calibration with MS detection in the 0.02 – 0.1 µg/mL range).

Recovery /Precision: see Table **4.1.2-48**

Limit of Quantification: 0.05 mg/kg for alpha-cypermethrin in both matrices.

Table 4.1.2-48: Primary and independent validation data (Smalley 2002)

Sample matrix.	Fortification level (mg/kg)	Recovery (%)		RSD (%)	n
		Individual	Mean		
Olive oil (primary validation)	0.05	77, 74, 98, 76, 84	82	12	5
	1.0	83, 81, 80, 81, 80	81	2	5
Olive (primary validation)	0.05	88, 107, 80, 81, 76	86	14	5
	1.0	67, 61, 71, 68, 69	**67**	6	5
Olive oil (independent validation)	0.05	84, 84, 80, 80, 81	82	3	5
	1.0	88, 90, 91, 91, 89	90	1	5
Olive (independent validation)	0.05	73, 104, 80, 108, 87	90	17	5
	1.0	75, 77, 75, 76, 77	76	1	5

Conclusion:	**Mean recoveries and RSD were within the acceptable limits (70 -110 % and < 20%) at the LOQ and higher fortification level for oil and olive oil except for olive at the higher level (1.0 mg/kg) where the mean recovery was slightly below 70% with an acceptable RSD. Independent laboratory validation was performed. The method can be considered to be "fit for purpose" to support residue trials in the case of the determination of alpha-cypermethrin at 0.05 and 1.0 mg/kg in olive oil and at 0.05 mg/kg in olive. Since the ILV confirmed acceptable results at 0.05 mg/kg and residue levels obtained in study AL-HE-99-530 (trial 99-530-01 referring to residue trial in olive – Trewhitt, 1999a, AL-714-001 – DocID 2000/7000996, the method is considered suitable for the quantification of alpha-cypermethrin in seeds, oil and press cake.**

Report:	CA 4.1.2/42 Walker B., Linkerhaegner M., 2000a Alphacypermethrin (AC900049): Validation of the DFG method S 19 (extended revision) for the determination of residues of Alphacypermethrin in/on grapes wheat grain cabbage and oilseed rape AL-244-008
Guidelines:	EEC 91/414, EEC 96/46 4.2.1,SANCO/825/00 rev. 6 (20 June 2000),BBA Guideline Residue Analytical Methods for Post-Registration Control Purposes of July 21 1998,EPA 860.1340
GLP:	Yes (Behoerde fuer Arbeit, Gesundheit und Soziales, Freie und Hasestadt Hamburg, Hamburg, Germany)

Report:	CA 4.1.2/43 Walker B., Linkerhaegner M., 2000b Alphacypermethrin (AC 900049): Validation of the DFG method S 19 (extended revision) for the determination of residues of Alphacypermethrin in processed fractions of cabbage and oilseed rape AL-244-009
Guidelines:	E EEC 91/414, EEC 96/46 4.2.1,SANCO/825/00 rev. 6 (20 June 2000),BBA Guideline Residue Analytical Methods for Post-Registration Control Purposes of July 21 1998,EPA 860.1340
GLP:	Yes (Behoerde fuer Arbeit, Gesundheit und Soziales, Freie und Hasestadt Hamburg, Hamburg, Germany)

Method: DFG S19 used in support of residue trials / processing studies

Principle of the method: DFG method S19 (extended revision):

- Multimethod L 00.00-34 of the Official Collection of Test Methods according to §35 LMBG with extraction according to module E1 (grapes, cabbage and processed cabbage) or E2 (wheat grain) or E7 (oilseed rape and oilseed rape press cake) or E6 (refined rapeseed oil)
- cleanup of extract using gel permeation chromatography (module GPC)
- supplemental cleanup using silica gel column chromatography (module C1)
- determination by capillary GC (DB1; 30 m x 0.25 mm i.d.; 0.25 µm) with ECD (module D1)

Specimen material (e.g. grapes, cabbage and wheat grain) is extracted with acetone. Water is added beforehand in an amount that takes full account of the natural water content of the specimen so that during extraction the acetone:water ratio remains constant at 2:1 (v:v). For liquid-liquid partition ethyl acetate/cyclohexane (1+1) and sodium chloride are added and after mixing excess water is separated. Fat or dry, high fat specimen material (e.g. oilseed rape) is intensively mixed with a suspension of synthetic calcium silicate in acetonitrile and acetone. The suspension is filtered and the volume of the filtrate (organic phase) is measured. The organic phase is evaporated and the residue dissolved in the GPC elution mixture. The evaporated residue of an aliquot of the organic phase is cleaned up by gel permeation chromatography on Bio Beads S-X3 polystyrene gel using a mixture of ethyl acetate/cyclohexane (1+1) as eluent. The residue-containing fraction is concentrated and after supplemental silica gel mini column chromatography analysed for residues of alphacypermethrin by gas chromatography using an electron capture detector (ECD) (DB-1, 30 m × 0.25 mm, 0.25 µ m). Plant and animal fat (e.g. rapeseed oil) is dissolved in the GPC elution mixture. An aliquot is cleaned up by gel permeation chromatography on Bio Beads S-X3 polystyrene gel using a mixture of ethyl acetate/cyclohexane (1+1) as eluent. The residue-containing fraction is concentrated and after supplemental silica gel mini column chromatography analysed for residues of alphacypermethrin be gas chromatography using an electron capture detector (ECD).

Specificity/Interferences: Representative chromatograms (standards, control, fortified samples) are shown; untreated control samples exhibit no significant interferences.

Linearity: The response of the GC-ECD system (= peak area) to alpha-cypermethrin was demonstrated to be linear (r = 0.9995) over a concentration range from 0.0010 to 1 µg/mL (n = 7). Calibration in toluene.

Recovery/Precision : Table **4.1.2-49**

Limit of Quantification: 0.01 mg/kg

Table 4.1.2-49: Validation of DFG S19 (extended revision)

Matrix	Analyte (fortification)	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
Walker & Linkerhägner, 2000a						
Grapes (acidic)	alpha-cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	93 – 99 96 – 106 93 – 106	96 101 99	2.4 4.0 3.8
wheat grain (dry)	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	90 – 105 95 – 106 90 – 106	96 100 98	5.9 4.9 5.6
Cabbage (high water)	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	85 – 109 89 – 98 85 – 109	98 93 96	9.0 3.8 7.1
oilseed rape (oily)	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	87 – 101 82 – 96 82 – 101	92 90 91	6.1 7.6 6.6
Walker & Linkerhägner, 2000b						
cabbage : outer leaves	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	78 – 98 79 – 92 78 – 98	85 87 86	9.3 6.7 7.8
cabbage : boiled leaves	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	84 – 95 84 – 94 84 – 95	88 89 89	4.9 4.0 4.3
Sauerkraut	alpha - cypermethrin	0.01 0.10	5 5 10	78 – 101 85 – 95 78-101	92 89 914	9.3 4.2 7.0
oilseed rape press cake	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	74 – 102 76 – 89 74 – 102	85 82 83	12 6.6 9.6
refined rapeseed oil	alpha - cypermethrin	0.01 0.10 0.01 – 0.10	5 5 10	95 – 103 89 – 101 89 - 103	99 93 96	3.6 4.9 5.0

Conclusion:	**DFG multi-residue Method S19 (extended revision) is suitable for determination of alpha-cypermethrin residues in/on grapes, wheat grain, cabbage (incl. processed fractions) and oilseed rape (incl. processed fractions) with a LOQ of 0.01 mg/kg. It can be proposed as enforcement method as well as data-generation method.**

The methods for which validation data are provided below were used for data generation in the submitted residue trials.

Remark: No stand-alone validation of an analytical method for bee-relevant matrices was conducted. Validation of bee-relevant matrices, such as nectar, pollen and flowers was accomplished as part of the respective residue trial (6.10/1 and 10.3.1.6/7; DocID 2014/1000203). A summary of the methods can be found in the respective section of Doc N, chapter 5.1.3.

Report:	CA 4.1.2/44 Rabe U., Mackenroth C., 2007 Validation of the analytical method No. 546/0: Method for the determination of Alphacypermethrin (BAS 310 I) in plant matrices 2004/1010543
Guidelines:	EPA 860.1340,EEC 96/46,SANCO/825/00 rev. 6 (20 June 2000),SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	Yes (Landesamt fuer Umweltschutzund Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/45 Rabe U., Schweda Z., 2003 Technical procedure: Method for the determination of BAS 310 I in plant matrices 2003/1001290
Guidelines:	not stated
GLP:	not stated

Method 546/0

Principle of the method

Alpha-cypermethrin is extracted with a mixture of methanol/water/hydrochloric acid (70:25:5). An aliquot of the extract is centrifuged and cleaned by liquid/liquid partition against cyclohexane. The organic phase is purified by solid phase extraction (SPE) on a silica gel column. The final determination of BAS 310 I is performed by HPLC-MS/MS on a YMC J'sphere L80 column with an acidified water/methanol gradient as mobile phase at a flow rate of 0.6 mL/min. Ionization was performed in the positive mode and ion transitions 416 → 191 (primary) and 418 → 193 (confirmatory) were monitored.

Recovery findings

Average recoveries over both fortification levels were between 70 and 110% using both the primary and the confirmatory transition for detection. The recovery data are corrected for interference from matrix compounds of the appropriate unfortified sample (primary transition: ±0.4-4.7% for wheat forage, ±0.9-8.7% for wheat grain, ±0% for wheat straw, oilseed rape whole plant and grape fruit, ±0.2-2.7% for oilseed rape seed, ±1.0-9.9% for orange fruit; confirmatory transition: ±2.0-20.6% for wheat forage, ±1.3-13.6% for wheat grain, ±0% for wheat straw, oilseed rape whole plant, oilseed rape seed and grape fruit, ±0.1-4.4% for orange fruit).
Average recoveries for each matrix are summarized in Table **4.1.2-50**.

Table 4.1.2-50: Results of method validation: BAS 310 I (alpha-cypermethrin) in wheat, oilseed rape, grape and orange specimens

Matrix	Analyte	m/z	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Wheat forage	BAS 310 I	416 → 191	0.05	5	96.5	7.8	8.1
			0.5	5	103.8	4.4	4.2
Wheat grain	BAS 310 I	416 → 191	0.05	5	92.3	4.3	4.7
			0.5	5	93.1	4.8	5.2
Wheat straw	BAS 310 I	416 → 191	0.05	5	97.6	4.1	4.2
			0.5	5	95.7	4.4	4.6
Oilseed rape whole plant	BAS 310 I	416 → 191	0.05	5	106.5	8.3	7.8
			0.5	5	91.8	4.0	4.4
Oilseed rape seed	BAS 310 I	416 → 191	0.05	5	90.3	7.3	8.1
			0.5	5	88.4	4.4	4.9
Grape fruit	BAS 310 I	416 → 191	0.05	5	99.2	4.3	4.4
			0.5	4	93.6	7.0	7.5
Orange fruit	BAS 310 I	416 → 191	0.05	5	89.4	5.7	6.4
			0.5	5	101.2	5.4	5.3
Wheat forage	BAS 310 I	418 → 193	0.05	5	86.2	9.5	11.0
			0.5	5	98.4	4.9	5.0
Wheat grain	BAS 310 I	418 → 193	0.05	5	86.7	5.9	6.8
			0.5	5	91.1	3.7	4.1
Wheat straw	BAS 310 I	418 → 193	0.05	5	94.1	5.8	6.2
			0.5	5	92.4	4.7	5.0
Oilseed rape whole plant	BAS 310 I	418 → 193	0.05	5	114.3	10.0	8.8
			0.5	5	96.8	5.2	5.4
Oilseed rape seed	BAS 310 I	418 → 193	0.05	5	90.1	7.5	8.4
			0.5	5	90.4	4.0	4.4
Grape fruit	BAS 310 I	418 → 193	0.05	5	98.1	9.0	9.2
			0.5	4	96.1	5.2	5.4
Orange fruit	BAS 310 I	418 → 193	0.05	5	95.2	11.7	12.3
			0.5	5	100.3	2.5	2.5

Linearity Linearity was tested using six calibration standards in the range of 0.25-10 ng/mL (0.25-10 mg/kg). Calibration curves gave acceptable correlation coefficients ≥ 0.99. Standard solutions are prepared in methanol.

Specificity Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity. Residues and chromatographic signal interference in all blank control specimens were below the LOQ.

Limit of Quantification The limit of quantification (LOQ) for BAS 310 I is 0.05 mg/kg for all investigated plant matrices.

Repeatability The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-50**.

Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The method 546/0 is suitable for the determination of residues of alpha-cypermethrin in wheat (forage, grain, straw), oilseed rape (whole plant, seed), grape (fruit) and orange (fruit).**

Report:	CA 4.1.2/46 Anonymous, 2006 Residue behaviour of Alpha-Cypermethrin in/on fennel, curly cale, cucumbers, carrots, radies, celery, celeriac and bunshing or green onions after outdoor application of Fastac SC (SC 100) in Germany, 2005 2006/1029533
Guidelines:	not stated
GLP:	not stated

Method SAA/C/PSM02.3

Principle of the method

Alpha-cypermethrin is extracted from homogenized plant material with a mixture of methanol/water/2 N hydrochloric acid (70:25:5). An aliquot of the extract is centrifuged and cleaned by liquid/liquid partition against cyclohexane. The cyclohexane is evaporated and the organic phase is re-diluted in methanol/water (80:20, v/v). The final determination of BAS 310 I is performed by LC-MS/MS Ion transitions 433 → 191 (primary) and 435 → 193 (confirmatory) were monitored.

Recovery findings

Average recoveries for each fortification level were between 70 and 110%. Average recoveries for each matrix are summarized in Table **4.1.2-51**.

Table 4.1.2-51: Results of method validation: BAS 310 I (alpha-cypermethrin) in cucumber specimens

Matrix	Analyte	m/z	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Cucumber	BAS 310 I	433 → 191 435 → 193	0.005	n.r.	92	n.r.	5.2
			0.01	n.r.	95	n.r.	4.0
			0.1	n.r.	97	n.r.	2.7

Linearity	Linearity was tested using six calibration standards in the range of 0.05-5.00 ng/mL. Standard solutions are prepared in methanol/water (80:20, v/v) from stock solutions prepared in acetone.
Specificity	Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity.
Limit of Quantification	The limit of quantification (LOQ) for BAS 310 I is 0.005 mg/kg for all investigated plant matrices. The limit of detection (LOD) is 0.001-0.0036 mg/kg.
Repeatability	The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-51**.

Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The method SAA/C/PSM 023 is considered to be fit for purpose for the determination of residues of alpha-cypermethrin in cucumber. The report indicates that the method is equivalent to analytical method 567/0, which was fully validated (see 2005/5000063, CA 4.1.2/49).**

Report:	CA 4.1.2/47 Stewart J., 2005a Method validation of BASF analytical method 567/0 entitled: Method for determination of Alphacypermethrin and Cypermethrin in plant matrices - Including standard stability and extract stability 2005/5000063
Guidelines:	EPA 860.1340, SANCO/825/00 rev. 6 (20 June 2000)
GLP:	yes (certified by United States Environmental Protection Agency)

Principle of the method

The analytical procedure described was developed to identify the residues of BAS 310 I (alpha-cypermethrin) and BAS 311 I (cypermethrin) in plant matrices. Fortified plant samples were analysed using BASF Analytical Method 567/0. BAS 310 I (alpha-cypermethrin) or BAS 311 I (cypermethrin) are extracted with a 70:25:5 methanol/water/hydrochloric acid mixture from matrices except from commodities with high fat content. After centrifugation, an aliquot of the extract is partitioned into cyclohexane. Silica Gel SPE column is used for further purification, if required (for example for wheat straw). Final determination is performed by LC-MS/MS using the ammonium adducts of cypermethrin. From matrices of commodities with high fat content, BAS 310 I (alpha-cypermethrin) or BAS 311 I (cypermethrin) is extracted with acetonitrile/n-hexane. After centrifugation and further partitioning with n-hexane, an aliquot of the acetonitrile phase is removed. Silica Gel SPE column is used for further purification, if required. Final determination is also performed by LC-MS/MS using the ammonium adducts of cypermethrin.

Recovery findings

BASF Analytical Method 567/0 was successfully validated for all eight plant matrices tested. Average recoveries over both fortification levels ranged from 82.8 to 112.9% using the primary transition (m/z 433 → m/z 191) for detection. Coefficients of variation (%RSD) for the average recoveries were all less than 20%. There were no residues detected in the control samples above the limit of detection, except in the secondary transition of one of the orange samples (0.001 m/kg – at the limit of detection).
Individual recoveries ranged from 71 to 121% using the primary transition for detection. One laboratory outlier was not included in the statistical analysis. Results obtained using the secondary transition for detection (435 → m/z 193) were comparable to those obtained using the primary transition.
Average recoveries for each matrix are summarised in Table **4.1.2-52**.

Table 4.1.2-52: Results of method validation: BAS 310 I (alpha-cypermethrin) and BAS 311 I (cypermethrin) in wheat, tomato, orange cotton, pea and potato specimens

Matrix	Analyte	m/z	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Wheat grain	BAS 310 I	433 → 191	0.01	5	90.1	3.1	3.4
			0.1	5			
Wheat plant w/o root	BAS 310 I	433 → 191	0.01	5	82.8	6.6	7.9
			1.0	5			
Wheat straw	BAS 310 I	433 → 191	0.01*	5	86.8	13.7	15.8
			1.0*	5			
Tomato fruit	BAS 310 I	433 → 191	0.01	5	89.5	7.8	8.7
			0.1	5			
Orange fruit	BAS 310 I	433 → 191	0.01	5	91.8	8.1	8.8
			0.1	5			
Cotton seed	BAS 310 I	433 → 191	0.01	5	87.1	8.4	9.7
			0.01*	4			
		433 → 191	0.1	5			
			0.1*	5			
Pea (succulent)	BAS 310 I	433 → 191	0.01	5	82.8	10.6	12.8
			0.1	5			
Potatoe tuber	BAS 310 I	433 → 191	0.01	5	92.8	11.2	12.0
			0.1	5			
Overall mean				**89**	**88**	**9**	**11**
Wheat plant w/o root	BAS 311 I	433 → 191	0.01	5	112.9	5.0	4.5
			1.0	5			
Wheat straw	BAS 311 I	433 → 191	0.01	5	98.2	5.7	5.8
			1.0	4			
Overall mean				**19**	**106**	**9**	**9**
Wheat grain	BAS 310 I	435 → 193	0.01	5	90.1	4.5	5.0
			0.1	5			
Wheat plant w/o root	BAS 310 I	435 → 193	0.01	5	92.5	3.4	3.7
			1.0	5			
Wheat straw	BAS 310 I	435 → 193	0.01*	5	94.6	18.6	19.7
			1.0*	5			
Tomato fruit	BAS 310 I	435 → 193	0.01	5	84.7	6.3	7.4
			0.1	5			
Orange fruit	BAS 310 I	435 → 193	0.01	5	93.3	10.2	10.9
			0.1	5			
Cotton seed	BAS 310 I	435 → 193	0.01	5	88.2	7.8	8.9
			0.01*	4			
		435 → 193	0.1	5			
			0.1*	5			
Pea (succulent)	BAS 310 I	435 → 193	0.01	5	81.9	11.2	13.7
			0.1	5			
Potatoe tuber	BAS 310 I	435 → 193	0.01	5	88.8	8.4	9.5
			0.1	5			
Overall mean				**89**	**89.2**	**10.1**	**11.3**
Wheat plant w/o root	BAS 311 I		0.01	5	119.0	22.6	19.0
			0.1	5			
Wheat straw	BAS 311 I		0.01	5	95.1	8.9	9.4
			0.1	4			
Overall mean				**19**	**107.7**	**21.0**	**19.5**

* Analysis conducted with optional clean-up column

Linearity	Calibration curves gave acceptable correlation coefficients ≥ 0.99.
Specificity	The method is specific for the determination of alpha-cypermethrin in plant matrices down to a level of 0.01 mg/kg.
Limit of Quantification	The limit of quantitation for BAS 310 I is 0.01 mg/kg for all investigated plant matrices.
Repeatability	The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-52**.
Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study
Extract stability	Recoveries obtained from stored grain and straw extracts did not diminish over the seven day storage period.
Standard stability	Recoveries obtained from stored chromatographic standard solutions of alpha-cypermethrin in acetonitrile/water (80:20 v,v) did not diminish after 33 days storage. The stock solution prepared in pure solvent (acetonitrile) was found to be stable for 94 days.
Conclusion	**The method 567/0 is suitable for the determination of residues of alpha-cypermethrin in wheat grain, wheat plant, wheat straw, tomato, orange, cotton seed, succulent pea and potato.**

Report: CA 4.1.2/48
Bretz M., 2007a
Technical procedure: Method for the determination of Alphacypermethrin in plant matrices by GC-MS - BASF method number 567/1
2007/1010254
Guidelines: none
GLP: no

Principle of the methods

BASF Method No 567/1 is an adaption of BASF Method No. 567/0 for the determination of alpha-cypermethrin and cypermethrin in various plant matrices. Data contained in the report on the technical procedure were not necessarily produced under GLP regulations. The document described in detail, is a technical procedure and no full validation report.
BAS 310 I (alpha-cypermethrin) is extracted with a 70:25:5 methanol/water/hydrochloric acid or a 95:5 methanol/hydrochloric acid mixture, depending on the matrix. After centrifugation, an aliquot of the extract is partitioned into cyclohexane. A Silica Gel SPE column is used for further purification, if required. Final determination is performed by GC-MS monitoring the three most intense ions. The two isomer peaks are determined together.

Recovery findings

Recoveries in barley matrices were determined in the context of the barley processing study 2007/1013068 (see Section 6, Point CA 6.5.3/2). All recovery data were determined under GLP and are reported in detail in DocID 2007/1013068.

The overall average recovery for all matrices over all fortification levels of BAS 310 I was 94.3% (%RSD = 12.0). Average recoveries for each matrix ranged from 82% (%RSD = 7.9) to 108% (%RSD = 3.8) and are summarised in Table **4.1.2-53**.

Table 4.1.2-53: Recoveries of alpha-cypermethrin in barley matrices

Crop	Matrix	Test Substance	Fortific. Level (mg/kg)	No. of Tests	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Barley	plant w/o root	BAS 310	0.1	5	100.7	8.5	8.4
			1.0	5	91.9	12.7	13.8
	malt culms	BAS 310	0.01	5	96.5	12.4	12.8
			0.1	5	82.0	6.5	7.9
	spent grain	BAS 310	0.1	5	91.0	12.8	14.1
	spent hops	BAS 310	0.01	4	93.1	10.8	11.6
			0.1	5	84.7	13.3	15.7
	yeast	BAS 310	0.01	5	108.0	4.1	3.8
			0.1	5	93.8	7.1	7.6
	beer	BAS 310	0.01	5	103.9	4.0	3.8
			0.1	5	86.5	6.6	7.7
	flour	BAS 310	0.1	5	99.2	5.4	5.4
			1.0	5	94.5	13.0	13.7
Overall mean				**64**	**94.3**	**11.3**	**12.0**

Linearity	Since the report describes the technical procedure, the linearity was not determined. However, in a residue report (BASF DocID 2009/1125197, see chapter 6.3) the detector response was reported to be linear within the range from 0.2 ng/mL to 70 ng/mL with a correlation coefficient >0.998.
Specificity	The GC-MS final determination for BAS 310 I is a highly selective detection. Three separate ions are monitored, one of which can be used for quantitative evaluation. The other ones can be used for confirmation of residue findings. Therefore no confirmatory technique is required.
Limit of Quantification	The limit of quantitation for BAS 310 I is 0.01 mg/kg for all investigated plant matrices.
Repeatability	For the plant matrices the relative standard deviations were below 20 % (4-16%).
Reproducibility	Reproducibility of the method was not determined.
Standard stability	Standards of BAS 310 I have been shown to be stable for up to 59 days in acetone and 62 days in acetone/H_20 (96/4) + 1 mg/ml L-gulono-ɤ-lacton.
Conclusion	**The successful validation of the method demonstrated that the BASF method number 567/1 is suitable to determine alpha-cypermethrin residues in barley and different processing matrices.**

Animal

Analytical methods for the determination of alpha-cypermethrin residues in animal matrices were evaluated in the context of the inclusion in Annex I of Directive 91/414/EEC. Methods evaluated previously are summarized in Table **4.1.2-54** for the reviewer´s convenience.

Table 4.1.2-54: Summary of peer-reviewed analytical methods for determination of alpha-cypermethrin residues in animal matrices

Method No.	Matrix	Method principle	Target analytes	LOQ mg/kg	year	DocID	EU reviewed
SAMS 461-01	Muscle, fat, kidney, liver	GC-ECD; GC-MS for confirmation	Alpha-cypermethrin	0.01	1988	AL-245-001	Yes
SAMS 456-01	Milk	GC-ECD; GC-MS for confirmation	Alpha-cypermethrin	0.001 mg/L	1988	AL-245-003	Yes
SAMS 461-1; SAMS 456-1	Muscle, fat, kidney, liver Milk	GC-ECD; GC/MS for specifity	Alpha-cypermethrin	0.05 0.01	1999	AL-245-006	Yes
DFG S 19	Milk, eggs	GC-ECD; GC-MS for confirmation GC/NPD as alternative determination method	Alpha-cypermethrin	0.01	2000	AL-245-007	Yes
DFG S 19	Blood and urine (Swine)	GC-ECD	Alpha-cypermethrin	0.005 mg/L	2000	AL-245-008	Yes
M 3499	Blood	GC-MS	Alpha-cypermethrin	0.005 mg/L	2001	AL-210-012 and RES 00-052 (AL-440-018)	No

Although already peer-reviewed during the previous Annex I-registration process, the previously submitted methods are summarised below for completeness of information.

The following method was not mentioned in the application but is also included in this dossier for completeness:
Method M3466 for determination of alpha-cypermethrin in hen egg had been validated as part of a poultry feeding study. A detailed description of the method is given in document M, chapter 6.4.1, Report CA 6.4.1/1 [redacted] 2001). Method 3466 as part of the respective poultry feeding study was not peer-reviews under directive 91/414*EEC earlier, as the entire study was regarded as not required due to an anticipated feed burden below the trigger value. Hence, method M3466 is not regarded as peer-reviewed and is described in detail in chapter CA 6.4.1/1 as part of the respective residue trial/feeding study.

Report:	CA 4.1.2/49 Anonymous, 1988 Determination of residues of Alphacypermethrin in animal tissues - Gas liquid chromatographic method AL-245-001
Guidelines:	not stated
GLP:	not stated

Principle of the method :
Tissue samples are chopped or minced finely, mixed with anhydrous sodium sulphate and extracted by boiling with acetone/hexane (1:2 v/v). The solvent is evaporated and the residue is re-dissolved in hexane. For fat and muscle samples, a portion of the extract is partitioned with acetonitrile by using an Extrelut extraction cartridge, while a normal hexane/acetonitrile partition is used for the liver and kidney extracts. All extracts are further cleaned up by liquid-solid chromatography on a Florisil cartridge. Alpha-cypermethrin residues are determined by packed column GC (GE-XE 60 1.2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD); quantification by external standardization. Residues of alpha-cypermethrin are confirmed by capillary GC-ECD (SE-54; 0.5 μ m) for resolution of the Cis-1, Trans-3 and Trans-4 isomers from alphacypermethrin (Cis-2) and GC-MS (CP SIL 5 CB), MS operated in negative ion chemical ionisation mode (ions monitored : m/e 207 and 209).

Specificity/Interference	Typical packed column GC-ECD chromatograms are shown; control samples exhibit no significant interfering peaks at the retention time of alpha-cypermethrin.
Recovery/Precision	see Table **4.1.2-55**
Limit of Quantification	0.1 mg/kg

Table 4.1.2-55: Validation of method SAMS 461-01 (no author specified, 1988)

Matrix	Analyte	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
fat	alpha-cypermethrin	0.1 0.2	6 2	80 - 100 100 - 115	94.2 107.5	8.5 (9.9)
		0.1 - 0.2	8	80 - 115	97.5	10.3
muscle	alpha-cypermethrin	0.1 0.2	3 1	85 - 100 90	90 -	9.6 -
		0.1 - 0.2	4	85 - 100	90	7.9
kidney	alpha-cypermethrin	0.1 0.2	2 1	85 - 95 90	90 -	(7.9) -
		0.1 - 0.2	3	85 - 95	90	5.6
liver	alpha-cypermethrin	0.1 0.2	2 1	80 - 95 95	87.5 -	(2.1) -
		0.1 - 0.2	3	80 - 95	90	9.6
overall	alpha-cypermethrin	0.1 0.2	13 5	80 - 100 90 - 115	91.5 98	8.5 10.6
		0.1 - 0.2	18	80 - 115	93.3	9.4

Conclusion **In terms of interfering blanks, accuracy (mean recoveries between 70 and 110%) and precision (RSD's lower than 20%), the method is suitable for alpha-cypermethrin residue analysis in animal tissues with a LOQ of 0.1 mg/kg.**

Report:	CA 4.1.2/50 Anonymous, 1988 Determination of residues of Alphacypermethrin in milk - Gas liquid chromatographic method
	AL-245-003
Guidelines:	not stated
GLP:	not stated

Principle of the method :
Milk samples are extracted by shaking with aqueous potassium oxalate solution, ethanol, diethyl ether and hexane. The extract is evaporated to dryness and the residue is redissolved in hexane, after which partition between acetonitrile and hexane is achieved rapidly by means of an Extrelut extraction column. Further clean-up is obtained by use of a cyano Bond Elut cartridge.
Alpha-cypermethrin is determined by packed column GC (GE-XE 60 1.2% (m/m) on Gas Chrom Q (100-120 mesh)) with electron capture detection (ECD); quantification by external standardization. Residues of alpha- cypermethrin are confirmed by capillary GC-ECD (SE-54; 0.5 µm) for resolution of the Cis-1, Trans-3 and Trans- 4 isomers from alphacypermethrin (Cis-2) and GC-MS (CP SIL 5 CB), MS operated in negative ion chemical ionisation mode (ions monitored : m/e 207 and 209).

Specificity/Interference	Typical packed column GC-ECD chromatograms are shown; control sample exhibits no significant interfering peaks at the retention time of alpha-cypermethrin.
Recovery/Precision	see Table **4.1.2-56**
Limit of Quantification	0.005 mg/L

Table 4.1.2-56: Validation of method SAMS 456-01 (no author specified, 1988)

Matrix	Analyte	Fortification level (mg/L commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
milk	alpha-cypermethrin	0.005 0.010 0.015 0.020	1 2 1 1	105 95 - 105 100 90	- 100 - -	- (7.1) - -
		0.005-0.020		90-105	99	6.6

Conclusion	**In terms of interfering blanks, accuracy (overall mean recovery between 70 and 110%) and of precision (overall RSD lower than 2 %), the method is suitable for residue analysis in milk.**

Report:	CA 4.1.2/51 Doran A.M., Mayer I. H., Khunachak A., 1999 Renegade Alphacypermethrin (CL 900049): Validation of analytical methods SAMS 461-1 and SAMS 456-1 for the determination of Alphacypermethrin residues in cattle tissues (muscle, fat, kidney and liver) and milk
	AL-245-006
Guidelines:	The Rules Governing Medicinal Products in the European Community - Volume VI
GLP:	Yes (Department of Health of the Government of the United Kingdom)

Remark: Study AL-245-006 is part of residue study RES 01-008.

Principle of the method:
The purpose of the study was to further validate methods SAMS 461-1 and SAMS 456-1. Method SAMS 461-1 for analysis of tissue samples and method SAMS 456-1 for analysis of milk samples are described in the draft monograph; deviations from these methods were minor. Quantification of the residues by both methods is achieved by capillary GC (SE 54; 15 m x 0.32 mm i.d.; 0.25 µm) with ECD. Confirmation by GC/MS, MS operated in CI-(methane) ionization mode (ions monitored: 207 and 209 amu). External calibration was used for purpose of quantification

Specificity/Interference	Specificity was demonstrated. Untreated control samples exhibit no significant interferences; representative chromatograms are shown.
Linearity:	The response of the GC-ECD system (= peak area) to alpha-cypermethrin was demonstrated to be linear over a range from 0.01 to 0.10 µ g (n = 4, single injection). The linear curve was provided in the study report with a correlation coefficient > 0.9997. Calibration seems to have been performed in solvent but this is not clearly mentioned in the study report. Matrix effects were not assessed. The extensive sample clean-up (liquid/liquid partitioning and/or SPE) removes the naturally occurring matrix effects.
Recovery/Precision	see Table **4.1.2-57**

Table 4.1.2-57: Validation of methods SAMS 461-1 and SAMS 456-1 (Doran, Mayer & Khunachak, 1999)

Matrix	Analyte (fortification)	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
Method SAMS 461-1						
muscle	α-cypermethrin	0.05	3	92 – 105	97	7.2
		0.10	3	106 – 109	108	1.4
		0.20	3	93 – 107	100	7.2
		0.05 – 0.20	9	92 – 109	102	6.8
fat	α-cypermethrin	0.05	3	78 – 89	84	6.6
		0.10	3	104 – 108	106	1.9
		0.20	3	95 – 97	96	1.0
		0.05 – 0.20	9	78 – 108	96	10
liver	α-cypermethrin	0.05	3	78 – 84	82	3.5
		0.10	3	99 – 101	100	1.1
		0.20	3	89 – 93	92	1.8
		0.05 – 0.20	9	78 – 101	91	8.9
kidney	α-cypermethrin	0.05	3	98 – 106	101	4.2
		0.10	3	82 – 85	84	1.9
		0.20	3	98 – 105	101	3.6
		0.05 – 0.20	9	82 – 106	95	9.7
Method SAMS 456-1						
milk	α-cypermethrin	0.01 mg/L	3	84 – 86	85	1.3
		0.02 mg/L	3	101 – 104	103	1.5
		0.04 mg/L	3	106 – 115	110	4.3
		0.01 – 0.04	9	84 – 115	99	12

Additional data from study RES 01-008:

Table 4.1.2-58: Procedural recoveries as reported in study RES 01-008

Matrix	analyte	Fortification range	Recovery %	Average recovery %
Muscle	Alpha-cypermehtrin	50-500 ppb (0.05 – 0.5 mg/kg)	65-102	85 (n=4, 2 at each level))
Fat	Alpha-cypermehtrin	50-500 ppb (0.05 – 0.5 mg/kg)	92-112	102 (n=4, 2 at each level)
Kidney	Alpha-cypermehtrin	50-500 ppb (0.05 – 0.5 mg/kg)	75-102	90 (n=4, 2 at each level)
Liver	Alpha-cypermehtrin	50-500 ppb (0.05 – 0.5 mg/kg)	82-104	92 (n=4, 2 at each level)
milk	Alpha-cypermehtrin	10-100 ppb (0.01 – 0.1 mg/L)	75-106	92 (n=4, 2 at each level)
Muscle	Alpha-cypermehtrin	50-500 ppb (0.05 – 0.5 mg/kg)	89-99	94 (n=2)
Fat	Alpha-cypermehtrin	50-1200 ppb (0.05 – 1.2 mg/kg)	76-117	97 +/- 19 (n=4)
Kidney	Alpha-cypermehtrin	50 ppb (0.05 mg/kg)	92-93	93 (n=2)
Liver	Alpha-cypermehtrin	50-500 ppb (0.05 – 0.5 mg/kg)	88-90	89 (n=2)
milk	Alpha-cypermehtrin	10-100 ppb (0.01 – 0.1 mg/L)	77-119	95 +/- 13 (n=30)

It should also be noted that a validation study performed by [redacted] was also presented and part of the study report RES 00-052: *CL 900049 (alpha-cypermethrin): Validation of SAMS Method 461-1 and Magnitude of the Residue of CL 900049 in Laying hen Tissues (meat, Liver and Abdominal Fat) – Analytical Phase Report –* [redacted] *2001, Study No. A054.010, GLP, Unpublished.* Minor modifications to the initial method were reported including slight amendment of the GC conditions (DB-5, 15 m× 0.53 mm, 1.5 µm). The extraction method remained unchanged.

The findings for the validation were as followed:

Specificity/Interference No significant interference has been observed at the retention time of alphacypermethrin. Chromatograms were provided for a standard solution, controls, fortified samples at LOQ and 10×LOQ and treated samples. GC-ECD is not a specific method. No information is provided about confirmation in that study report but GC-MS can be used as previously described.

Linearity: Method was found to be linear in the 0.0015 – 0.015 µg/mL range with a correlation coefficient of 0.9992 (n=5). The calibration curve and the regression equation were provided. Calibration in solvent was used for quantification.

Recovery/Precision: The method was validated at two fortification levels (0.05 and 0.5 mg/kg) in hen eggs. For each fortification level, five replicates were analyzed. Additionally, two replicates of unfortified samples were examined. Results are presented in table below.

Table 4.1.2-59: Validation of method SAMS 461-1 for determination of alpha-cypermethrin in laying hen tissues (Meat, Liver and Abdominal Fat)

Matrix	Analyte (fortification)	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
Method SAMS 461-1						
Meat	α-cypermethrin	0.05	5	95-115	104	7.9
		0.50	5	74-102	84	16.2
		Overall	10	74-115	94	
Liver	α-cypermethrin	0.05	5	79-87	83	3.5
		0.5	5	74-89	82	6.7
		Overall	10	74-89	83	
Fat	α-cypermethrin	0.05	5	86-109	102	7.6
		0.5	5	93-108	98	6.3
		Overall	10	89-109	100	

Remark: Recoveries run concurrently with samples showed also acceptable recoveries:
- Meat at 0.05 mg/kg: 115% (n=1)
- Liver at 0.05 – 0.5 mg/kg: 78 and 88 % (n=1 each)
- Fat at 0.05 mg/kg: 87 and 107% (n=2) and at 0.5 mg/kg: 86% (n=1).

Limit of Quantification	0.05 mg/kg for each matrix.
Conclusion:	**Methods SAMS 461-1 and 456-1 have been used in support of feeding studies in poultry and ruminants and the results of these studies can be considered reliable.**

Report:	CA 4.1.2/52 Hausmann S., 2000 Alphacypermethrin (CL 900049): Validation of the multi-residue method DFG S19 with modified extraction for the determination of Alphacypermethrin residues in milk and eggs AL-245-007
Guidelines:	EEC 91/414 Annex II 4.2.1, EU Guideline 8064/VI/97 rev. 4 15.12.1998
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

Principle of the method:
Water is added to the samples in an amount that takes the natural water content of the samples into account so that during extraction, the water/acetone ratio remains constant (1/2 v/v). Thus water/acetone (1/2 v/v, 300 mL) is used for extraction followed by partition in acetone/ethyl acetate/cyclohexane (which replaced dichloromethane). GPC is used for elimination of fat and macro-molecules. Adsorption chromatography on silica gel with precisely defined fractionation (the silica gel cleanup is performed according to DFG S19 with some modifications). The analysis is performed by GC-ECD (non-polar 5% phenyl methyl silicone fused silica capillary column, stationary phase, 30 m × 0.25 mm, 0.25 µm film thickness, oven: 100°C hold 2 min., rate 25°C/min. to 260°C hold 4 min., rate 25°C/min to 280°C hold 6 min (milk) and 10 min (eggs)). External calibration. GC/MS was used as a confirmation method (using a 5 % phenyl methyl siloxane fused silica capillary column, non polar stationary phase: 30 m × 0.25 mm, 0.25 µm film thickness, SIM : m/z 181, 163 and 165 for monitoring and identification and m/z 181 taken for quantification). GC-NPD was also demonstrated as an alternative method for determination of alpha-cypermethrin in milk and eggs (5 % phenyl methyl silicone fused silica capillary column, 30 m × 0.25 mm, 0.25 µm film thickness, oven: 70°C hold 1.3 min, rate 25°C/min to 280°C hold 10 min), detection at 320°C).

Specificity/Interference	No interference has been observed at the retention time of alphacypermethrin. GC-ECD Chromatograms were provided for standard solutions, controls, fortified samples at LOQ and 10×LOQ, GC-MS chromatograms were provided for calibrations, controls and fortified samples at LOQ and GC-NPD chromatograms were provided for a calibration and fortified samples. Confirmation of the GC-ECD occurred by GC-MS monitoring three m/z ions 181, 163 and 165, with m/z 181 used for quantification.
Linearity:	The response of the detector was found quadratic in the 0.02 – 1.0 µg/mL range (n = 6, single injection) for the GC-ECD (eggs), for the GC-MS (for milk and eggs) and linear in the same concentration range for the GC-ECD (in milk). It was not clearly stated if the calibration occurred with matrix-matched standards. Two calibration curves were provided (one quadratic and one linear) both with a r2 > 0.99 (0.9988 for milk by GC-ECD and 0.99959 for milk and eggs by GC-MS).
Recovery/Precision:	Milk and egg samples have been fortified with alpha-cypermethrin at two levels (0.01 mg/kg and 0.1 mg/kg).

Table 4.1.2-60: Validation of method DFG S19 for the determination of alpha-cypermethrin in milk and eggs

Matrix	Analyte (fortification)	Fortification level (mg/kg commodity)	Recovery		
			Number of samples Range (%)	Mean (%)	RSD (%)
milk	α-cypermethrin	0.01 0.1	5 5	93 100	9 6
		0.01 (confirmation by GC-MS)	1	78	-
		0.1 (confirmation by GC-MS)	1	104	-
eggs	α-cypermethrin	0.01 0.1	5 5	97 79	3 3
		0.01 (confirmation by GC-MS)	1	103	-
		0.1 (confirmation by GC-MS)	1	96	-

Limit of Quantification 0.01 mg/kg

Conclusion: The method is considered sufficiently validated in regards of SANCO/3029/99 rev. 4. The method allows the determination of alpha-cypermethrin in milk and egg samples with a validated LOQ of 0.01 mg/kg in each matrix. However the method is not used in support of residue studies considered for the purpose of the renewal.

Report:	CA 4.1.2/53 Walker B., Linkerhägner M., 2000 Alphacypermethrin (AC 900049): Validation of the DFG method S 19 (extended revision) for the determination of residues of Alphacypermethrin in blood and urine of animal origin (swine) AL-245-008
Guidelines:	EEC 91/414, EEC 96/46 4.2.1, SANCO/825/00 rev. 6 (20 June 2000),BBA Guideline Residue Analytical Methods for Post-Registration Control Purposes of July 21 1998,EPA 860.1340
GLP:	Yes (Behoerde fuer Arbeit, Gesundheit du Soziales, Frei und Hansestadt, Hamburg, Germany)

Principle of the method

The extraction of alpha-cypermethrin from swine blood and urine was performed according to extraction module E1 (DFG method S19): specimen material is extracted with acetone. Water is added beforehand in an amount that takes full account of the natural water content of the specimen so that during extraction the acetone/water ratio remains constant at 2:1 (v/v). For liquid/liquid partition ethyl acetate/cyclohexane (1:1) and sodium chloride are added and after repeated mixing excess water is separated. The evaporated residue of the organic phase is cleaned up by gel permeation chromatography (GPC) and silica gel column chromatography (module C1) according to DFG method S19 (extended revision). All specimens are analyzed by capillary gas chromatography with electron capture detection (GC-ECD) on a fused silica capillary column (DB1; J&W) with argon/methane (95:5) as carrier gas at a flow rate of 1.0mL/min.

Recovery findings

Average recoveries for all fortification levels were between 70 and 110%. Average recoveries for each matrix are summarized in Table **4.1.2-61**.

Table 4.1.2-61: Results of method validation: BAS 310 I (alpha-cypermethrin) in swine blood and urine specimens

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Blood	BAS 310 I	0.005	5	105	4.3	4.1
		0.05	5	101	3.4	3.4
Urine	BAS 310 I	0.005	5	96	6.0	6.3
		0.05	5	95	2.7	2.8

Linearity	Linearity was tested using six calibration standards in the range of 0.00250-0.0998 µg/mL, covering the working range, with a correlation coefficient >0.99. Standards were prepared in toluene.
Specificity	A confirmatory method has not been performed; however, no significant interferences of matrix or labware have been reported. Control specimens yielded no residues above the limit of detection (0.001 mg/kg), i.e.<30% LOQ. Good validation results have been achieved with the primary method.
Limit of Quantification	The limit of quantification (LOQ) for BAS 310 I is 0.005 mg/kg for blood and urine.
Repeatability	The relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table **4.1.2-61**.
Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The method DFG S19 is considered sufficiently validated for the determination of residues of alpha-cypermethrin in blood and urine.**

Report:	CA 4.1.2/4 Xu B., 2001 BAS 310 I (Alpha-Cypermethrin): Validation of method M 3499 for the confirmation of BAS 310 I residues in water soil and blood by GC/MS
	AL-210-012
Guidelines:	Guidance Document on Residue Analytical Methods (SANCO/825/00rev.6)
GLP:	Yes (United States Environmental Protection Agency)

Report:	CA 4.1.2/54 [redacted] 2001 FASTAC Insecticide (Alphacypermethrin BAS 310 I): Magnitude of BAS 310 I residues in Laying Hen Eggs, Muscle, Liver and Abdominal Fat After Oral Administration of BAS 310 I for 28 Consecutive Days
	AL-440-018
Guidelines:	Guidance Document on Residue Analytical Methods (SANCO/825/00rev.6 EPA 860.1480, EEC 91/414,EEC 7031/VI/95 Appendix G rev. 4,SANCO/3029/99 rev. 4 (11 July 2000),8046/VI/97-rev 4 (15/12/1998))
GLP:	Not stated

Principle of the method:

Residue of alpha-cypermethrin are extracted from homogenized samples of eggs with a mixture of hexane and tetrahydrofuran (THF) 1:2 v:v. Aliquots of the organic extract are evaporated and the residue is dissolved in a mixture of hexane and dichloromethane 1:1 v:v. Extracts are purified using adsorption chromatography on a one gram Silica SPE cartridge, followed by liquid/liquid partitioning using hexane/acetonitrile. Quantitative determination is carried out by gas chromatography/Electron Capture Negative Ion Chemical Ionization Tandem Residue of alpha-cypermethrin are extracted from homogenized samples of eggs with a mixture of hexane and tetrahydrofuran (THF) 1:2 v:v. Aliquots of the organic extract are evaporated and the residue is dissolved in a mixture of hexane and dichloromethane 1:1 v:v. Extracts are purified using adsorption chromatography on a one gram Silica SPE cartridge, followed by liquid/liquid partitioning using hexane/acetonitrile. Quantitative determination is carried out by gas chromatography/Electron Capture Negative Ion Chemical Ionization Tandem.

Specificity/Interference	No significant interference has been observed at the retention time of alphacypermethrin. Chromatograms were provided for a standard solution, control, fortified samples at LOQ and 10×LOQ and treated samples. Method is specific by monitoring several m/z ions. However, only one ion fragment (m/z: 207) was considered here for quantification.
Linearity:	Method was found to be linear in the 0.0025 – 0.030 µg/mL range with a correlation coefficient of 0.9997 (n=5). The calibration curve and the regression equation were provided. However, it is not indicated in the study report if the calibration was performed with matrix-matched standard solutions or with calibration standards in solvent. The presence or absence of matrix effect was not reported. However, the extensive sample clean-up (liquid/liquid partitioning and/or SPE) would remove the naturally occurring matrix effects.

Recovery/Precision The method was validated at two fortification levels (0.01 and 0.1 mg/kg) in hen eggs. For each fortification level, five replicates were analyzed. Additionally, two replicates of unfortified samples were examined. Results are presented in Table **4.1.2-62**.

Limit of Quantification: 0.01 mg/kg.

Table 4.1.2-62: Validation of method M3466 for determination of alpha-cypermethrin in hen eggs

Matrix	Analyte (fortification)	Fortification level (mg/kg commodity)	Recovery			
			Number of samples	Range (%)	Mean (%)	RSD (%)
Method M3466						
Hen egg	α-cypermethrin	0.01 0.10	5 5	102 – 119 79 – 98	110 92	7 11
Blood	α-cypermethrin	0.005	3	Retention times: ≤0.1% difference from bracketing standards Ion ratios: ≤5.5% difference compared to the standards		

Stability: Residue sin extracts are shown to be stable for 7 days.

Conclusion: **Method M3466 is validated according to SANCO/3029/99 rev. 4. The method is suitable for the quantification of alpha-cypermethrin and is suitable to support the feeding study in poultry. The results obtained can be considered reliable.**

(f)Methods in soil, water, sediment, feed and any additional matrices used in support of ecotoxicology studies

No stand-alone analytical methods were validated in support of ecotoxicological studies. Methods for the determination of concentration, whenever necessary, are reported along with the respective ecotoxicological study.

No stand-alone analytical methods were validated in support of ecotoxicological studies. Methods for the determination of concentration, whenever necessary, are reported along with the respective ecotoxicological study as well as summarised below. Validation parameters are summarised under the DocID of the ecotoxicological studies as analytical phase reports have not been issued. In such cases where a method was used for a large number of studies, validity criteria are addressed in overview tables to facilitate review.

Report:	CA 4.1.2/55 [redacted] 2009 BAS 310 I – Acute toxicity in the zebra finch (Taniopygia guttata) after single oral administration (LD50)
	2009/1114317
Guidelines:	EPA 540/9-82-024, EPA 540/9-85-007,EPA 850.2100,EPA 712-C-96-139,EPA 71-1
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the method:
Samples were transferred with approximately 100 mL acetonitrile/highly deionized water 1/1 (v/v) into 500 mL measuring flasks and were made up to the calibration marc with acetonitrile and dissolved under sonication. Aliquots of the dilutions were used for HPLC-UV analysis (column: Synergy Fusion RP 80 4 µm, 250 m x 3 mm, eluent 80% acetonitrile + formic acid (1,000 + 1 mL)/15 % highly deionized water + formic acid (1,000 + 1 mL, detection at 198 nm). External calibration. Matrix analysed was 0.5 % carboxymethylcellulose (CMC) in drinking water.

Dose verification and stability for 6h at room temperature were verified.

Specificity/Interferences:	No interferences occurred at the retention time of the analyte in the control sample (0.5 % CMC in drinking water). Chromatograms were provided for standard solutions, blank and samples. The identity was confirmed on the basis of the selected wavelength and the retention time. The method is used for dose verification of known substances and known nominal concentrations. Hence, no additional confirmatory technique is necessary.
Linearity:	Method was found to be linear: in the ~ 8.112 – 50.7 mg/100mL range (3 different concentrations, duplicate infection). Calibration in acetonitrile/highly deionized water 1/1 v/v, further diluted with acetonitrile. The calibration curve was provided but not the regression equation and the correlation coefficient. However, since the calibration curve showed good linearity, no further data are required. The concentration of the samples meet the calibration range.
Limit of Quantification:	8.112 mg/100 mL (0.08112 mg/mL) corresponding to 4.056 % w/w (lowest calibration concentration).

Table 4.1.2-63: Dose verification and stability data for alpha-cypermethrin in 0.5% CMC in drinking Water

Sample matrix/analyte	Nominal concentration % w/w (corresponding concentration in mg/100 mL)	Nominal concentration in mg/kg bdw	% of the nominal concentration	n*
Dose verification				
0.5% CMC in drinking water/alpha-cypermehtrin	5 (10 mg/100 mL)	500	102.1	1 × 2
	10 (20 mg/100 mL)	1000	101.3	1 × 2
	20 (40 mg/100 mL)	2000	100.2	1 × 2
Stability				
0.5% CMC in drinking water/alpha-cypermethrin	1.092 (0h) (~ 2 mg/100 mL)	-	99.5	1 × 2
	1.060 (6h) (~ 2 mg/100 mL)	-	95.4	1 × 2

Note: for the stability determination, a separate analytical report was done and was part of the study KCA 8.1.1.1/001. In that analytical report, a new calibration curve was performed and chromatograms were also provided.
* one replicate at each level with duplicate injections.

Conclusion: The method is suitable for the determination of alpha-cypermethrin in 0.5% CMC in drinking water used in gavage studies on birds. The method has been used in study KCP 10.1.1.2/001. The LOQ and the validated working range meet the ecotox endpoint.

Report:	CA 4.1.2/56 [redacted] 2009 BAS 310 I - Early life-stage test on the fathead minnow (Pimephales promelas) with pulse dose exposure 2009/1031203
Guidelines:	OECD 210, EPA 72-4 (a), EPA 850.1400,EPA 540/9-86-138
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/57 Bergtold M., 2007 Chronic toxicity of BAS 310 I to Daphnia magna STRAUS in a 21 day semi-static test - A time to effect study 2007/1016502
Guidelines:	OECD 211,EPA 850.1300
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/58 Hoffmann F., 2009a Effect of BAS 310 I (Reg.No. 4078193) on the growth of the fresh water diatom Navicula pelliculosa - A limit test 2009/1109081
Guidelines:	OECD 201, EPA 850.5400
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/59 Hoffmann F., 2009b Effect of BAS 310 I (Reg.No. 4078193) on the growth of the blue-green alga Anabaena flos-aquae - A limit test 2009/1109080
Guidelines:	OECD 201, EPA 850.5400
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/60 Hoffma Effect of BAS 310 I (Reg.No. 4078193) on the growth of the marine diatom Skeletonema costatum - A limit testnn F., 2009c 2009/1109079
Guidelines:	OECD 201,EPA 850.5400
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/61 Hoffmann F., 2009d Effect of BAS 310 I (Reg.No. 4078193) on the growth of Lemna gibba - A limit test 2009/1108874
Guidelines:	OECD 221,EPA 850.4400,ASTM E 1415-91
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 4.1.2/62 Hoffmann F., 2009d Effect of BAS 310 51 I on the growth of the green alga Pseudokirchneriella subcapitata 2009/1011443
Guidelines:	OECD 201
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the method APL0445/01:

Method applied here was method APL0445/01 developed for analysis of alpha-cypermethrin in soil, water and blood and validated under study 130428 (method validation M3499-01 – Xu B. 2001 - BAS 310 I (alpha- cypermethrin): GC-MS confirmatory method for BAS 310 I residues in water, soil and blood). Samples were extracted by solvent partition with hexane and diluted to be within the calibration range with hexane after evaporation to dryness. To improve the performance of the method, n-heptane was used. Therefore, trace amounts of alpha- cypermethrin dissolved or suspended in water were enriched by liquid/liquid extraction with n- heptane, the organic layers were combined and evaporated to dryness using a rotary evaporator under vacuum. The residue was then re-dissolved in L-gluconic acid-Lactone 1 mg/mL in acetone in an appropriate range for analysis. Analysis occurred by GC-MS (DB-5 column, 30 mm x 0.32 mm i.d, 0.25 µm particle size; ion monitored: m/z = 207). External calibration was used for quantification purposes.

Validation was not done for each available water type. If the method is shown to be suitable for quantification purposes in the most difficult water matrix, e.g. in terms of salt or matrix load, the easier matrices were not fully validated again, as the matrix is mainly water.

Table 4.1.2-64: Validation data for method L0045/01 (M3499) to determine alpha-cypermethrin in water

Study/ Matrix and Analyte	LOQ (µg/L)	Linearity* Range (µg/L) (n) Correlation coefficient r	Fortification levels	Specificity and interferences
CA 4.1.2/56 2009/1031203 Drinking water/alpha-cypermethrin	0.02	0.02 – 5 (n=5, duplicate injection) r ≥ 0.999 Calibration curve and regression equation provided	Samples of nominal concentrations between 0.019 and 1.2 µ g/L were prepared, analysed directly and after 7 days storage at room temperature and deep frozen. Mean recovery at each fortification level remains within the 70-110% range. Analysis of retained samples (stored for 7 days at room temperature or deep frozen) showed some losses in concentration.	Blank control was not tested during the fortification experiment but blank controls during the ecotoxicity study showed no significant interferences occurring at the retention time of alpha-cypermethrin except in one sample for which contaminations during sample preparation were suspected. Some typical chromatograms were provided for a blank control, a standard, a sample at 0.15 µ g/L at day 0 and day 4, stock solution at 1.4 µ g/L. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
CA 4.1.2/57 2007/1016502 Reconstituted water (M4 medium)/alpha-cypermethrin	0.01	3.2 – 26 (n=4, duplicate injection) $r^2 \geq 0.999$ Calibration curve and regression equation provided	Fortified samples were prepared at levels in the range of the test samples and atlevel of the stock solution. Mean recovery at each level is presented in the table below.	No significant interference from the blank control was stated in the study report. Chromatograms were provided for the blank solution (M4 medium), standard solutions and test sample at 37.2 ng/L at initiation of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
CA 4.1.2/58 2009/1109081 Algal medium (OECD medium according to OECD guideline 201 /alpha-cypermethrin	2.0	0.05 – 5 (n=4, duplicate injection) $r^2 \geq 0.998$ Calibration curve and regression equation provided. The concentration of the samples meet the calibration range, otherwise dilution occurred to meet the calibration range	2– 50 Mean recovery at each level is presented in the table below.Two controls were tested concurrently with the test. Degradation of test item occurred below the LOQ after 4 days at room temperature.	No significant interference from the blank control was stated in the study report. Chromatograms were provided for the blank solution (OECD medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard

CA 4.1.2/59 2009/1109080 Algal medium (1×APP medium)/alpha-cypermethrin	2.0	0.05 – 5 (n=4, duplicate injection) $r^2 \geq 0.999$ Calibration curve and regression equation provided. The concentration of the samples meet the calibration range, otherwise dilution occurred to meet the calibration range	2– 50 Mean recovery at each level is presented in the table below. Two controls were tested concurrently with the test. Degradation of test item occurred below the LOQ after 4 days at room temperature.	No significant interference from the blank control was observed. Chromatograms were provided for the blank solution (algal medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
CA 4.1.2/60 2009/1109079 Algal medium (ESW medium)/alpha-cypermethrin	2.0	0.05 – 5 (n=4, duplicate injection) $r^2 \geq 0.999$ Calibration curve and regression equation provided. The concentration of the samples meet the calibration range, otherwise dilution occurred to meet the calibration range.	2– 50 Mean recovery at each level is presented in the table below. Two controls were tested concurrently with the test. Degradation of test item occurred below the LOQ after 4 days at room temperature.	No significant interference from the blank control was observed. Chromatograms were provided for the blank solution (algal medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the Standard.
CA 4.1.2/61 2009/1108874 20×APP medium/alpha-cypermethrin	2.0	0.06 – 6 (n=4, duplicate injection) $r^2 \geq 0.999$ Calibration curve and regression equation provided. The concentration of the samples meet the calibration range, otherwise dilution occurred to meet the calibration range.	2– 120 Mean recovery at each level is presented in the table below. Two controls were tested concurrently with the test. Stability test in the carrier for 120 µ g/L sample showed slightly degradation of the test item after 7 days	No significant interference from the blank control was observed. Chromatograms were provided for the blank solution (algal medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard
CA 4.1.2/62 2009/1011443 OECD algal medium according to OECD guideline 201 / alpha-cypermethrin	0.029 mg/L BAS 310 51I. However, based on the data reported, it appears that it should be 0.025 mg/L BAS 310 51 I	5 – 100 for BAS 310 51 I (5 concentrations, duplicate injection), corresponding to $r^2 \geq 0.998$ Calibration curve and regression equation provided. The concentration of the samples meet the calibration range, otherwise dilution occurred to meet the calibration range.	25-93000 for BAS 310 55 I corresponding to ~ 1.2 µ g a.s./L and 4600 µ g a.s./L, respectively. Two controls were tested concurrently with the test. After three days (room temperature), slightly degradation was observed.	No significant interference from the blank control was observed. Chromatograms were provided for the blank solution (algal medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard

Table 4.1.2-65: Mean recoveries and RSD obtained for method L0045/01 for the determination of alpha-cypermethrin in water

Matrix/analyte	Fortification levels (µg/L)	Mean recovery % (range)	RSD %	n
CA 4.1.2/56 2009/1031203 Drinking water/alpha-cypermethrin	0.019 (directly measured at day 0)	100.7	-	1×2**
	0.296 (directly measured at day 0)	98.5	-	1×2**
	0.161 (directly measured at day 0)	95.4	-	1×2**
	0.321 (directly measured at day 0)	104.0	-	1×2**
	1.205 (directly measured at day 0)	100.5	-	1×2**
	0.019 – 1.205	99.8*	3.17*	5×2
CA 4.1.2/57 2007/1016502 Reconstituted water (M4-edium)/alpha-cypermethrin	See table below in more detail			
CA 4.1.2/58 2009/1109081 Algal medium (OECD medium according to OECD guideline 201 /alpha-cypermethrin	2.14	99.1* (99.2-98.9)	-	2×2**
	49.5	98.7* (98.8, 98.5)	-	2×2**
CA 4.1.2/59 2009/1109080 Algal medium (1×APP medium)/alpha-cypermethrin	2.14	100.4* (99.5-101.3)	-	2×2**
	49.5	97.9* (98.2, 97.5)	-	2×2**
CA 4.1.2/60 2009/1109079 Algal medium (ESW medium)/alpha-cypermethrin	2.14	97.2* (98.2-96.2)	-	2×2**
	49.5	95.4* (96.3, 94.5)	-	2×2**
CA 4.1.2/61 2009/1108874 20×APP medium/alpha-cypermethrin	2.0	100.9* (96.7-105.2)	-	2×2**
	120.0	98.8* (98.4, 99.3)	-	2×2**
CA 4.1.2/62 2009/1011443 OECD algal medium according to OECD guideline 201 / alpha-cypermethrin	25 µg formulation/L (corresponding to 1.2 µg/L alpha-cypermethrin)	84.3 (-)	-	1×2**
	93000 µg formulation/L (corresponding to 4600 µg/L alphacypermethrin)	84.9 (-)	-	1×2**

Table 4.1.2-66: Alpha-cypermethrin validation results from water (study 2007/1016502, CA 4.1.2/57)

Sample matrix/analyte	Fortification level (nominal)	Alpha-cypermehtrin recovery (mean %) Day 0 – at concentration in the range of test samples (peak concentration)	RSD (%)	n**
Reconstituted water (M4 medium)/alpha-cypermethrin	28.7 ng/L	84.7	-	2×2
	67.1 ng/L	74.1	-	2×2
	95.8 ng/L	69.1	-	2×2
	134 ng/L	61.6	-	2×2
	overall	72.4	13.4	8×2
	Day 7 – at concentration in the range of test samples (peak concentration)			
	29.4 ng/L	117.0	-	2×2
	58.7 ng/L	148.3	-	2×2
	88.1 ng/L	118.4	-	2×2
	117 ng/L	104.8	-	2×2
	Day 0- 3- 5 at concentration representative for the stock solution			
	4.79 µ g/L	51.2	-	2×2
	5.49 µ g/L	82.3	-	2×2
	Day 7 at concentration representative for the stock solution			
	5.87 µ g/L	95.7	-	2×1
	Day 10 and 12 at concentration representative for the stock solution			
	4.61 µ g/L	115.5	-	2×2
	Day 14 at concentration representative for the stock solution			
	5.48 µ g/L	105.2	-	2×2
	Day 17 and 19 at concentration representative for the stock solution			
	4.87 µ g/L	71.9	-	2×2

* recoveries found during the test (test samples) were all within the 70-110% range except for the 37.2 ng/L after day 0 and day 7 (recovery > 110%), for the 74.4 ng/L level for day 7 (recovery > 110%), for stock solutions at 5 µg/L at day 0 and day 7 (recovery < 70%).
** n = 2 corresponds to two replicates, each injected two times.

Limit of Quantification: The proposed limit of quantification is 0.0287 µg/L which was confirmed by n=4 fortification samples in M4-enriched medium. 0.019 µg/L are achievable, but this has been confirmed by n=2 replicates only.

Conclusion: **The method is considered suitable for the quantification of alpha-cypermethrin in various aqueous media using GC-ECD.**

Report:	CA 4.1.2/63 Janson G.-M.,Weltje L., 2009 Acute toxicity of BAS 310 I (Reg.No. 4078193) to larvae of the phantom midge Chaoborus crystallinus in a 48 hour static test 2009/1085205
Guidelines:	OECD 202, SANCO 3029/99 rev.4
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Note: the report on the analytical phase written by Class T. 2009 was part of the Study by Janson G-M. and Weltje L. 2009.

Principle of the method APL0445/01:

Water samples were extracted in centrifuge vials or separatory funnels 3-times after adding 3 mL of a saturated sodium chloride solution by liquid/liquid partition with n-hexane (each time 3 mL). After phases separation aided by centrifugation (5 min., 3000 rpm), the combined organic phases were concentrated to near dryness adding about 1 mL of toluene as keeper. The final extract volume was then adjusted to 0.25, 0.5, 1 or 2 mL for GC-ECD injection (Varian VF-1701 MS fused silica capillary column, 30 mm x 0.32 mm i.d, 0.25 µm particle size; oven program: 90°C, 2.0 min. hold, ramp 30°C/min to 300°C, 4 min. hold at 300°C). External calibration. The column used is able to distinguish the isomers of cypermethrin.
Matrix: Mesocosm water. Characteristics of the water: 2.23 mmol/L total hardness, 2.2 mmol/L alkalinity up to pH 4.3, pH = 7.97, conductivity of 552 µS/cm. For validation tap water from the laboratory.

Specificity/Interference	No interferences occurred at the retention time of alpha-cypermethrin. Chromatograms were provided for calibration solutions, tap water as blank control, test samples, BASF pond water, BASF water blank control, solvent control and fortified samples at 5 ng/L. Identification was performed by comparison of the retention time in the test sample with the standard .
Linearity:	Method was found to be linear : $r^2 \geq 0.999$ in the 0.25 – 25.0 ng/mL range (7 concentrations, duplicate injection). Calibration occurred in toluene. The water was extracted by liquid-liquid partitioning using n-hexane. Analysis of the respective untreated water samples confirm the absence of any interference; additionally, due to the partitioning, naturally occurring interferences would have been removed. The corresponding linear regression plot and equation were provided. The prepared samples meet the calibration range.
Accuracy:	Samples of water were fortified at different concentration levels. Mean recoveries are provided in the table below.
Repeatability:	Relevant RSDs are reported in the table below.
Limit of Quantification:	5 ng/L

Table 4.1.2-67: Alpha-cypermehtrin validation results from water in a mesocosm study

Sample matrix/analyte	Fortification level (ng/L)	Alpha-cypermehtrin recovery (%) Mean (range)		RSD (%)	n
Tap water/alpha-cypermethrin	5	110 (100-117)*		7	6
	5000	98 (95-101)		-	2
	1250	89		-	1
	470	80		-	1
Tap water data overall		**Range: 80 – 117**	**102**	**12**	**10**

* Three from the individual recoveries were equal to 117% (> 110%).

Conclusion: **The method is considered fit for purpose to support the data generated. The propose LOQ is considered validated and covers the relevant ecotoxicological endpoints.**

Report:	CA 4.1.2/64 Janson G.-M., Dorner S., Weltje L., 2009 Acute toxicity of Alpha-Cypermethrin (BAS 310 I) and of Cypermethrin (BAS 311 I) to the non-biting midge Chironomus riparius in a 48 hour static test 2009/1102214
Guidelines:	OECD 202
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the method:

Ecotox water samples were extracted in separatory funnels 3-times with n-hexane. The combined organic extract was transferred into a tapered 25-mL or 50-mL flask, concentrated using a rotary evaporator to near dryness using a gentle stream of nitrogen. The final extract volume was then adjusted to 0.25 or 0.2 mL with 10-ng/mL IS solution (vortex and sonicate) for GC/ECD injection (Varian VF-1701 MS fused silica capillary column 30 m ×0.32 mm, 0.25 µm, internal calibration using lambda-cyhalothrin). The column used is able to distinguish the isomers of cypermethrin. Matrix**:** reconstituted water (M4 medium according to Elendt: prepared on the basis of ultrapure deionized water and containing Tween 80 at 2 µ/L), characteristics: 2.45 mmol/L total hardness, 0.88 mmol/L alkalinity up to pH 4.3, 7.99 pH, 665 µs/cm conductivity. Fortification were performed on tap water.

Specificity/Interference	No significant interference from the blank controls was stated in the study report. Chromatograms were provided for the blank solution (tap water but not for the blank of the test: M4 medium / Tween 80), standard solutions, fortified samples at LOQ and 100 ng/L and test samples for both analytes. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
Linearity:	Method was found to be linear: $r^2 \geq 0.998$ in the 0.5 – 50.0 ng/mL (n = 5) for alpha-cypermethrin and $r^2 \geq 0.998$ in the 0.5 – 100.0 ng/mL (n = 6) for cypermethrin. Calibrations were prepared in toluene. The water/control media were extracted by liquid-liquid partitioning using n-hexane. Analysis of respective untreated water samples confirm the absence of any interference; additionally, due to the portioning, naturally occurring interference would have been removed. The corresponding linear regression plots and equations were provided.
Accuracy:	Samples of tap water were fortified with 5 and 100 ng/L of each of the analyte. Mean recovery at each fortification level and for each analyte is presented in the Table below.
Repeatability:	RSD was reported in table below and is lower than 20%.
Limit of Quantification:	5 ng/L

Table 4.1.2-68: Alpha-cypermethrin validation results from water

Sample matrix/analyte	Fortification level (ng/L)	Alpha-cypermehtrin recovery (%) Mean (range)	RSD (%)	n
Tap water/alpha-cypermethrin	5	106 (102-109)	4	3
	100	99 (98-99)	1	3
	5	103 (95-109)	6	6
	100	82 (70-92)	11	3

Conclusion: **The method using a capillary column allows the distinction of the four isomers of cypermethrin and the method is also considered as validated with an LOQ of 5 ng/L for cypermethrin (sum of all isomers). For the GC-ECD method (LOQ = 5 ng/L and validated working range of 5 – 500 ng/L), the LOQ covers the endpoints and the validated working range covers the mean measured concentrations except the lowest level but this level (4 ng/L) is very close to 5 ng/L. Therefore, the method is considered sufficiently "fit for purpose" and the ecotox results are therefore considered as reliable.**

Report:	CA 4.1.2/65 Backfisch K.,Weltje L., 2011 Chronic toxicity of Reg.No. 4078193 (BAS 310 I; Alpha-Cypermethrin) to the non-biting midge Chironomus riparius - A spiked sediment study 2011/1124187
Guidelines:	OECD 218 (2004)
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the method:

Alpha-cypermethrin was determined in the water samples (overlaying water and sediment pore water samples) by liquid/liquid partition into hexane and GC/ECD determination based on internal calibration (same method as described in KCA 8.2.5.3/001). Alpha-cypermethrin was determined in the sediment samples by modified DFG S19 method and GC/MS (Varian VF-5 MS, 30 m × 0.32 mm, 0.25 µm or Machery-Nagel OPTIMA – 17 ms, 30 m × 0.32 mm, 0.25 µm, monitored ion: m/z 207) based on internal calibration (lambda cyhalothrin). At 2.5 g of a homogenized specimen, 10 mL of water was added and let the sample soak for 10 min. 40 mL of acetone were added and the mixture shaken for 30 min. with the horizontal shaker. 7.0 g of sodium chloride were added to the homogenate and 20 mL of GPC eluting mixture (ethyl acetate/cyclohexane 1/1 v/v). The sample was shaken for 30 min. When the phases were clearly separated, an aliquot of 6.0 mL of the upper organic phase was taken and evaporated to dryness using constant stream of nitrogen. The final volume was then adjusted to 0.2 mL or 1.0 mL (for higher fortification level) with 10 ng/mL IS solution (vortex and sonicate) for GC-MS injection.

Matrix:

- reconstituted water (M4 medium according to Elendt: prepared on the basis of ultrapure deionized water), characteristics: 2.53 mmol/L total hardness, 0.9 mmol/L alkalinity up to pH 4.3, 7.94 pH, 678 µs/cm conductivity. Fortification were performed on tap water or M4 medium.
- Sediment: artificial substrate as described in OECD guideline 218 (2004).

Specificity/Interference	No significant interference from the blank controls (M4 water, tap water, sediment, solvent) was observed. Chromatograms were provided for the blank solution (tap water, M4 water, sediment), standard solutions, internal standard, fortified samples at LOQ and higher fortification levels and test samples at the initiation and at the end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
Linearity:	For water: Method was found to be linear: $r^2 \geq 0.999$ in the 0.5 – 50.0 ng/mL (n = 7) for alpha-cypermethrin. Calibrations were prepared in toluene. The corresponding linear regression plots and equations were provided. For sediment: Method was found to be linear: $r^2 \geq 0.999$ in the 0.25 – 50.0 or 100 ng/mL (n ≥ 7) for alpha-cypermethrin. Matrix-matched calibrations were prepared. The corresponding linear regression plots and equations were provided. The samples meet the calibration. Further dilutions occurred to meet the calibration range if required.

Accuracy: Samples of M4 water or tap water were fortified with 5 and 5000 ng/L of alpha-cypermethrin. Samples of untreated sediment were fortified at 1.0 µg/kg and 400 µg/kg. Mean recovery at each fortification is presented in the Table below.

Repeatability: RSD was reported in table below and is lower than 20%..

Limit of Quantification: 5 ng/L (water), 1 µg/kg (sediment)

Table 4.1.2-69: Alpha-cypermethrin validation results from water and sediment

Sample matrix/analyte	Fortification level	Alpha-cypermethrin recovery (%) Mean (range)	RSD (%)	n
M4 water or tap water)/alphacypermethrin	5 ng/L	106 893-121)	11	6*
	50 ng/L	79 (-)	-	1
	500 ng/L	99 (96-101)	3**	4*
	5000 ng/L	117 (-)	-	1
Sediment/ alpha-cypermethrin	1 µg/kg	98 (93-102)	4	6
	400 µg/kg	110 (106-109)	5	5

For determination in water : three blank controls (two M4 water and one tap water) were tested. For determination in sediment: three blank controls (sediment) were tested.
* Among the 6 replicates, 5 were performed with M4 water and one with tap water. Among the 4 replicates, 3 were performed with M4 water and one with tap water.
** Calculated based on the results presented in the study report.

Conclusion: **The GC-MS (modified DFG S19) method for the determination of alpha-cypermethrin in sediment with an LOQ of 0.0 01 mg/kg is fully validated according to SANCO/3029/99 rev. 4. Mean recoveries and RSD are within the acceptable limits according to SANCO/3029/99 rev.4. Only one individual recovery was found to be higher than 110% at 400 µg/kg but the mean recovery remained acceptable. The GC-MS method is considered to fully support the ecotox results. For study KCA. 8.5.1.2.6, only the endpoints for sediment are used in the risk assessment.**

Report:	CA 4.1.2/66 Hoess S., 2013 Chronic toxicity of Alpha-Cypermethrin to Caenorhabditis elegans exposed via spiked sediment according to ISO guideline 10872 (2010) 2013/1250848
Guidelines:	ISO 10872 (2010)
GLP:	Yes (Landesamt fuer Umwelt, Messungen und Naturschutz Baden-Wuerttemberg, Karlsruhe Germany)

Report:	CA 4.1.2/67 Anonymouns, 2016 Excerpt of raw data supporting analytics used in ecotoxicological studies) 2016/1232573
Guidelines:	not stated
GLP:	not stated

Principle of the method:

The method is almost identical to the method described in study KCA 8.2.5.3/002 03 (see Study no. 11 here above) with the exception that the aliquot is filtered before concentration and injection. The amount of sample and solvent were also adapted and the GC-MS (monitored ions : m/z 207, 209 and 211) conditions are slightly different (injection temperature, temperature program).

- **Matrix:** Formulated sediment according to ISO 10872.

Specificity/Interference	No significant interference from the blank control was observed. Chromatograms were provided for the control (sediment), a standard solution, fortified sample at LOQ and test samples. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
Linearity:	Method was found to be linear: r2 ≥ 0.996 in the 3.0 – 500 ng/mL (n = 7). Calibrations were prepared in toluene. In the raw data (*DocID 2016/1232573 – Excerpt of raw data supporting analytics used in ecotoxicological studies)*, matrix matched quality control samples (QC) have been analysed during the quantitative analysis to confirm the absence of matrix effects. Levels tested were 3 and 500 ng/L. No significant matrix effects were observed. The corresponding linear regression plots and equations were provided. The samples meet the calibration range. Further dilutions occurred to meet the calibration range if necessary. For water: Method was found to be linear: $r^2 \geq 0.999$ in the 0.5 – 50.0 ng/mL (n = 7) for alpha-cypermethrin. Calibrations were prepared in toluene. The corresponding linear regression plots and equations were provided. For sediment: Method was found to be linear: $r^2 \geq 0.999$ in the 0.25 – 50.0 or 100 ng/mL (n ≥ 7) for alpha-cypermethrin. Matrix-matched calibrations were prepared. The corresponding linear regression plots and equations were provided. The samples meet the calibration. Further dilutions occurred to meet the calibration range if required.

Accuracy: Samples of sediment were fortified with 0.1 and 50 mg/kg of alphacypermethrin. Mean recovery at each fortification level is presented in the Table below.

Repeatability: RSD was reported in table below and is lower than 20%.

Limit of Quantification: 0.1 mg/kg sediment

Table 4.1.2-70: Alpha-cypermethrin validation results from sediment by GC-MS

Sample matrix/analyte	Fortification level	Alpha-cypermethrin recovery (%) Mean (range)	RSD (%)	n
Sediment/ alpha-cypermethrin	1 µg/kg	105 (100-109)	5	3
	50 µg/kg	110 (101-116)	7	3

One blank control has been tested

Conclusion: **The GC-MS method is considered suitable for the quantification of alpha-cypermethrin in sediment matrices with an LOQ of 0.1 mg/kg. The method supports the endpoints derived from study KCA 8.2.5.4/002.**

Report:	CA 4.1.2/68 Gilbert D, Goth M., Class T., 2012 Alpha-Cypermethrin (BAS 310 I): A study on the chronic toxicity to the sediment dweller Lumbriculus variegatus 2012/1205915
Guidelines:	OECD 203 (1992), OECD 225 Sediment-water Lumbriculus toxicity test using spiked sediment (October 2007)
GLP:	Yes (Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden, Germany)

Remark: CA 4.1.2/68 (2016/123573) is also considered here.

Principle of the method:

Alpha-cypermethrin was determined in the water samples (overlaying water samples) by liquid/liquid partition into dichloromethane and GC/MS determination (column Agilent VF-5ms, 30 m ×0.32 mm, 0.25 µm, SIM mode, monitored ion: m/z 207) based on internal calibration (lambda cyhalothrin). The extraction method is similar to the one described in study KCA 8.2.5.3/001-and 002 03 (studies No. 10 and 11) despite that amounts were adapted and that dichloromethane has been used instead of hexane. Water samples (75 mL) are extracted in separatory funnels 3-times with each 10 mL of dichloromethane. The combined organic extract was filtered through a funnel fitted with glass wool and anhydrous sodium sulfate. The sodium sulphate was washed two times with each 5 mL of dichloromethane. The extract was concentrated using a rotary evaporator to about 5 mL. The 5 mL-residue was transferred into 15 mL glass centrifuge vial, the evaporation flask was rinsed with 5 mL of dichloromethane and
0.25 mL of the 10-ng/mL IS solution was added. The extract was concentrated using a N2-evaporator at about 40°C water bath temperature to about 1 mL. The extract was transferred into a 1 mL autosampler vial and the remaining dichloromethane was evaporated. Alpha-cypermethrin was determined in the sediment samples by modified DFG S19 method and GC/MS (Agilent VF-5ms, 30 m × 0.32 mm, 0.25 µm, SIM mode, monitored ion: m/z 207) based on internal calibration (lambda cyhalothrin). The extraction method is identical to the one described in study KCA 8.2.5.3/002 03 (study No. 11) despite that amounts were adapted.

Matrix:

- reconstituted water according to OECD guideline No. 203, characteristics: 2.53 mmol/L total hardness, 0.9 mmol/L alkalinity up to pH 4.3, 7.94 pH, 678 µs/cm conductivity. Fortification were performed on tap water or M4 medium.
- Sediment: artificial substrate as described in OECD guideline 225.

Specificity/Interference	No significant interference from the blank controls was observed. Chromatograms were provided for the blank solutions (tap water, sediment, untreated overlying water), standard solutions, internal standard, fortified samples at LOQ and higher fortification levels and test samples at the initiation and at the end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.

Linearity: For water and sediment: Method was found to be linear : r2 ≥ 0.999 in the 0.3– 100 ng/mL (n = 7) for alpha-cypermethrin. Calibrations were prepared in toluene. No matrix-matched calibration was used. In the raw data (*DocID 20161232573 – Excerpt of raw data supporting analytics used in ecotoxicological studies*), matrix effects were assessed by analysis of matrix-matched quality control samples (QC) prepared for the test item and the internal standard; by calculating the ratio of both, any occurring matrix effect is accounted for and the ratio obtained is comparable to the solvent standards. The corresponding linear regression plot and equation were provided. The samples meet the calibration. Further dilutions occurred to meet the calibration range when required.

Accuracy: Samples of tap water were fortified with 5 and 500 ng/L of alpha-cypermehtrin. Samples of untreated sediment were fortified at 17.0 µg/kg and 1667 µg/kg dry sediment weight. Mean recovery at each fortification is presented in the Tablebelow.

Repeatability: RSD was reported in table below and is lower than 20%..

Limit of Quantification: 5 ng/L for water and 17 µg/kg sediment (dry)

Table 4.1.2-71: Alpha-cypermethrin validation results from water and sediment by GC-MS (modified DFG S19)

Sample matrix/analyte	Fortification level	Alpha-cypermethrin recovery (%) Mean (range)	RSD (%)	n
Tap water)/alpha-cypermethrin	5 ng/L	87 (71-95)	16	[3]
	500 ng/L	101 (99-102)	2	3
Sediment/ alpha-cypermethrin	17 µg/kg	106 (103-108)	3	3
	1667 µg/kg	102 (100-106)	3	3

Stability of standard solutions about 13 days and extracts during the course of the study was demonstrated by consistent GC-MS results.
For determination in water : one blank control (tap water) was tested. For determination in sediment: two blank controls (sediment) were tested.

Conclusion: **The modified DFG S19 method (GC-MS) for sediment was already considered as fully validated in the 0.001 – 0.4 mg/kg range (see KCA 8.2.5.3/03). Acceptable validation data were also provided in the 0.1 – 50 mg/kg (3 replicates at each level) (see KCA 8.2.5.4/002 – Study No. 12) and here acceptable validation data are obtained in the 0.017 – 1.7 mg/kg (3 replicates at each level). Overall, the method shows suitable mean recoveries and RSD although the number of replicates at some fortification levels is only 3. Therefore, the method is considered suitable for the determination of alpha-cypermethrin in sediment with an LOQ of 0.001 mg/kg and in a range from 0.001 to 50 mg/kg. The method is therefore considered "fit for purpose" and the ecotox results in study KCA 8.2.5.4/001 are reliable.**

Report:	CA 4.1.2/69 Janson G.-M., 2008 Acute toxicity of BAS 310 51 I to Daphnia magna STRAUS in a 48 hour static test 2008/1010445
Guidelines:	OECD 202, EPA 850.1010
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

The report on the analytical phase (method APL0528/02) was part of the ecotox study report.
Matrix**:** Reconstituted water (M4 medium according to Elendt, prepared based on ultrapure deionized water); characteristics: 2.4 1 mmol/L total hardness, 0.96 mmol/L alkalinity up to pH 4.3, pH = 8.03, 663 µS/cm conductivity.

Report:	CA 4.1.2/70 [redacted] 2008 BAS 310 51 I - Acute toxicity study with the rainbow trout (Oncorhynchus mykiss) in a static system over 96 hours 2008/1009839
Guidelines:	OECD 203, EEC 92/69 A V C 1,EPA 72-1,EPA 850.1075,EEC 86/609,EPA 712-C-96-118,EPA 540/9-82-024
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Note: the report on the analytical phase by Obermann M. 2008, 302191_1, BASF Agricultural Center Limburgerhof, Ecology and Environmental Analytics, Germany, GLP was part of the ecotox study.
Matrix**:** Mixing-water Reconstituted water (M4 medium according to Elendt, prepared based on ultrapure deionized water); characteristics: 1.0 mmol/L total hardness, pH = 7.5-8.5, 250 µS/cm conductivity

Principle of the method APL0528/02:
The samples with a concentration of 5 µg/L (500 mL) were filled into a separating funnel. The glass bottles were washed with 50 mL dichloromethane and were added to the sample in the funnel. After vigorously shaking for 3 minutes, the phases are allowed to separate. The lower phase was run through a filter funnel with sodium sulfate, in a 100 mL round bottom flask. After repeating this procedure with the remaining water phase, the combined organic phases were reduced to dryness by rotary evaporation by 30°C water bath and 615 mbar. The residue in the flask was eluted with 1 mL ethanol, shaken carefully and 4 mL water added. The solution was then injected in HPLC-MS/MS (Waters X-Bridge C18, 50 mm x 3 mm i.d, 3.5 µm particle size; mobile phase A: water/formic acid 1000/1 + 4 mM HCOONH4 and mobile phase B: methanol/formic acid 1000/1 + 4 mM HCOONH4, monitoring: m/z 433_ 191). External calibration was used for quantification.

Table 4.1.2-72: Validation data for method APL0528/02 (LC-MS/MS) to determine alpha-cypermethrin in water

Study/Matrix/ analyte	LOQ	Linearity Range (µg/L) (n) Correlation coefficient r	Fortification levels	Specificity and interferences
CA 4.1.2/69 Reconstituted water (M4 medium according to Elendt)/alpha-cypermethrin	0.0004 mg formulation/L (= 0.00002 mg a.i./L)	0.001 – 0.02 mg BAS 310 51 I/L Range (5 concentrations, Duplicate injection).	Fortified samples were prepared at levels in the 0.0004 – 0.01 mg BAS 310	No significant interference from the blank control was observed. Chromatograms were provided for
		r ≥ 0.998 The corresponding linear regression plot and equation were provided. The concentration of the samples are within the calibration range, otherwise dilution or concentration steps occurred to meet the calibration range	51 I/L range. Mean recovery at each fortification level is presented in the Table below. RSD was not reported but was calculated based on the available results.	the blank solution (M4 algal medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard
CA 4.1.2/70 Mixing-water Reconstituted water (M4 medium according to Elendt)/alpha-cypermethrin	0.01 mg formulation/L (= 0.0005 mg a.i./L)	0.004 – 0.04 mg BAS 310 51 I/L range (4 concentrations, duplicate injection). r ≥ 0.992 Calibration curve and regression equation provided. The concentration of the samples are within the calibration range, otherwise dilution occurred to meet the calibration range	Fortified samples were prepared at levels in the 0.01 – 0.2 mg BAS 310 51 I/L range. Mean recovery at each fortification level is presented in the Table below. RSD was not reported but calculated from the available results.	No significant interference from the blank control was observed. Chromatograms were provided for the blank solution (mixing-water medium), a standard solution and test samples at initiation and end of the test. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard

Note: the absence of matrix effects was not demonstrated but any naturally occurring matrix would have been removed by the extensive liquid-liquid partitioning of the water samples.

Table 4.1.2-73: Mean recovery and RSD for method APL0528/02 (LC-MS/MS) to determine alphacypermethrin in water

Study/Matrix/Analyte	Fortification levels (mg formulation/L)	Mean* recovery % (range)	RSD %*	n*
CA 4.1.2/69 Reconstituted water (M4 medium according to Elendt)/alpha-cypermethrin	0.0004**	110.2 (108.7, 112.5)	1.5	4
	0.01**	98.7 (81.7, 115.6)	18	4
CA 4.1.2/70 Mixing-water Reconstituted water (M4 medium according to Elendt)/alpha-cypermethrin	0.01**	95.6 (88.4-103.9)	8.3	4
	0.2**	92.7 (84.7-100.6)	8.7	4

In KCP 10.2.1/002: The stability in M4 medium over a period of 2 days at room temperature could not be confirmed since slightly degradation was observed. In KCP 10.2.1/001, the stability in water over a period of 4 days at room temperature could not be confirmed since slightly degradation was observed. Two controls were tested concurrently with the test.
* results of fortified samples at the initiation and at the end of the test, each injected in duplicated. Mean recovery and RSD recalculated based on the results at the initiation and at the end of the test.
** corresponding to ~ 0.00002 mg a.s./L and 0.0005 mg a.s./L, respectively in KCP 10.2.1/001 and corresponding to ~ 0.0005 mg a.s./L and 0.01 mg a.s./L, respectively in KCP 10.2.1/001.

Conclusion: **Mean recoveries and RSD at each fortification level tested were within the acceptable limits according to SANCO/3029/99 rev.4. for the determination of alpha-cypermethrin by LC-MS/MS according to method APL0528/02. The method is therefore considered as sufficiently validated and supports the corresponding ecotox results.**

Report:	CA 4.1.2/71 [redacted] 2008 BAS 310 51 I - Acute toxicity in the bobwhite quail (Colinus virginianus) after single oral administration (LD50)
	2008/1051526
Guidelines:	EPA 71-1, EPA 540/9-85-007,EPA 540/9-82-024,EPA 850.2100
GLP:	Yes (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the method:

Samples were diluted with acetonitrile/highly deionized water 1/1 v/v. Aliquots of the dilutions were used for HPLC-UV analysis (column: Luna 2.5 µm C18(2)-HST, 50 m x 2 mm for determination during the test and Gemini 3 µm C18, 50 ×2 mm in the analytical phase report, eluent 65% acetonitrile + formic acid (1,000 + 1 mL)/35 % highly deionized water + formic acid (1,000 + 1 mL, detection at 202 nm). External calibration was used for quantification. Matrix was drinking water.

Specificity/Interference	No interferences occurred at the retention time of the analyte in the control sample (drinking water). Chromatograms were provided for standard solutions, blank and samples. The identity was confirmed on the basis of the selected wavelength and the retention time. The method is used for dose verification of known substances and known nominal concentrations. Hence, no additional confirmatory technique is necessary.
Linearity:	Method was found to be linear : in the ~ 0 – 150 mg/L and ~ 0 – 475 mg/L range (3 different concentrations, duplicate injection). Calibration in acetonitrile/highly deionized water 1/1 v/v, further diluted with acetonitrile. The calibration curve was provided but not the regression equation and the correlation coefficient. However, since the calibration curve showed good linearity, no further data are required.
Limit of Quantification:	83.5 mg/L (lowest calibration standard concentration, corresponding to 4.1% w/w).

Table 4.1.2-74: Dose verification and stability data for BAS 310 51 I in drinking water

Sample matrix/analyte	Nominal concentraton %w/w (g formulation/100g)	Nominal concentration in mg formulation/kg bdw Mean (range)	% of nominal concnetration	N*
Drinking water /BAS 310 51I	**Dose verification**			
	5 (corr. to 100 mg/L)	500	96.6	2
	10 (corr. to 200 mg/L)	1000	96.2	2
	20 (corr. to 4000 mg/L)	2000	96.0	2
Sediment/ alpha-cypermethrin	**Stability**			
	0.99 (0h)	-	100.0	2
	0.99 (6h)	-	.102.0	2

Conclusion: **83.5 mg/L (lowest calibration standard concentration, corresponding to 4.1% w/w the method is considered to be "fit for purpose" taken into account the overall results as the absence of interferences and the good recoveries obtained. Additionally, it is noted that the same method has been used in study KCA 8.1.1.1/01 for the determination in 0.5% CMC in drinking water and with similar validation data at similar levels. The results of study KCP 10.1.1.2/001 are therefore supported by the analytical method.**

Report:	CA 4.1.2/72 Liepold K., 2010 Assessment of side effects of BAS 310 55 I on the honey bee (Apis mellifera L.) in the field 2009/1050236
Guidelines:	OEPP/EPPO Guideline No. 170 (3) (2001)
GLP:	Yes (Landesamt fuer Umwelt, Messungen und Naturschurz, Baden-Wuerttemberg, Karlsruhe, Germany)

Report:	CA 4.1.2/73 Schmitzer S., 2010 Toxicity testing BAS 310 55 I on honey bees (Apis mellifera L.) in the field 2009/1050237
Guidelines:	OEPP/EPPO Guideline No. 170 (3) (2001)
GLP:	Yes (Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden)

Remark: CA 4.1.2/69 (2016/123573) is also considered here.

Report:	CA 4.1.2/74 Bertram S., 2014 Determination of residues of BAS 310 55 I in nectar, pollen and flowers of winter oil seed rape after one application in Germany 2013 2014/1000203
Guidelines:	EEC 7029/VI/95 rev. 5,EU Regulation 1107/2009 with Regulation 283/2013,EU Regulation 1107/2009 with Regulation 284/2013, SANCO/3029/99 rev 4, SNACO/825/00 rev. 8.1; ENV/JM/MONO(2007)/17
GLP:	Yes (Landesamt fuer Umwelt, Messungen und Naturschurz, Baden-Wuerttemberg, Karlsruhe, Germany)

Report:	CA 4.1.2/75 Mack P., 2014 Determination of residues of BAS 310 55 I (Alpha-Cypermethrin) in nectar, pollen and flowers of winter oilseed rape after one application in a semi-field residue study with honeybees (Apis mellifera L.) in Germany 2014 2015/1000382
Guidelines:	EEC 91/414 (1607/IV/97 Rev. 2), EU Regulation 1107/2009 with Regulation 283/2013,EU Regulation 1107/2009 with Regulation 284/2013,SANCO/3029/99 rev. 4 (11 July 2000), SANCO/3029/99 rev 4
GLP:	Yes (Landesamt fuer Umwelt, Messungen und Naturschurz, Baden-Wuerttemberg, Karlsruhe, Germany)

Principle of the method L0020/01 = 567/0 (Method for determination of Alpha-cypermethrin in plant matrices):

All matrices except for commodities with high fat content:
Alpha-cypermethrin or cypermethrin is extracted with a 70:25:5 methanol/water/hydrochloric acid (2N) mixture. After centrifugation, an aliquot of the extract is partitioned into cyclohexane. Silica gel SPE column is used for further purification, if required. Final determination is performed by LC-MS/MS after reconstitution in methanol/water 80/20 v/v using the ammonium adduct of cypermethrin (Phenomenex Columbus C18 column, 2.0 mm × 100 mm, 5 µm particle size, mobile phase A: 4 mM ammonium formate + 0.1% formic acid in water and mobile phase B: 4mM ammonium formate + 0.1% formic acid in methanol with gradient elution [0 _ 0.5 min: 70/30, 0.6 → 4.0: 5/95, 4.2 → 6.0: 70/30], ESI positive mode for matrices containing little to no matrix interferenceor Phenomenex Synergi Max-RP column, 4.0 mm × 20 mm, 2 µm particle size, mobile phase A: 4 mM ammonium formate + 0.1% formic acid in water and mobile phase B: 4mM ammonium formate + 0.1% formic acid in methanol with gradient elution [0 min: 40/60, 0.6; 4 → 5 min: 0/100, 5.1 → 7.0: 40/60], ESI positive mode for matrices requiring more separation of matrix interferences). External calibration was used for quantification.
For commodities with high fat content:
Alpha-cypermethrin or cypermethrin is extracted with acetonitrile/n-hexane. After centrifugation and further partitioning step with n-hexane, the acetonitrile phase is diluted for LC-MS/MS determination (LC-MS/MS characteristics: see above). Due to the low amount of sample material the following changes to the method were made in studies KCP 10.3.1.6/001 and /005: Pollen: 0.5 g of pollen was extracted with 10 mL acetonitrile and 4 mL iso-hexane. Nectar: 1 g of nectar was extracted with 10 mL methanol/water 80/20. An aliquot of 1 mL of the extract was partitioned into 5 mL cyclohexane.
For study KCP 10.3.1.6/007, the method for all matrices except for commodities with high fat content was used;
no SPE clean-up was necessary. Minor modifications were used compared to the actual method:

- 0.25g pollen and nectar and 2.5g flowers was used instead of 5g in method 567/0
- The same extraction solvent was used but different volumes because of the different weight. The ratio between volume and weight was retained.
- The principle of the following clean-up is the same; only used volumes are different: 2 mL aliquot of the extract was used instead of 1 mL. In both cases 5 mL cyclohexane were used for liquid/liquid extraction. 5 mL of the cyclohexane phase was used for further steps instead of 3 mL. The cyclohexane was evaporated to dryness and taken up with 2 mL methanol/water instead of 1 mL.
- A similar but quiet different mobile phase was used. In the actual method water + 0.1% acetic acid, methanol + 0.1% acetic acid and 5 mM ammonium acetate in methanol were used. In method 567/0 ammonium formate + 0.1% formic acid in water and 4 mM ammonium formate + 0.1% formic acid in methanol was used.

Mobile phase composition and gradient elution at 0.35 mL/min as follows:

Time (min)	Mobile phase A : water+ 0.1% acetic acid	Mobile phase B: 5 mM ammonium acetate in methanol	Mobile phase C: methanol + 0.1% ascetic acid
0.00	11%	4%	85%
7.01	1%	4%	95%
9.01	1%	4%	95%
9.03	11%	4%	85%
11.00	11%	4%	85%

- Calibration standards were prepared in acetonitrile/water 1/1 v/v instead of methanol/water 80/20 v/v in 567/0.
- In 567/0, two possible columns were presented: Phenomenex Columbus C18 100 mm ×2 mm, 5 µm or Phenomenex Synergi Max RP 20 mm ×4 mm, 2 µm. In study KCP 10.3.1.6/007, the column Phenomenex Ultracarb 5 µm ODS 150 mm × 3.2 mm, 5 µm was used.
- For Study KCP 10.3.1.6/008, the SPE clean up step only occurred for pollen samples. Modifications were made to the original method 567/0 in order to adapt the method to the needs of the study (improvement of the extraction and reduction of matrix effects):
- The stock and fortification solutions were prepared in acetonitrile + 0.1% formic acid instead of acetone in 567/0.
- The whole nectar material (0.07 – 1.6g), approx. 0.2 g of pollen and 2.0 g of flower material were used for the extraction instead of 5 g in 567/0.
- The same extraction solvent was used. The ratio between the weight of the sample and extraction solvent volume was retained (1/20). Extraction (for pollen and flower samples) was done twice instead of one extraction with 567/0. Nectar samples were extracted one time and the ratio between the weight of the sample and extraction solvent volume was 1/40.
- The principle of the clean-up by liquid/liquid partition with cyclohexane was the same; only volumes of the extracts were different: 2 mL aliquot was taken instead of 1 mL. In both cases, 5 mL cyclohexane were used for liquid/liquid partition and 3 mL of the cyclohexane phase were used for the further steps. The cyclohexane was evaporated to dryness and reconstituted in 3 mL methanol/water 80/20 v/v instead of 1 mL.
- The column Phenomenex Ultracarb 5 µm ODS (30) 50 mm ×4.6 mm was used. The column is similar to Phenomenex Columbus C18 100 mm ×2 mm, 5 µm used in 567/0.

Mobile phase composition and gradient elution at 0.35 mL/min as follows:

Time (min)	Mobile phase A :4 mM ammonium formate in water +0.1% formic acid	Mobile phase B: 4 mM ammonium formate in methanol + 0.1% formic acid
0.00	10%	90%
7.00	1%	99%
10.00	1%	99%
10.01	10%	90%
12.00	10%	90%

Matrices: Nectar, pollen, flowers

Specificity/Interference:	No significant interference from the blank controls (nectar [combs, forager bees], pollen [combs and forager bees], flowers, nectar and pollen) was observed except in one sample of pollen (forager bees) in the study No. 365576 (part of study S09-00811 – KCP 10.3.1.6/001). A "retain" control specimen was therefore analysed and no residues of alpha-cypermethrin were obtained. Chromatograms were provided for control samples (nectar only), a standard solution, a fortified sample at the LOQ for nectar and pollen and test samples of nectar and pollen for study No. 365576 (part of study S09-00811) and for control samples (nectar and pollen), a standard solution, a fortified sample at the LOQ for nectar and pollen and test samples of nectar and pollen for study No. 365577 (part of study 50391040 – KCP 10.3.1.6/005). In study S13-01463, chromatograms were provided for a standard solution, controls (pollen, nectar and flower), fortified nectar/pollen/flower at 0.01 mg/kg and treated nectar/pollen/flower. In these chromatograms, two peaks are visible whereas they were not visible in the chromatograms of the other studies. The second peak is interference/matrix separated from the analyte and this was already observed in the original validation of method 567/0 in dry and fat matrices. In study S14-00950, chromatograms were provided for standards (calibration in solvent and matrix-matched calibration), controls (nectar/pollen/flower), fortified nectar/pollen/flower at 0.01 mg/kg and treated nectar/pollen/flower at both mass transitions. The retention times of the reference item standards matched the retention times in extracts from fortified samples and from treated samples. The method is highly specific by the monitoring of two m/z transitions.

Linearity:	In study No. 365576, method was found to be linear: r ≥ 0.999 for both transitions in both matrices. Correlation coefficients together with regression equations were provided for both transitions and for both matrices. A typical calibration curve was only presented for the 433 _ 191 transition in nectar in the ~ 0.005 – 2.5 ng/mL range (n = 6, duplicate injection). Calibration occurred in solvent. Matrix-matched quality control samples match solvent standard at a concentration of 0.005 mg/kg and no significant matrix effects were therefore observed. The concentration of the samples were within the calibration range. In study No. 365577, a typical calibration curve was also provided for the 433 → 191 m/z transition in the 0.005 – 2.5 ng/mL range (n = 6, duplicate injection, r > 0.999). Calibration occurred in solvent. The absence of matrix effects was not demonstrated within the study. The analysis was conducted using the same analytical method as in Study No. 365576 in which matrix effects were addressed. In study S13-01463, method was found to be linear in the 0.02 – 100 ng/mL range with correlation coefficient > 0.999 (n = 10, single injection, calibration in acetonitrile/water 1/1 v/v, calibration curve and regression equation provided). The report mentioned that no significant matrix effects were observed (matrix effects < 20%) allowing the use of calibration in solvent. The raw data were however not reported. The concentration of the samples are within the calibration range. Dilution may occur if necessary. In study S14-00950, method was found to be linear in the 0.03 – 100 ng/mL for nectar (n=9, single injection, calibration in methanol/water 80/20 v/v, r =0.9998, m/z 433→191), in the 0.03 – 100 ng/mL for pollen (n=9, single injection, matrix-matched calibration, r = 0.9995, m/z 433 → 191) and in the 0.03 – 100 ng/mL in flowers (n = 9, single injection, matrix-matched calibration, r = 0.9994, m/z 433→191). Matrix effects (based on the mean response factor obtained with calibration in solvent and matrix-matched calibration at different levels) were not found for nectar samples. In the contrary, significant matrix effects were observed for analysis of pollen and flower samples (24 and 20%, respectively) leading to the use of matrix matched calibrations for these matrices.

Accuracy: Fortified samples were prepared at levels in the 0.01 – 0.1 mg/kg range. Mean recovery at each fortification level is presented in the Table below. Some procedural recoveries were also performed in the course of the study No. 365576 (KCP 10.3.1.6/001): recoveries in nectar (combs) were found to be 88.8 (n = 1) and 96% (n = 1) at 0.01 and 0.1 mg/kg, respectively and recoveries in pollen (combs) were found to be 60.8 and 83.6 % (n = 2, mean = 72.2%) at 0.01 mg/kg and 75 and 79.6% (n = 2, mean = 77.3%) at 0.1 mg/kg. Procedural recoveries were also reported in study No. 365577 (KCP 10.3.1.6/005): recoveries in nectar were found to be 71.3 (n = 1) and 86 % (n = 1) at 0.01 and 0.1 mg/kg, respectively and recoveries in pollen were found to be 75.2 and 70.0 % (n = 2, mean = 72.6%) at 0.01 mg/kg and 74.2 and 80.2% (n = 2, mean = 77.2%) at 0.1 mg/kg.

Repeatability: The RSD was below 20% in each case.

Limit of Quantification: 0.01 mg/kg in each matrix

Table 4.1.2-75: Alpha-cypermethrin validation results from nectar and pollen by method L0020/01 (Study CA 4.1.2/73)

Sample/analyte	Fortification level (mg/kg)	m/z 433 ⟶ 191			m/z 435 ⟶ 193		
		Mean recovery % (range)	RSD %	n	Mean recovery % (range)	RSD %	n
Nectar/alpha-cypermehtrin	0.01	88.3 (86.8-89.5)	1.2	5	89.2 (84.3-92.0)	3.3	5
	0.1	91.3 (88.0-98.0)	4.3	5	91.0 (87.5-96.0)	3.5	5
Pollen/alpha-cypermehtrin	0.01	82.5 (76.8-88.8)	5.8	5	89.7 (87.2-92.0)	2.3	5
	0.1	81.0 (76.2-85.8)	5.1	5	81.7 (76.4-86.2)	4.4	5

Control specimens of pollen and nectar were received and analysed. The residue concentration in ine control specimen (pollen from forager bees) equaled the concentration of residues detected in the treated specimen. A “retain” control specimen was therefore analysed and no residues of alpha-cypermethrin were obtained.

Table 4.1.2-76: Procedural recoveries of alpha-cypermethrin (quantifier and qualifier transitions) in nectar, pollen and flowers (CA 4.1.2/75 – study S13-01463

Sample/analyte	Fortification level (mg/kg)	m/z 433 ⟶ 191			m/z 435 ⟶ 193		
		Mean recovery % (range)	RSD %	n	Mean recovery % (range)	RSD %	n
Nectar*/alpha-cypermehtrin	0.01	75 (56-85)	16	5	73 (51-84)	18	5
	0.1	102 (102)	-	4	104 (100-106)	2	4
	1	86 (70-106)	18	5	88 (69-108)	20	5
Pollen/alpha-cypermehtrin	0.01	78 (70-92)	11	5	88 (77-99)	11	5
	0.1	101 (99-103)	2	4	100 (97-103)	3	4
	15	90 (75-98)	10	5	89 (73-97)	11	5
Flowers/alpha-cypermehtrin	0.01	70 (56-91)	19	5	78 (61-96)	16	5
	0.1	85 (78-91)	6	4	86 (78-90)	6	4
	10	71 (60-84)	16	5	71 (61-83)	15	5

* prepared by dilution of honey with water (honey/water 1/2 v/v).
The most sample extracts were analysed within 24 hours after extraction and stability was not checked. Some sample extracts were analysed within 72 hours. The recoveries of these samples show values approx. 100% and therefore were regarded as stable.

Table 4.1.2-77: Procedural recoveries of alpha-cypermethrin (quantifier transition, results for qualifier transition were not reported) in nectar, pollen and flowers (CA 4.1.2/77 – study S14-00950)

Sample/analyte	Fortification level (mg/kg)	m/z 433 →191		
		Mean recovery % (range)	RSD %	n
Nectar*/alpha-cypermethrin	0.01	93 (82-101)	8	5
	5	93 (86-102)	8	5
Pollen/alpha-cypermethrin	0.01	81 (71-87)	8	5
	15	70 (67-74)	5	5
Flowers/alpha-cypermethrin	0.01	77 (61-91)	17	5
	15	71 (51-83)	20	5

* prepared by dilution of honey with water (honey/water 1/3 v/v).
Three control samples for pollen and two control samples for nectar and flowers.
Nectar and flower extracts were analysed within 24 hours after extraction and stability was not checked. A few pollen extracts were analysed within 96 hours storage at 15°C in the autosampler vials. The mean recovery obtained after 96 hours storage is in the same range as the mean recovery obtained for fresh samples. Samples were therefore considered stable for at least 96 hours.

Conclusion: **The method L0020/01 (567/0) LC-MS/MS appears to be fully validated according to SANCO/3029/99 rev. 4 with an LOQ of 0.01 mg/kg in pollen and in nectar, each. Mean recoveries and RSD are all within the acceptable limits and additional validation data (procedural recoveries) from studies S13-01463 (CA 4.1.2/75=KCP 10.3.1.6/007) and S14-00950 (CA 4.1.2/77=KCP 10.3.1.6/008) confirm that the method is suitable and supports the residue trials. For flowers, mean recoveries and RSD obtained are within the acceptable limits. It should however be noted that some individual recoveries (two to three at LOQ and at 10 mg/kg level) were found to be below 70%. Nevertheless, since the mean recovery for these higher levels and RSD remain acceptable even if sometimes close to the limit, the method is considered as sufficiently validated and supporting the residue trials.**

Report:	CA 4.1.2/76 Class T., 2007 Residues of Alpha-Cypermethrin in honey and pollen 2007/1013272
Guidelines:	EEC 91/414 Annex II (Part A Section 4.2), EEC 91/414 Annex III (Part A Section 5),SANCO/825/00 rev. 7 (17 March 2004),SANCO/3029/99 rev. 4 (11 July 2000), SANCO/3029/99 rev.4
GLP:	Yes (Landesamt fuer Umwelt, und Verkehr Baden-Wuerttemberg, Stuttgart, Germany)

Report:	CA 4.1.2/77 Class T., Richter S., 2016 Expert Statement - Residues of Alpha-Cypermethrin in honey and pollen 2016/1232650
Guidelines:	not stated
GLP:	not stated

Principle of the method:
Pollen (0.5 g) was extracted overnight by 10 mL of acetone/dichloromethane 1/1 v/v, the extract was filtered over sodium sulfate, concentrated to dryness, and re-dissolved in methanol/water 8/2 v/v. Honey (nectar) (1 g) was extracted with 30 mL of water/acetone 1/2 v/v, NaCl and 10 mL of ethyl acetate/cyclohexane 1/1 v/v were added to partition the analyte into methanol/water 8/2 v/v. LC-MS/MS monitored two parent-daughter ion MRMS for quantitation (433 to 191) and confirmation (435 to 95) (Phenomenex Synergy Max RP, 20 mm ×2.0 mm, 2 µm; pre-column: Phenomenex C18, 4 mm ×3 mm, 5µm). External calibration was used for quantification.
Matrix: Pollen and Nectar.

Mobile phase composition at a flow rate of 600 µL/min:

Time (min)	Mobile phase A :4 mM ammonium formate in water +0.1% formic acid	Mobile phase B: 4 mM ammonium formate in methanol + 0.1% formic acid
0.00	40%	60%
1.00	40%	60%
4.00	0%	100%
5.00	0%	100%
5.10	40%	60%
8.00	40%	60%

Specificity/Interference: No significant interference from the blank control (nectar and pollen) was observed Chromatograms were provided for control samples (nectar and pollen), standard solutions in solvent, fortified samples at the LOQ and 10 ×LOQ for nectar and pollen and treated samples of pollen. Chromatograms were also provided for calibration in honey/nectar and in pollen. The method is highly specific by the monitoring of two m/z transitions.

Linearity:	Method was found to be linear : r ≥ 0.9998 for both transitions when calibration was prepared in solvent in the 0.1 – 10 ng/mL range (n= 3, duplicate injection). The study reports also mentioned that calibration were prepared with matrix-matched standards in the same concentration range but the correlation coefficient, together with the calibration curves and regression equations were not provided. It is also not clearly stated in the study report if the matrix-matched calibration was used to determine the concentration of alphacypermethrin in the samples and the matrix effects were not considered in order to allow the use of the calibration in solvent. In the expert statement (BASF DocID 20161232650), states that for honey/nectar, samples injected were all evaluated via matrix-matched standards whereas for pollen both calibrations (in solvent for initial injections and with matrix-matched standards for later injections) were used. Matrix effects in honey/nectar at a concentration of 10 ng/mL were found to be not significant (< 20%) but for pollen, the matrix effects were insignificant for the 433 to 191 m/z transition but not for for the 435 _ 193 m/z transition for a later injection series. Since the transitions 433 to 191 was used to quantify the pollen samples, standards in solvent as well as matrix-matched standards were considered appropriate. Correlation coefficients- together with the calibration curves and regression equations were finally also provided for matrix-matched (honey/nectar and pollen) calibrations for both transitions (n = 5 in the 0.1 – 100 ng/mL range, r ≥ 0.9969). The concentration of the samples was within the calibration range after appropriate dilution if required.
Accuracy:	Fortified samples were prepared at levels in the 0.01 – 0.1 mg/kg range. Mean recovery at each fortification level in each matrix is presented in the Table below.
Repeatability:	RSD was below 20 % except in one case. See Table below.
Limit of Quantification:	0.01 mg/kg for pollen and nectar.

Table 4.1.2-78: Alpha-cypermethrin validation results from nectar and pollen by LC-MS/MS

Sample/analyte	Fortification level (mg/kg)	m/z 433 →191			m/z 435 →193		
		Mean recovery % (range)	RSD %	n	Mean recovery % (range)	RSD %	n
Nectar (honey)/alpha-cypermethrin	0.01	97 (88-113)	11	4	87 (67-110)	23	4
	0.1	108 (103-113)	4	4	106 (99-112)	5	4
Pollen/alpha-cypermethrin	0.01	84 (68-95)	15	5*	92 (81-114)	13	7
	0.1	82 (66-92)	17	3	84 (72-94)	13	3

Control specimens of pollen and nectar were received and analysed.
* Two samples from the five were not taken into account to calculate the recovery based on the average and RSD as the response for this mass transition (but not for the 2nd transition) was presumably suppressed by an exceptional matrix effect.

Conclusion: **Together with the levels found in pollen and nectar in the corresponding residue studies (levels between 0.003 – 0.026 mg/kg for pollen, no residue found for nectar in studies performed by IBACON), the method can be considered as sufficiently accurate and precise and can be considered suitable to support the residue results on pollen and nectar.**

Report:	CA 4.1.2/78 Kleebaum K., 2014 Chronic toxicity of BAS 310 I honeybee larvae Apis mellifera L. under laboratory conditions (in vitro) 2014/1162697
Guidelines:	OECD 237 (2013) Honey bee (Apis mellifera) larval toxicity test single exposure, OECD Draft Test Guideline on Honey bee (Apis mellifera) Larval Toxicity Test Repeated Exposure (February 2014)
GLP:	Yes, (Saechsisches Staatsministrerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Principle of the method:

Verification of the test stock solution's concentration of alpha-cypermethrin in acetone was performed by reversed phase HPLC-UV with detection at 277 nm (Macherey Nagel Nucleoshell RP18, 2.7 mm × 100 mm, 2.7 µm). External calibration was used for quantification.

Mobile phase composition at a flow rate of 400 µL/min:

Time (min)	Mobile phase A: water + 0.1% (v/v) phosphoric acid (85%)	Mobile phase B: acetonitrile + 0.1% (v/v) phosphoric acid (85%)
0.00	40%	60%
7.00	10%	90%
8.00	10%	90%
8.01	40%	60%

Specificity/Interference:	No significant interference from the blank control was observed Chromatograms were provided for blank, standard solutions, fortified samples at the lowest and highest levels and sample solution to analyse. Identity was confirmed by comparison of the retention time and UV spectrum of the reference with the test item.
Linearity:	Method was found to be linear: r = 0.999 in the 3.77 – 11.42 mg/mL range (n= 5, single injection). The calibration curve and the regression equation were provided. The concentration of the samples were within the calibration range after appropriate dilution.
Accuracy:	Fortified samples were prepared at levels in the 71.4 – 154 mg/L range (corresponding to 4.75 mg/L and 9.54 mg/L after dilution). Mean recovery at each fortification level in each matrix is presented in the Table below.
Repeatability:	RSD was below 20 % except in one case. See Table below.
Limit of Quantification:	71.4 mg/L (4.75 mg/L after dilution).

Table 4.1.2-79: Alpha-cypermethrin validation results from acetone by HPLC-UV

Matrix/analyte	Fortification level (mg/L)	Mean recovery % (range)	RSD %	n
Acetone/alpha- cypermethrin	71.4	101 (100-102)	0.6	5
	154	102 (102-103)	0.7	5

Two blank samples were analysed and test item was not detected (< 30% of the LOQ).

Conclusion: **The HPLC-UV method is considered validated according to SANCO/3029/99 rev. 4 and is suitable to determine the content of alpha-cypermethrin in acetone with an LOQ of 71.4 mg/L (4.75 mg/L after dilution). The method was used for concentration control of the stock solution used to prepare feed administrated to bees in support of toxicity studies on the bees.**

Report:	CA 4.1.2/79 Class T., 2014 Outdoor aquatic mesocosm study with ME solo-formulation of Alpha-Cypermethrin (BAS 310 55 I, nominal 50 g/L) - Preparation and verification of dose solutions, analysis of water and sediment samples 2014/1246534
Guidelines:	SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	Yes, (Landesamt fuer Umwelt, Messungen und Naturschutz, Baden-Wuerttemberg, Karlsruhe, Germany)

Report:	CA 4.1.2/80 Class T., 2014 Expert Statement on aquatic mesocosm study, PTRL Ulm Germany: Outdoor aquatic mesocosm study with ME Solo-Formulation of Alpha-Cypermethrin (BAS 310 55 I, nominal 50 g/L) - Preparation and verification of dose solutions, analysis of water and sediment samples 2016/1232658
Guidelines:	not stated
GLP:	nit stated

Principle of the method:

One objective of the study was to prepare and verify dose solutions to be used for two application events in the Mesocosm GmbH Study No. 233-01 and ti verify the decline of alpha-cypermethrin in the mesocosm study. A second objective was to determine alpha-cypermethrin and its epimers in water and sediment samples originating from the above mentioned mecocosm study. For dose verification, aliquots were extracted 3-times with n-hexane or toluene, the combined extracts were concentrated to dryness and then redissolved in an appropriate volume of 10 ng/mL internal standard (IS) solution in toluene for final determination by GC/MS(NCI). For mecocosm enclosure water 1.0L of water samples were extracted in separatory funnels 4-times, each time with 50 mL of dichloromethane, also used to rinse sample bottles with screw-cap in order to extract any adsorbedanalyte. The combined organic extract was transferred into a 250-mL round bottom flask and concentrated using a rotary evaporator to about 5 mL. The remaining extract volume was transferred into a 15-mL glass centrifuge vial, another 5 mL of dichloromethane were used to rinse the round bottom flask. Using a stream of nitrogen with water bath at 40°C the extract was further concentrated to about 1 mL and transferred completely into an autosampler vial and concentrated with a stream of nitrogen to dryness. The final extract volume was then adjusted to 50 µL or 1.0 mL with 10 ng/mL IS solution (vortex and sonicate) for GC/MS (NCI) injection (Varian VF-5ms, 30 m ×0.32 mm, 0.25 µm, oven temperature: 90°C, 2 min hold, ramp with 40°C/min to 250°C, ramp with 5°C/min to 280°C, ramp with 100°C/min to 320°C, 4 min hold, monitored ion for alpha-cypermehtrin : 207 m/z and for internal standard lambda-cyhalothrin: 241 m/z). The method is nearly the same as in KCA 8.2.5.4/001 (sudy 12CT1LA – Study No. 13 here above). For sediment, a modified DFG S19 extraction/partition method approach was used. For mesocosm sediment samples, 10g of wet sediment was weighed and 10 mL of water was added. This mixture was then extracted with 20 mL of acetone and shaken for 30 min. with a horizontal shaker. Then about 3.5 g of sodium chloride and exactly 10 mL of ethyl acetate/cyclohexane 1/1 v/v were added for salting out and homogeneous liquid/liquid partition by shaking for another 30 min and centrifugation for 2 min. at 4000 rpm. Following phase separation after about 5 min, an aliquot of 14 mL of the upper organic phase was dried by filtration over anhydrous sodium sulfate and evaporated to about 1.5 mL.

The 1.5 mL were transferred into a 2-mL autosampler vial and subsequently concentrated to dryness using a N2-evaporator. The final extract volume was then adjusted to 0.2 mL with 10 ng/mL IS solution (vortex, sonicate and centrifugation using micro centrifuge filter) for GC/MS (NCI) injection (Varian VF-5ms, 30 m ×0.32 mmm, 0.25 µm, oven temperature: 90°C, 2 min hold, ramp with 40°C/min to 250°C, ramp with 5°C/min to 280°C, ramp with 100°C/min to 320°C, 4 min hold, monitored ion for alpha-cypermethrin: 207 m/z and for internal standard lambda-cyhalothrin: 241 m/z). The method was previously used and validated in study 383065 (KCA 8.2.5.3/002). The GC/MS(NCI) is able to distinguish the epimers (Cis-I) originating from epimerisation of alpha-cypermethrin
Matrix**:** natural water and sediment.

Specificity/Interference:	No significant interference from the blank control water was observed. Chromatograms were provided for the blank solution (tap water), standard solutions, fortified samples at 0.05 and 500 ng/L and mesocosm water samples. No significant interference from the blank control sediment was observed. Chromatograms were provided for the blank control (sediment), standard solutions (same as above), fortified sediment at 0.001 and 0.1 mg/kg and a mesocosm sediment sample. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
Linearity:	Method was found to be linear: r^2 = 0.9982 in the 0.3 – 100 ng/L range (5 concentrations, single/duplicate/triplicate injections depending on the concentration). Calibration occurred in solvent (same calibration for water and sediment) and the absence of matrix effects was not reported in the report but it has been stated that "*Calibration via internal standardization, where the IS is present in the extracts as well as in the calibration solutions always in nominally the same concentration, compensates in GC/MS for the following effects: Varying evaporation in the GC injection system caused e.g. by matrix components, thus it is not necessary to prepare matrix-matched standards. Thus due to the use of the IS very similar to the actual analyte, matrix-effects were not specifically assessed during the study. Furthermore, acceptable recoveries obtained throughout the study for water as well as for sediment samples indicate that the use of the IS in the final extracts and calibration solutions completely compensate for any varying matrix effect on GC injection or NCI detector response*". The corresponding linear regression plot and equation were provided. The concentration of the samples are within the calibration range, otherwise dilution occurred to meet the calibration range (mainly for higher levels in sediment samples).

Accuracy: Verification of dose solutions for mesocosm applications event 1 to 3 (extraction with n-hexane or toluene) lead to 86 to 105% (mean = 93%) of the nominal concentrations (0.19 – 140 ng/mL). Fortified samples of water were prepared at levels in the 0.05 – 500 ng/L range. Mean recovery at each fortification level is presented in the Table below. Fortified samples of sediment were prepared at levels in the 0.001 – 0.1 mg/kg range. Mean recovery at each fortification level is presented in the Table below.

Repeatability: RSD was below 20 %.

Limit of Quantification: 0.05 ng/L for water; 0.1 µg/kg for sediment (wet weight)

Table 4.1.2-80: Alpha-cypermethrin validation results from water and sediment by GC-MS(NCI)

Matrix/analyte	Fortification level	Recovery (5)	RSD %	n
Tap /pond water / alpha-cypermethrin	0.05 ng/L	105 (79-127)	11	10
	500 ng/L	87 /73-99)	13	4
Sediment /alpha-cypermethrin	0.0001 mg/kg	102 (95-116)	8	5
	0.1 mg/kg	87 (82-92)	-	2

Eight blank controls were analysed for water.
Two blank controls were analysed for sediment.

Conclusion: **The modified DFG S19 method (GC/MS(NCI)) has already been considered as fully validated in the 0.001 – 0.4 mg/kg range (see KCA 8.2.5.3/002). The validation results obtained here at the 0.1 mg/kg level confirmed the previous obtained validation results. Additionally, the current validation data indicates that a LOQ of 0.0001 mg/kg instead of 0.001 mg/kg is achievable. Both methods support the ecotox results.**

Report:	CA 4.1.2/81 Schulz L., 2012 Effects of BAS 310 55 I on earthworms under field conditions 2012/1000062
Guidelines:	SO/CD 11268-3 (1999), Kula et al. (2006) - Technical Recommendations for the Update of the ISO Earthworm Field Test Guideline (ISO 11268-3), SANCO/3029/99 rev.4
GLP:	Yes, (Saechsisches Staatsministerium fuer Umwlet undLandwirtschaft, Dresden, Germany)

Principle of the method:

The scope of analytical method was to quantify alpha-cypermethrin in soil after application of BAS 310 55 I. Alpha-cypermethrin was quantified after extraction from soil using a mixture of acetone and hexane according to slightly modified method SAMS 354-2 "Determination of Residues of alpha-cypermethrin in soils – Gas chromatographic method" and "Alphacypermethrin (CL 900049): validation of Method SAMS 354-2 for the determination of Residues in soil" (BASF DocID 1999/7001547). The scale of the method was reduced by a factor 2.5 (e.g. 20 g soil instead of 50 g soil, 120 mL extraction solvent instead of 300 mL) and the clean-up using a Florisil column was omitted because not necessary. Quantification occurred by GC-MS (Rxi 5 SIL ms by Restek, 20 m ×0.18 mm, 0.18 µm, oven program: 120°C hold 1 min, ramp 40°C/min to 240 °C hold 0.00 min then ramp of 10°C/min to 290°C, hold 4.00 min, ions 209, 207 and 171)
Matrix: Soil.

Specificity/Interference:	No significant interference from the control soil was observed. Chromatograms were provided for the three ions for the control (soil), a standard solution, fortified samples at 0.005 mg/kg and treated samples. The identity was confirmed by comparison of the retention time in the test sample with the retention time of the standard.
Linearity:	Method was found to be linear: r > 0.9994 for the three ions in the 0.125 – 5.99 ng/mL range (6 or 7 concentrations). Matrix-matched calibration was used. The corresponding linear regression plots and equations were provided.
Accuracy:	Method was found to be linear: r > 0.9994 for the three ions in th Procedural recoveries were performed at 0.005 mg/kg and 0.05 mg/kg. Mean recovery at each fortification level is presented in the Table below.
Limit of Quantification:	0.005 mg/kg dry soil.

Table 4.1.2-81: Alpha-cypermethrin validation results from soil by GC/MS (method SAMS 354-2, monitored ions: m/z 209 for quantitation, 207 and 171 for confirmation) – procedural recoveries

Sample matrix/analyte	Fortification level (mg/kg dry soil)	m/z 209			m/z 207			m/z 171*		
		Mean recovery % (range)	RSD (%)	n	Mean recovery % (range)	RSD (%)	n	Mean recovery % (range)	RSD (%)	n
Soil/alpha-cypermethrin	0.005	81.3 (72-87)	10	3	79.0 (76-84)	5.6	3	66.3 (57-75)	13.6	3
	0.05	94.3 (87-105)	10.1	3	96.3 (90-106)	8.8	3	92.0 (85-105)	12.3	3

Four controls were analysed.
Dilute solutions of the stock solution are stable on storage for at least 3 months when stored in dark at ambient temperature.
* m/z 171 was of very low intensity explaining the difficulties to obtained acceptable recoveries at the lowest level in this case.

Conclusion: **A slightly modified method compared to the method SAMS 354-2 was used here. SAMS 354-2 (GC-ECD) was previously considered as sufficiently validated according to SANCO/3029/99 rev. 4 in the 0.05 – 0.5 mg/kg range. The validation results provided here by GC-MS at the 0.05 mg/kg level confirmed the previously acceptable obtained results and the results obtained at 0.005 mg/kg indicates that the method can achieve a LOQ of 0.05 mg/kg. The method is considered fit for purpose and supports the ecological data.**

Report:	CA 4.1.2/82 Weltje L., Janson G.-M., 2013 The influence of humic acid and green algae on the toxicity of BAS 310 55 I to Daphnia magna STRAUS 2013/1404157
Guidelines:	OECD 202, EPA 850.1010 draft April 1996
GLP:	Yes, (Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the method:

Alpha-cypermethrin with its epimer (Cis-I cypermethrin) were determined in water samples by liquid/liquid extraction using dichloromethane and GC-MS determination based on internal calibration (use of lambdacyhalothrin) (Macherey Nagel Optima-5HT column, 30 m × 0.25 mm, 0.25 µm film thickness, splitless injection, injector temperature of 230 °C, oven temperature: 80°C, 1 min hold, ramp with 50°C/min to 260°C, hold for 8 min, ramp with 50°C/min to 300°C, hold for 2 min, He, 1.2 ml/min flow, SIM mode, negative chemical ionisation, monitored ion for quantitation: 207 m/z, for confirmation m/z ions 209 and 211, for internal standard: m/z 241).

Matrix:
- reconstituted water (M4 according to Elendt)
- M4- water + humic acid
- M4-water + green algae

Specificity/Interference:	No interference of the matrices with the test substance could be observed under the analytical conditions used. Chromatograms have been provided for calibration standards, controls, test samples and fortified samples at the lowest fortification level. Identity of the analysed compound was confirmed by monitoring 3 m/z ions.
Linearity:	Six standard solutions in the concentration range from 0.2 to 150 or 300 ng alpha-cypermethrin/mL (corresponding to 10 ng/L and 7500 or 14000 ng/L) were used for the linearity. The response of the method was found to be linear in this range with 2 = 0.9994. The resulting calibration graph was provided together with the regression equation. It is noted that the lowest calibration level corresponds exactly to the lowest fortification level. This will not influence negatively the results. Calibration occurred in toluene, however the absence of matrix effect was not demonstrated. The concentration of the prepared samples meet the calibration range (samples are concentrated before injection, the 10 ng/L fortified level corresponds to 0.2 ng/mL and the 1000 ng/L fortified level corresponds to 20 ng/mL). During the ecotox test, the Cis- I isomer was determined from the calibration performed with alphacypermethrin and taking into account for the response factor of the Cis-I isomer.
Accuracy:	Fortifications occurred with alpha-cypermethrin at 10 and 1000 ng/L in three types of water (M4-water, M4-water with humic acid and M4-water with green algae. Mean recovery at each fortification level is presented in the Table below.

Repeatability: RSD is provided in the table below.

Limit of Quantification: 10 ng alpha-cypermethrin/L

Table 4.1.2-82: Alpha-cypermethrin recoveries obtained from various fortified water types

Fortified samples					
Analyte	**Matrix**	**Fortified level ng/L**	**% mean recovery (range)**	**% RSD**	**n***
Alpha-cypermethrin	M4-water	10	88 (72 – 105)	16	5
		1000	102 (89 – 116)	11	5
	M4- water with humic acid	10	84 (72 – 110)	18	5
		1000	87 (78 – 101)	11	5
	M4-water with greenalgae	10	83 (70 – 106)	17	5
		1000	93 (69 – 107)	16	5

* Most of the replicates was injected twice.
Four or five controls were checked..

Conclusion: **The GC-MS method is considered sufficiently validated according to SANCO/3029/99 rev. 4. The LOQ of 10 ng a.i./L is validated. The endpoints determined in study KCA 8.2.4.1/02 are therefore considered reliable.**

Remark: A series of methods used in support of old ecotoxicity tests is also presented and assessed here below. These older methods were all used in support of ecotox studies which have been considered as relied upon in the frame of the renewal with some of them leading to critical endpoints.

Report:	CA 4.1.2/83 [redacted] 1981 WL 85871 and Cypermethrin: A comparison of their acute toxicity to Salmo gairdneri, Daphnia magna and Selenastrum capricornutum AL-511-001 Report No. SBGR.81.27
Guidelines:	EPA 850.1075, EPA 850.1010,EPA 72-1,EPA 72-2
GLP:	not stated,

Report:	CA 4.1.2/84 [redacted] 983 WL85871 and Cypermethrin: A comparative study of their toxicity to the fathead minnow Pimephales promelas (RAFINESQUE) AL-512-002 Report No. SBGR.82.298
Guidelines:	EPA 850.1075, EPA 850.1400,EPA 72-1,EPA 72-4
GLP:	not stated

Principle of the method:
Extraction of the samples was carried out with hexane (2 × 10 mL) in the separating funnels. For daphnia and algal experiments, the hexane extractant was used to rinse the test vessels. Hexane extracts of the samples were dried by passage through anhydrous sodium sulphate. The combined extracts for a sample were diluted if required and analysed by packed column GC-ECD (GC condition 1 for quantitation of most results: Varian 2 m × 6 mm ×2 mm id packed with 3% OV-1 on mesh 100-120 GCQ, column T°: 260°C, injector T°: 260°C, detector T°: 350°C, carrier gas N2, flow 0.045 l/min; for WL 85871 (alpha-cypermethrin) some determinations were made with GC condition 2: HP 3 × 6 mm × 2 mm id packed with 3% OV-1 on mesh 100-120 GCQ, column T°: 250°C, injector T°: 250°C, detector T°: .350°C, carrier gas: Argon/Methane 90/10, flow 0.033 l/min). External calibration. In study SBGR.82.298, the method used is almost identical to the method used in Study SBGR.81.277 except that 2 × 15 mL hexane were used and where necessary, the combined hexane extracts were evaporated to volume (5 mL) suitable for alpha-cypermehtrin or cypermehtrin determination using the GC-ECD. Two sets of GC operating conditions were also used, slightly amended compared to here above (GC conditions 1: HP 1.5 m × 6 mm × 2 mm id glass column packed with 3% OV-& on 100-120 mesh GCD, column T°: 270°C, injector T°: 250°C, detector T°: 300°C, carrier gas Ar/Metane 90/10, flow: 0.03 l/min; GC conditions 2: Varian 1 m × 6 mm × 4 mm id glass column packed with 31% OV-1 on 100-120 mesh GCQ, column T°: 235°C, injector T°: 260°C, detector T°: 300°C, carrier gas N2, flow: 0.04 l/min).
Matrix: drinking water

Specificity/Interference:	Cypermethrin or alpha-cypermethrin have not been identified in the control test vessels above 0.01 µg/L for study SBGR.81.277 and above 0.03 µg/L in Study SBGR.82.298. No chromatograms were provided.

Linearity:	The analytical standards for alpha-cypermethrin were made in hexane in the 2 – 200 µg/L range and for cypermehtrin in the 1 – 200 µg/L range. No other data were provided regarding the calibration and the linearity of the response for the method.

Accuracy: Fortifications occurred at 0.05 and 5.0 µg/L for cypermethrin and WL 85871. Mean recovery at each fortification level is presented in the Table below. Fortifications occurred with alpha-cypermethrin at 10 and 1000 ng/L in three types of water (M4-water, M4-water with humic acid and M4-water with green algae. Mean recovery at each fortification level is presented in the Table below.

Repeatability: RSD is also presented in the Table below and has been re-calculated based on the available results.

Limit of Quantification: In study SBGR.82.298, the LOQ is stated to be 0.03 µg/L but occasionally as low as 0.01 µg/L for cypermethrin and 0.01 or 0.02 µg/L for alphacypermethrin) but these values were not substantiated by the available data. Based on the available results, an LOQ of 0.1 µg/L can be considered as achievable for cypermethrin and 0.5 µg/L for alpha-cypermethrin but can however not be considered as fully validated.

Table 4.1.2-83: Recovery data for cypermethrin and WL 85871 in water (study SBGR.81.277) and recovery experiments from the chronic and acute toxicity studies (study SBGR.82.298)

analyte	Fortification levels µ g/L	Recoveries (range in %)	Mean recoveries (%)	RSD (%)	n
Study SBGR.81.277					
cypermethrin	0.05	80	80	0.0	3
	0.5	80	80	0.0	3
	5.0	88 - 92	92	4.3	3
WL 85871 (alpha-cypermethrin)	0.05	80 - 160	113	36.8	3
	0.5	80	80	0.0	3
	5.0	93 - 105	97	6.9	3
Study SBGR. 82.298					
cypermehtrin	0.1 – 1.0 ***	78 - 133	97	-*	26
	0.5 – 5.0****	73 - 100	90	-*	10
WL 85871 (alpha-cypermethrin)	0.1 - 1.0 ***	73 – 117	95	-*	24
	2 – 10****	103 - 106	105	-**	2

In controls, concentration found was below 0.01 µg/L
The study report does not mention clearly if n well corresponds to 3 replicates or if it correspond to three injections.
* the overall RSD cannot be calculated since the individual recoveries were not displayed in the study report.
** not relevant since the number of replicates is only 2.
*** for chronic tests.
**** for acute tests.

Table 4.1.2-84: Recovery data for cypermethrin in algae/water medium (concentration added 100 µg/L for each analyte)

Concentration of selenastrum capricornutum (cells ml^{-1})	Recovery cypermethrin (%)	Recovery WL 85871 (%)
5×10^3	59	97
5×10^3	75	103
5×10^6	68	94

Conclusion: **The method was developed and validated prior to SANCO/3029/99 rev. 4 coming into force. The method is deemed fit for purpose to quantify alpha-cypermethrin at a LOQ of 0.05µg/L, bearing in mind that a newly developed method would probably yield better repeatability at low concentration levels. The method is still considered fit for purpose as 0.1 µg/L levels were assessed but no detailed data was reported. Furthermore, the endpoint in Daphnia is covered by a more recent study (KCS 8.2.4.1/02, DocID 2013/1404157).**

Report:	CA 4.1.2/85 Garforth B.M., 1982 WL 85871 and Cypermethrin: Chronic toxicity to Daphnia magna AL-523-001 Report No. SBGR.82.119
Guidelines:	US-ECO 96-325, US-954-96-325,US-305AX-446-004,US-96178/02-ASCr
GLP:	not stated

Principle of the method:
Alpha-cypermethrin and cypermethrin were extracted from water with hexane. The hexane extracts were dried by passage through a bed of anhydrous sodium sulphate, combined, evaporated to small volume, purified by HPLC and analysed by GC-ECD (1.5 m × 6 mm × 2 mm id glass column packed with 3% m-OV-1 on 100-120 mesh GCQ, column temperature: 225°C, injector temperature: 250°C, detector temperature: 300°C, flow rate 22 ml/min). External calibration. The method seems to be similar to the method described in studies No. 26 and 27 here above with some modifications. Matrix**:** water (from which algal cells were removed).

Specificity/Interference:	Chromatograms were not available in the report. Recovery rates support the fact that no significant interference was present
Linearity:	Standard solutions in the concentration range from 0.001 to 0.02 µg/mL were prepared for both alpha-cypermethrin and cypermethrin. However, the linearity of the response has not been shown (number of standard solutions prepared unknown, correlation coefficients not provided, calibration graphs and regression equations not provided). Calibrations occurred in hexane.
Accuracy:	Fortifications occurred with alpha-cypermethrin and cypermethrin at 0.1 and 0.3 µg/L in water from which algal cells had been removed. Mean recovery at each fortification level is presented in the Table below.
Repeatability:	RSD was not provided.
Limit of Quantification:	Not stated in the study report. However, based on the available results, a LOQ of 0.1 µg/L for alpha-cypermethrin or cypermethrin seems to be achievable.

Table 4.1.2-85: Recoveries obtained for alpha-cypermethrin and cypermethrin in the fortified samples of water

Fortified samples						
Analyte	**Matrix**	**Fortified level (µg/L)**	**% mean recovery**	**% mean recovery*** **	**% RSD***	**n****
Alpha-cypermethrin	Water from which algal cells had been removed	0.1	80, 80, 80, 90	82.5	6.06	4
		0.3	95	95	-	1
cypermethrin		0.1	150, 110, 80, 100	110	24.1	4
		0.3	190, 115, 115	140	30.9	4

* RSD was not provided in the study report. When present, it has been re-calculated based on the available results and assuming that the number n was well the number of replicates.
** It is not clearly stated in the study report if it is the true number of replicates or if there were several injections of a same replicate.
*** Calculated based on the available results. However, It is not clearly stated in the study report if the number of replicates is true replicates or if there were several injections of a same replicate.

Conclusion: **The method is suitable for quantification of alpha-cypermethrin in aqueous media with a limit of quantification of 0.1 µg/L. A nearly identical method was used in study CA 4.1.2/84 with a proposed LOQ of 0.05 µg/L.**

Report:	CA 4.1.2/86 Heintze A., 1997 Alphacypermethrin (AC 900049): Effects on the development of sediment-dwelling larvae of Chironomus riparius in a water-sediment system AL-523-002 Report No. 96178/02-ASCr
Guidelines:	US-ECO 96-325, US-954-96-325,US-305AX-446-004,US-96178/02-ASCr 4
GLP:	not stated

Principle of the method:
Aliquots of acetone samples were evaporated with a gentle stream of nitrogen. The residues were dissolved with acetonitrile/ultra pure water 50:50 (v/v). The analysis of alpha-cypermethrin was performed by HPLC on a C8 reversed phase column (Shandon Hypersil MOS C8, 125 mm × 4 mm, 5 µm particle size) with acetonitrile/ultra pure water (65:35 v:v) as mobile phase and UV detection at 210 nm. External calibration was used for quantification.
Matrix: acetonic stock solutions

Specificity/Interference:	No interference could be observed under the analytical conditions used in the acetone control samples at the retention time of alpha-cypermethrin. Chromatograms have been provided for a calibration standard, acetone control and a test sample (0.06 mg/L). Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
Linearity:	Eight standard solutions in the concentration range from 0.03 to 5.0 µg/mL (mg/L) were used for the linearity (duplicate injection at each concentration). The response of the method was found to be linear in this range with a correlation coefficient = 0.999991. The resulting calibration graph was provided together the corresponding regression equation. The concentration of the prepared samples meet the calibration range except for the highest fortification level (40.0 mg/L) unless appropriate dilution occurred. Calibration occurred in acetonitrile/ultra pure water 50:50 (v/v).
Accuracy:	In the current report, it was mentioned that in the validation report 96478/01-RVF, fortifications occurred at 0.04, 0.4, 4.0 and 40.0 mg/L with alphacypermethrin and that the overall mean recovery was found to be 93% (duplicate injection of each sample) (97 +/- 4 for the 0.04 mg/L level). No details of these validation data were reported, only the results for the dose verification were reported (see Table below).
Repeatability:	RSD was not provided.
Limit of Quantification:	Stated to be 0.02 mg/L however this value seems to be not supported by the validation data of the fortification experiment.

Table 4.1.2-86: Recoveries obtained for alpha-cypermethrin in the fortified samples of acetone solutions by HPLC-UV

Concentration verification of the stock solutions for the first definitive test (mg/L)	Recovery (% of nominal)	n*
0.06	**124****	1
0.12	99	1
0.24	100	1
0.48	100	1
0.96	100	1
1.92	100	1
3.84	99	1
Concentration verification of the stock solutions for the second definitive test (mg/L)	**Recovery (% of nominal)**	**n***
0.48	94	1
0.96	95	1
1.92	97	1
3.84	94	1
7.68	96	1
15.36	96	1
30.72	96	1
Overall	**93 +/- 2.9**	

* Duplicate injection. No alpha-cypermethrin detected in the control samples.
** This value is outside the acceptable limit of 110%. The report however stated that since no blanks for alpha-cypermethrin were found in acetone controls, acceptable test substance recoveries of 97% (with a standard deviation of 4%) were found at low level (i.e. 0.04 mg/L determined for the validation of the analytical method reported in Study 96478/01) and the calibration curve was reliable even at 0.03 mg/L, the increased test substance concentration is not caused to a great extent by methodical drawbacks but rather to accidental mistakes in sampling handling. Assuming that the method has been correctly validated, this explanation seems acceptable.

Conclusion: **The method is suitable for quantification of alpha-cypermethrin in acetone solutions at a limit of quantification of 0.04 mg/L.**

Report:	CA 4.1.2/87 Klumpp M., 1998 Alpha-cypermethrin (CL 900049): Validation of a HPLC method for the determination in acetone solutions
	AL-249-002
Guidelines:	not stated
GLP:	Yes (Ministrium fuer Umwelt und Verkehr Baden-Wuerttemberg, Stuttgart, Germany)

Principle of the method

An analytical method was validated for the determination of alpha-cypermethrin in acetone solutions of alpha-cypermethrin test substance used for a toxicity test with *Chironomus riparius*. Dilutions (0.04-40.00 mg/L) for recovery experiments were prepared in duplicate with acetone from a stock solution (alpha-cypermethrin in acetonitrile). The dilutions were placed into a water bath at about 40°C and the acetone was removed with a stream of nitrogen. The residue was made up with acetonitrile/water (50:50/v/v). The analysis was performed on a C8 reversed-phase column (Shandon Hypersil MOS C8) with acetonitrile/ultrapure water (65:35, v/v) as mobile phase at a flow rate of 1.0 mL/min and UV (photodiode array) detection at 210 nm.

Recovery findings

The overall mean recovery was 93% for alphacypermethrin test substance. Average recoveries for each matrix are summarized in Table **4.1.2-87**.

Table 4.1.2-87: Results of method validation: BAS 310 I (alpha-cypermethrin) in acetone solutions

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Acetone solution	BAS 310 I	0.04	2	97	N/A	N/A
		0.40	2	92	N/A	N/A
		4.00	2	92	N/A	N/A
		40.00	2	93	N/A	N/A
		Overall	8	93	2.6	2.8

N/A Not applicable

Linearity Linearity was tested using eight calibration standards in the range of 0.03-5.00 µg/mL. Calibration curves gave acceptable correlation coefficients ≥ 0.99. Standard solutions were prepared in acetonitrile/ultrapure water (50:50, v/v).

Specificity A confirmatory method has not been performed; however, no significant blank signals or unspecific interferences in untreated control samples were observed. Good validation results have been achieved with the primary method. The limit of detection is ≤50% LOQ (0.02 mg/kg).

Limit of Quantification	The validated limit of quantification (LOQ) for alpha-cypermethrin is 0.04 mg/kg for acetone solutions.
Repeatability	The relative standard deviation (RSD, %) over all fortification levels was below 20%. The detailed values are shown in Table **4.1.2-87**.
Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The analytical method (96178/01-RVF) is considered to be fit for purpose for the determination of residues of alpha-cypermethrin in acetone solutions.**

Report:	CA 4.1.2/88 Fuchsbichler G., 1999 Validation of method SAMS 469-2 for the determination of Alphacypermethrin (AC 900049) in pond water and treatment solutions AL-243-004
Guidelines:	EU Guideline 8064/VI/97 rev. 4 15.12.1998
GLP:	Yes (certified by Bayerisches Staatsministerium fuer Arbeit, Familie und Sozialordnung, Muenchen)

Principle of the method

An analytical method was validated for the determination of alpha-cypermethrin (CL 900049) in spray solutions. Dilutions (0.050-0.200 mg/L) for recovery experiments were prepared with acetonitrile/water (50:50, v/v) from a stock solution (formulation solutions in water). The analysis was performed with HPLC, equipped with a C_8 reversed-phase column (Shandon Hypersil MOS C8), with acetonitrile/ultrapure water (65:35, v/v) as mobile phase and UV (photodiode array) detection at 210 nm.

Recovery findings

The overall mean recoveries were 96-99% for alphacypermethrin formulation solutions. Average recoveries for each matrix are summarized in Table **4.1.2-88**.

Table 4.1.2-88: Results of method validation: BAS 310 I (alpha-cypermethrin) in spray solutions

Matrix	Analyte	Fortific. Level (mg/kg)	No. of replicates	Average Recovery (%)	Standard Deviation (±)	Coefficient of Variation (%RSD)
Fastac 10EC	BAS 310 I	0.050	2	97	N/A	N/A
		0.200	2	97	N/A	N/A
		Overall	4	97	1.7	1.8
Fastac SC	BAS 310 I	0.050	2	95	N/A	N/A
		0.200	2	98	N/A	N/A
		Overall	4	96	2.1	2.1
Fastac WG	BAS 310 I	0.050	2	98	N/A	N/A
		0.200	2	99	N/A	N/A
		Overall	4	99	1.3	1.3

N/A Not applicable

Linearity Linearity was tested using seven calibration standards in the range of 1-30 µg/mL. Calibration curves gave acceptable correlation coefficients ≥ 0.99. Standard solutions were prepared in acetonitrile/water (50:50, v/v).

Specificity A confirmatory method has not been performed; however, no significant blank signals or unspecific interferences in untreated control samples were observed. Good validation results have been achieved with the primary method. The limit of detection is ≤40% LOQ (0.02 mg/kg). Blanks did not exceed 30% LOQ.

Limit of Quantification	The validated limit of quantification (LOQ) for alpha-cypermethrin is 0.05 mg/kg for spray solutions.
Repeatability	The relative standard deviations (RSD, %) over all fortification levels were below 20% for all formulation solutions tested. The detailed values are shown in Table **4.1.2-88**.
Reproducibility	Reproducibility, in terms of inter-day precision, of the method was not determined within this validation study.
Extract stability	Stability of extracts was not determined within this validation study.
Standard stability	Stability of standards was not determined within this validation study.
Conclusion	**The analytical method is considered to be fit for purpose for the determination of residues of alpha-cypermethrin in spray solutions.**

Report:	CA 4.1.2/89 Jatzek H.-J., 2002 BAS 310 I - Determination of the inhibitory effect on the cell multiplication of unicellular green algae 2002/1004851
Guidelines:	EEC 92/69 A V C 3,OECD 201,EPA 850.5400
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz, Rheinland-Pflanz, Mainz, Germany)

Principle of the method AL-243-008 corresponding to SAM-469-2:
The analytical method describes the determination of alpha-cypermethrin in aqueous solutions. The method is based on GC-ECD and quantitation is performed by external calibration. The sample preparation in the current study has been slightly adapted. The sample volumes to be extracted were modified and the clean-up step was omitted since in the matrix effected no interference with the measured signal. The suitability of the adaptation of the method AL-243-008 (SAMS-469-2) for the determination of alpha-cypermethrin in OECD medium was confirmed by the analyses of fortified solutions with concentrations of 0.1, 1.0, 10, 50, 100 and 1000 µg/L. Samples with nominal concentrations below 100 µg/L were extracted fivefold with 25 mL portions of hexane. Samples with nominal concentrations higher than 100 µg/L were extracted with 5 extractions of 100 mL portions. 2 µL of the combined extracts were injected. External calibration was used for quantification.
Matrix: water (OECD-medium)

Specificity/Interference:	No interference could be observed under the analytical conditions used in the acetone control samples at the retention time of alpha-cypermethrin. Chromatograms have been provided for a control and a test sample (1.0 mg/L). Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
Linearity:	Five standard solutions in the concentration range from 2.0 to 40 mg/L (mg/L) were used for the calibration (each concentration injected twice). The response of the method was found to be quadratic in this range with a $r^2 \geq 0.9996$. Then resulting calibration graph was provided together with the corresponding regression equation. The accuracy of the standard solution was checked by ab separated second weight.
Accuracy:	Fortifications occurred at different levels with alpha-cypermethrin (see Table below)
Repeatability:	RSD was not provided.
Limit of Quantification:	Not stated in report; proposed LOQ based on the available data of 0.05 mg/L.

Table 4.1.2-89: Recoveries obtained for alpha-cypermethrin in the fortified samples of water (OECD medium) by the GC-ECD method SAMS-469-2

Fortified samples						
Analyte	**Matrix**	**Fortified level µg/L**	**% mean recovery % range**	**% mean recovery**	**% RSD**	**n***
Alpha-cypermethrin	Water (OECD-medium)	0.1	-	83.9	-	1 × 2
		1.0	-	89.2	-	1 × 2
		10.3	-	98.3	-	1 × 2
		49.9	80.8 – 83.8	82.3	-	1 × 2
		103.0	-	87.9	-	1 × 2
		997.8	-	90.0	-	1 × 2
Test samples at test initiation						
Alpha-cypermethrin	Water (OECD-medium)	50.0	-	84.1	-	1 × 2
		100.0	-	95.7	-	1 × 2
		220.0	-	80.4	-	1 × 2
		500.0	-	87.5	-	1 × 2
		1000.0	-	90.3	-	1 × 2
Overall						
Alpha-cypermethrin	Water (OECD-medium)	**Overall (0.1 – 1000 µg/L)**	**82.3 – 98.3**	**88.0**	**6.4**	**11 × 2**

Alpha-cypermethrin not detectable in controls
* at test initiation
** identical organic extract analysed three days after sample extraction. 51.5% found after four days storage in the aqueous sample and stated
to be related to presumable decomposition of test substance.
** Calculated based on the results obtained with the fortified and test (Day 0) samples.

Conclusion: **The method has already been partly validated and considered suitable as a pre-registration method (but not for monitoring) for determination of alpha-cypermethrin in different water types (surface, source) with an LOQ of 0.05 µg/L. Therefore, the method is considered fit for purpose.**

Report:	CA 4.1.2/90 Kwasniok A. et al., 1991 Analysis of samples from a pond enclosure study with AC900049 (Alphacypermethrin)
	AL-123-029
Guidelines:	not stated
GLP:	Yes (Freie und Hansestadt Hamburg, Behoerde fuer Arbeit, Gesundheit und Soziales, Hamburg, Germany)

Principle of the method AL-243-008 corresponding to SAM-469-2:
The report stated that the analytical method used is the modified method SAMS-469-2 for which partial validation data (in the 0.01 – 10 µg/mL range) in pond water were generated under report ECO 97-138 – *Validation of Method SAMS-469-02 for the Determination of Alpha-cypermethrin Applied as formulation (FASTAC TM 100g/L OESC insecticide) in Pond Water*. However, in the current study, determination of alpha-cypermethrin occurred in stock solutions and not in pond water. Samples were extracted with hexane and directly injected for analysis by capillary GC-ECD without any clean-up step (J&W DB1 column, 30 m fused silica, 0.53 mm, 0.25 µm, Ar/methane: 1ml/min, oven: 60°C hold for 2 min, ramp at a rate of 25°C/min to 200°C, then a ramp at a rate of 10°C/min to 280°C, hold 5 min). External calibration.
Matrix: stock solutions.

Results of the stock solutions and a recovery sample analysed concurrently with the stock solutions are presented here below:

Analyte	Concentration in µg/mL	% recovery	n
Alpha-cypermethrin	1.27 (stock solution)	87.8	1
	127.45 (stock solution)	82.2	1
	523 (recovery sample)	106	1

Conclusion:	**No validation data for the determination of alpha-cypermethrin in the stock solutions have been provided. However, the method SAMS-469-02 (GC-ECD using capillary column) has been validated for determination in stock solutions in the Study by Huber W. 2000.**

Report:	CA 4.1.2/91 Huber W. et al., 2000 Evaluation of possible effects of a 100 g/L SC formulation (CF 06677) of AC 900049 (Alphacypermethrin) on macroinvertebrates, zooplankton and algae in pond-enclosures and determination of the ecologically acceptable concentration (EAC)
Guidelines:	AL-560-023 not stated
GLP:	Yes (Landeanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

Report:	CA 4.1.2/92 Fuchsbichler G., 1999 Validation of method SAMS 354-2 for the determination of Alphacypermethrin (AC 900049) in sediment
Guidelines:	AL-243-007 Report ETX-99-407 EU Guideline 8064/VI/97 rev. 4 15.12.1998
GLP:	Yes (Bayrisches Staatsministerium fuer Arbeit, Familie und Sozialordnung, Muenchen, Germany)

Report:	CA 4.1.2/93 Fuchsbichler G., 1999 Validation of method SAMS 469-2 for the determination of Alphacypermethrin (AC 900049) in pond water and treatment solutions
Guidelines:	AL-243-008 Report ETX-99-407 EU Guideline 8064/VI/97 rev. 4 15.12.1998
GLP:	Yes (Bayrisches Staatsministerium fuer Arbeit, Familie und Sozialordnung, Muenchen, Germany)

Analysis of aqueous treatment solutions:

The method used to analyse alpha-cypermethrin in the aqueous treatment solutions is a slightly modified version of method SAMS-469-02 (GC-ECD, capillary column), specifically the column clean-up step was eliminating due to the high concentrations of alpha-cypermethrin in solutions. The method is therefore identical to the method SAMS-469-02 described here above in Study KCA 8.2.8/05 (Study No. 31). Validation occurred in Study: *Validation of Method SAMS-469-2 for the Determination of alpha-cypermethrin (AC900049) in Pond Water and Treatment Solutions*, Report ETX-99-405, HVA 17/99. A summary of the validation data (only recoveries) obtained in that study has been reported in Appendix C of the Study KCA 8.2.8/06. The method even not fully validated appeared suitable for the determination of alpha-cypermethrin in stock solutions in the ~ 0.7 to 22250 µg/L range.

Matrices: Aqueous treatment solutions, pond water, sediment

Specificity/Interference: Not reported.

Linearity: Not reported.

Accuracy: Fortifications occurred at different levels with alpha-cypermethrin (see Table below)

Repeatability: Summary of recoveries from Study ETX-99-405 (HVA 17/99) has been provided (see Table below).

Table 4.1.2-90: % of nominal concentration of alpha-cypermethrin determined the aqueous treatment solutions with GC-ECD method SAMS-469-2

Analyte	Matrix	Concentration µg/L	% recovery (data taken from HVA 20-99 – ETX-99-408 and reported in the present Study HVA ETX-99-101,)	% recovery (data from Study HVA 17/99)
Alpha-cypermethrin	Aqueous treatment solutions	0.712	87	102
		7.12	88	92
		35.6	83	90
		178	85	88
		890	88	93
		4450	88	96
		22248	88	93
		Overall (n = 7)	**87 +/- 2 (RSD = 2.3%)**	**93 +/- 4.5 (RSD = 4.9%)**

Analysis of pond water samples:

The method used is method SAMS-469-02 (GC-ECD using capillary column). For details on the method, please refer to KCA 8.2.8/05 (Study No. 31) here above or in section B.5.2.4. The report stated that validation occurred in Study: *Validation of Method SAMS-469-2 for the Determination of alpha-cypermethrin (AC900049) in Pond Water and Treatment Solutions*, Report ETX-99-405, HVA 17/99. A summary of the validation data (only recoveries) obtained in that study has been reported in Appendix C of the Study KCA 8.2.8/06. It should be noted that the same method has also been partly validated.

Specificity/Interference: Interferences were below 30%. Example chromatograms were provided in the validation report AL-243-008.

Linearity: Linearity was good with r^2>0.9975. (AL-243-008)

Accuracy: Fortifications occurred at different levels with alpha-cypermethrin (see Table below)

Repeatability: Summary of recoveries from Study ETX-99-405 (HVA 17/99) has been provided (see Table below).

Limit of Quantification: 0.01 µg/L

Table 4.1.2-91: Recoveries obtained for alpha-cypermethrin determined in pond water with GC-ECD method SAMS-469-2

Analyte	Matrix	Concentration µg/L	% recovery obtained during validation (data taken from HVA 17/99 – ETX-99-405 and presented in Appendix C ofStudy ETX-99-101) (% range) [n] %RSD	Procedural recoveries (data from Study HVA 20/99)
Results obtained during validation and procedural recoveries				
Alpha-cypermethrin	Pond water	0.01	97+/- 3 (94, 96, 94, 100, 100) [n = 5] %RSD = 3.1*	97 +/- 7* (90, 96, 102, 110, 100, 86, 100, 91) [n = 8] %RSD = 7.2*
		0.05	-	82
		0.1	-	89.2 (92, 87) [n = 2]
		0.5	-	80
		1.0	-	95
		2.0	85 +/- 3 (84, 81, 84, 87, 87) [n = 5] %RSD = 3.0*	99.3 (94, 100, 104) [n = 3]
Results for determination in pond water at Day 0				
Alpha-cypermethrin	Pond water	0.015 (corresponding to treatment with the aqueous treatment solution of 178 µ g/L)	113 [n = 2]	-
		0.075 (corresponding to treatment with the aqueous treatment solution of 890 µ g/L)	92 [n = 2]	-
		0.375 (corresponding to treatment with the aqueous treatment solution of 4450 µ g/L)	109 [n = 2]	-
		1.875 (corresponding to treatment with the aqueous treatment solution of 22248 µ g/L)	111 [n = 1]	-

* Calculated based on the available results.

Stability of the samples:
Storage stability in pond water was investigated after 3 and 6 day-storage at 2 fortification levels 0.1 and 2 µg/L). Results showed that residues were stable for 6 day-storage under refrigerated conditions.

Analysis of sediment samples:
The method used is method SAMS-354-02, more than likely in the GC-ECD mode. SAMS-354-02 GC-ECD was considered suitable for the determination in soil with a validated working from 0.05 to 0.5 mg/kg and an LOQ = 0.05 m /kg. However, in the current study, validation in sediment occurred at lower levels (0.01 – 0.5 mg/kg). The report stated that validation occurred in Study: *Validation of Method SAMS-354-2 for the Determination of alpha-cypermethrin (AC900049) in Sediment*, Report ETX-99-407, HVA 19/99. A summary of the validation data (only recoveries) obtained in that study has been reported in Appendix C of the Study KCA 8.2.8/06.

Specificity/Interferences: Interferences were below 30% of the LOQ (AL-243-007). Example chromatograms are shown in AL-243-007.

Linearity: Calibration over a range of 0.005-0.02 µg/mL. Regression coefficients were good with r^2>0.9948.

Accuracy: Summary of recoveries obtained in Study ETX-99-405 (HVA 17/99) has been provided (see Table below)

Repeatability: See Tables below

Limit of Quantification: 0.01 mg/kg

Table 4.1.2-92: Recoveries obtained for alpha-cypermethrin determined in sediment with GC-MS method SAMS-354-2

Analyte	Matrix	Concentration mg/kg	% recovery obtained during validation (data taken from HVA 19/99, ETX-99-407 and presented in Appendix C of Study ETX-99-101) (% range) [n] %RSD	Procedural recoveries (data from Study HVA 20/99)
Alpha-cypermethrin	Sediment	0.01	83+/- 9	89 +/- 7
			(97, 86, 75, 77, 81) [n = 5] %RSD = 10.8*	(82, 88, 86, 100, 88) [n = 5] %RSD = 7.9*
		0.05	-	88
		0.1	-	100 (100, 100) [n = 2]
		0.2	-	80 (78, 81) [n = 2]
		0.5	86 +/- 12 (93, 103, 76, 76, 82) [n = 5] %RSD = 13.9*	-

* Calculated based on the available results.

Conclusion: **The method is considered suitable for the quantification of alpha-cypermethrin in pond water as well as in sediment. Although this particular method does not cover the lowest test concentration of 0.6 ng/L, the relevant ecotoxicological endpoints are covered by a more recent study using the method described in 2014/2356534 and 2016/123268. The LOQ of this more recent methods covers the derived NOEC in the mesocosm studies. The method described was state of the art in the year 1999.**

Report:	CA 4.1.2/94 [redacted] 2001 Alphacypermethrin (BAS 310 I) - Dietary toxicity (LC50) to the northern bobwhite (Colinus virginianus) AL-534-003 Report ETX-00-182
Guidelines:	EPA 71-2, EPA 850.2200, OECD 205, EEC 91/414 Annex II 8.1.2,EEC 96/12
GLP:	Yes (Department of Health of the Government oft he United Kingdom, United Kingdom)

Principle of the method HRC/FCH/M27/00 (based on SAMS 354-2):
Extraction from diet occurred with hexane (under heating), then concentration of the extracts occurred followed by dilution in the mobile phase and analysis by HPLC-UV (Zorbax ODS, 5 µm, 150 × 4.6 mm id, mobile phase: acetonitrile/aq. 0.005M ammonium formate (75/25 v/v), detection at 230 nm). External calibration.

Specificity/Interferences:	No interference was observed at the retention time of the analyte. Chromatograms were provided for control diet, reagent blank, calibration solution, fortified sample and test sample. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
Linearity:	Five standard solutions in the concentration range from 0.5 to 2.5 µg/mL were used for the calibration (in total three calibrations). The response of the method was found to be linear in this range with a r2 = 0.99948. The resulting calibration graph was provided together with the corresponding regression equation.
Accuracy:	Fortifications occurred at different levels with alpha-cypermethrin (see Table below). Results for procedural recoveries but also for homogeneity and dose verification are given in the Table below.
Repeatability:	See Tables below
Limit of Quantification:	set at 1.125 ppm (based on signal to noise ratio)

Table 4.1.2-93: Dose verification, homogeneity and procedural recoveries

Procedural recoveries					
Matrix	**Analyte**	**Level (ppm)**	**Mean recovery (%)**	**% RSD**	**n**
Diet	Alpha-cypermethrin	156	100.5 (Day 1) 99.5 (Day 2)	0.35 (Day 1) 1.26 (Day 2)	2 6
		Overall 156 (Day 1 + Day 2)	99.7	1.16	8
		5000	99.7 (Day 1) 100.0 (Day 2)	0.5 (Day 1) 0.91 (Day 2)	2 6
		Overall 5000 (Day 1 + Day 2)			8
Dose verification					
Diet	Alpha-cypermethrin	156	100	-	2
		313	99	-	2
		625	101	-	2
		1250	100	-	2
		2500	99	-	2
		5000	99	-	2
Homogeneity					
Diet	Alpha-cypermethrin	156	98	0.96	6 (n =3×2, top, middle, bottom)
		5000	99.2	0.63	6 (n =3×2, top, middle, bottom)

Conclusion: **The method is validated according to SANCO/3029/99 rev.4. However, based on the fortification experiments, a LOQ of 156 ppm is proposed instead of the LOQ stated in the report (determined by the signal/noise approach). The method (LOQ = 156 ppm and working range of 156 – 5000 ppm) is therefore considered "fit for purpose" and covers the endpoints determined in the study (LC50 of 5000 ppm and NOEC of 625 ppm).**

Report:	CA 4.1.2/95 [redacted] 2002 CL 912554 (metabolite of BAS 310 I, Alpha-Cypermethrin): Acute toxicity study on the bluegill sunfish (Lepomis macrochirus) in a static system over 96 hours 2002/1004682
Guidelines:	EPA 72-1, EEC 92/69 A V C 1, OECD 203, EPA-SEP 540/9-85-006
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

An analytical method determination of the test substance in the test water was developed by the Agricultural Center Limburgerhof of BASF Aktiengesellschaft. The analyses were carried out as a separate study at the test facility Agricultural Center Limburgerhof of BASF Aktiengesellschaft under the responsibility of the Study Director of this test facility. The study was carried out in compliance with the Principles of Good Laboratory Practice (GLP certificate provided and dated from 1998). Analytical phase: Concentration Control Analyses of CL912554 (Reg. No. 4080830) - DCVA, metabolite of BAS 310I alpha-cypermethrin) in Water According to HPLC-Method CP389 GV/TC – Project Code 14F0420/015033, Study code PCP06571 (Guenter Fietz, 2002).

Report:	CA 4.1.2/96 [redacted] 2002 CL 206128 (metabolite of BAS 310 I, Alpha-Cypermethrin): Acute toxicity study on the bluegill sunfish (Lepomis macrochirus) in a static system over 96 hours 2002/1004683
Guidelines:	EPA 72-1, EPA-SEP 540/9-85-006, EEC 92/69 A V C 1, OECD 203
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

An analytical method determination of the test substance in the test water was developed by the Agricultural Center Limburgerhof of BASF Aktiengesellschaft. The analyses were carried out as a separate study at the test facility Agricultural Center Limburgerhof of BASF Aktiengesellschaft under the responsibility of the Study Director of this test facility. The study was carried out in compliance with the Principles
of Good Laboratory Practice (GLP certificate provided and dated from 1998). Analytical phase: Concentration Control Analyses of CL 206128 (Reg. No. 130213 – 3-PBA, metabolite of BAS 310I alpha-cypermethrin) in Water According to HPLC-Method CP389 GV/TC – Project Code 14F0418/015034, Study code PCP06572 (Guenter Fietz, 2002).

Report:	CA 4.1.2/97 Jatzek H.-J., 2001 CL 206128 (metabolite of BAS 310 I, Alpha-Cypermethrin) - Determination of the acute effect on the swimming ability of the water flea Daphnia magna STRAUS 2001/1014673
Guidelines:	EEC 92/32 A V C 2, OECD 202, EPA 850.1010, ISO 6341, ISO/DIS 10706
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

The analytical phase report was part of the current study report under Appendix – Concentration control analysis of Reg. No. 4080830 (metabolite DCVA) in M4-water (ZH/TC-project 01/0420/50/1) by HPLC method CP389 – PCP0641, Fietz G. (2001), BASF Ecology and Environmental Analytics, DE, GLP. Matrix: M4-medium (water)

Report:	CA 4.1.2/98 Jatzek H.-J., 2001 Reg.No. 4080830 - Determination of the acute effect on the swimming ability of the water flea Daphnia magna STRAUS 2001/1017462
Guidelines:	EEC 92/32 A V C 2, OECD 202, EPA 850.1010, ISO 6341, ISO/DIS 10706
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

The analytical phase report was part of the current study report under Appendix – Concentration control analysis of Reg. No. 130213 (metabolite 3-PBA) in M4-water (ZH/TC-project 01/0418/50/1) by HPLC method CP389 – PCP06410, Fietz G. (2001), BASF Ecology and Environmental Analytics, DE, GLP. Matrix: M4-medium (water)

Report:	CA 4.1.2/99 Werner D.I., 2002 CL 912554 (metabolite of BAS 310 I, Alpha-Cypermethrin) - Determination of the inhibitory effect on the cell multiplication of unicellular green algae 2002/1004139
Guidelines:	EEC 92/69 A V C 3, OECD 201, EPA 850.54006
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

Analyses were carried out as a separate study at the test facility Ecology and Environmental Analytics, BASF, DE - Concentration control analysis of Reg. No. 4080830 (metabolite DCVA) in water according to method CP389 (GV/T-project 01/0420/60/2)– PCP06466, Fietz G. (2002), BASF Ecology and Environmental Analytics, DE, GLP (GLP certificate provided). Matrix: OECD-medium (water)

Report:	CA 4.1.2/100 Werner D.I., 2002 CL 206128 (metabolite of BAS 310 I, Alpha-Cypermethrin) - Determination of the inhibitory effect on the cell multiplication of unicellular green algae 2002/1004140
Guidelines:	EEC 92/69 A V C 3, OECD 201, EPA 850.54006
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

Analyses were carried out as a separate study at the test facility Ecology and Environmental Analytics, BASF, DE - Concentration control analysis of Reg. No 130213 (metabolite 3-PBA) in water according to method CP389 (GV/T-project 01/0418/60/2)– PCP06475, Fietz G. (2002), BASF Ecology and Environmental Analytics, DE, GLP (GLP certificate provided). Matrix: OECD-medium (water)

Report:	CA 4.1.2/101 N.N. Certificate of Analysis in Support of KCA 8.2.1/10 (BAS 310I) 2017/1125983
Guidelines:	not stated
GLP:	not stated

Note: This document provides a valid certificate of analysis as the one reported in study KCA 8.2.1/10 apparently showed an expiry date prior to study finalization. No data for the summary of the studies below is in this document.

Principle of the method CP 389 (HPLC-UV):
The analytical method CP 389 describes the determination of Reg. No. 130213 (metabolite 3-PBA) and Reg. No. 4080830 (DCVA) in aqueous matrices by means of HPLC. The method is based on reversed phase chromatography using a YMC J'sphere ODS-H80 4 µm. Quantification was achieved by UV detection at 212 nm and external calibration. A 2 mL aliquot of the 100 mg/L sample was transferred into a 20 mL volumetric flask. The sample was diluted with 6 mL acetonitrile, subsequently 100 µL sulfuric acid 0.5 mol/l was added. The flask was made up to the calibration mark with water. 50 µL of these solutions were injected and analysed by HPLC-UV. For analysis in OECD-medium, all samples were dissolved that the resulting solution contains 30% (v/v) acetonitrile and sulfuric acid in the concentration 500 µL/100 mL. 50 µL of these solutions were injected and analysed by HPLC-UV.

Table 4.1.2-94: Validation data for method CP389 (HPLC-UV) to determine metabolites 3-PBA and DCVA in water

Metabolite 3-PBA (CL 206128, Reg. No. 130213)				
Matrix	**LOQ**	**Linearity** **Range** **Correlation coefficient**	**Fortification levels**	**Specificity/interferences**
CA 4.1.2/96 2002/ 1004683 Tap water	Not stated. Based on the available results: 100 mg/L.	Linear in the concentration range from 0.2 to 20 mg/L (n = 5, duplicate injection). r ≥ 0.9999. The resulting calibration graph was provided but not the corresponding regression equation. The accuracy of the standard solution was checked by a separate second weight. The concentration of the prepared samples (~ 10 mg/L) meet the calibration range. The absence of matrix effects was not demonstrated.	Analysis of fortified solutions of tap water were performed. Mean recovery is presented in the Table below. RSD is also presented in the Table below and has been re-calculated based on the available results.	No interference of the matrix with the test substance could be observed under the analytical conditions used. Chromatograms have been provided for a calibration standard, control, test sample and fortified sample. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.

CA 4.1.2/98 2001/ 1014673 Water (M4-medium)	Not stated. Based on the available results: 6 mg/L seems to be achievable but not considered as fully validated (n = 3 if the overall results are considered). The level of 50 mg/L can be considered as sufficiently validated taking into account the overall results of studies KCA 8.2.1/10 and KCA 8.2.4.1/05	Linear in the concentration range from 0.2 to 20 mg/L (n = 5, duplicate injection). r ≥ 0.9999. The resulting calibration graph was provided but not the corresponding regression equation. The accuracy of the standard solution was checked by a separate second weight. The concentration of the prepared samples (~ 0.6 mg/L for the lowest fortified level and ~ 10 mg/L for the highest fortified level) meet the calibration range. The absence of matrix effects was not strictly demonstrated.	Fortifications of M4-water occurred with alpha-cypermethrin at 6 and 50.6 mg/L. Mean recovery at each fortification level is presented in the Table below. No RSD has been calculated (n = 2).	No interference of the matrix with the test substance could be observed under the analytical conditions used. Chromatograms have been provided for controls and test samples at the lowest and highest concentrations. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
CA 4.1.2/100 2002/ 1004140 Water (OECD-medium)	Not stated.	Linear in the concentration range from 1.0 to 20 mg/L (n = 5, duplicate injection). r ≥ 0.9999. The resulting calibration graph was provided but without the corresponding regression equation. The accuracy of the standard solution was checked by a separated second weight. The absence of matrix effect was not reported.	Fortifications of OECD-medium occurred with the metabolite at 6.2 and 51.7 mg/L (see Table below). No data on RSD has been reported.	No interference could be observed. Chromatograms have been provided for control and test samples only. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.

Metabolite DCVA (CL 912554, Reg. No. 4080830)				
CA 4.1.2/95 2002/ 1004682 Tap water	Not stated. Based on the available results: 100 mg/L.	Linear in the 0.4 to 40 mg/L (n = 5, duplicate injection) r ≥ 0.9999 The resulting calibration graph was provided but not the corresponding regression equation. The accuracy of the standard solution was checked by a separate second weight. The concentration of the prepared samples (~ 10 mg/L) meet the calibration range.	Analysis of fortified solutions of tap water were performed. Mean recovery is presented in the Table below.	No interference of the matrix with the test substance could be observed under the analytical conditions used. Chromatograms have been provided for a calibration standard, control, test sample and fortified sample. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
CA 4.1.2/97 2001/1017462 Water (M4-medium)	Not stated. Based on the available results: 6 mg/L seems to be achievable but not considered as fully validated (n = 3 if the overall results are considered). The level of 50 mg/L can be considered as sufficiently validated taking into account the overall results of studies KCA 8.2.1/08 and KCA 8.2.4.1/04.	Linear in the concentration range from 0.2 to 20 mg/L (n = 5, duplicate injection). r ≥ 0.9999. The resulting calibration graph was provided but not the corresponding regression equation. The accuracy of the standard solution was checked by a separate second weight. The concentration of the prepared samples (~ 1.2 mg/L for the lowest fortified level and ~ 10 mg/L for the highest fortified level) meet the calibration range. The absence of matrix effects was not strictly demonstrated.	Fortifications of M4-water occurred with alpha-cypermethrin at 6 and 50.4 mg/L. Mean recovery at each fortification level is presented in the Table below. No RSD has been calculated (n = 2).	No interference of the matrix with the test substance could be observed under the analytical conditions used. Chromatograms have been provided for controls and test samples at the lowest and highest concentrations. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.

CA 4.1.2/99 2002/ 1004139 Water (OECD-medium)	Not stated.	Linear in the concentration range from 1.0 to 20 mg/L (n = 5, duplicate injection). r ≥ 0.99998. The resulting calibration graph was provided but not the corresponding regression equation. The accuracy of the standard solution was checked by a separated second weight. The absence of matrix effects was not reported	Fortifications of OECD-medium occurred with the metabolite at 6 and 50 mg/L (see Table below). No data on RSD has been reported.	Absence of interference has not been demonstrated within this study. Reference was made to controls a nalysed in study 01/0418/60/2 (Study No. 37). Typical chromatograms have been provided for test samples only. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.

Table 4.1.2-95: Recoveries obtained for metabolite DCVA in the fortified samples of tap water, M4-water and OECD-water and the test samples

	Fortified samples				
Matrix (Study)	*Fortified level mg/L*	*% range*	*% mean recovery*	*% RSD*	*n**
Tap water CA 4.1.2/96 2002/ 1004683	101.3 – Day 0 101.3 – 2 day storage 101.3 – 4 day storage **overall**	- - - -	101.2 101.5 97.5 **100.1**	- - - -	1×2 1×2 1×2 3×2
	101.6	-	100.4	-	1×2
	99.91	-	99.4	-	1×2
	Test samples at Day 0				
	100		104	-	1×2
	100		102	-	1×2
	100		101.8	-	1×2
	Overall				
	Overall (100 mg/L)**	101.2 - 104	101.3	1.5	6×2
	Fortified samples				
M4-water CA 4.1.2/ 97 2001/1017462	6.05 – Day 0 6.05 – 3 day storage **Overall**	- - -	98.6 101.2 **99.9**	- - -	1 × 2 1 × 2 2 × 2
	50.4 – Day 0 50.4 – 3 day storage **Overall**	- - -	100.8 100.8 **100.8**	- - -	1 × 2 1 × 2 2 × 2
	Test samples at Day 0				
	6.25	-	97.2	-	1 × 2
	50.0	-	98.7	-	1 × 2
	Overall				
	Overall (6.0 mg/L)**	97.2 – 98.6	97.9	-	2 × 2
	Overall (50.0 mg/L)**	98.7 – 100.8	99.8	-	2 × 2
	Overall (all levels during fortification experiment at Day 0 storage and test at Day 0)				
	Overall (6 – 100 mg/L)**	97.8 – 100.8	99.04	1.1	7 × 2
	Fortified samples				

OECD-water CA 4.1.2/992002/ 1004139	6.0	-	98.8	-	1 × 2
	50.2	-	99.7	-	1 × 2
		Test samples at Day 0			
	6.25	-	100.4	-	1 × 2
	50.0	-	100.1	-	1 × 2
			Overall		
	Overall (6.0 mg/L)**	98.8 – 100.4	99.6	-	2 × 2
	Overall (50.0 mg/L)**	99.7 – 100.1	99.9	-	2 × 2
			Overall (all levels during fortification experiment at Day 0 storage and test at Day 0)		
	Overall (6 - 125 mg/L)**	98.8 – 102.1	100.7	1.1	8 × 2

Metabolite not detectable in controls
* number of replicates, each injected twice.
** Calculated based on the results obtained with the fortified (0 Day storage) and test (Day 0) samples.

Notes:
- In tap water, the analytical results of the fortified samples after storage for two days resp. four days confirm the stability of the test substance in tap water. Recoveries at the end of the test remain within +/- 10% of the nominal concentration.
- In M4-medium, the analytical results of the fortified samples after storage for three days confirm the stability of the test substance in M4-water. Recoveries at the end of the test remain within +/- 10% of the nominal concentration.
- In OECD-medium, recoveries after 72hrs (end of the test) remain within the +/- 10% of the nominal concentration.

Table 4.1.2-96: Recoveries obtained for metabolite 3-PBA in the fortified samples of tap water, M4-water and OECD-water and the test samples

	Fortified samples				
Matrix	*Fortified level mg/L*	*% range*	*% mean recovery*	*% RSD*	*n**
Tap water CA 4.1.2/96 2002/1004683	102.4 – Day 0 102.4 – 2 day storage 102.4 – 4 day storage **overall**	- - - -	100.5 99.42 99.62 **99.8**	- - - -	1× 2 1× 2 1× 2 3 × 2
	100.2	-	100.4	-	1× 2
	100.7	-	100.1	-	1× 2
	Test samples at Day 0				
	100	-	103.4	-	1× 2
	100	-	102	-	1× 2
	100	-	101.4	-	1× 2
	Overall				
	Overall (100 mg/L)**	99.42 – 103.4	101.3	1.2	6 × 2
	Fortified samples				
M4-water CA 4.1.2/98 2001/1014673	3.04 – Day 0 3.04 – 3 day storage **Overall**	- - -	97.7 99.4 **98.6**	- - -	1 × 2 1 × 2 2 × 2
	50.6 – Day 0 50.6 – 3 day storage **Overall**	- - -	100.2 101.3 **100.8**	- - -	1 × 2 1 × 2 2 × 2
	Test samples at Day 0				
	3.13	-	96.1	-	1 × 2
	50.0	-	98.9	-	1 × 2
	Overall				
	Overall (3.0 mg/L)**	96.1 – 97.7	96.9	-	2 × 2
	Overall (50.0 mg/L)**	98.9 – 100.2	99.6	-	2 × 2
	Overall (all levels during fortification experiment at Day 0 storage and test at Day 0)				
	Overall (3 - 100 mg/L)**	96.1 – 100.2	98.7	1.5	8 × 2

	Fortified samples				
OECD-water CA 4.1.2 /100 2002/104140	6.2	-	101.1	-	1 × 2
	51.7	-	102.7	-	1 × 2
	Test samples at Day 0				
	6.25	-	99.3	-	1 × 2
	50.0	-	100.0	-	1 × 2
	Overall				
	Overall (6.2 mg/L)**	99.3 – 101.1	100.2	-	2 × 2
	Overall (~ 50.0 mg/L)**	100.0 – 102.7	101.4	-	2 × 2
	Overall (all levels during fortification experiment at Day 0 storage and test at Day 0)				
	Overall (6.2 – 125 mg/L)**	99.3 – 102.7	100.3	1.1	8 × 2

Metabolite not detectable in controls.
* number of replicates, each injected twice.
** Calculated based on the results obtained with the fortified (Day 0 storage) and test (Day 0) samples.

Notes:
- In tap water, analytical results of the fortified sample after storage for two days resp. four days confirm the stability of the test substance in tap water. The recoveries obtained at the end of the test remain within +/- 10% of the nominal concentration.
- In M4-medium, the analytical results of the fortified samples after storage for three days confirm the stability of the test substance in M4-water. Recoveries at the end of the test remain within +/- 10% of the nominal concentration.
- In OECD-medium, recoveries after 72hrs (end of the test) remain within the +/- 10% of the nominal concentration.

Conclusion: **Considering the results available for the fortified samples together with the results of the test samples, the method can be considered "fit for purposes". The LOQ in the various aqueous media support the derived ecotoxicological endpoints.**

Report:	CA 4.1.2/102 Jatzek H.-J., 2002 CL 206969 - Determination of the acute effect on the swimming ability of the water flea Daphnia magna STRAUS 2002/1004857
Guidelines:	EEC 92/32 A V C 2, OECD 202 Part I (1984),EPA 850.1010,ISO 6341,ISO/DIS 10706
GLP:	Yes (Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz, Germany)

The analyses were carried out as a separate study at the test facility Ecology and Environmental Analytics, BASF, DE - Concentration control analysis of REG. No. 4080665 (CL 206969-(metabolite 3-Phenoxybenzaldehyde) in water by HPLC method CP389 (ZH/TC-project01/0419/50/2)– PCP06427, Fietz G. (2001), BASF Ecology and Environmental Analytics, DE, GLP (GLP certificate dated from 1998, end of the analytical report: 2001).

Principle of the method CP390 (HPLC-UV):
The analytical method CP390 describes the determination of Reg. No. 4080665 (3-phenoxybenzaldehyde) in aqueous solutions by means of HPLC-UV. The method is based on reversed phase chromatography using a YMC J'sphere ODS-H80. Quantification was achieved by UV detection at 224 nm and external calibration. The samples were appropriately diluted with acetonitrile/water so that the final solutions had an acetonitrile content of approx. 20%. 100 µL of these solutions were injected and analysed by HPLC-UV. External calibration.
Matrix**:** water (M4-medium).

Specificity/interferences:	No interference of the matrix with the test substance could be observed under the analytical conditions used. Chromatograms have been provided for a calibration standard, control and test samples. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
Linearity:	Five standard solutions in the concentration range from 0.08 to 10 mg/L were used for the linearity. The response of the method was found to be linear in this range with a correlation coefficient = 0.99985. The resulting calibration graph was provided together the corresponding regression equation. The accuracy of the standard solution was checked by a separate second weight. It cannot be checked if the concentration of the prepared samples meet the calibration range but it is stated that dilution occurred to meet the calibration range. The absence of matrix effects was not strictly demonstrated.
Accuracy*:*	Fortifications of M4-water occurred with the metabolite at 0.77 and 10.77 mg/L. Mean recovery at each fortification level is presented in the Table below.
Repeatability*:*	No RSD has been calculated (insufficient replicates).

Limit of Quantification: Not stated in the study report but proposed at 0.77 mg/l based on the lowest fortification level assessed.

Table 4.1.2-97: Recoveries obtained for the metabolite 3-phenoxybenzaldehyde (CL 206969) in the fortified samples of M4-water by the HPLC-UV method CP390

Fortified samples						
Analyte	**Matrix**	**Fortified level mg/L**	**% mean recovery % range**	**% mean recovery**	**% RSD**	**n***
CL 206969 (3-phenoxybenzaldehyde)	M4-water	0.77	-	86.4	-	1 × 2
		10.77	-	101.3	-	1 × 2
Test samples at test initiation						
CL 206969 (3-phenoxybenzaldehyde)	M4-water	10.0	-	74.1	-	1 × 2
		Overall (10) **	74.1 – 101.3	87.7	-	2 × 2

* at initiation only since at the end, a considerable decrease of the recoveries is obtained.
** Calculated based on the results obtained with the fortified and test (Day 0) samples.
Note: the recoveries obtained after three days of storage decreased considerably to levels as low as 5.6% at the lowest level and at non detectable
level at the highest level tested.

Conclusion: **The method allows the determination of the metabolite 3-phneoxybenzoic acid in aqueous media by HPLC-UV detection.**

Report:	CA 4.1.2/103 Fietz G., 2001 Bestimmung der Alpha-Cypermethrin Metabolite Reg.No. 4 080 830 und Reg.No. 130 213 in Wasser mittels HPLC 2001/1015123
Guidelines:	none stated
GLP:	Yes

Principle of the method CP390 (HPLC-UV):
The analytical method CP390 describes the determination of Reg. No. 4080665 (3-phenoxybenzaldehyde) and Reg.No. 130213 (DCVA) in aqueous solutions by means of HPLC-UV. The validation was done in OECD-medium as representative aqueous medium for other aqueous media.
The method is based on reversed phase chromatography using a YMC J'sphere ODS-H80. Quantification was achieved by UV detection at 224 nm and external calibration ra. The samples were appropriately diluted with acetonitrile/water so that the final solutions had an acetonitrile content of approx. 30%. Prior to injection, samples were acidified with 0.5M sulphuric acid. 100 µL of these solutions were injected and analysed by HPLC-UV. External calibration.
Matrix**:** Water (OECD-medium)

Specificity/interferences:	No interference of the matrix with the test substance could be observed under the analytical conditions used. Preparation of a blank water sample followed by analysis did not show any interferences. Chromatograms have been provided for a calibration standard, control and test samples. Identity of the analysed compound was confirmed by comparison of the mean retention time of the reference substance with the retention time of the corresponding peaks of the test item.
Linearity:	Five standard solutions in the concentration range from 0.2 to 20.0 mg/L were used. Preparation was done in in a mixture of acetonitrile/pure water/0.5 M H_2SO_4 (300/700/5 v/v/v). The response of the method was found to be linear in this range with a correlation coefficient of at least 0.9999 for both analytes. The resulting calibration graph was provided together the corresponding regression equation. The accuracy of the standard solution was checked by a separate second weight. It cannot be checked if the concentration of the prepared samples meet the calibration range but it is stated that dilution occurred to meet the calibration range. The absence of matrix effects was not strictly demonstrated.
Accuracy*:*	Fortifications of OECD-water occurred with the two metabolite at 0.4, 10.0, and 100 mg/L. Mean recovery at each fortification level is presented in the Table below. 5 replicates at each concentration level was prepared.
Repeatability*:*	RSD was below 20% for both analytes at all concentration levels.

Limit of Quantification: 0.4 mg/L for 3-PBAS as wll as for DCVA.

Table 4.1.2-98: Recoveries obtained for the metabolites 3 in the fortified samples of OECD-medium by the HPLC-UV method CP390

Fortified samples						
Analyte	**Matrix**	**Fortified level mg/L**	**% mean recovery % range**	**% mean recovery**	**% RSD**	**n***
Reg.No4080830 (3-phenoxybenzaldehyde)		0.4	99.7-100.7	100.4	0.4	5
	OECD	10.0	99.0-100.6	99.9	0.6	5
		100.0	101.4-102	101.8	0.2	5
Reg.No. 130213 (DCVA)		0.4	97.3-99.8	98.4	1.1	5
		10.0	99.1-100.3	99.7	0.5	5
	OECD	100.0	101.7-102.4	102.2	0.3	5

Conclusion: **The method allows the determination of the metabolites 3-phneoxybenzoic acid and DCVA in aqueous media by HPLC-UV detection with a limit of quantification of 0.4 mg/L**

(g)Methods in water, buffer solutions, organic solvents and any additional matrices resulting from the physical and chemical properties tests

Where necessary, these methods are reported along with the respective studies.

Report:	CA 4.1.2/104 Bohle J. F., 1991 Alphacypermethrin (FASTAC): Development of an analytical method AL-210-004
Guidelines:	None
GLP:	No

The analytical method was used to support the results of the solubility in organic solvents determined in the study by Bohle 1991 which was considered for the first approval of alpha-cypermethrin. Description of the method and validation were however not initially reported and are presented here to be complete after the study has been submitted during the course of the assessment.

Principle of the method:
The content of alpha-cypermethrin is quantified by using a gas chromatographic (GC) procedure that employs an internal standard (octacosane). The samples are dissolved in acetone at an exactly known concentration of approx. 1 g/L and diluted with acetone/n-hexane (90:10% v/v) in order to obtain the desired concentrations. The analysis is performed on a CP-Sil 8-CB capillary column (25 m x 0.32 (I.D.) mm, d = 0.25 µm; or equivalent) with helium as carrier gas (temperature program: initial temperature 265°C, then 3°C/min, final temperature 290°C, hold 2 min). The analyte is detected using a flame ionization detector (FID).

Specificity/Interferences:	Identification by mean of retention time. Chromatograms were provided for a standard solution, blank solvent, blank containing the internal standard, samples: no interferences occurred at the retention time of the analyte.
Linearity:	The response of the GC-FID system (= peak area) to alpha-cypermethrin was demonstrated to be linear over a concentration range from 0.03 to 0.25 g/L (n = 6, each solution injected in duplicate); r = 0.9999. Samples to be analysed for determination of the solubility in organic solvents were further diluted appropriately in order to meet the calibration range.
Accuracy:	Not assessed
Repeatability:	Not assessed
Limit of Quantification:	Not stated in the study report

Table 4.1.2-99: Validation of analytical method (AL-210-004) used for the determination of the solubility in organic solvents

Analyte	Linearity, concentration range, correlation coefficient	Accuracy (Recovery) / Precision (Repeatability)	Specificity / Interference
Alpha-cypermethrin	0.03 – 0.25 g/L (n = 6, each solution injected in duplicate); correlation coefficient: r = 0.9999	Not demonstrated	Not highly specific. Identification by mean of retention time. No interferences occurred at the retention time of the analyte.

Conclusions **From the linearity data and the absence of interference, it should be expected that the method will also be accurate and precise. Therefore, the method is considered suitable for the quantification of alpha-cypermethrin in organic solvents.**

CA 4.2 Methods for post-approval control and monitoring purposes

New methods were developed to allow the separation and hence separate quantification of the diastereomeric forms of Cypermethrin in different plant and animal as well as in environmental matrices, except for air, in which quantification is limited to alpha-Cypermethrin.

(a) Methods for the analysis in food and feed of plant and animal origin

Two multiresidue methods are already peer-reviewed and are hence not again discussed in detail in this section. Both DFG S19 multiresidue methods AL-244-008 and AL-244-009 are listed in the overview Table **4.1.2-35** in CA 4.1.2. They allow the determination of alpha-Cypermethrin in grapes, wheat grain, cabbage (also addressing cabbage leaves and boiled cabbage), oilseed rape (also addressing oilseed rape press cake and refined rapeseed oil) as well as sauerkraut at a LoQ of 0.01mg/kg using GC-ECD.

Report:	CA 4.2/1 Class T.,Bendig P., 2014a Validation of the BASF analytical method L0245/01: Method for the determination of individual or combined diastereomeric forms of BAS 311 I (Cis I, Cis II (Alpha-Cypermethrin, BAS 310 I), Trans III and Trans IV) in plant matrices 2013/1361966
Guidelines:	SANCO/3029/99 rev. 4 (11 July 2000), SANCO/825/00 rev. 8.1 (16 November 2010), OECD Guidance Document on Pesticide Residue Analytical Methods (ENV/JM/MONO(2007)17 - 13-Aug-07, EPA 860.1340
GLP:	yes (certified by Landesanstalt fuer Umwelt, Messungen und Naturschutz Baden-Wuerttemberg, Karlsruhe, Germany)

Remark: The title has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study in a more appropriate manner. A BASF-method number was also allocated to unequivocally identify the method. The originally assigned title was “Development and Validation of an Analytical Method for the Determination of alpha-Cypermethrin (BAS 310 I) in 5 Crop Types”; however by the assigned DocID 2013/1361966 the study can be unequivocally identified.

Principle of the method

The Cis II isomer pair of Cypermethrin (= alpha-Cypermethrin) and the other 3 isomer pairs, i.e. the diastereomeric forms of BAS 311 I (Cypermethrin) are analysed using the QuEChERs multi-residue method approach with acetonitrile extraction and d-SPE clean-up.

Sample extracts were analysed by reversed-phase (RP-) liquid chromatography and tandem mass spectrometry (LC-MS/MS) and quantified using matrix-matched standards.

The LC-MS/MS method allows to quantify and to confirm the four diastereomeric forms of Cypermethrin, using two MRM transitions (common to all analytes) based on the formation of ammonium adducts (433 m/z -> 191 m/z and 435 m/z -> 193 m/z).

The limit of quantification (LOQ) of the method is ≤0.01 mg/kg per pair of enantiomers.

Recovery findings

For method validation, plant materials were fortified: Lettuce (high in water), whole orange (high in acid), wheat grain (dry, high in starch), dry bean (high in protein), and oil seed rape seed (high in oil) (5 replicates per level and crop type). Solutions containing all diastereomeric forms of Cypermethrin (i.e. Cis I/II and Trans III/IV) in appropriate ratios to obtain fortifications levels at LOQ and at 10 x LOQ were added.

Additionally, duplicate samples per crop type were kept untreated as blank controls to show that no significant chromatographic signal interference was observed. Furthermore, two samples per crop type were fortified with alpha-Cypermethrin (Cis II) only at 10 x LOQ and examined for significant conversion, e.g. the formation of the Cis I diastereomeric form.

Average recoveries for each analyte peak, for both ion transitions monitored, and for all five crop materials were within the acceptable range of 70 % to 110 % with relative standard deviations (RSD) of ≤ 20 %.
Residues and chromatographic signal interference in all blank control specimens were below 20 % of the LOQ (one exception of Cis II in grain at ≤ 0.003 mg/kg). Transformation of alpha-Cypermethrin (Cis II) e.g. to its Cis I diastereomeric form was not significant during analysis.

Matrix effects were significant (i.e. > ±20%) especially for wheat grain and oilseed rape seed were more than 20 % response enhancement was observed. Thus quantitative determination was always carried out using calibration solutions in matrix, with lambda-cyhalothrin added as internal standard.

Detailed results of recoveries for each mass transition and crop are given in Table 4.2-1. Effects of matrix on response are demonstrated in Table 4.2-2. Results of the examination of conversion of the cis II-isomer to the cis I-isomer are presented in Table 4.2-3.

Table 4.2-1: BASF analytical method L0245/01, summary of validation results

Fortification Levels mg/kg	Transition Ion (m/z) Recovery	Cis I		Cis II		Trans III		Trans IV	
		433 -> 191	435 -> 193	433 -> 191	435 -> 193	433 -> 191	435 -> 193	433 -> 191	435 -> 193
Lettuce									
0.010	Average	80%	79%	79%	79%	78%	80%	78%	84%
	RSD	3%	3%	3%	4%	7%	6%	5%	7%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	100%	100%	96%	97%	102%	104%	96%	94%
	RSD	1%	2%	1%	0%	4%	6%	4%	2%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	90%	90%	88%	88%	90%	92%	87%	89%
	RSD	12%	13%	10%	11%	15%	15%	11%	7%
	Replicates	10	10	10	10	10	10	10	10
Orange									
0.010	Average	104%	104%	95%	95%	104%	101%	96%	98%
	RSD	3%	2%	12%	12%	2%	5%	4%	3%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	103%	104%	102%	103%	106%	103%	95%	97%
	RSD	2%	2%	2%	2%	4%	4%	2%	3%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	104%	104%	98%	99%	105%	102%	95%	97%
	RSD	2%	2%	9%	9%	3%	4%	3%	3%
	Replicates	10	10	10	10	10	10	10	10
Wheat Grain									
0.010	Average	85%	85%	90%	85%	78%	73%	72%	72%
	RSD	4%	6%	8%	6%	10%	6%	6%	3%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	91%	91%	96%	95%	97%	102%	81%	80%
	RSD	6%	5%	12%	13%	3%	5%	4%	5%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	88%	88%	93%	90%	88%	87%	76%	76%
	RSD	6%	6%	10%	11%	13%	18%	8%	7%
	Replicates	10	10	10	10	10	10	10	10
Dry Bean Seed									
0.010	Average	87%	87%	85%	85%	90%	90%	92%	89%
	RSD	3%	5%	15%	18%	15%	9%	3%	12%
	Replicates	5	5	3	3	5	5	5	5
0.10	Average	90%	90%	84%	87%	94%	90%	85%	83%
	RSD	3%	3%	11%	11%	5%	2%	1%	4%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	88%	88%	85%	86%	92%	90%	89%	86%
	RSD	3%	4%	13%	14%	10%	6%	4%	10%
	Replicates	10	10	8	8	10	10	10	10
Oilseed Rape Seed									
0.010	Average	82%	80%	89%	81%	78%	77%	92%	75%
	RSD	2%	5%	2%	2%	8%	9%	5%	5%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	84%	80%	84%	82%	92%	88%	80%	80%
	RSD	2%	2%	4%	4%	4%	7%	4%	3%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	83%	80%	86%	81%	85%	82%	86%	78%
	RSD	3%	3%	4%	3%	10%	10%	9%	5%
	Replicates	10	10	10	10	10	10	10	10

Table 4.2-2: BASF analytical method L0245/01, matrix effect on response

A positive value means enhancement and a negative value means suppression of the analyte response.

P2983G: Cypermethrin in 5 Plant Types

Matrix Effects for Cypermethrin Isomers by LC/MS/MS

Matrix	Solution			LC/MS Run	Cis I				Cis II				Trans III				Trans IV			
	ID	Conc.	Type		433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*
	K2983 -	ng/mL		P2983																
Lettuce	17	10	S	001	1.15E+06	10%	7.57E+05	8%	8.59E+05	15%	5.61E+05	13%	3.07E+05	-7%	1.86E+05	4%	1.30E+05	23%	8.99E+04	16%
	159	10	MMS	015	1.26E+06		8.14E+05		9.84E+05		6.35E+05		2.85E+05		1.93E+05		1.60E+05		1.04E+05	
Orange	17	10	S	027	1.04E+06	13%	6.73E+05	12%	7.48E+05	14%	4.83E+05	16%	2.21E+05	10%	1.52E+05	-1%	1.04E+05	9%	6.63E+04	8%
	68	10	MMS	043	1.17E+06		7.57E+05		8.51E+05		5.58E+05		2.43E+05		1.51E+05		1.13E+05		7.17E+04	
Wheat Grain	17	10	S	055	6.96E+05	126%	4.45E+05	129%	4.90E+05	186%	3.24E+05	176%	1.35E+05	293%	8.58E+04	328%	6.14E+04	327%	3.90E+04	333%
	120	10	MMS	078	1.57E+06		1.02E+06		1.40E+06		8.94E+05		5.31E+05		3.67E+05		2.62E+05		1.69E+05	
Dry Bean	17	10	S	083	1.00E+06	1%	6.68E+05	0%	9.64E+05	-4%	6.14E+05	-4%	2.65E+05	-4%	1.85E+05	-1%	1.24E+05	27%	7.86E+04	35%
	128	10	MMS	099	1.01E+06		6.66E+05		9.23E+05		5.90E+05		2.75E+05		1.84E+05		1.57E+05		1.06E+05	
Rape Seed	17	10	S	168	5.85E+05	50%	3.71E+05	53%	4.28E+05	148%	2.72E+05	153%	1.42E+05	84%	9.74E+04	79%	6.72E+04	217%	4.21E+04	228%
	211	10	MMS	165	8.80E+05		5.66E+05		1.06E+06		6.87E+05		2.61E+05		1.74E+05		2.13E+05		1.38E+05	

Calibration used for quantitation: Calibration solutions in solvent ("S"). MMS: Matrix-matched standards.

*A positive value indicates enhancement, a negative value suppression.

Table 4.2-3: Presence of Cis I diastereomer of alpha-Cypermethrin (Cis II) in stored sample extracts from samples fortified with alpha-Cypermethrin only

Alpha-Cypermethrin (cis II) isomerisation to cis I (epimer)

Matrix	Sample ID P2983-	Isomer	LC/MS Run P2983-	Peak Area	Isomerisation*
Cypermethrin					
m/z 433 -> m/z 191					
Lettuce	155	Cis I	023	8.88E+04	3%
		Cis II		2.60E+06	
	156	Cis I	024	5.49E+04	2%
		Cis II		2.32E+06	
Orange	63	Cis I	051	1.59E+04	1%
		Cis II		1.86E+06	
	64	Cis I	052	6.33E+03	0%
		Cis II		1.79E+06	
Wheat Grain	86	Cis I	079	3.12E+04	3%
		Cis II		1.21E+06	
	87	Cis I	081	1.25E+04	1%
		Cis II		8.95E+05	
Dry Bean	101	Cis I	107	2.24E+04	2%
		Cis II		1.23E+06	
	102	Cis I	108	1.70E+04	1%
		Cis II		1.16E+06	
Rape Seed (3rd set)	207	Cis I	163	1.53E+03	0%
		Cis II		3.84E+05	
	208	Cis I	164	6.99E+02	0%
		Cis II		4.50E+05	
* % Isomerisation = Peak Area (Cis I) / (Peak Area (Cis I) + Peak Area (Cis II))					

Linearity

Internal calibration using matrix matched standards was used for quantitation of the analytes by LC-MS/MS. Calibration functions ranging from (approx. concentrations exemplified for Cis I and Trans III Cypermethrin) 0.40 ng/mL to 30 ng/mL for lettuce and orange, from 0.20 ng/mL to 30 ng/mL for wheat grain and dry bean and from 0.1 ng/mL to 30 ng/mL for oilseed rape seed, always with ≥ 5 concentration levels were used to evaluate the final extracts.

Linear regression equations were generated using the Analyst software with 1/x weighting, resulting in internal calibration functions with correlation coefficients r ≥ 0.99.

Specificity	The method allows the specific determination of the diastereomeric forms of BAS 311 I (Reg. No. 127266) by using two specific mass transitions for quantitation and confirmation: 433 m/z → 191 m/z and 435 m/z → 193 m/z (ammonia adduct). Residues and chromatographic signal interference in all blank control specimens were below 20 % of the LOQ (one exception of Cis II in grain at ≤ 0.003 mg/kg). Transformation of alpha-Cypermethrin (Cis II) e.g. to its Cis I diastereomeric form was not significant during analysis.
Limit of Quantitation	The method achieves an LOQ of ≤ 0.01 mg/kg per analyte. The LOD (limit of detection) is set to about ≤ 0.002 mg/kg per analyte.
Repeatability	The overall relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table 4.2-1.
Reproducibility	Reproducibility of the method was not determined within this validation study.

Stability in Solutions and Extracts

Solutions with the analytes were used throughout the study and gave consistent results and recoveries. Final sample extracts acidified with formic acid were stored prior to final LC/MS/MS analyses for several weeks refrigerated. Examination of sample extracts where only alpha-Cypermethrin (Cis II) was fortified resulted in no or very little conversion e.g. formation of the analyte to the Cis I peak after several weeks of refrigerated storage. Acceptable recoveries of all analytes in crop sample extract (acidified acetonitrile) stored refrigerated for several weeks demonstrate the stability of the analytes during refrigerated storage.

Thus, extracts and solutions are considered stable for at least 4 weeks.

Conclusion	**It was proven that the method L0245/01 is suitable to determine residues of the diastereomeric forms of BAS 311 I (Cypermethrin, Reg. No. 127266) in the 5 following crop types: lettuce (high in water), whole orange (high in acid), wheat grain (dry, high in starch), dry bean (high in protein), and oil seed rape seed (high in oil). The residue methods fulfils the registration requirements of SANCO/3029/99 rev.4, 11/07/2000; SANCO/825/00 rev. 8.1, 16/11/2010, OECD ENV/JM/MONO(2007)17, 13-Aug-2007, and EPA OPPTS 860.1340, Aug-1996.**

Remark: Assessment of the extraction efficiency of the residue method 567/1, as well as multimethods QuECHERS and DFG-S19 was accomplished within the metabolism study in lettuce, representing crop commodities of high water content. The extraction schemes were shown to give comparable extraction recoveries. Results are discussed in detail in chapter M-CA 6.2.1/1 ("Metabolism of ^{14}C-Alpha-Cypermethrin in lettuce", Schreiner D., Possienke M.).

As in due course of gathering scientific information it was deemed scientifically adequate to also assessed extraction efficiency for other crop commodities, hence an additional, separate study using unlabeled samples from field trials was conducted: "BAS 310 I (alpha-Cypermethrin): Bridging Extractability of BAS 310 I (alpha-Cypermethrin) from Barley Grain, Olive Fruit and Orange Whole Fruit using Acetonitrile/Water (4 Methods, incl. QuEChERS) or Methanol/Water/HCl (BASF Method 567/1) as Extraction Solvents"). This study is presented and described in detail in chapter MCA 4.2/11.

Report:	CA 4.2/2 Austin R., 2014a Independent laboratory validation (ILV) of the BASF analytical method L0245/01: Method for the determination of individual or combined diastereomeric forms of BAS 311 I (Cis I, Cis II [Alpha-Cypermethrin, BAS 310 I], Trans III and Trans IV) in plant matrices 2014/1145880
Guidelines:	SANCO/825/00 rev. 8.1 (16 November 2010), OECD Guidance Document on Pesticide Residue Analytical Methods (ENV/JM/MONO(2007)17 - 13-Aug-07, EPA 860.1340
GLP:	yes (certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Remark: The title of 2014/1145880 has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study a more appropriate manner. A BASF-method number was also allocated to unequivocally identify the method. The originally assigned title was "ILV of BAS 310 I and its isomers in plant matrices"; however by the assigned DocID 2014/1145880 the study can be unequivocally identified.

Principle of the methods

BAS 311 I (Cis I, Cis II [alpha-Cypermethrin: BAS 310 I], Trans III and Trans IV) were analysed using the QuEChERs multi-residue method approach with acetonitrile extraction and d-SPE clean-up. Sample extracts were analysed by liquid chromatography and tandem mass spectrometry (LC-MS/MS) and quantified using matrix-matched standards. The LC-MS/MS method allows to quantify and to confirm the four diastereomeric forms of Cypermethrin, using two MRM transitions (common to all analytes) based on formation of ammonium adducts (433 m/z -> 191 m/z and 435 m/z -> 193 m/z). The limit of quantification (LOQ) of the method is ≤ 0.01 mg/kg per pair of enantiomers.

Recovery findings

The validation study was carried out using lettuce (high in water), orange (high in acid), wheat grain (high in starch), oil seed rape seed (high in oil) and dry bean seed (high in protein). Plant materials were fortified (5 replicates per level and crop type) with solutions containing all diastereomeric forms of Cypermethrin (i.e. Cis I/II and Trans III/IV) in appropriate ratios to obtain fortifications levels at LOQ and at 10×LOQ.

Average recoveries for each analyte peak, for both ion transitions monitored, and for all five crop materials were within the acceptable range of 70 to 120% with relative standard deviations (RSD) of ≤ 20%.

Matrix effects were significant (i.e. > ±20%) for orange, wheat grain, oilseed rape seed and dry bean seed, where greater than 20 % response suppression was observed. Thus quantitative determination was always carried out using calibration solutions in matrix, with lambda-cyhalothrin added as internal standard.

Detailed results of recoveries for each mass transition and crop are given in Table 4.2-4. Effects of matrix on response are demonstrated in Table 4.2-5.

Table 4.2-4: BASF analytical method L0245/01, summary of validation results (ILV)

Fortification Level (mg/kg)	Analyte	Cis I		Cis II		Trans III		Trans IV	
	Recoveries (%)	m/z 433 - 191	m/z 435 - 193	m/z 433 - 191	m/z 435 - 193	m/z 433 - 191	m/z 435 - 193	m/z 433 - 191	m/z 435 - 193
Lettuce									
≤ 0.01	Average	104	105	106	106	102	101	102	100
	RSD	3.8	3.4	2.8	4.4	6.0	5.5	6.4	4.5
	Replicates	5	5	5	5	5	5	5	5
≤ 0.1	Average	94	93	95	95	95	93	97	95
	RSD	3.0	2.7	2.3	3.1	4.9	3.1	2.6	2.3
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	99	99	100	101	99	97	100	98
	RSD	6.5	6.7	6.4	6.7	6.6	6.0	5.5	4.5
	Replicates	10	10	10	10	10	10	10	10
Orange									
≤ 0.01	Average	104	103	112	112	107	110	113	113
	RSD	1.1	1.2	2.3	3.2	4.8	2.8	2.3	3.7
	Replicates	5	5	5	5	5	5	5	5
≤ 0.1	Average	97	96	105	105	105	104	106	107
	RSD	3.3	3.4	2.7	1.9	0.91	3.1	1.7	1.6
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	100	99	109	108	106	107	109	110
	RSD	4.1	4.0	4.2	4.3	3.4	4.2	3.9	3.9
	Replicates	10	10	10	10	10	10	10	10
Wheat Grain									
≤ 0.01	Average	95	95	92	92	97	97	106	105
	RSD	5.0	5.8	19	19	8.2	12	4.4	4.2
	Replicates	5	5	5	5	5	5	5	5
≤ 0.1	Average	86	86	88	89	93	90	96	95
	RSD	4.0	3.4	3.9	5.4	4.2	4.3	2.4	1.4
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	90	91	90	90	95	93	101	100
	RSD	6.7	7.1	14	13	6.7	9.3	6.5	6.2
	Replicates	10	10	10	10	10	10	10	10

Fortification Level (mg/kg)	Analyte	Cis I		Cis II		Trans III		Trans IV	
	Recoveries (%)	m/z 433 - 191	m/z 435 - 193	m/z 433 - 191	m/z 435 - 193	m/z 433 - 191	m/z 435 - 193	m/z 433 - 191	m/z 435 - 193
Oilseed Rape Seed									
≤ 0.01	Average	78	78	82	81	81	79	84	83
	RSD	3.7	3.4	4.2	3.3	3.8	2.4	4.8	5.0
	Replicates	5	5	5	5	5	5	5	5
≤ 0.1	Average	82	81	80	80	81	81	82	81
	RSD	6.3	6.2	5.0	5.7	4.0	4.6	3.5	4.3
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	80	79	81	81	81	80	83	82
	RSD	5.9	5.3	4.6	4.5	3.7	3.9	4.2	4.6
	Replicates	10	10	10	10	10	10	10	10
Dry Bean Seed									
≤ 0.01	Average	94	96	98	100	95	93	99	101
	RSD	5.2	6.4	4.5	4.7	4.1	5.1	2.8	5.4
	Replicates	5	5	5	5	5	5	5	5
≤ 0.1	Average	98	98	98	98	97	99	97	99
	RSD	14	14	8.3	7.7	7.3	6.5	2.9	3.9
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	96	97	98	99	96	96	98	100
	RSD	10	11	6.3	6.1	5.7	6.7	3.0	4.7
	Replicates	10	10	10	10	10	10	10	10

Table 4.2-5: BASF analytical method L0245/01, matrix effect on response (ILV)

Matrix	Standard Concentration (ng/mL)	Compound	Transition *m/z*	Standards	Average Peak Area	Matrix Effect (%)
Lettuce	2	Cis I	433/191	Solvent	295772	-5
				Matrix-Matched	281867	
			435/193	Solvent	201748	-4
				Matrix-Matched	192862	
		Cis II	433/191	Solvent	103468	1
				Matrix-Matched	104068	
			435/193	Solvent	66388	1
				Matrix-Matched	67003	
		Trans III	433/191	Solvent	93644	11
				Matrix-Matched	104302	
			435/193	Solvent	62607	12
				Matrix-Matched	69862	
		Trans IV	433/191	Solvent	31618	3
				Matrix-Matched	32506	
			435/193	Solvent	20412	8
				Matrix-Matched	22106	
Orange	2	Cis I	433/191	Solvent	295772	-84
				Matrix-Matched	46647	
			435/193	Solvent	201748	-84
				Matrix-Matched	32310	
		Cis II	433/191	Solvent	103468	-82
				Matrix-Matched	18109	
			435/193	Solvent	66388	-81
				Matrix-Matched	12441	
		Trans III	433/191	Solvent	93644	-81
				Matrix-Matched	17741	
			435/193	Solvent	62607	-81
				Matrix-Matched	11657	
		Trans IV	433/191	Solvent	31618	-81
				Matrix-Matched	5872	
			435/193	Solvent	20412	-81
				Matrix-Matched	3980	
Wheat Grain	2	Cis I	433/191	Solvent	174108	-83
				Matrix-Matched	30102	
			435/193	Solvent	119523	-83
				Matrix-Matched	20064	
		Cis II	433/191	Solvent	65543	-79
				Matrix-Matched	13940	
			435/193	Solvent	43564	-79
				Matrix-Matched	9100	
		Trans III	433/191	Solvent	56457	-82
				Matrix-Matched	10266	
			435/193	Solvent	36953	-80
				Matrix-Matched	7354	

Matrix	Standard Concentration (ng/mL)	Compound	Transition *m/z*	Standards	Average Peak Area	Matrix Effect (%)
		Trans IV	433/191	Solvent	20189	-82
				Matrix-Matched	3547	
			435/193	Solvent	13227	-82
				Matrix-Matched	2395	
Oilseed Rape Seed	4	Cis I	433/191	Solvent	556633	-89
				Matrix-Matched	63496	
			435/193	Solvent	371810	-89
				Matrix-Matched	42600	
		Cis II	433/191	Solvent	198475	-89
				Matrix-Matched	22591	
			435/193	Solvent	127820	-88
				Matrix-Matched	15081	
		Trans III	433/191	Solvent	172599	-87
				Matrix-Matched	22004	
			435/193	Solvent	118065	-88
				Matrix-Matched	14703	
		Trans IV	433/191	Solvent	60411	-88
				Matrix-Matched	7046	
			435/193	Solvent	39497	-89
				Matrix-Matched	4347	
Dry Bean Seed	2	Cis I	433/191	Solvent	174108	-62
				Matrix-Matched	66427	
			435/193	Solvent	119523	-63
				Matrix-Matched	44437	
		Cis II	433/191	Solvent	65543	-62
				Matrix-Matched	25077	
			435/193	Solvent	43564	-63
				Matrix-Matched	16129	
		Trans III	433/191	Solvent	56457	-60
				Matrix-Matched	22680	
			435/193	Solvent	36953	-58
				Matrix-Matched	15449	
		Trans IV	433/191	Solvent	20189	-62
				Matrix-Matched	7741	
			435/193	Solvent	13227	-64
				Matrix-Matched	4752	

Average matrix effect (%) = (average matrix-matched standard peak area - average solvent standard peak area) ÷ average solvent standard peak area × 100. A positive value means enhancement and a negative value means suppression of the analyte response.

Linearity	Good linearity (regression coefficients ≥ 0.999) was observed at a concentration > 0.1 ng/mL for Cis I and Trans III.
Specificity	Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity. Residues and chromatographic signal interference in all blank control specimens were below 20 % of the LOQ.
Limit of Quantification	The limit of quantification (LOQ) of the method was ≤ 0.01 mg/kg per pair of enantiomers.
Repeatability	For both ion transitions monitored, and for all five crop materials, the relative standard deviations (RSD) were always below 20%. The detailed values are shown in Table 4.2-4.
Reproducibility	In context of this ILV study, the reproducibility of the method L0245/01 was estimated. As can be seen from the results, a high reproducibility was determined.
Conclusion	**The method L0245/01 was successfully independently validated with a limit of quantification of ≤ 0.01 mg/kg per analyte (pair of enantiomers) in the 5 following crop types: Lettuce (high in water), whole orange (high in acid), wheat grain (dry, high in starch), dry bean (high in protein), and oil seed rape seed (high in oil).** **The residue method fulfils the registration requirements of SANCO/825/00 rev. 8.1, OECD ENV/JM/MONO(2007)17 and EPA OPPTS 860.1340.**

Two multiresidue methods for animal matrices are already peer-reviewed and are hence not again discussed in detail in this section. Both DFG S19 multiresidue methods AL-245-007 and AL-245-008 are listed in the overview Table 4.1.2-16 in CA 4.1.2.

A newly developed method allowing the separation to allow the separation and quantification of the diastereomeric forms of Cypermethrin in different plant and animal as well as in environmental matrices.

Report:	CA 4.2/3 Class T., Bendig P., 2014a Validation of the BASF analytical method L0231/01: Method for the determination of individual or combined diastereomeric forms of BAS 311 I (cis I, cis II (Alpha-Cypermethrin, BAS 310 I), trans III and trans IV) in animal matrices 2013/1361967
Guidelines:	SANCO/3029/99 rev. 4 (11 July 2000), SANCO/825/00 rev. 8.1 (16 November 2010), EPA 860.1340, OECD-ENV/JM/MONO/(2007)17
GLP:	yes (certified by Landesanstalt fuer Umwelt, Messungen und Naturschutz Baden-Wuerttemberg, Karlsruhe, Germany)

Remark: The title has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study a more appropriate manner. A BASF-method number was also allocated to unequivocally identify the method. The originally assigned title was "Validation of the BASF Analytical Method L0231/01: Method for the determination of Individual or Combined Diastereomeric Forms of BAS 311 I (Cis I, Cis II [alpha-Cypermethrin, BAS 310I], Trans III and Trans IV) in Animal Matrices"; however by the assigned docID 2013/1361967 the study can be unequivocally identified.

Principle of the method

The Cis II isomer pair of Cypermethrin (= alpha-Cypermethrin) and the other 3 isomer pairs, i.e. the diastereomeric forms of BAS 311 I, are analysed using the QuEChERs multi-residue method approach with acetonitrile extraction and d-SPE clean-up, with the exception of fat where the DFG S19 module was used for extraction (E7) and clean-up by gel permeation chromatography (GPC) for clean-up. In addition to the QuEChERS approach an acetonitrile based extraction was performed for egg and d-SPE was applied for extract clean-up.

Sample extracts were analysed by reversed-phase (RP-) liquid chromatography and tandem mass spectrometry (LC-MS/MS) and quantified using matrix-matched standards.

The LC-MS/MS method allows to quantify and to confirm the four diastereomeric forms of Cypermethrin, using two MRM transitions based on ammonium adduct ions (433 m/z -> 191 m/z and 435 m/z -> 193 m/z) for quantitation and confirmation.

The limit of quantification (LOQ) of the method is ≤0.01 mg/kg per analyte (i.e. pair of enantiomers).

Recovery findings

The analytical method L0231/01 was validated in the PTRL study no. P 2984 G. For method validation, animal materials (whole milk, eggs, bovine meat, liver, kidney and fat) were fortified with solutions containing all diastereomeric forms of Cypermethrin (i.e. Cis I/II and Trans III/IV) in appropriate ratios to obtain fortifications levels at LOQ (≤0.01 mg/kg per peak) and at 10xLOQ, each with 5 replicates per level and matrix type.

Additionally, two samples were kept untreated as blank controls to show that no significant chromatographic signal interference caused by the analytical method was observed.

Furthermore, two samples per matrix type were fortified with alpha-Cypermethrin (Cis II) only at 10xLOQ (0.1 mg/kg) and examined for significant formation of the Cis I diastereomeric form.

Average recoveries for each analyte, for both ion transitions monitored, and for all six matrix types were within the acceptable range of 70 % to 110 % with relative standard deviations (RSD) of ≤ 20 %.

Residues and chromatographic signal interference in all blank control specimens were below 20 % of the LOQ. Transformation of alpha-Cypermethrin (Cis II) to its Cis I diastereomeric form was not significant.

Matrix effects were significant (i.e. > ±20%) for all matrices except for egg (acetonitrile extracts).

Thus quantitative determination was always carried using calibration solutions in matrix, with lambda-cyhalothrin used as internal standard for all matrices except egg (acetonitrile extracts).

Detailed results of recoveries for each mass transition and matrix are given in Table 4.2-6. Effects of matrix on response are demonstrated in Table 4.2-7. Results of the examination of conversion of the cis II-isomer to the cis I-isomer are presented in Table 4.2-8.

Table 4.2-6: BASF analytical method L0231/01, summary of validation results

Fortification Levels	Fragment Ion (m/z)	Cis I		Cis II		Trans III		Trans IV	
mg/kg	Recovery	191	193	191	193	191	193	191	193
Whole Milk									
0.010	Average	91%	90%	86%	91%	102%	94%	80%	84%
	RSD	5%	5%	4%	5%	3%	8%	7%	6%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	94%	93%	87%	88%	104%	93%	80%	79%
	RSD	2%	2%	2%	1%	3%	2%	3%	3%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	92%	91%	86%	89%	103%	94%	80%	82%
	RSD	4%	4%	3%	4%	3%	5%	5%	6%
	Replicates	10	10	10	10	10	10	10	10
Meat									
0.010	Average	72%	73%	74%	74%	79%	77%	75%	73%
	RSD	2%	2%	2%	3%	4%	6%	2%	3%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	82%	82%	79%	78%	94%	93%	74%	74%
	RSD	3%	3%	4%	5%	5%	3%	4%	4%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	77%	78%	76%	76%	86%	85%	75%	73%
	RSD	7%	6%	5%	5%	10%	10%	3%	3%
	Replicates	10	10	10	10	10	10	10	10
Kidney									
0.010	Average	77%	77%	83%	82%	80%	82%	76%	75%
	RSD	3%	2%	8%	8%	7%	8%	2%	1%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	84%	85%	86%	84%	94%	96%	72%	74%
	RSD	11%	7%	4%	3%	5%	6%	3%	3%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	80%	81%	85%	83%	87%	89%	74%	75%
	RSD	9%	7%	6%	6%	10%	11%	4%	3%
	Replicates	10	10	10	10	10	10	10	10
Liver									
0.010	Average	77%	79%	77%	77%	77%	81%	79%	83%
	RSD	9%	8%	7%	7%	10%	12%	10%	4%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	100%	100%	97%	98%	100%	99%	98%	95%
	RSD	1%	2%	2%	2%	2%	3%	1%	3%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	88%	90%	87%	88%	88%	90%	89%	89%
	RSD	14%	13%	13%	13%	15%	13%	13%	8%
	Replicates	10	10	10	10	10	10	10	10

Table 4.2-6, continued: BASF analytical method L0231/01, summary of validation results

Fortification Levels	Fragment Ion (m/z)	Cis I		Cis II		Trans III		Trans IV	
mg/kg	Recovery	191	193	191	193	191	193	191	193
Egg – QuEChERS									
0.010	Average	75%	74%	85%	87%	73%	73%	100%	100%
	RSD	1%	2%	3%	3%	6%	3%	3%	2%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	83%	83%	82%	82%	92%	91%	83%	80%
	RSD	2%	2%	2%	3%	5%	2%	3%	2%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	79%	78%	84%	84%	83%	82%	92%	90%
	RSD	6%	6%	3%	4%	13%	12%	10%	12%
	Replicates	10	10	10	10	10	10	10	10
Egg - ACN Extraction									
0.010	Average	104%	103%	109%	107%	107%	107%	94%	93%
	RSD	2%	2%	1%	2%	1%	3%	4%	4%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	104%	105%	107%	106%	107%	108%	95%	94%
	RSD	1%	1%	2%	1%	1%	1%	1%	1%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	104%	104%	108%	107%	107%	108%	95%	93%
	RSD	2%	2%	1%	1%	1%	2%	3%	3%
	Replicates	10	10	10	10	10	10	10	10
Fat - DFG S19									
0.010	Average	83%	74%	105%	108%	87%	87%	110%	106%
	RSD	6%	6%	8%	10%	8%	9%	5%	6%
	Replicates	5	5	5	5	5	5	5	5
0.10	Average	83%	83%	94%	92%	91%	87%	88%	87%
	RSD	19%	18%	13%	12%	12%	16%	11%	11%
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	83%	78%	99%	100%	89%	87%	99%	96%
	RSD	13%	14%	12%	13%	10%	12%	14%	13%
	Replicates	10	10	10	10	10	10	10	10

Table 4.2-7: BASF analytical method L0231/01, matrix effect on response

A positive value means enhancement and a negative value means suppression of the analyte response.

Matrix Effects for Cypermethrin Isomers by Reversed Phase LC-MS/MS

Matrix	Solution			LC/MS Run	Cis I				Cis II				Trans III				Trans IV			
	ID	Conc.	Type		433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*
	K2984-	ng/mL		P2984																
Milk	17	10	S	001	7.68E+05	-83%	4.84E+05	-82%	5.64E+05	-79%	3.72E+05	-79%	2.19E+05	-80%	1.37E+05	-78%	9.98E+04	-74%	6.47E+04	-74%
	107	10	MMS	024	1.33E+05		8.86E+04		1.17E+05		7.75E+04		4.42E+04		3.08E+04		2.55E+04		1.65E+04	
Meat	17	10	S	029	8.23E+05	-65%	5.28E+05	-66%	5.97E+05	-59%	3.96E+05	-61%	2.61E+05	-66%	1.61E+05	-63%	1.08E+05	-50%	7.15E+04	-48%
	115	10	MMS	052	2.87E+05		1.80E+05		2.44E+05		1.56E+05		8.98E+04		5.89E+04		5.44E+04		3.71E+04	
Kidney	17	10	S	057	1.03E+06	-56%	6.92E+05	-57%	7.83E+05	-49%	5.00E+05	-49%	3.16E+05	-52%	2.12E+05	-52%	1.38E+05	-32%	9.53E+04	-38%
	123	10	MMS	080	4.57E+05		3.00E+05		3.96E+05		2.57E+05		1.53E+05		1.02E+05		9.32E+04		5.89E+04	
Liver	17	10	S	085	7.80E+05	-89%	5.18E+05	-89%	5.63E+05	-87%	3.64E+05	-87%	2.24E+05	-87%	1.55E+05	-87%	1.02E+05	-83%	6.42E+04	-82%
	154	10	MMS	108	8.87E+04		5.88E+04		7.40E+04		4.65E+04		2.95E+04		2.02E+04		1.73E+04		1.18E+04	
Egg (QuEChERS)	17	10	S	113	7.29E+05	56%	4.72E+05	57%	5.31E+05	105%	3.49E+05	103%	2.19E+05	69%	1.35E+05	84%	9.29E+04	187%	6.12E+04	192%
	223	10	MMS	139	1.14E+06		7.39E+05		1.09E+06		7.08E+05		3.70E+05		2.48E+05		2.67E+05		1.79E+05	
Egg (ACN)	296A	10	S	311	2.17E+06	-7%	1.40E+06	-7%	1.38E+06	-19%	8.89E+05	-19%	1.12E+06	-15%	7.46E+05	-15%	4.49E+05	-1%	2.90E+05	-2%
	289	10	MMS	306	2.01E+06		1.30E+06		1.12E+06		7.22E+05		9.55E+05		6.34E+05		4.43E+05		2.84E+05	
Fat	17	10	S	174	6.94E+05	-64%	4.45E+05	-62%	4.85E+05	-29%	3.26E+05	-31%	1.53E+05	-41%	9.38E+04	-36%	6.86E+04	5%	4.56E+04	5%
	215	10	MMS	169	2.50E+05		1.68E+05		3.46E+05		2.25E+05		9.01E+04		6.02E+04		7.21E+04		4.79E+04	

Calibration used for quantitation: Calibration solutions in solvent ("S"). MMS: Matrix-matched standards.
*A positive value indicates enhancement, a negative value suppression.

Table 4.2-8: Presence of Cis I of alpha-Cypermethrin in stored sample extracts from samples fortified with alpha-Cypermethrin only

alpha-Cypermethrin (Cis II) Isomerisation to Cis I (Epimer)

Matrix	Sample ID P2984-	Isomer	LC/MS Run P2984-	Peak Area	Isomerisation
Cypermethrin					
m/z 433 -> m/z 191					
Milk	43	Cis I	025	5.61E+03	2%
		Cis II		3.11E+05	
	44	Cis I	026	3.45E+03	2%
		Cis II		2.01E+05	
Meat	73	Cis I	053	3.04E+04	2%
		Cis II		1.80E+06	
	74	Cis I	054	2.58E+04	1%
		Cis II		1.86E+06	
Kidney	88	Cis I	081	4.24E+03	3%
		Cis II		1.55E+05	
	89	Cis I	082	2.97E+03	2%
		Cis II		1.50E+05	
Liver	103	Cis I	109	2.80E+04	3%
		Cis II		1.09E+06	
	104	Cis I	110	3.17E+04	2%
		Cis II		1.34E+06	
Egg (QuEChERS)	232	Cis I	176	0.00E+00	0%
		Cis II		4.75E+05	
	233	Cis I	177	1.63E+03	0%
		Cis II		4.37E+05	
Egg (ACN Extr.)	276	Cis I	308	1.16E+04	1%
		Cis II		8.01E+05	
	276 (stored 15days)	Cis I	333	8.63E+03	1%
		Cis II		6.72E+05	
	277	Cis I	309	1.29E+04	2%
		Cis II		8.23E+05	
Fat	234	Cis I	170	0.00E+00	0%
		Cis II		2.56E+05	
	235	Cis I	171	4.35E+03	0%
		Cis II		1.10E+06	

* Isomerisation = Peak Area (Cis I) / (Peak Area (Cis I) + Peak Area (Cis II))

Linearity

Linear regression equations were generated using the Analyst software with 1/x weighting, resulting in internal calibration functions with correlation coefficients r ≥ 0.99.

Internal calibration was used for quantitation of the analytes by LC-MS/MS using the QuEChERS and DFG S19 approach. External calibration was used for the acetonitrile extraction of egg. Calibration functions were established with matrix matched standards. Calibration functions ranging from 0.20 ng/mL to 30 ng/mL for whole milk, bovine meat, kidney, liver, from 0.20 ng/mL to 20 ng/mL for fat, from 0.10 ng/mL to 30 ng/mL for egg (QuEChERS) and from 0.10 ng/mL to 5.0 ng/mL for egg (acetonitrile extraction), using ≥ 5 concentration levels, were used to evaluate the final extracts.

Specificity

The method allows the specific determination of the diastereomeric forms of BAS 311 I (Reg. No. 127266) by using two specific mass transitions for quantitation and confirmation: 433 m/z → 191 m/z and 435 m/z → 193 m/z (ammonia adduct). Apparent residues or interferences in blank control specimens were below 20 % of the LOQ. Transformation of alpha-Cypermethrin (Cis II) to its Cis I diastereomeric form was not significant.

Limit of Quantitation

The method achieves an LOQ of ≤0.01 mg/kg per analyte. The LOD (limit of detection) is set to ≤0.002 mg/kg per analyte.

Repeatability

The overall relative standard deviations (RSD, %) for all fortification levels were ≤ 20%. The detailed values are shown in Table 4.2-6.

Reproducibility

Reproducibility of the method was not determined within this validation study.

Stability in Solutions and Extracts

Solutions with the analytes were used throughout the study and gave consistent results and recoveries. Final sample extracts acidified with formic acid were stored prior to final LC/MS/MS analyses for several weeks refrigerated. Acetonitrile extracts of egg were stable for at least 15 days when stored refrigerated.

Examination of sample extracts where only alpha-Cypermethrin (Cis II) was fortified after several weeks of refrigerated storage by LC-MS/MS resulted in no or very little formation of the Cis I peak.

Acceptable recoveries of the analytes in sample extracts demonstrate thus the stability during refrigerated storage.

Conclusion	**It was proven that the method L0231/01 is suitable to determine residues of the diastereomeric forms of BAS 311 I (Cypermethrin, Reg. No. 127266) in in various animal types: Milk, egg, bovine meat, kidney, liver and fat. The residue methods fulfils the registration requirements of SANCO/3029/99 rev.4, 11/07/2000; SANCO/825/00 rev. 8.1, 16/11/2010, OECD ENV/JM/MONO(2007)17, 13-Aug-2007, and EPA OPPTS 860.1340, Aug-1996.**

Report:	CA 4.2/4 Schernikau N., 2014a Independent laboratory validation of the BASF analytical method L0231/01: Determination of individual or combined diastereomeric forms of BAS 311 I (Cis I, Cis II (Alpha-Cypermethrin, BAS 310 I), trans III and trans IV) in animal matrices 2014/1145882
Guidelines:	EC 1107/2009 of the European Parliament and of the Council of 21 Oct 2009, SANCO/3029/99 rev. 4 (11 July 2000), SANCO/825/00 rev. 8.1 (16 November 2010), OECD-ENV/JM/MONO/(2007)17, EPA 860.1340
GLP:	yes (certified by Freie und Hansestadt Hamburg, Behoerde fuer Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg, Germany)

Remark: The title has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study a more appropriate manner. A BASF-method number was also allocated to unequivocally identify the method. The originally assigned title was “Independent Laboratory Validation of BAS 310 I and Isomers in Animal Matrices”; however, by the assigned DocID 2014/1145882 the study can be unequivocally identified.

Principle of the method

The objective of the study was to independently validate BASF method L0231/01 (PTRL validation study P 2984, see MCA 4.2/3) for the determination of individual or combined diastereomeric forms of BAS 311 I (cis I, cis II [alpha-Cypermethrin, BAS 310I], trans III and trans IV) in matrices of animal origin.

Samples of milk, meat, liver, kidney and egg were extracted with acetonitrile after addition of water. After addition of a buffer salt mixture, containing magnesium sulphate, sodium chloride and sodium citrate, the extract was shaken. After centrifugation, an aliquot of the acetonitrile phase was cleaned by dispersive solid phase extraction, using primary secondary amine (PSA). Additionally, samples of egg were extracted with acetonitrile without addition of water and buffer salt mixture. Samples of fat were dissolved in cyclohexane/ethylacetate and cleaned over GPC. Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity. Quantification was performed by using the linear regression with 1/x weighting and internal standards (λ-cyhalothrin).

Recovery findings

The samples were fortified with solutions containing all diastereomeric forms of Cypermethrin (i.e. Cis I/II and Trans III/IV) in appropriate ratios to obtain fortification levels at LOQ (≤ 0.01 mg/kg per isomer) and at 10xLOQ. All mean recovery values at fortification levels of 0.01 mg/kg and 0.1 mg/kg for both ion mass transitions were within the acceptable range of 70 - 120%.
The effect of all matrices on the LC-MS/MS response was assessed by comparing peak areas of matrix-matched standards with solvent standards at equivalent concentrations (at LOQ, 10xLOQ). Matrix effects on the detection of Cypermethrin isomers in extracts of all matrices were found to be significant (> 20 %). Therefore, matrix-matched standards were used for calibration for all matrices.

Detailed results of recoveries for each mass transition and matrix are given in Table 4.2-9. Matrix effects are demonstrated in Table 4.2-10.

Table 4.2-9: BASF method L0231/01, summary of validation results (ILV)

Fortification Level (mg/L)	Transition Ion (m/z) Recovery	Recovery							
		Cis I		Cis II		Trans III		Trans IV	
		433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193
Milk									
0.01	Average	75	79	82	77	75	82	99	103
	RSD	2	2.8	6.6	9.2	5.4	6	13	3.9
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	80	81	82	81	79	79	84	79
	RSD	3.4	4.4	3.2	4.8	3.9	4.9	4.5	4.6
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	77	80	82	79	77	80	91	91
	RSD	4.6	3.8	4.9	7.2	5.3	5.6	13	14
	Replicates	10	10	10	10	10	10	10	10
Meat									
0.01	Average	79	80	78	78	77	78	75	82
	RSD	3	4.3	4.2	3	3.4	7.7	5.4	13
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	76	76	76	76	78	78	77	76
	RSD	5	4.7	3.8	4.4	4.7	2.8	2.2	2.9
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	78	78	77	77	78	78	76	79
	RSD	4.4	5.1	4.2	3.7	4	5.5	4.1	9.7
	Replicates	10	10	10	10	10	10	10	10

Fortification Level (mg/L)	Transition Ion (m/z) Recovery	Recovery							
		Cis I		Cis II		Trans III		Trans IV	
		433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193
Kidney									
0.01	Average	85	85	84	84	85	84	75	73
	RSD	7.1	7.8	4.8	6.4	9.3	8.5	3.8	3.8
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	81	83	79	79	83	87	71	75
	RSD	8.7	8	6.5	7.5	11	9.5	3.1	7.5
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	83	84	82	82	84	86	73	74
	RSD	7.9	7.5	6.2	7.2	9.9	8.7	4.1	5.8
	Replicates	10	10	10	10	10	10	10	10
Liver									
0.01	Average	83	83	82	82	86	87	76	75
	RSD	5.3	5.5	3.5	4.8	4.4	6.4	4.9	5.1
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	84	83	82	83	85	80	71	74
	RSD	4.1	4.3	3.2	3.2	6.5	8.9	3.1	3.8
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	84	83	82	83	85	83	74	74
	RSD	4.5	4.6	3.1	3.9	5.3	8.3	5.1	4.3
	Replicates	10	10	10	10	10	10	10	10
Egg with water									
0.01	Average	85	86	83	82	89	93	86	81
	RSD	4.2	2.9	5.9	4.4	14	4.5	9.2	4.2
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	88	89	87	86	94	90	86	84
	RSD	10	10	9.6	7	8.6	14	7.6	10
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	86	88	85	84	91	92	86	83
	RSD	7.8	7.4	7.9	5.9	11	9.7	7.9	7.8
	Replicates	10	10	10	10	10	10	10	10

Fortification Level (mg/L)	Transition Ion (m/z) Recovery	Recovery							
		Cis I		Cis II		Trans III		Trans IV	
		433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193
Egg without water									
0.01	Average	88	90	85	83	88	90	79	83
	RSD	5.6	6.5	7.3	7.3	7.5	7	8.6	5.2
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	86	87	79	82	84	86	83	82
	RSD	3.6	5	3	6.2	8.1	8.3	4.3	7.2
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	87	88	82	83	86	88	81	82
	RSD	4.6	5.9	6.5	6.4	7.7	7.5	6.8	6
	Replicates	10	10	10	10	10	10	10	10
Fat									
0.01	Average	95	92	93	91	95	100	83	79
	RSD	6.3	5.7	4.3	6.7	11	7.2	8.4	2.6
	Replicates	5	5	5	5	5	5	5	5
0.1	Average	94	93	96	95	98	106	77	75
	RSD	11	12	4.8	7.2	3.5	3.4	5.9	8
	Replicates	5	5	5	5	5	5	5	5
Overall	Average	94	93	95	93	97	103	80	77
	RSD	8.7	8.7	4.5	7	7.5	6.1	8.1	6.2
	Replicates	10	10	10	10	10	10	10	10

Table 4.2-10: BASF method L0231/01, matrix effect on response (ILV)

Matrix / Commodity	Standard Conc. (ng/mL)	Mean Matrix Effect							
		Cis I (%)		Cis II (%)		Trans III (%)		Trans IV (%)	
		433→191 *m/z*	435→193 *m/z*	433→191 *m/z*	435→193 *m/z*	433→191 *m/z*	435→193 *m/z*	433→191 *m/z*	435→193 *m/z*
Milk	1.0 / 10	(-) 99	(-) 99	(-) 100	(-) 100	(-) 99	(-) 99	(-) 99	(-) 99
Meat	1.0 / 10	(+) 45	(+) 46	(+) 27	(+) 28	(+) 66	(+) 51	(+) 45	(+) 49
Liver	1.0 / 10	(-) 93	(-) 93	(-) 93	(-) 93	(-) 92	(-) 91	(-) 95	(-) 90
Kidney	1.0 / 10	(-) 42	(-) 42	(-) 42	(-) 43	(-) 32	(-) 30	(-) 31	(-) 31
Egg	1.0 / 10	(-) 74	(-) 74	(-) 74	(-) 74	(-) 72	(-) 63	(-) 68	(-) 69
Egg w/o water	1.0 / 10	(+) 58	(+) 58	(+) 136	(+) 146	(+) 163	(+) 165	(+) 97	(+) 87
Fat	1.0 / 10	(-) 65	(-) 64	(-) 58	(-) 57	(-) 54	(-) 63	(-) 63	(-) 63

(+) matrix enhancement; (-) matrix suppression

Linearity	The linearity of the detector response of Cypermethrin isomers in matrix were demonstrated by single determination of matrix-matched calibration standards at at least seven (7) concentration levels ranging from 0.2 ng/mL to 30 ng/mL for Cis I and Trans III (Cis II and Trans IV in the given ratios). The calibration curves obtained for both ion mass transitions in all matrices were linear with coefficients of determination (r^2) greater than 0.99. The linearity of the detector response of Cypermethrin isomers in egg was demonstrated by single determination of matrix-matched calibration standards at eight (8) concentration levels ranging from 0.1 ng/mL to 20 ng/mL for Cis I and Trans III (Cis II and Trans IV in the given ratios). The calibration curves obtained for both ion mass transitions in egg were linear with coefficients of determination (r^2) greater than 0.99. The linearity of the detector response of Cypermethrin isomers in egg without water was demonstrated by single determination of matrix-matched calibration standards at eight (8) concentration levels ranging from 0.05 ng/mL to 10 ng/mL for Cis I and Trans III (Cis II and Trans IV in the given ratios). The calibration curves obtained for both ion mass transitions in egg were linear with coefficients of determination (r^2) greater than 0.99. Linear regression was performed with 1/x weighting.
Specificity	Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity. No significant interference above 30 % of LOQ was detected in any of the control specimen extracts of each matrix, so that a high level of selectivity was demonstrated.
Limit of Quantitation	The limit of quantification is ≤ 0.01 mg/kg per isomer in all matrices.
Repeatability	The overall relative standard deviations (RSD, %) for all fortification levels were ≤ 20%. The detailed values are shown Table 4.2-9.
Reproducibility	In context of this ILV study, the reproducibility of the method L0231/01 was estimated. As can be seen from the results, a high reproducibility was determined.
Conclusion	**The method was found to be valid according to the guidance documents SANCO/3029/99 rev. 4 and SANCO/825/00 rev. 8.1 for the determination of Cypermethrin isomers in milk, meat, liver, kidney, egg and fat with a LOQ of ≤ 0.01 mg/kg for Cis I and Trans III (Cis II and Trans IV in the given ratios).**

Report:	CA 4.2/5 [redacted] 2014a Investigation of the extractability of BAS 310 I in samples from animal metabolism studies based on method L0231/01 2014/1145878
Guidelines:	OECD Test Guideline 503 - Metabolism in livestock, SANCO/825/00 rev. 8.1 (16 November 2010), OECD-ENV/JM/MONO/(2007)17
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the methods

Selected samples from hen (BASF DocID 2014/1140293, see M-CA 6.2) and goat (BASF DocID 2014/1083330, see M-CA 6.2) metabolism studies performed with radiolabelled BAS 310 I were extracted with extraction method L0231/01, which is based on the QuEChERS multimethod and the DFG S19 E6 method. All extraction procedures were compared to the extractions used in the animal metabolism studies, except for the 'hen egg yolk' extraction with acetonitrile (method L0231/01), since it is similar to the extraction procedure used in the animal metabolism study.

L0231/01 - Extraction Procedure Based on QuEChERS Multimethod:

For the extraction of muscle, egg yolk and milk matrices:

Aliquots of the homogenized samples (5 g for muscle and milk, 2.5 g for egg yolk) were extracted once with an extraction mixture (5 mL water for muscle and milk / 8 mL water for egg yolk and 10 mL acetonitrile) by vigorous shaking. Subsequently, a blend of buffer salts was added ($MgSO_4$, NaCl, disodium citrate and trisodium citrate). The mixture was shaken strongly and centrifuged. The supernatant was cleaned up using a dispersive SPE tube (Agilent) and concentrated. The residue was redissolved in a mixture of acetonitrile, water and Triton X-100 (for muscle and milk 300/280/20, v/v/v; for egg yolk 600/280/20, v/v/v). For each matrix, one replicate per extraction was generated.

L0231/01 - Extraction Procedure Based DFG S19 E6 Method (fat matrices):

For the extraction of fat matrices:

5 g of the homogenized sample were supplemented with 50 mL cyclohexane/ethyl acetate (1/1, v/v). The emulsion was decanted over cotton wool and again supplemented with 50 mL cyclohexane/ethyl acetate (1/1, v/v). Thereafter, the sample was concentrated and redissolved in 35 mL isohexane. The sample was partitioned four times using a separating funnel, each time with 30 mL acetonitrile. The pooled acetonitrile phase was subsequently concentrated and redissolved in a mixture of acetonitrile, water and Triton X-100 (approximate ratio of 9/6/1, v/v/v).

The extracts were analysed by LSC and ^{14}C-BAS 310 I was quantified by HPLC method LC01 (identical to method LC02 from the goat metabolism study). The identification of BAS 310 I was based on retention time comparison of the ^{14}C signals of the quantitative HPLC analyses with those of the application solution used in a reference metabolism study.

The extraction efficiency was calculated as percentage of the mg/kg values obtained from the extractions in the reference metabolism studies. For the calculation of general extractability of radioactive residues, the LSC measurements of unconcentrated extracts have been used.

Limit of Quantification

The validated LOQ of the extraction method L0231/01 is 0.01 mg/kg per analyte.

Extractability

The extractability of the matrix muscle with method L0231/01 (QuEChERS based part) was 47.4 % TRR for hen and 75.7 % TRR for goat. In fat, 87.9 % TRR (hen) and 96.9 % TRR (goat) were extracted with method L0231/01 (DFG S19 E6 based part). The extraction of milk with method L0231/01 (QuEChERS based part) resulted in 59.0 % TRR. Egg yolk was extracted with method L0231/01 (QuEChERS based part), whereby the extractability was low and accounted for 27.8 % TRR.

Compared with the reference metabolism studies, the extractabilities of the tested extraction protocol were comparable for goat muscle, hen fat and goat fat and ranged from 78.6 % to 95.1 % of the metabolism studies extractions. For hen muscle and goat milk, the extractabilities were somewhat lower and ranged from 63.3 % to 69.2 % Met. The extractability of egg yolk was significantly lower and accounted for 34.7 % of the metabolism study extractions.

Detailed data of the extractability of radioactive residues with the extraction methods from the reference metabolism studies are summarized in Table 4.2-11.

HPLC Results for Extracts of Animal Matrices

Muscle Extracts: In the hen muscle extract, 0.006 mg/kg of BAS 310 I (33.7 % TRR) were identified and in the goat muscle extract, 0.019 mg/kg of BAS 310 I (64.9 % TRR) were identified. The amount of BAS 310 I in goat muscle is identical to the amount of BAS 310 I, which was identified in the reference metabolism study (100.0 % of the metabolism study extractions). For hen muscle, the amount of BAS 310 I is higher in this study (150.0% of the metabolism study extractions). The relative increase of BAS 310 I in hen muscle in this study might be due to the low absolute amount of BAS 310 I.

Fat Extracts: The extraction of fat matrices resulted in 0.044 mg/kg (52.2 % TRR, hen fat) and 0.084 mg/kg (59.5 % TRR, goat fat) of BAS 310 I. The extractability of BAS 310 I was comparable to the extractabilities of the reference metabolism studies (115.8 % Met for hen fat and 65.1 % Met for goat fat).

Milk Extracts: The extraction of the matrix milk yielded 0.033 mg/kg (61.3 % TRR) of BAS 310 I. The extractability of BAS 310 I was comparable with the reference metabolism study (76.7 %).

Egg Yolk Extracts: In the extract of egg yolk (QuEChERS based part), 0.027 mg/kg of BAS 310 I (18.0 % TRR) were identified. Compared with the HPLC results of the reference metabolism study, lower amounts of BAS 310 I were extracted (52.9 % of the metabolism study extractions).

The HPLC results of the extracts of animal matrices and the HPLC results of the extracts of the reference metabolism studies are summarized in Table 4.2-11.

Table 4.2-11: Extractability of animal matrices based on Method L0231/01

Extraction Method	ERR			BAS 310 I		
	[mg/kg]	[% TRR]	[% Met]	[mg/kg]	[% TRR]	[% Met]
Hen Muscle						
L0231/01, QuEChERS based part	0.009	47.4	69.2	0.006	33.7	150.0
Acetonitrile and Water (Hen Metabolism Study)	0.013	69.0	n.a.	0.004	20.9	n.a.
Goat Muscle						
L0231/01, QuEChERS based part	0.022	75.7	78.6	0.019	64.9	100.0
Acetonitrile and Water (Goat Metabolism Study)	0.028	97.5	n.a.	0.019	66.0	n.a.
Hen Fat						
L0231/01, DFG S19 E6 based part	0.074	87.9	89.2	0.044	52.2	115.8
Acetonitrile, Isohexane and Water (Hen Metabolism Study)	0.083	98.9	n.a.	0.038	45.4	n.a.
Goat Fat						
L0231/01, DFG S19 E6 based part	0.136	96.9	95.1	0.084	59.5	65.1
Acetonitrile, Isohexane and Water (Goat Metabolism Study)	0.143	101.4	n.a.	0.129	91.6	n.a.
Goat Milk						
L0231/01, QuEChERS based part	0.031	59.0	63.3	0.033	61.3	76.7
Acetonitrile (Goat Metabolism Study)	0.049	92.3	n.a.	0.043	80.8	n.a.
Hen Egg Yolk						
L0231/01, QuEChERS based part	0.041	27.8	34.7	0.027	18.0	52.9
Acetonitrile, Isohexane and Water (Hen Metabolism Study)	0.118	78.8	n.a.	0.051	34.3	n.a.

% Met = Extraction efficiency compared to the extraction method used in the respective metabolism study
n.a. = not applicable

Conclusion

Generally, the extractabilities in this study were comparable to the extractabilities of the reference metabolism studies (63.3-95.1 % Met); only for hen egg yolk (QuEChERS based part, 34.7 % Met) the relative extractability was lower.

For all investigated matrices and extraction methods of this study, the amount of BAS 310 I was comparable to the results of the reference metabolism studies (76.7 to 125.0 % of the metabolism studies extractions). Only for hen egg yolk (QuEChERS based part), the relative extractability was somewhat lower (52.9 % of the metabolism studies extractions).

However, for the acetonitrile based extraction of eggs in L0231/01 an identical extraction efficiency can be assumed in comparison to the reference metabolism study, since the majority of BAS 310 I was determined to be in the acetonitrile fraction in the metabolism study (~100 % Met).

Report:	CA 4.2/6 [redacted] 2014b Investigation of the extractability of BAS 310 I in samples from animal metabolism studies 2014/1145877
Guidelines:	OECD Test Guideline 503 - Metabolism in livestock, SANCO/825/00 rev. 8.1 (16 November 2010), OECD-ENV/JM/MONO/(2007)17
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Principle of the methods

Selected samples from hen (BASF DocID 2014/1140293, see M-CA 6.2) and goat (BASF DocID 2014/1083330, see M-CA 6.2) metabolism studies performed with radiolabeled BAS 310 I were extracted. Three different extraction procedures designed for the determination of BAS 310 I were tested, which are based on the residue methods SAMS 461-1 (AL-245-001), SAMS 456-1 (AL-245-003) and DFG S19 (AL-245-007). All extraction procedures were compared to the extractions used in the animal metabolism studies.

Method 1 - SAMS 461-1 (AL-245-001):

Aliquots of the homogenized samples were mixed with Na_2SO_4 in an approximate ratio of 1/2 (w/w) in case of the matrix fat or 1/12 (w/w) in case of the matrix muscle. The sample/salt mixture was extracted by boiling in acetone/isohexane (1/2, v/v) in an approximate ratio of 1/10 (sample/solvents, w/v). The extraction procedure was performed three times. The supernatants were decanted over cotton wool, pooled and made up to an adequate volume with isohexane. The extracts were concentrated and redissolved in isohexane. The samples were partitioned with acetonitrile in a separating funnel. The acetonitrile supernatants were pooled, concentrated and redissolved in a mixture of acetonitrile/water/Triton X-100 (approximately 1150/650/30, v/v/v) in case of the matrix fat and water/acetonitrile/Triton X-100 (690/300/10, v/v/v) in the case of the matrix muscle.

Method 2 - SAMS 456-1 (AL-245-003):

The homogenized milk sample was consecutively mixed with a 5 % solution of aqueous potassium oxalate, ethanol, diethyl ether and isohexane in a ratio of 10/1/10/20/10 (v/v/v/v/v). The supernatant was concentrated, made up with isohexane to an adequate volume and partitioned with acetonitrile in a separating funnel. The acetonitrile supernatants were pooled, concentrated and redissolved in a mixture of water/acetonitrile/Triton X-100 (680/300/20, v/v/v).

Method 3 - DFG S19 (AL-245-007):

Prior to extraction, 25.0 g of the sample were mixed with an adequate amount of water to result in an approximate ratio of water/acetone 1/2 (v/v) after the addition of acetone in the subsequent extraction step. After the addition of water, the mixture was homogenized using a Polytron. A volume of 100 mL acetone (final water/acetone ratio 1/2, v/v) and 17.5 g NaCl were added and the mixture was homogenized again. Thereafter, 50 mL ethyl acetate/cyclohexane (1/1, v/v) were added and the mixture was again homogenized. The homogenate was centrifuged, decanted over cotton wool and partitioned in a separating funnel (organic phase/ aqueous phase). The organic phase was made up with acetone to an adequate volume. The extract was concentrated, redissolved in isohexane and partitioned with acetonitrile in a separating funnel. In case of the matrix egg yolk, the acetonitrile supernatants were pooled, concentrated and redissolved in a mixture of water/acetonitrile/Triton X-100 (700/700/50, v/v/v).

The sample was centrifuged prior to measurement. In case of the matrix milk, the acetonitrile supernatants were redissolved in a mixture of water/acetonitrile/Triton X-100 (680/600/20, v/v/v) after the concentration step.

The extracts were analysed by LSC and ^{14}C-BAS 310 I was quantified by HPLC method LC01 (identical to HPLC method LC02 from goat metabolism study). The identification of BAS 310 I was based on retention time comparison of the ^{14}C signals of the quantitative HPLC analyses with those of the application solution used in a reference metabolism study

Limit of Quantification

The validated LOQs of the extraction methods SAMS 461-1 and SAMS 456-1 are 0.05 mg/kg and 0.01 mg/kg, respectively; the method based on DFG S19 was developed with an LOQ of 0.01 mg/kg

Extractability

The extractability of the matrix muscle with extraction method 1 was 55.7 % TRR for hen and 69.2 % TRR for goat. In fat, 88.9 % TRR (hen) and 98.7 % TRR (goat) were extracted with extraction method 1. Milk was extracted with methods 2 and 3 resulting in 94.3 % TRR and 90.7 % TRR, respectively. Egg yolk was extracted with method 3, whereby the extractability accounted for 73.0 % TRR.

Compared with the reference metabolism studies, the extractabilities of the tested extraction protocols were comparable for all matrices and ranged from 71.4 % to 102.0 % of the metabolism studies extractions.

Detailed data of the extractability of radioactive residues with the extraction methods from the reference metabolism studies are summarized in Table 4.2-12.

HPLC Results for Extracts of Animal Matrices

Muscle Extracts: In the hen muscle extract, 0.006 mg/kg of BAS 310 I (30.9 % TRR) were identified and in the goat muscle extract, 0.015 mg/kg of BAS 310 I (50.2 % TRR) were identified. The amount of BAS 310 I in hen and goat muscle was comparable to the amount of BAS 310 I in the reference metabolism studies. For goat muscle, 78.9 % of the metabolism studies extractions were identified and for hen muscle, 150.0 % of the metabolism studies extractions were identified. The relative increase of BAS 310 I observed in hen muscle might be due to the low absolute amounts of BAS 310 I.

Fat Extracts: The extraction of fat matrices with method 1 resulted in 0.053 mg/kg (63.2 % TRR, hen fat) and 0.120 mg/kg (85.2 % TRR, goat fat) of BAS 310 I. The extractability of BAS 310 I was comparable to the extractabilities of the reference metabolism studies (139.5 % Met for hen and 93.0 % Met for goat).

Milk Extracts: The extraction of the matrix milk with methods 2 and 3 yielded 0.041 mg/kg (76.8 % TRR) and 0.037 mg/kg (69.4 % TRR) of BAS 310 I. The extractability of BAS 310 I was comparable with the reference metabolism study (95.3 % for extraction method 2 and 86.0 % for extraction method 3).

Egg Yolk Extracts: In the extract of egg yolk (method 3), 0.034 mg/kg of BAS 310 I (22.8 % TRR) were identified. Although the overall extractability of the radiolabelled material was significantly lower, the amount of freely extractable BAS 310 I was largely comparable (66.7 % of the metabolism study extractions).

The HPLC results of the extracts of animal matrices and the HPLC results of the extracts of the reference metabolism studies are summarized in Table 4.2-12.

Table 4.2-12: Extractability of animal matrices

Extraction	ERR			BAS 310 I		
	[mg/kg]	[% TRR]	[% Met]	[mg/kg]	[% TRR]	[% Met]
Hen Muscle						
Method 1 (Acetone / Isohexane)	0.011	55.7	84.6	0.006	30.9	150.0
Hen Metabolism Study (Acetonitrile and Water)	0.013	69.0	n.a.	0.004	20.9	n.a.
Goat Muscle						
Method 1 (Acetone / Isohexane)	0.020	69.2	71.4	0.015	50.2	78.9
Goat Metabolism Study (Acetonitrile and Water)	0.028	97.5	n.a.	0.019	66.0	n.a.
Hen Fat						
Method 1 (Acetone / Isohexane)	0.075	88.9	90.4	0.053	63.2	139.5
Hen Metabolism Study (Acetonitrile, Isohexane and Water)	0.083	98.9	n.a.	0.038	45.4	n.a.
Goat Fat						
Method 1 (Acetone / Isohexane)	0.139	98.7	97.2	0.120	85.2	93.0
Goat Metabolism Study (Acetonitrile, Isohexane and Water)	0.143	101.4	n.a.	0.129	91.6	n.a.
Goat Milk						
Method 2 (Potassium Oxalate, Ethanol, Diethyl Ether and Isohexane)	0.050	94.3	102.0	0.041	76.8	95.3
Method 3 (Water / Acetone)	0.048	90.7	98.0	0.037	69.4	86.0
Goat Metabolism Study (Acetonitrile)	0.049	92.3	n.a.	0.043	80.8	n.a.
Hen Egg Yolk						
Method 3 (Water / Acetone)	0.109	73.0	92.4	0.034	22.8	66.7
Hen Metabolism Study (Acetonitrile, Isohexane and Water)	0.118	78.8	n.a.	0.051	34.3	n.a.

[1] % Met = Extraction efficiency compared to the extraction method used in the respective metabolism study

n.a. = not applicable

Conclusion	**For all investigated matrices and extraction methods of this study, the amount of BAS 310 I was comparable to the results of the reference metabolism studies (66.7 to 116.3 % of the metabolism studies extractions)** **Generally, the extractabilities in this study were comparable to the extractabilities of the reference metabolism studies (71.4-102.0 % Met).**

(b) Methods for the analysis in soil and water

Analytical methods for the determination of alpha-Cypermethrin and its relevant metabolites for post-approval and monitoring purposes in soil are identical to the ones validated for pre-registration purposes. Respective methods are described in detail in document M, chapter 4.1.2; hence they are not described again in detail in this chapter.

The newly developed method for the determination of the diastereomeric forms of Cypermethrin and its metabolites as well as its ILV is summarised below.

Report:	CA 4.2/7 Andrews, R.S., 2014a Validation of Method D1413/01 (L0258/01): Method for the Determination of the Diastereomers of BAS 311 I and Metabolites 3-Phenoxybenzoic acid (Reg. No. 130213), DCVA (Cis and Trans Isomers, Reg. No. 180011) and 3-Phenoxybenzaldehyde (Reg. No. 4080665) in Water by LC-MS/MS 2014/7002180
Guidelines:	OPPTS 850.7100, OPPTS 835.6100, PR Notice 2011-3, SANCO/3029/99 rev. 4 (11 July 2000, pre-registration residue methods) SANCO/825/00 rev. 8.1(16 November 2010, post-registration residue methods) OCSPP 850.6100: Environmental Chemistry Methods and Associated Independent Laboratory Validation
GLP:	yes

Remark: The title has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study a more appropriate manner as analysis addressed all four isomers; hence BAS 311 was used in the updated study title. The originally assigned title was "Validation of Method D1413/01 (L0258/01): Method for the Determination of the Diastereomers of BAS 310 I and Metabolites 3-Phenoxybenzoic acid (Reg. No. 130213), DCVA (Cis and Trans Isomers, Reg. No. 180011) and 3-Phenoxybenzaldehyde (Reg. No. 4080665) in Water by LC-MS/MS", however by the assigned DocID 2014/7002180, the study can be unequivocally identified.

Principle of the method

Cypermethrin (BAS 311 I) and the four isomers of Cypermethrin (cis I, cis II, trans III, trans IV):

Using BASF Analytical Method No. D1413/01 (L0258/01), residues of parent Cypermethrin in water samples (500 mL each) are acidified and then cleaned-up and concentrated on an Oasis® HLB solid phase extraction (SPE) column eluted with acetonitrile followed by dichloromethane. The residues in the eluent are evaporated to dryness, re-dissolved in acidified acetonitrile:water (50:50, v/v), and then determined by high performance liquid chromatography (HPLC) with detection using positive electrospray ionization tandem mass spectrometry (MS/MS-ESI), monitoring at m/z 433→191 (proposed as the primary transition for quantitation) and m/z 435→193 (typically used for confirmatory purposes) for each of the four isomers. The isomers are separated by their retention times on the HPLC column, enabling quantitation of the contribution of each analyte. The limit of quantitation (LOQ) of the method is 0.75 ng/L for parent Cypermethrin and the four isomers in water.

The metabolites in water are extracted and determined in two additional, separate analyses:

DCVA (cis and trans) and PBA:

DCVA and PBA residues in water samples (60 mL each) are acidified and partitioned by shaking with ethyl acetate:cyclohexane (40:60, v/v). Residues in the organic layer are evaporated to dryness, re-dissolved and diluted with ethyl acetate and acidified water, and then concentrated to an aqueous remainder. The residues are then diluted to a final volume of acidified methanol:water (20:80, v/v) and analyzed by HPLC/MS/MS, monitoring in the negative mode ion transitions at m/z 213→93 (quantitation) and m/z 213→169 (confirmation) for PBA. The determination of DCVA is conducted without any fragmentation of the parent ion, due to the nature of the ionization, monitoring at m/z 207 or 209 in the negative mode for both isomers, which are separated by retention time and quantitated as separate species. An alternate chromatographic method is used for DCVA for confirmatory purposes. The limit of quantitation (LOQ) of the method is 50 ng/L for DCVA (cis and trans) and PBA in water.

POAL:

POAL residues in water samples (100 mL each) are acidified, and then cleaned-up and concentrated on a styrene-divinylbenzene polymeric (SDB-L) SPE column eluted with acetonitrile. The residues are then diluted to a final volume of acetonitrile:water (50:50, v/v) and determined by HPLC/MS/MS monitoring ion transitions at m/z 199→171 (for quantitation) and m/z 199→65 (for confirmation) in the negative mode. The limit of quantitation (LOQ) of the method is 50 ng/L for POAL in water.

All results are calculated by direct comparison of the sample peak responses to those of external standards.

Recovery findings

Method L0258/01 was proved to be suitable to determine residues of Cypermethrin (cis I, cis II, trans III, trans IV) and its metabolites PBA, DCVA (cis and trans) and POAL to the respective limit of quantification in drinking and surface water.

Mean recoveries of Cypermethrin residues (cis I, cis II, trans III, trans IV) from drinking and surface water samples fortified with each isomer at 0.75 ng/L and 7.5 ng/L ranged from 77 to 109% (relative standard deviation, ≤20%), considering results obtained using both the primary and secondary ion transitions.

Mean recoveries of PBA residues from drinking and surface water samples fortified at 50 ng/L and 500 ng/L ranged from 94 to 109% (RSD, ≤13%) for both the primary and secondary ion transitions.

Mean recoveries of residues of cis- and trans-DCVA from drinking and surface water samples fortified with each isomer at 50 ng/L and 500 ng/L ranged from 74 to 110% (RSD, ≤16%), considering results obtained using both the primary and secondary ion.

Mean recoveries of residues of POAL from drinking and surface water samples fortified at 50 ng/L and 500 ng/L were in the acceptable range (actual 73 to 92%, RSD ≤12%) for both transitions, except for surface water at the limit of quantitation (50 ng/L), where mean recoveries were 57% (RSD 8%) and 60% (RSD 10%), monitoring at the primary and secondary ion transition, respectively. Although on the margins of the acceptable range, the results were consistent, with low relative standard deviations; therefore, the data obtained are considered of acceptable accuracy and precision for this metabolite.

The detailed results are given in Table 4.2-13.

Table 4.2-13: Results of the method validation for the determination of Cypermethrin (cis I, cis II, trans III, trans IV) and its metabolites PBA, DCVA (cis and trans) and POAL in surface water and drinking water

Analyte	m/z	Matrix	Replicates	Fortification level [ng L^{-1}]	Mean recovery [%]	RSD [%]
Cis I Cypermethrin	433->191	drinking water	5	0.75	89	5
			5	7.5	85	4
		surface water	5	0.75	91	4
			5	7.5	84	5
	435->193	drinking water	5	0.75	90	2
			5	7.5	87	7
		surface water	5	0.75	94	5
			5	7.5	85	1
Cis II Cypermethrin	433->191	drinking water	5	0.75	93	6
			5	7.5	89	5
		surface water	5	0.75	96	3
			5	7.5	84	5
	435->193	drinking water	5	0.75	93	4
			5	7.5	90	6
		surface water	5	0.75	92	5
			5	7.5	83	4
Trans III Cypermethrin	433->191	drinking water	5	0.75	93	9
			5	7.5	84	4
		surface water	5	0.75	99	7
			5	7.5	86	4
	435->193	drinking water	5	0.75	101	16
			5	7.5	82	3
		surface water	5	0.75	96	8
			5	7.5	77	5

Analyte	m/z	Matrix	Replicates	Fortification level [ng L^{-1}]	Mean recovery [%]	RSD [%]
Trans IV Cypermethrin	433->191	drinking water	5	0.75	109	15
			5	7.5	86	2
		surface water	5	0.75	91	6
			5	7.5	85	3
	435->193	drinking water	5	0.75	104	20
			5	7.5	88	4
		surface water	5	0.75	100	2
			5	7.5	82	10
Cis-DCVA	207	drinking water	5	50	75	12
			5	500	79	14
		surface water	5	50	92	13
			5	500	95	8
	209	drinking water	5	50	79	16
			5	500	82	16
		surface water	5	50	82	14
			5	500	93	9
Trans-DCVA	207	drinking water	Acceptable recovery data could not be obtained for this analyte monitoring ion m/z 207; therefore, the alternate ion (m/z 209, shown below) was used.			
		surface water	5	50	110	14
			5	500	102	7
	209	drinking water	5	50	93	11
			5	500	86	14
		surface water	5	50	93	8
			5	500	107	9
PBA	213->93	drinking water	5	50	109	11
			5	500	99	9
		surface water	5	50	95	13
			5	500	103	7
	213->169	drinking water	5	50	109	9
			5	500	97	7
		surface water	5	50	94	13
			5	500	101	9
POAL	199->171	drinking water	5	50	92	11
			5	500	80	6
		surface water	5	50	57	8
			5	500	73	6
	199->65	drinking water	5	50	90	12
			5	500	79	8
		surface water	5	50	60	10
			5	500	75	6

Linearity

Acceptable linearity was observed for the standard range and the two mass transitions tested for each analyte: The method-detector response was linear over the nominal 0.25-0.5 ng/mL range (r = ≥0.9949) for Cypermethrin (each isomer), the 0.6-10 ng/mL range (r = ≥0.9920) for DCVA (each isomer) and PBA, and the 0.08-2.0 ng/mL range (r = ≥0.9980) for POAL.

Specificity

The method successfully determines residues of Cypermethrin in water by LC-MS/MS. This was demonstrated on the basis of two different water types (surface water and drinking water). No interfering peaks were found at the retention times for these analytes, or if they were encountered, as with DCVA, the interferences were resolved using the available alternate (optional) chromatography methods. The experiment to evaluate any potential matrix effects showed that the matrix load in the samples from the each water type had no significant influence on analysis (matrix effects < 20%); therefore, the validation samples were analyzed using solvent-based calibration standard solutions.

Limit of Quantification

The method has a limit of quantification of 0.75 ng/L for parent Cypermethrin (for each of the four isomers) and 50 ng/L for the metabolites DCVA (cis and trans), PBA and POAL.

Repeatability

The relative standard deviations (RSD, %) for all fortification levels were ≤ 20%.

Stability in Solutions and Extracts

The substance solutions and final sample extracts were stored in a refrigerator and were used within the time period of stability. The stability of POAL was not tested in the final volume solution of acetonitrile:water (1:1, v/v) due to an oversight.

Conclusion

It was demonstrated that BASF Analytical Method No. D1413/01 (L0258/01) fulfils the requirements with regard to specificity, repeatability, limit of quantification, and recoveries and is, therefore, applicable to correctly determine residues of the Cypermethrin in drinking (well) water and surface (pond) water.

Report:	CA 4.2/8 Perez S., Perez R., Adams J, 2015 Independent Laboratory Validation of Method D1413: Method for the Determination of Diasteromers of BAS 311 I and Metabolites 3-Phenoxybenzoic acid (Reg. No. 130213), DCVA (Cis and Trans Isomers, Reg. No. 180011) and 3-Phenoxybenzaldehyde (Reg. No. 4080665) in Water by LC-MS/MS. 2014/7002181
Guidelines:	OCSSP 860.6100, SANCO/825/00 rev 8.1 (Nov.16, 2010), SANCO/3029/99 rev. 4 (11/07/00), SANCO/10684/2009, ENV/JMONO (2007)17
GLP:	yes

Remark: The title has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study a more appropriate manner as analysis addressed all four isomers; hence BAS 311 was used in the updated study title. The originally assigned title was "Independent Laboratory Validation of Method D1413/01 (L0258/01): Method for the Determination of the Diastereomers of BAS 310 I and Metabolites 3-Phenoxybenzoic acid (Reg. No. 130213), DCVA (Cis and Trans Isomers, Reg. No. 180011) and 3-Phenoxybenzaldehyde (Reg. No. 4080665) in Water by LC-MS/MS", however by the assigned DocID 2014/70021801 the study can be unequivocally identified.

The presented OECD-summary of the study is based on under GLP audited data presented in a fully audited draft report. However, as the report was not finalized at time of submission of the electronic version of the dossier, a copy of the original, signed final report of the study will be submitted upon availability, which is estimated to be early February.

Principle of the method

For the analysis of BAS 311 I (Cypermethrin) isomers Cis I, Cis II, Trans III, and Trans IV, a sample aliquot (500 mL) is concentrated on an Oasis HLB® SPE column. The eluent is concentrated to dryness and reconstituted for analysis by LC-MS/MS.

For the analysis of DCVA (*cis* and *trans*) and 3-PBA, a sample aliquot (60 mL) is extracted with a mixture of cyclohexane:ethyl acetate using a liquid–liquid partition. The organic partition is evaporated in the presence of an aqueous keeper and diluted for analysis by LC-MS/MS.

Method D (utilizing an Xbridge PFP column) of the analytical method serves as an alternative chromatographic technique for the analysis of DCVA in the case that chromatographic interferences prevents the use of Method B (utilizing an Xbridge phenyl column).

The validation trials for drinking and surface water were run using Method B with the phenyl column; however, low recoveries were obtained for cis-DCVA in surface water. The validation trials for surface and drinking water were injected a second time using the alternative chromatographic technique described in Method D with the PFP column and obtained good results.

For the analysis of POAL, a sample aliquot (100 mL) is concentrated onto a polymeric SDB-L column. The eluent is diluted for analysis by LC-MS/MS.

Recovery findings

Method D1413/01 (L0258/01) was proved to be suitable to determine residues of Cypermethrin (cis I, cis II, trans III, trans IV) and its metabolites 3-PBA, DCVA (cis and trans) and POAL to the respective limit of quantification in drinking and surface water.

Mean recoveries of Cypermethrin residues (cis I, cis II, trans III, trans IV) from drinking and surface water samples fortified with each isomer at 0.75 ng/L and 7.5 ng/L ranged in total from 51 to 126% (relative standard deviations per analyte and transition were always ≤20%; except for trans III cypermethrin (2^{nd} transition), trans IV cypermethrin (1^{st} + 2^{nd} transition) in surface water, for which RSDs of slightly above 20% were determined), considering results obtained using both the primary and secondary ion transitions.

Mean recoveries of 3-PBA residues from drinking and surface water samples fortified at 50 ng/L and 500 ng/L ranged from 65 to 113% (RSD, ≤14%) for both the primary and secondary ion transitions.

Mean recoveries of residues of cis- and trans-DCVA from drinking and surface water samples fortified with each isomer at 50 ng/L and 500 ng/L (Method B and D) ranged from 47 to 243% (RSD, ≤47%), considering results obtained using both the primary and secondary ion.

Mean recoveries of residues of POAL from drinking and surface water samples fortified at 50 ng/L and 500 ng/L ranged from 80 to 114% (RSD, ≤10%), considering results obtained using both the primary and secondary ion.

The detailed results are given in Table 4.2-14.

Table 4.2-14: Results of the method validation for the determination of Cypermethrin (cis I, cis II, trans III, trans IV) and its metabolites 3-PBA, DCVA (cis and trans) and POAL in surface water and drinking water

Analyte	m/z	Matrix	Replicates	Fortification level [ng L^{-1}]	Mean recovery [%]	RSD [%]
Cis I Cypermethrin	433->191	drinking water	5	0.75	97	6.8
			5	7.5	95	11.1
		surface water	5	0.75	82	17.5
			5	7.5	87	12.0
	435->193	drinking water	5	0.75	94	5.8
			5	7.5	95	7.1
		surface water	5	0.75	79	18.7
			5	7.5	84	11.3
Cis II Cypermethrin	433->191	drinking water	5	0.75	96	8.5
			5	7.5	89	10.1
		surface water	5	0.75	73	17.2
			5	7.5	83	12.0
	435->193	drinking water	5	0.75	93	6.7
			5	7.5	90	12.5
		surface water	5	0.75	77	17.0
			5	7.5	81	13.3
Trans III Cypermethrin	433->191	drinking water	5	0.75	74	9.6
			5	7.5	82	11.0
		surface water	4	0.75	104	17.6
			5	7.5	101	17.1
	435->193	drinking water	5	0.75	76	8.6
			5	7.5	85	11.7
		surface water	4	0.75	91	22.3
			5	7.5	89	21.4
Trans IV Cypermethrin	433->191	drinking water	5	0.75	86	13.5
			5	7.5	83	8.0
		surface water	4	0.75	109	20.5
			5	7.5	98	16.0
	435->193	drinking water	5	0.75	92	5.8
			5	7.5	87	15.0
		surface water	4	0.75	105	20.4
			5	7.5	96	16.1

Analyte	m/z	Matrix	Replicates	Fortification level [ng L^{-1}]	Mean recovery [%]	RSD [%]
Cis-DCVA	207	drinking water (Method B)	5	50	97	20.8
			4	500	85	19.4
		drinking water[1] (Method D)	5	50	77	11.1
			4	500	81	8.6
		surface water[1] (Method B)	5	50	107	11.3
			5	500	83	14.7
		surface water[1] (Method D)	5	50	76	7.0
			5	500	98	23.3
	209	drinking water (Method B)	5	50	76	6.8
			4	500	102	13.6
		drinking water[1] (Method D)	5	50	81	9.9
			4	500	88	19.1
		surface water[1] (Method B)	5	50	78	25.4
			5	500	57	7.1
		surface water[1] (Method D)	5	50	111	7.7
			5	500	86	13.8
Trans-DCVA	207	drinking water (Method B)	5	50	93	4.0
			4	500	98	14.5
		drinking water[1] (Method D)	5	50	74	15.7
			4	500	82	3.5
		surface water (Method B)	5	50	78	22.3
			5	500	85	5.5
		surface water[1] (Method D)	5	50	180	42.3
			5	500	103	15.3
	209	drinking water (Method B)	5	50	103	6.3
			4	500	83	10.6
		drinking water[1] (Method D)	5	50	76	20.9
			4	500	80	13.7
		surface water (Method B)	5	50	93	18.7
			5	500	80	16.3
		surface water[1] (Method D)	5	50	98	29.9
			5	500	84	10.0

Analyte	m/z	Matrix	Replicates	Fortification level [ng L^{-1}]	Mean recovery [%]	RSD [%]
3-PBA	213->93	drinking water	5	50	90	9.4
			4	500	91	7.9
		surface water	5	50	84	16.5
			5	500	85	7.3
	213->169	drinking water	5	50	100	5.8
			4	500	97	10.8
		surface water	5	50	97	17.2
			5	500	93	11.3
POAL	199->171	drinking water	5	50	92	9.7
			5	500	101	1.6
		surface water	5	50	109	5.6
			5	500	97	3.1
	199->65	drinking water	5	50	89	12.2
			5	500	100	4.4
		surface water	5	50	96	10.6
			5	500	85	3.6

Statistical calculations in this table are based on unrounded values; therefore, slight differences may be noted in calculations performed using rounded numbers.

Linearity

Good linearity ($r > 0.99$) was observed in the range of 0.0005 to 0.0200 ng injected in the standard solutions for Cypermethrin isomers (Cis I/II and Trans III/IV) for all mass transitions analyzed.

Good linearity ($r > 0.99$) was observed in the range of 0.0180 to 0.3000 ng injected in the standard solutions for DCVA cis/trans mixture and 3-PBA for all mass transitions analyzed.

Good linearity ($r > 0.99$) was observed in the range of 0.0048 to 0.1200 ng injected in the standard solutions for POAL for all mass transitions analyzed.

Specificity

The method determines residues of Cypermethrin, DCVA, 3-PBA and POAL. No interfering peaks were found at the retention times of the analyte. The use of matrix-matched standards was not necessary.

Limit of Quantification

The method has a limit of quantification of 0.75 ng/L for parent Cypermethrin (for each of the four isomers) and 50 ng/L for the metabolites DCVA (cis and trans), 3-PBA and POAL.

Repeatability

The relative standard deviations (RSD, %) for all fortification levels were mostly ≤ 20%. For some fortification series RSDs slightly above 20% were determined.

Reproducibility

In context of this ILV study, the reproducibility of this analytical method was estimated. As can be seen from the results, a high reproducibility was determined.

Conclusion

The method D1413/01 (L0258/01) for the analysis of Cypermethrin (cis I, cis II, trans III, trans IV) and its metabolites 3-PBA, DCVA (cis and trans) and POAL) in surface water and drinking water used LC-MS/MS for final determination, with limit of quantification of 0.75 ng/L for parent Cypermethrin (for each of the four isomers) and 50 ng/L for the metabolites DCVA (cis and trans), 3-PBA and POAL.
It was demonstrated that BASF Analytical Method No. D1413/01 (L0258/01) fulfils the requirements with regard to specificity, repeatability, limit of quantification, and recoveries and is, therefore, applicable to correctly determine residues of the Cypermethrin in drinking (well) water and surface (pond) water.

(c) Methods for the analysis in air

Report:	CA 4.2/9 Bendig P., Class T., 2013a Development and validation of an analytical method for the determination of Alpha-Cypermethrin (BAS 310 I) in air 2013/1281809
Guidelines:	SANCO/825/00 rev. 8.1 (16 November 2010), SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	yes (certified by Landesamt fuer Umwelt, Messungen und Naturschutz Baden Wuerttemberg, Karlsruhe, Germany)

The objective of this study was to validate an analytical method for the determination of alpha-Cypermethrin in air (warm: approx. 35°C, humid: relative humidity approx. 95%), with a limit of quantification (LOQ) of 1.4 µg m^{-3} alpha-Cypermethrin in air.

Principle of the method

Air is sucked through XAD adsorption tubes at about 1.0 L min^{-1} for 6 hours (total air sampling volume ~0.36 m^3). Subsequently, the adsorption material is extracted with ethyl acetate, the internal standard lambda-cyhalothrin is added to the extract, and finally analyzed by gas chromatography with mass spectrometric detection in the negative chemical ionisation mode (GC/MS(NCI)), monitoring three fragment ions.

The limit of quantification is defined as the lowest fortification level used in the validation process corresponding to a concentration of 1.4 µg m^{-3} alpha-Cypermethrin in air.

Recovery findings

The analytical method was validated with warm, humid air (approx. 35°C, approx. 95% relative humidity) at fortification levels of 0.50 µg and 5.0 µg per adsorption tube, corresponding to about 1.4 µg m^{-3} (LOQ) and about 14 µg m^{-3} (10xLOQ) and the results showed that recoveries were between 70% and 110%. Extraction efficiency was demonstrated with average recoveries between 98 to 99%. Average recoveries from the frozen storage stability test were between 95 to 97%. The average recoveries for both fortification levels and three ions after sampling of warm, humid air ranged between 86% and 102%. For each fortification level the RSD values were <20%. The detailed results are given in Table 4.2-15.

Table 4.2-15: Results of method validation of alpha-Cypermethrin in air

Specimen Type	Fortified alpha-Cypermethrin [µg]	Average C_{Air} [µg m^{-3}]	m/z 207		m/z 209		m/z 171		n
			Average Recovery [%]	RSD [%]	Average Recovery [%]	RSD [%]	Average Recovery [%]	RSD [%]	
Extraction Efficiency	5.0	--	99	3	98	2	99	2	4
Storage Stability: 6 Days, Freezer Temperature (approx. -20°C)	5.0	--	95	--	95	--	97	--	2
Warm, Humid Air	0.5	1.39	102	6	102	6	102	7	5
	5.0	13.9	86	12	87	11	88	12	5
	Overall		94	12	95	11	95	12	10

Linearity

Good linearity ($r > 0.99$) was observed in the range of 10 ng mL^{-1} to 300 ng mL^{-1} for three mass transitions of alpha-Cypermethrin.

Specificity

Under the described conditions the method is specific for the determination of alpha-Cypermethrin in air. Significant interferences (> 30% of LOQ) were not observed at the retention time and mass transitions of alpha-Cypermethrin.

Due to the high selectivity and specificity of GC/MS(NCI) an additional confirmatory technique was not necessary. Three mass transitions of alpha-Cypermethrin were quantified.

Limit of Quantification

The method has a limit of quantification (LOQ) of 1.4 µg m^{-3} alpha-Cypermethrin in air.

Repeatability

The relative standard deviations (RSD, %) for all fortification levels were below 20%.

Reproducibility

Reproducibility of the method was not determined within this validation study.

Storage Stability:

The average recovery after storage at -20 °C for 6 days on the adsorber material was 95%, hence test tubes can be stored for this period of time after air sampling until analysis.

Conclusion

Based on the results obtained, an analytical method for the determination of alpha-Cypermethrin in air using highly selective GC/MS(NCI) determination (monitoring three fragment ions) was successfully validated with a limit of quantification of 1.4 µg m^{-3}. The method fulfils the requirements of SANCO/825/2000 rev. 8.1, 16/11/2010 and SANCO/3029/99 rev. 4, 11/07/2000.

Report:	CA 4.2/10
	Maas X., Bendig P., 2015
	Validation of Analytical Method L0265/02 for the Determination of alpha-Cypermethrin (BAS 310 I) in Air
	2015/1225669
Guidelines:	SANCO/825/00 rev. 8.1 (16 November 2010), SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	yes
	(certified by Landesamt fuer Umwelt, Messungen und Naturschutz Baden Wuerttemberg, Karlsruhe, Germany)

The objective of this study was to validate an analytical method L0265/02 for the determination of alpha-Cypermethrin (BAS 310 I) in air using GC/MS, with a target limit of quantitation (LOQ) of 0.06 µg/m^3. The target LOQ is based on the new (reduced) $AOEL_{inhalative}$ of 0.0004 mg alpha-cypermethrin /kg body weight which is based on a value of 0.25 mg/kg and applying a safety factor of 300 and another factor of 2 resulting in an overall safety factor of 600.

Principle of the method

Air was sucked through XAD adsorption tubes at about 1.0 L/min for 6 hours (total air sampling volume of about 0.36 m^3). Subsequently, the adsorption material is extracted with ethyl acetate, the internal standard lambda-cyhalothrin is added to the extract, and finally analysed by gas chromatography with mass spectrometric detection in the negative chemical ionisation mode (GC/MS), monitoring three fragment ions.

The validation was performed at LOQ and at 10 times LOQ fortification levels, each with 5 replicates and 2 untreated control samples.

The matrix effects were tested for each fragment ion. No significant matrix effects (i.e. > 20% suppression or enhancement) on GC-MS response were observed. Thus, calibration solutions in solvent were used for evaluation of the results. The following negative fragment ions of alpha-Cypermethrin were used for the analysis by GC-MS on a capillary GC-column (Optima 5 HT, 30 m, 0.25 mm i.d. 0.25 µm film thickness):

-207 m/z for quantification

-209 m/z for confirmation

-171 m/z for confirmation

Recovery findings

The analytical method was validated with warm, humid air (approx. 35°C, ≥80% relative humidity) at fortification levels of 0.06 µg m^{-3} (LOQ) and about 0.6 µg m^{-3} (10xLOQ) and the results showed that recoveries were between 70% and 110%. Blank samples showed results <LoD and break-through samples were below 2% of the fortification level, hence absence of break-through can be postulated.

Extraction efficiency was demonstrated to range between 103 and 106%. Average recoveries from the frozen storage stability test were between 92 and 103%. The average recoveries for both fortification levels and three ions after sampling of warm, humid air ranged between 94% and 98%. For each fortification level the RSD values were <20%. The detailed results are given in Table 4.2-16.

Table 4.2-16: Results of method validation of alpha-Cypermethrin in air

Specimen Type	Average C_{Air} [µg/m^3]	Fortified alpha-Cypermethrin [µg]	m/z 207		m/z 209		m/z 171		n
			Average Recovery [%]	RSD [%]	Average Recovery [%]	RSD [%]	Average Recovery [%]	RSD [%]	
Extraction Efficiency	0.06	0.0216	106	4	106	4	103	1	2
	0.6	0.216	103		103		103		
	Overall		97	7	97	8	98	7	2
Storage Stability Sampling cartridge: 11 Days, at < - 18°C in the dark	0.06	0.0216	92	5	90	4	94	8	5
	0.6	0.216	103	2	103	3	102	2	5
Fortifications (warm, humid Air)	0.06	0.0216	94	12	95	12	95	10	5
	0.6	0.216	97	4	98	4	98	4	5
	Overall		96	8	97	8	97	7	10

Linearity

Good linearity ($r^2 > 0.99$) was observed in the range of 0.4 ng mL^{-1} to 50 ng mL^{-1} for three fragments of alpha-Cypermethrin. Matrix effects were assessed and found to be not significant for the XAD resign. Hence, calibration was accomplished in solvent without matrix load in pure ethylacetate containing the internal standard.

Specificity

Under the described conditions the method is specific for the determination of alpha-Cypermethrin in air. Significant interferences (> 30% of LOQ) were not observed at the retention time and mass transitions of alpha-Cypermethrin.

Due to the high selectivity and specificity of GC/MS(NCI) an additional confirmatory technique was not necessary. Three mass transitions of alpha-Cypermethrin were quantified.

Limit of Quantification	The limit of quantification (LOQ) of the analytical method is 0.06 µg/m^3. The limit of detection (LOD) of the method was defined as the lowest analyte concentration injected as a calibration solution, resulting in an LOD of 0.012 µg/m^3 (equal. 20 % of the LOQ).
Repeatability	The method L0265/02 is suitable for the determination of alpha-cypermethrin in air. The mean recovery values were between 70 % and 110 % of the nominal values. The relative standard deviations for all fortification levels were below 20%.
Reproducibility	Reproducibility of the method was not determined within this validation study.
Storage Stability:	The average recovery after storage at -20 °C for 11 days on the XAD adsorber material was 97%, hence test tubes can be stored for this period of time after air sampling until analysis. Extracts, as well as stock solutions in ethylacetat and calibration solutions have been confirmed to be stable over a time period of 14 days.
Conclusion:	**It could be demonstrated that method L0265/02 fulfils the requirements with regard to specificity, repeatability, limit of quantification and recoveries and is therefore applicable to correctly determine residues of alpha-cypermethrin in air with a limit of quantification of 0.6 µg/m^3 air. The method is considered fully validated according to SANCO/825/00 rev. 8.1.**

(d) Methods for the analysis in body fluids and tissues

Analytical methods for the determination of alpha-Cypermethrin residues in body fluids were evaluated in the context of the inclusion in Annex I of Directive 91/414/EEC. Methods evaluated previously are summarized in Table 4.2-17 for the reviewer´s convenience.

Table 4.2-17: Peer-reviewed methods for body fluids and tissues

Method No.	Matrix	Method principle	Target analytes	LOQ	year	DocID	EU reviewed
DFG S 19	Blood, urine (swine)	GC/ECD	Alpha-Cypermethrin	0.005 mg/l	2000	AL-245-008	yes
M 3499	blood	GC/MS (confirmatory method)	Alpha-Cypermethrin	0.005 mg/kg	2001	AL-210-012	yes
AMS 887-1	aerosols	HPLC/UV	Alpha-Cypermethrin	Min. detectable conc.: 3 µg/m^3 for 15L air sample; 0.1 µg/m^3 for 480 L air sample	Not reported	AL-210-002	yes

Report:	CA 4.2/11 Bendig P., 2014a Validation of BASF analytical method L0246/01: Method for the determination of individual or combined diastereomeric forms of BAS 311 I (Cis I, Cis II (alpha-Cypermethrin, BAS 310 I), Trans III and Trans IV) in body fluids 2014/1145911
Guidelines:	SANCO/825/00 rev. 8.1 (16 November 2010), SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	yes (certified by Landesamt fuer Umwelt, Messungen und Naturschutz Baden Wuerttemberg, Karlsruhe, Germany)

Remark: The title has slightly changed compared to the one originally submitted in the application. The title as presented now in the dossier reflects the purpose of the study a more appropriate manner. A BASF-method number was also allocated to unequivocally identify the method. The originally assigned title was "Development and validation of an Analytical Method for the Determination of alpha-Cypermethrin, BAS 310 I in body fluids", however by the assigned DocID 2014/1145911 the study can be unequivocally identified.

Principle of the methods

Urine: Acetonitrile extraction of urine with consecutive dilution and LC-MS/MS determination (with positive ionisation: ESI) was used for quantitation and confirmation.

Blood plasma: A small scale DFG S19 extraction module E7 was used for extraction of the blood plasma. An aliquot of the extract was evaporated to dryness and then redissolved in acetonitrile/water (1/1, v/v + 0.1% formic acid) followed by LC-MS/MS determination (with positive ionisation: ESI) for quantitation and confirmation.

The limit of quantification (LOQ) of both methods is < 0.050 mg/L per analyte.

Recovery findings

The urine and blood plasma (5 replicates) were fortified at ≤ 0.050 mg/L per isomer (LOQ) with solutions containing all 4 Cypermethrin isomers. The fortification level of Cis II isomer (alpha-Cypermethrin) was 0.035 mg/L. The average recoveries for the two parent-daughter ion transitions monitored were within the acceptable range of 70 % to 110 %.

Matrix effects were not significant (i. e. < 20 %). Nevertheless, quantitative determination of final urine and blood plasma extracts was carried out by external standardization using matrix matched standard solutions.

Detailed results of recoveries for each mass transition and crop are given in Table 4.2-18. Effects of matrix on response are demonstrated in Table 4.2-19.

Table 4.2-18: BASF analytical method L0246/01, summary of validation results

Fortification Level (mg/L)	Fragment Ion (m/z)	Recovery							
		Cis I		Cis II		Trans III		Trans IV	
		433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193	433 ->191	435 ->193
Urine									
0.050	Average	100%	99%	99%	99%	97%	95%	95%	96%
	RSD	2%	1%	2%	2%	2%	2%	1%	2%
	Replicates	5	5	5	5	5	5	5	5
Blood Plasma									
0.050	Average	100%	98%	104%	103%	99%	95%	100%	100%
	RSD	2%	3%	4%	3%	3%	3%	2%	4%
	Replicates	5	5	5	5	5	5	5	5

Table 4.2-19: BASF analytical method L0246/01, matrix effect on response

Matrix	Solution ID K3412-	Solution Conc. ng/mL	Type	LC/MS Run P3412	Ion Mass Transition 433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*	433 m/z -> 191 m/z	*	435 m/z -> 193 m/z	*
					Cis I				Cis II			
Urine	24	1.0	S	022	1.57E+05	-13%	1.02E+05	-12%	1.06E+05	-15%	6.53E+04	-10%
	40	1.0	MMS	009	1.37E+05		9.00E+04		9.06E+04		5.88E+04	
Blood Plasma	24	1.0	S	066	1.65E+05	-3%	1.09E+05	-3%	1.11E+05	-2%	7.16E+04	1%
	55	1.0	MMS	054	1.60E+05		1.06E+05		1.09E+05		7.22E+04	
					Trans III				Trans IV			
Urine	24	1.0	S	022	7.71E+04	-11%	4.89E+04	-9%	2.87E+04	-6%	1.83E+04	-6%
	40	1.0	MMS	009	6.87E+04		4.43E+04		2.71E+04		1.72E+04	
Blood Plasma	24	1.0	S	066	8.31E+04	-2%	5.61E+04	-6%	3.54E+04	-10%	2.22E+04	-10%
	55	1.0	MMS	054	8.15E+04		5.27E+04		3.18E+04		1.99E+04	

Calibration used for quantitation: Calibration solutions in solvent ("S"). MMS: Matrix-matched standards.
*A positive value indicates enhancement, a negative value suppression.

Linearity	Good linearity (regression coefficients ≥ 0.999) was observed in the range of 0.20 ng/mL to 5.00 ng/mL or 10.00 ng/mL for Cis I and Trans III (Cis II and Trans IV in the given ratios).
Specificity	Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity. No significant interference above 30 % of LOQ was detected in any of the control specimen extracts, so that a high level of selectivity was demonstrated.
Limit of Quantification	The limit of quantification for both methods was ≤ 0.050 mg/L per analyte.
Repeatability	For each fortification level, for urine and blood plasma and for both MS/MS transitions monitored, the relative standard deviations (RSD) were always below 20%. The detailed values are shown in Table 4.2-18.
Reproducibility	Reproducibility of the method was not determined within this validation study.
Conclusion	**The LC-MS/MS based methods were successfully validated for the determination of all 4 Cypermethrin isomers in human urine and blood plasma with an LOQ of ≤ 0.050 mg/L and thus demonstrated to be applicable for enforcement and monitoring purposes.** **The method fulfills the registration requirements of SANCO/825/00 rev. 8.1 16/11/2010 and SANCO/3029/99 rev.4 11/07/2000.**

Report:	CA 4.2/12 Richter S., 2015 BAS 310 I (alpha-Cypermethrin): Bridging Extractability of BAS 310 I (alpha Cypermethrin) from Barley Grain, Olive Fruit and Orange Whole Fruit using Acetonitrile/Water (3 Methods, incl. QuEChERS) or Methanol/Water/HCl (BASF Method 567/1) as Extraction Solvents DocID 2014/1162676
Guidelines:	SANCO/825/00 rev. 8.1 (16 November 2010), SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	yes (certified by Landesamt fuer Umwelt, Messungen und Naturschutz Baden Wuerttemberg, Karlsruhe, Germany)

Remark:
The presented OECD-summary of the study is based on under GLP audited data. However, as the report was not finalized at time of submission of the electronic version of the dossier, a copy of the original, signed final report of the study will be submitted upon availability, which is estimated to be early February.

Principle of the methods

The objective of this study is to bridge alpha-Cypermethrin's extractability when extracted with various solvent mixtures from acidic, dry, and oily crops.

QuEChERS based residue method for all 3 crop types (BASF-method No. L0245/01)
The QuEChERS method as validated in a previous study (BASF DocID 2013/1361966) was used for all crops, all untreated samples (single extraction) and all treated samples analysed in duplicate. Sample aliquots are extracted with a mixture of water and acetonitrile for subsequent LC/MS/MS analysis.

Residue method 567/1 for all 3 crop types (BASF DocIDs 2007/1010254 and 2005/500006)
Sample aliquots are extracted with a mixture of water and methanol (acidified with HCl) for subsequent LC-MS/MS analysis.

Residue method 567/0 for olive fruit (BASF DocIDs 2005/5000063 and 2005/500006)
Sample aliquots are extracted with acetonitrile and cleaned-up with n-hexane liquid-liquid partition for subsequent LC-MS/MS analysis.

Extraction for olive and orange as used in metabolism study AL-640-001

10 g of crop material are extracted twice, each time with 30 mL of acetonitrile/water (70/30 v/v), followed by centrifugation and LC-MS/MS analysis.

Extraction for barley grain as used in metabolism study AL-640-004

10 g of grain material are soaked with 5 mL of water, followed by 2-times extraction with each 40 mL of acetonitrile (0.1% formic acid), centrifugation and LC-MS/MS analysis.

The LC-MS/MS method allows to quantify and to confirm alpha-Cypermethrin, using two MRM transitions based on the formation of ammonium adducts (433 m/z -> 191 m/z and 435 m/z -> 193 m/z).

The limit of quantification (LOQ) of all methods is 0.01 mg/kg, the limit of detection was 0.002 mg/kg.

Residue results

Unlabeled samples from field trials were analysed using the various solvent mixtures.

For whole orange, the 3 extraction methods QuEChERS (BASF method No. L0245/01), BASF method 567/1 and the extraction method from the metabolism study AL-640-001 resulted in almost similar residue results for alpha-Cypermethrin.

Residue results of alpha-Cypermethrin in olive fruit are comparable for the extraction methods QuEChERS (BASF method No. L0245/01), BASF method 567/0 and the extraction method from the metabolism study AL-640-001. Results obtained with BASF method 567/1 were corrected for the low recovery results obtained from the concurrent fortified samples and are similar to those residue results found with the other extraction methods.

Residue results for barley grain obtained with the QuEChERS method (BASF method No. L0245/01) and the extraction method from the metabolism study AL-640-004 are comparable. The extraction method 567/1 resulted in lower residues (in the same order of magnitude).

A summary of residue results of treated samples is given in Table 4.2-20.

Table 4.2-20: Extraction methods in plant materials; summary of residues results

Methods		QuEChERS (L0245/01)	567/0	567/1		AL-640-004	AL-640-001
Sample Type	Sample No.	Alpha-Cypermethrin residues (mg/kg)					
Orange (sweet) / Whole Fruit	L1203930005	0.55	na	0.59		na	0.65
Orange (sweet) / Whole Fruit	L1203930013	0.34	na	0.47		na	0.57
Orange (sweet) / Whole Fruit	L1203930017	0.73	na	0.87		na	1.2
Olive / Fruit	L1203860005	2.2	2.2	0.78	1.9*	na	2.1
Olive / Fruit	L1203860009	2.0	2.1	0.69	1.7*	na	1.9
Olive / Fruit	L1203860017	1.4	1.4	0.49	1.2*	na	1.2
Barley / Grain	L1300290023	0.081	na	0.048		0.091	na
Barley / Grain	L1300300027	2.4	na	1.3		2.4	na

na: not applicable

* Residue results are corrected for low recoveries of concurrent fortified samples.

Recovery findings

For concurrent method validation, plant materials were fortified (3 replicates per level and crop type) with solutions containing alpha-Cypermethrin to obtain fortifications levels at LOQ and at a higher level (100xLOQ).

Average recoveries for all three crop materials were within the acceptable range of 70 % to 110 % with relative standard deviations (RSD) of ≤ 20 % except for olive extracted with extraction method 567/1 with an average recovery of about 40 %. This low recovery was to be expected as the extraction method was not designed for alpha-Cypermethrin in oily matrices.

Detailed results of recoveries are given in Table 4.2-21.

Table 4.2-21: Extraction methods in plant materials; summary of validation results

Matrix	Fortification Level (mg/kg)	Alpha-Cypermethrin (433 m/z -> 191 m/z)			
		Average (%)	SD (%)	RSD (%)	n
QuEChERS (BASF method No. L0245/01)					
Orange	0.01, 1.0	91.8	17	18	6
Olive	0.01, 1.0	101	5.8	5.7	6
Grain	0.01, 1.0	97.3	8.1	8.3	6
Overall:		**96.7**	**11**	**12**	**18**
Method 567/1					
Orange	0.01, 1.0	78.9	4.4	5.6	6
Olive*	0.01, 1.0	41.6	5.3	13	6
Grain	0.01, 1.0	87.3	15	17	6
Overall:**		**83.1**	**11**	**14**	**12**
Method 567/0					
Olive	0.01, 1.0	72.9	1.8	2.4	6
Method AL-640-001					
Orange	0.01, 1.0	79.2	8.4	11	6
Olive	0.01, 1.0	79.3	7.4	9.3	6
Overall:		**79.2**	**7.6**	**10**	**12**
Method AL-640-004					
Grain	0.01, 1.0	101	7.8	7.8	6

SD=Standard Deviation, RSD=Relative Standard Deviation, n=Number of results included in calculation

* Low recovery as expected since the extraction method was not designed for alpha-Cypermethrin in oily matrices.

** Olive recoveries excluded.

Linearity Good linearity (correlation coefficients ≥ 0.999) was observed in the range of 0.01 ng/mL to 25 ng/mL.

Specificity Quantification was performed by use of highly selective LC-MS/MS detection. Two selected ion mass transitions were evaluated in order to demonstrate that the method achieves a high level of selectivity. No significant interference above 20% of LOQ was detected in any of the control specimens.

Limit of Quantification The limit of quantification (LOQ) of the methods is 0.01 mg/kg.

Repeatability The relative standard deviations (RSD) were always below 20%. The detailed values are shown in Table 4.2-21.

Stability in Solutions and Extracts

Standard solutions and final extracts were stored refrigerated in the dark when not in use. Solutions with the analyte were used throughout the study and gave consistent results and recoveries. Acceptable recoveries for alpha-Cypermethrin in crop sample extracts stored refrigerated for several days demonstrate the stability of the analyte during refrigerated storage.

Conclusion

For alpha-Cypermethrin, the methods showed good results with regard to the parameters studied. No significant differences among the validated residue methods for the extraction of alpha-Cypermethrin were found. Solely in the case of method 567/1 lower recoveries for oily matrices were found compared to the extraction scheme of the metabolism studies. However, method 567/1 was not specifically developed for matrices of high oil content, whereas method 567/0 which shows satisfactory high extraction efficiency of > 70%.

BASF
The Chemical Company

Alpha-Cypermethrin

Document M-CA, Section 5

TOXICOLOGICAL AND METABOLISM STUDIES ON THE ACTIVE SUBSTANCE

Compiled by:

[redacted]

[redacted]

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
October 09, 2015	CA 5.8.1/37 and CA 5.8.1/38 were amended with analytical results.	
January 20, 2016	DNT study has been amended with analytical results, pathological examinations of dams and pups and weight, length and width data of pups brain. Data on alphacypermethrin content in the pups brain and carcass is included. Diverse analytical methods belonging to divers studies (a.o. CA 5.7.1/3, CA 5.8.3/50 and CA 5.8.1/38) were amended	M-CA 5.7.1/3 2015/1001621 M-CA-4.1.2/3 – 4.1.2/13

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 5 TOXICOLOGICAL AND METABOLISM STUDIES ON THE ACTIVE SUBSTANCE

Introduction

This chapter is preceded with a

- **Bridging situation** for alpha-cypermethrin
- **Bridging rational** to compare alpha-cypermethrin with cypermethrin, zeta-cypermethrin and beta-cypermethrin and to conclude on the consequences of bridging between the data.

The data are presented as follows:

- **Studies already evaluated in the draft monograph of alpha-cypermethrin combined with data from other DARs belonging to the group of cypermethrins:** for completeness short summaries of key investigations from the divers DARs are presented in case where the data are no longer data protected (with headers presented in the original monograph). These studies are highlighted in grey. In case of data protection we kindly refer to the original DAR and give only summary tables.
- **Submission of not yet peer-reviewed studies in this AIRIII-Dossier:** new data are presented and discussed in detail.

Bridging situation for alpha-cypermethrin:

Alpha-cypermethrin has been investigated in a huge package of regulatory studies and data from other cypermethrins were already used in the first Annex I registration to supplement the regulatory datapackage and were regarded as acceptable for the Renewal process:

- the rat carcinogenicity study has been addressed with data generated for cypermethrin and
- the multi-generation data with alpha-cypermethrin were supplemented with cypermethrin data

New data to account for the potential developmental neurotoxicity as requested for pyrethroids in general in the Review report of alpha-cypermethrin (SANCO/4335/2000 final) are now addressed via data from zeta-cypermethrin that are no longer data protected.

- a developmental neurotoxicity study with zeta-cypermethrin (CA 5.7)

Lactational and placental transfer as well as milk concentration is addressed with the following studies:

- Dietary placental transfer and lactation transfer of zeta-cypermethrin at 50, 125, 300 ppm in rats from GD 6-20, or GD6-LD21 (see CA 5.8.2)
- Measurement of zeta-cypermethrin in milk following dietary administration at 125 and 375 ppm from GD 6 to LD 17 (see CA 5.8.2)

A new DNT study with direct pup dosing as requested by the rapporteur is presented:

- Targeted developmental neurotoxicity study with direct pup dosing addressing the most sensitive endpoints found in the study described in the DAR of beta-cypermethrin (see CA 5.7)

Important to mention: Based on the proven lactational transfer of cypermethrins on the one side, and the artificial exposure type of direct pup dosing on the other side, the findings of this study are considered not appropriate and not relevant to be used in a human risk assessment for infants which are mainly exposed via breast or bottle fed.

The exposure via rat breast milk is exceeding the human exposure by several orders of magnitude (see CA 5.8.2) and therefore the dietary DNT study with zeta-cypermethrin is considered as the relevant study to derive an endpoint for human risk assessment.

The generation of a dietary developmental neurotoxicity study with alpha-cypermethrin itself is considered not necessary based on the huge database available for pyrethroids and the principal similarity of toxicity as outlined in the following bridging rationale.

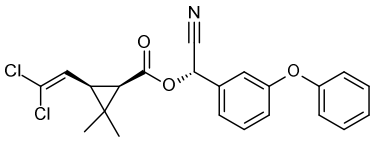
Further information from other international authorities:
The US EPA evaluated the existing database for Pyrethroids in the Memorandum “Pyrethroids: Evaluation of Data from Developmental Neurotoxicity Studies and consideration of Comparative Sensitivity” [see KCA 5/6 2010/1232203], including zeta-cypermethrin and three other Typ II Pyrethroids. It was concluded that guideline DNT studies with dietary administration are not appropriate to provide new information which is relevant for risk assessment.

Also on European level within the Biocides Technical Meetings (TM III 09, TM I 10, TM II 2010) the relevance of DNT studies with pyrethroids were discussed. The following conclusion was drawn: *”As neurotoxic effects are critical effects after acute or medium-term exposure and the available data indicate that DNT effects are induced at higher LOAELs, it is unlikely that, in the absence of DNT studies, the potential DNT effects are not covered by AELs set on neurotoxic effects observed in acute and mediumterm studies. The data also indicate that an additional assessment factor for species sensitivity is not required. It was concluded that additional DNT studies according to OECD TG 426, if such a study is not present, is not necessary.” (TM II 2010, page 4-5)*

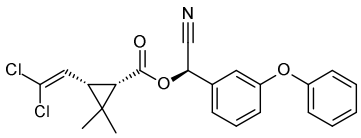
Bridging rationale

Alpha-cypermethrin is an insecticide that belongs to the class of Typ II pyrethroids, which contain the □-cyano-3-phenoxybenzyl moiety whereas Type I compounds are lacking the □-cyano-3-phenoxybenzyl group. Alpha-cypermethrin consists of 2 of the 8 stereo isomers that comprise cypermethrin, the Cis2 Isomers. For clarity, the two stereo isomers of alpha-cypermethrin (1R cis □S and 1S cis □R) are shown in Figure 5-1.

Figure 5-1: Stereoisomers of alpha-cypermethrin



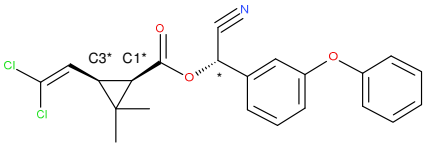
1R cis αS



1S cis αR

Cypermethrin in comparison is a racemic mixture of 8 isomers (with 3 chiral centres as shown in Figure 5-2) and contains around 22% of alpha-cypermethrin. In Table 5-1 all cypermethrins are listed with regard to their isomer composition and biological activity.

Figure 5-2: Structure of cypermethrin and location of its chiral centres



*: denotes the positions of chiral carbon centres

Table 5-1: Composition of the different cypermethrins

Isomers of Cypermethrin C1 α-carbon		Receptor Inhibition[a]	Mouse toxicity[b]	Cyper-methrin [%]	Alpha-cypermethrin [%]	Beta-cypermethrin [%]	Theta-cypermethrin [%]	Zeta-cypermethrin [%]
CIS1	1R cis αR	0	0	14	-	-	-	3
	1S cis αS	0	0	14	-	-	-	22
CIS2	1R cis αS	100	100	11	**50**	20	-	22
	1S cis αR	0	0	11	**50**	20	-	3
TRANS3	1R trans αR	0	0	14	**-**	-	-	3
	1S trans αS	0	0	14	**-**	-	-	22
TRANS4	1R trans αS	43	54	11	**-**	30	50	22
	1S trans αR	0	0	11	**-**	30	50	3
1R:1S ratio at C1				50/50	**50/50**	50/50	50/50	50/50
S/R ratio at α-carbon				50/50	**50/50**	50/50	50/50	89/11
Cis/trans ratio				50/50	**100/0**	40/60	0/100	50/50
Content of active isomers				22	**50**	50	50	44

[a] Lawrence and Casida (1983), DocID 1983/1002378, inhibition of [35S]t-butylbicyclophosphorothionate binding to rat brain synaptic membranes relative to the potency of 1R cis αS isomer at 5µM set to 100%

[b] Lawrence and Casida (1983), DocID 1983/1002378, Toxicity is relative to 1R cis αS isomer with a mouse intracerebral LD_{50} of 1.4 nmole per gram of brain tissue determined by injection in 3 µl of methoxytriglycol into the third ventricle.

The bridging concept of cypermethrins is based on the shared toxicity profile relevant for Reference value setting, which is acute neurotoxicity. Even though there have been a lot of investigations within the last decades about possible targets of mammalian intoxication as summarized in the reviews of Soderlund et al., 2002 [see KCA 5/1 2002/1027257] and Soderlund, 2012 [see KCA 5/2 2012/1367222], there is still a broad agreement that **voltage-gated sodium channels are the primary target sites for the neurotoxic action of pyrethroids.** The stereospecific interaction of different isomers of pyrethroids with voltage-gated sodium channel was extensively investigated in vitro (Narahashi, 1986, KCA 5/3 1986/1002771) and also in vivo (Lawrence and Casida, 1983 and 1985, [see KCA 5/4 1983/1002378 and KCA 5/5 1985/1002192]).

Other stereospecific mode of actions like blockage of voltage-sensitive calcium and chloride channels, associated with the production of salivation (Soderlund, 2012, [see KCA 5/2 2012/1367222]; Meijer et al., 2014, [see KCA 5/7 2014/1242699]), have been implicated as alternative or secondary sites of action but, like other potential neuroactive targets (voltage-sensitive potassium-channels, ligand-gated ion channels (i.e. GABA- and Nicotine acetylcholine-receptors as well as excitatory glutamate receptors), peripheral-type benzodiazepine receptors, ion channel and receptor regulation as well as mitochondrial electron transport) failed to demonstrate relevance to pyrethroid intoxication and are therefore not considered in the literature search of alpha-cypermethrin as relevant.

Based on the most relevant mode of action, the lesions exerted by cypermethrins are primarily manifested as acute and reversible pharmacological actions resulting from nerve excitability (Ray & Frey, 2006, [see KCA 5/8 2006/1051137]; Ray, 2001, [see KCA 5/9 2001/1032003]), and although the different cypermethrins are separate active ingredients, they are considered as similar in their primary toxicity profile of neurotoxicity based on the following considerations:

1. The neurotoxicity of pyrethroids to mammals depends on structural characteristics:

   - The stereochemical configuration at cyclopropane C-1:

     only esters of 1R cyclopropanecarboxylates are neurotoxic in mammals, whereas the corresponding 1S is without measurable toxicity even when administered at high doses directly to the CNS (Soderlund et al, 2002, [see KCA 5/1 2002/1027257]).
     The 1R to 1S ratio is similar for all cypermethrins, indicating that no relevant difference concerning neurotoxicity is expected by this property only.

   - the presence of an□□-cyano substituent in S configuration in the 3-phenoxybenzyl alcohol moiety:

     A pyrethroid with an □-cyano substituent in S configuration also greatly enhanced acute neurotoxicity while the R epimer was not toxic (Soderlund et al, 2002, [see KCA 5/1 2002/1027257]). In a paper published by Casida and Lawrence (1985, [see KCA 5/5 1985/1002192]) the inhibitory potency of α-cyano 3-phenoxybenzyl pyrethroids at the butylcyclophosphorothionate (TBPS) receptor on fresh rat brain membranes was studied. In parallel, toxicity was evaluated on intracerebrally treated mice and the potency of the different isomers was compared to that of Cis R α-S cypermethrin which is normalized to 100. This study demonstrated the potency of the Cis R α-S isomer of cypermethrin.
     The S/R ratio at □-carbon is 50/50 for all cypermethrins except for the isomeric improved zeta-cypermethrin, which is 89/11.

   - The cis/trans ratio at the cyclopropyl ring:

     In mammals, the absolute configuration at cyclopropane C-3 of cyclopropane carboxylate esters of primary alcohols also strongly influences toxicity. Pyrethroids having the 1R, cis configuration are both insecticidal and toxic to mammals, whereas the corresponding pyrethroids having the 1R, trans-configuration, though similar in insecticidal potency, lack measurable acute toxicity to mammals. The low toxicity of these 1R, trans-pyrethroids was once ascribed solely to their rapid hydrolytic detoxication by liver esterases. However, intra-cerebral dosing experiments have demonstrated that these compounds had low intrinsic toxicities even when the impact of biotransformation was removed (Soderlund et al, 2002, [see KCA 5/1 2002/1027257]). For the cypermethrin isomers 1R cis □S and 1R trans □S a factor of 2 to 3 difference between the cis and trans isomer was shown for the LD50 values after intracerebral injection in mice.

     The cis/trans ratio of cypermethrin and zeta-cypermethrin is 50/50 whereas that of alpha-cypermethrin is 100/0 and beta-cypermethrin is 40/60. Thereby alpha-cypermethrin can be considered as slightly more toxic than cypermethrin, beta- or zeta-cypermethrin.

Conclusion: Differences in neurotoxic potential at comparable plasma peak levels might occur to some extent based on the different active isomers.

2. The neurotoxicity of cypermethrins depends on the substance concentration at the voltage-gated sodium channels as primary target sites for the neurotoxic action of pyrethroids

   Bioavailability is determined by identical physico-chemical properties (low solubility in water and high solubility in fat, pH-dependent hydrolytic stability and molecular weight) and similar toxicokinetic properties. This is substantiated by data generated with cypermethrin, alpha-cypermethrin and zeta-cypermethrin:

   - Absorption and excretion is rapid and comparable. The neurotoxic symptoms are correlated with the plasma concentration, which increase and decrease rapidly showing a short peak plasma effect at Tmax within 3-9 hours

   - Distribution of cypermethrins takes place in well perfused (liver, lung, kidney and heart) or lipophilic organs (fat). Storage within the lipophilic cell membrane apart from voltage-gated sodium channels will not contribute to neurotoxic symptoms but might induce other non-specific effects like for example oxidative stress. The terminal half-life for elimination of alpha-cypermethrin from the fat of rats is 17-26 days, compared to 18.9 days for cis-cypermethrin (WHO, 1992, [see KCA 5/10 1992/1005337]).

   Conclusion: Absorption and excretion are comparable, accumulation in fat is not considered to crucially contribute to neurotoxic symptoms.

3. Neurotoxicity is terminated via metabolic degradation.

   All Cypermethrins are primarily metabolized by cleavage of the ester bond releasing two common metabolites:
   - 3-phenoxybenzoic acid (PBA) and
   - 3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecaroxylic acid (DCVA).
   - common metabolites are furthermore conjugated and hydroxylated.

   Conclusion: Neurotoxic effects will occur mainly after bolus administration where plasma peak levels are reached by exceedance of the metabolic capacity. At human relevant doses reflected by the experimental dietary administration studies systemic toxicity is mainly characterized by the toxicity of the common metabolites.

In addition, it has been shown that the magnitude of the induced toxicity in bolus studies can vary by several orders, and is modulated by factors like: species and strain, vehicle, age of animals, intrinsic binding, fasted or unfasted animals. This is comprehensively described in the open literature for pyrethroids, see Crofton et al. (1995, [see KCA 5/11 1995/1008154]) and Wolansky et al. (2007, [see KCA 5/12 2007/1070388]). Therefore the variability within the test system dominates the variability of substance specific toxicity.

Therefore, although there might be differences in structure activity factors (R/S ratio at C1, cis/trans ratio) that influence mammalian (neuro-) toxicity of the Typ II-pyrethroids cypermethrin, alpha-, beta-and zeta-cypermethrin, overall no relevant differences in mammalian toxicity is expected in correlation to biological variability between the different cypermethrins when focusing on risk related studies and not on hazard related acute lethal toxicity studies. This is confirmed by a number of studies performed with different cypermethrins and presented within the following tables for acute neurotoxicity and repeated dose studies.

The following Table 5-2 and Table 5-3 demonstrate that differences in acute neurotoxicity study results are rather due to biological variability than due to a different toxicological profile of the cypermethrins:

Table 5-2: Acute neurotoxicity studies

Compound studied	Strain*	Age of animals	Dose [mg/kg bw] vehicle	Neuro NOAEL [mg/kg/d]	Neuro LOAEL [mg/kg/d]	Reference
Alpha-cypermethrin[1]	SD (Crl:CD:BR)	6-7 weeks	0-4-20-40 in 10 ml/kg corn oil	4	20	[redacted] 1993b [see KCA 5/13 AL-451-004]
Zeta-cypermethrin[4]	LE	Not given	0-10-50-250 mg/kg bw undiluted	10	50	[redacted] 1998
Cypermethrin[3]	LE	10-13 weeks	0-20-60-120 in 1 mL/kg corn oil	M: <20 F: 20	M: 20 F: 60	[redacted] 1993
Cypermethrin[4]	SD (Crl:CD:BR)	Not given	0-30-100-200 mg/kg bw in 5% corn oil	30	100	[redacted] 993
Beta-cypermethrin[2]	SD (Crl:CD:BR)	6-7 weeks	0-20-100-500 mg/kg bw in aqueous CMC	M: <20 F: 20	M: 20 F: 100	1998

* LE = Long Evans; SD = Sprague-Dawley;
1 Study reported in the DAR alpha-cypermethrin (1999)
2 Study reported in the DAR beta-cypermethrin (2013)
3 Study reported in the DAR cypermethrin (1999)
4 Study reported in the DAR zeta-cypermethrin and Final Addendum (2008)

These data are supplemented by acute oral toxicity studies published by Wolansky et al. (2006, [see KCA 5/14 2006/1051134]) and Weiner et al. (2009, Doc ID [see KCA 5/15 2009/1131083]) using several doses and a relatively high number of animals per dose group.

Table 5-3: Acute neurotoxicity studies from peer-reviewed literature

Compound studied	Strain* Number/group Age of animals	Study Type	Doses [mg/kg bw] vehicle	NOAEL [mg/kg bw]	LOAEL [mg/kg/d]	Reference
Cypermethrin	LE 8-18/dose 9 weeks	Acute Oral Toxicity – Motor Activity	0.1-0.4-1-10-40 120 1 ml/kg corn oil	1	10	Wolansky et al., 2006, [see KCA 5/14 2006/1051134]
Cypermethrin	SD (Crl:CD:BR) 10/dose 6 weeks	Acute Oral Toxicity – FOB	65-100-150 5 ml/kg corn oil	<65	65	Weiner et al., 2009, [see KCA 5/15 2009/1131083]

*Threshold dose: estimate of the highest no-effect dose level at which treated rats would not display any decrease in motor activity.

Overall, the acute neurotoxicity studies on cypermethrin reflect the huge variability of NOAELs (1-30 mg/kg bw/day) and LOAELs (10-100 mg/kg bw/day). It has to be noted that the variability of effect and no effect levels for cypermethrin alone is higher than the differences observed for the whole group of cypermethrins.

The same is true also for repeated dose toxicity studies performed with the different cypermethrins. In these studies neurotoxic symptoms are no longer critical for NOAEL setting but appear together with further symptoms of toxicity or even at higher doses.

Table 5-4: Comparison of repeated-dose toxicity studies performed with Cypermethrin, Zeta-cypermethrin, Alpha-cypermethrin and Beta-cypermethrin

Compound studied	Strain*	Study Type	Dose [mg/kg bw or ppm] vehicle	Neuro NOAEL [mg/kg/d]	Neuro LOAEL [mg/kg/d]	Study NOAEL [mg/kg/d]	Study LOAEL [mg/kg/d]	Time to LOAEL Neuro	Reference
Alpha-cypermethrin3	SD	Subchronic Neurotoxicity (feeding)	0-50-250-500 ppm	M: 36.1 F: 42	M: >36.1 F: >42	M: 3.7 F: 4.2	M: 17.9 F: 21	No neurotoxic effects	KCA 5/16 AL-425-007
Zeta-cypermethrin4	LE	Subchronic Neurotoxicity (feeding)	0-75-400-750 ppm	M: 5 F: 55.6	M: 26.3 F: >55.6	M: 5 F: 31.5	M: 26.3 F: 55.6	8 weeks	[redacted] 1999b
Cypermethrin4	SD	Subchronic Neurotoxicity (feeding)	0-500-1300-1700 ppm	30.5	76.6	30.5	65	3 weeks	[redacted] 1993a
Cypermethrin2	SD	Subchronic Neurotoxicity (feeding)	0-100-400-800 ppm	40	80	<10	10	Week 7	1998
Ranking of Toxicity based on available NOAEL Subchronic Neurotoxicity (feeding) studies in rats: Alpha ~ Zeta ~ Cypermethrin (all within one order of magnitude), Alpha was not neurotoxic. Ranking of Toxicity based on available LOAEL Subchronic Neurotoxicity (feeding) studies in rats: Cypermethrin > Alpha > Zeta (all within one order of magnitude)									
Alpha-cypermethrin1	W	Subchronic Toxicity (feeding)	0-20-60-180-540 ppm	M: 12.5 F: 27	M: 27 F: >27	M: 12.5 F: 14.9	M: 27 F: 27	14-28 days	KCA 5/17 AL-425-003
Beta-cypermethrin2	SD	Subchronic Toxicity (feeding)	0-160-320-640 ppm	M: 16.7 F: <10	M: 33.1 F: 10	M: 16.7 F: <10	M: 33.1 F: 10	Week 7	Sanitized, 1998c
Zeta-cypermethrin4	F	Subchronic Toxicity (feeding)	0-10-50-150-250-500-900 ppm	M: 33.7 F: 38.4	M: 68.0 F: 79.5	M: 16.7 F: 19.7	M: 33.7 F: 38.4	High dose 4 days	[redacted] 1990
Cypermethrin4	AP	Subchronic Toxicity (feeding)	0-75-150-1500 ppm 1/10 conversion see DAR	>150	>150	15	150	None reported	[redacted] 1980
Cypermethrin4	SD	Subchronic Toxicity (feeding)	0-150-500-1500 ppm	37.5	M: 118 F: 130	37.5	M: 118 F: 130	7 days	[redacted] 1983
Ranking of Toxicity based on available NOAEL Subchronic Toxicity (feeding) studies in rats: Beta > Alpha ~ Zeta > Cypermethrin (all within one order of magnitude) Ranking of Toxicity based on available LOAEL Subchronic Toxicity (feeding) studies in rats: Beta > Cypermethrin > Alpha > Zeta (all within one order of magnitude)									

Compound studied	Strain*	Study Type	Dose [mg/kg bw or ppm] vehicle	Neuro NOAEL [mg/kg/d]	Neuro LOAEL [mg/kg/d]	Study NOAEL [mg/kg/d]	Study LOAEL [mg/kg/d]	Time to LOAEL Neuro	Reference
Alpha-cypermethrin1	dog	Subchronic Toxicity (feeding)	0-30-90-270 ppm	M: 3.5 F: 3.8	M: 6.75 F: 6.75	M: 3.5 F: 3.8	M: 6.75 F: 6.75	From day 2 or 3 onwards	KCA 5/18 AL-425-005
Beta-cypermethrin2	dog	Subchronic Toxicity (capsul)	0-1-10-20 mg/kg bw/d	1	10	1	10	4h postdosing	Sanitized 1998f
Cypermethrin4	dog	Subchronic Toxicity (feeding)	0-300-600-800-1100 ppm	M: 20.7 F: 25.4	M: 24.6 F: 34.3	M: 20.7 F: 25.4	M: 24.6 F: 34.3	Day 49-67	[redacted] 1994
Ranking of Toxicity based on available NOAEL Subchronic Toxicity studies in dogs: Beta ~ Alpha > Cypermethrin (all within one order of magnitude) Ranking of Toxicity based on available LOAEL Subchronic Toxicity studies in dogs: Beta ~ Alpha > Cypermethrin (all within one order of magnitude)									
Alpha-cypermethrin1	dog	Chronic Toxicity (feeding)	0-60-120-240 ppm	M: 2.0 F: 2.2	M: 4.1 F: 4.3	M: 2.0 F: 2.2	M: 4.1 F: 4.3	Skin irritation from week 2	KCA 5/19 AL-427-001
Beta-cypermethrin2	dog	Chronic Toxicity (capsul)	0-0.3-1-3-6-10-15 mg/kg bw/d	1	3	0.3	1	Tremors, convulsion 2h postdosing	sanitized (2004)
Cypermethrin5	dog	Chronic toxicity (capsule)	0-1-5-15 mg/kg bw	5	15	5	15	Tremors, gait abnormalities, uncoordination,	MRID 00112909 (1982)
Ranking of Toxicity based on available NOAEL Chronic Toxicity studies in dogs: Alpha ~ Beta ~ Cypermethrin (within one order of magnitude) Ranking of Toxicity based on available LOAEL Chronic Toxicity studies in dogs: Alpha ~ Beta ~ Cypermethrin (within one order of magnitude)									
Alpha-cypermethrin1	CF	Multigeneration (oral gavage, vehicle not mentioned)	M: 0-2.5-10-25 mg/kg F: 0-5-10-20 mg/kg bw/d	20 see Review report	>20	20	>20	No neurotoxic effects	[redacted] 1989 Gharda study
Zeta-cypermethrin4	SD	Multigeneration (feeding)	0-7.5-25-100-375-750 ppm	5.9	22	5.9	22	12 days	[redacted] 1991
Cypermethrin1	W	Multigeneration (feeding)	0-10-100-500 ppm	Approx. 50 Calculated 44.8	Approx. >50 Calculated>44.8	Approx. 10 Calculated: 9	Approx. 50 Calculated44.8	No neurotoxic effects	[redacted] 1978
Cypermethrin4	W	Multigeneration (feeding)	0-50-150-750/1000 ppm (conversion factor 1/20 changed to 1/10. See CA 5.6)	75 (conversion factor 1/10)	100	5	15	After 3 days at 100 mg/kg bw	[redacted] 1982b
Beta-cypermethrin2	SD	Multigeneration (gavage, CMC)	0-3-12-30/40 mg/kg bw/d	30	> 30	30	> 30	No effects	Sanitized, 2005
Ranking of Toxicity based on available NOAEL Multigeneration studies in rats: Zeta ~ Cypermethrin ~ Alpha > Beta (within one order of magnitude) Ranking of Toxicity based on available LOAEL Multigeneration studies in rats: Alpha ~Zeta ~ Cypermethrin (within one order of magnitude)									

Compound studied	Strain*	Study Type	Dose [mg/kg bw or ppm] vehicle	Neuro NOAEL [mg/kg/d]	Neuro LOAEL [mg/kg/d]	Study NOAEL [mg/kg/d]	Study LOAEL [mg/kg/d]	Time to LOAEL Neuro	Reference
Alpha-cypermethrin1	SD	Developmental Toxicity (gavage, corn oil)	0-3-9-18/15-15 mg/kg bw	9	15	Developmental: 9 Maternal: 9	Developmental: 15 Maternal: 15	9 days	[redacted] 1994; KCA 5/20 AL-432-002
Zeta-cypermethrin4	SD	Developmental Toxicity (gavage, corn oil)	0-5-12.5-25-35 mg/kg bw	12.5	25	Developmental: > 35 Maternal: 12.5	Maternal: 25 (clinical signs)	6 days	[redacted] 1990
Cypermethrin4	SD	Developmental Toxicity (gavage, aqueous mixture with corn oil)	0-17.5-35-70 mg/kg bw in corn oil, 1% w/v solution	35	70	Developmental: > 70 Maternal: 17.5	Maternal: 35 (body weight)	6 days	[redacted] 1978
Beta-cypermethrin2	SD	Developmental Toxicity (gavage in CMC)	0-30-100-300 mg/kg bw/d	30	100	Developmental: 300 Maternal: 30	Maternal: 100	8 days	Sanitized, 1998a
Ranking of Toxicity based on available NOAEL in Developmental toxicity studies in rats: Alpha > Zeta > Cypermethrin > Beta Ranking of Toxicity based on available LOAEL in Developmental toxicity studies in rats: Alpha > Zeta ~ Cypermethrin ~ Beta									
Zeta-cypermethrin4	SD	Developmental neurotoxicity (dietary)	0-50-125- 300 ppm 0-3.6-9-21.1	21.1 (slight effects on Motor activity & FOB at 21.1)	>21.1	Developmental: 9 Maternal: 9	Developmental:21 Maternal: 21	No direct neurotoxic effects	[redacted] 2005b
Beta-cypermethrin2	SD	Developmental neurotoxicity (gavage-lactational transfer)	0-3-12-30 mg/kg bw	Developmental: 12	Developmental: 30	Developmental: 12 Maternal: 3	Developmental: 30 Maternal: 12	↑ motor activity at maternal toxic doses	Sanitized, 2004a
Ranking of Toxicity based on available NOAEL in Developmental Neurotoxicity studies in rats: Beta ~ Zeta Ranking of Toxicity based on available LOAEL in Developmental toxicity studies in rats: Beta ~ Zeta									

* AP = Alderley Park; CF = Charles Foster; F = Fischer 344; LD = Long Evans; SD = Sprague-Dawley; W = Wistar
1 Study reported in the DAR alpha-cypermethrin (1999); 2 Study reported in the DAR beta-cypermethrin (2013); 3 Study newly submitted within this Annex III dossier
4 Study reported in the DAR zeta-cypermethrin Final Addendum (2008); 5 Study reported in the Alpha-Cypermethrin Human Health Risk Assessment report, EPA; DP No D37609, 2012

Comparing the NOAELs and LOAELs from the repeated dose studies, neurotoxicity does not seem to progress with prolonged exposure, which is consistent with the kinetic profile for pyrethroids. Differences in toxicity are less than one order of magnitude and follow no substance-specific ranking but are mainly explainable via substance unrelated parameters as dietary vs. capsule feeding vs. gavage in different vehicles, different strains and age of animals.

Similar toxicity is also seen when comparing other endpoints: All cypermethrins are of no acute toxicity via the dermal route, are harmful via inhalation, were evaluated as being non-irritants to skin and eye (persistent discolouration led to classification of beta-cypermethrin), induce no skin sensitization except for zeta-cypermethrin. Furthermore alpha-, beta-, zeta-cypermethrin and cypermethrin itself show no genotoxicity or carcinogenicity. Reproductive and developmental effects were restricted to reduced litter size and reduced pup body weight at maternally toxic doses.

Concerning this general similarity, no difference is expected between alpha-cypermethrin and any other cypermethrin with regard to the developmental neurotoxicity study and the lactational and/or placental transfer. The same applies for ADME data, where studies were taken from cypermethrin and alpha-cypermethrin already for the first Annex I inclusion. The same is true for the multi-generation study which was submitted by Gharda with alpha-cypermethrin as test substance, and supported by cypermethrin data.

A more in depth discussion of the toxicological similarity of cypermethrin, zeta-cypermethrin and alpha-cypermethrin was submitted to the US EPA in 2009 [see KCA 5/21 2009/7003352] to waive the need of a dietary DNT study with alpha-cypermethrin. US EPA accepted the waiver and evaluated the hazard of alpha-cypermethrin in combination with cypermethrin and zeta-cypermethrin due to structural and toxicological similarities and comparable toxicity profiles (US EPA, 2012; [see KCA 5/22 2012/1367302]).

Conclusion with regard to bridging consequences:

Based on the substantial similarity of alpha-, zeta- , beta-, and cypermethrin as discussed based on comparable data (see
Table 5-4: Comparison of repeated-dose toxicity studies performed with Cypermethrin, Zeta-cypermethrin, Alpha-cypermethrin and Beta-cypermethrinTable 5-2 -
Table 5-4), no additional assessment factor is considered necessary when bridging study results within the group of cypermethrins. Furthermore all relevant studies for endpoint derivation were performed with alpha-cypermethrin itself.

CA 5.1 Studies on Absorption, Distribution, Metabolism and Excretion in Mammals

CA 5.1.1 Absorption, distribution, metabolism and excretion by oral exposure

Studies evaluated in the draft monograph of alpha-cypermethrin (1999): Alpha-cypermethrin is a component of cypermethrin, and there is no indication that it would have different toxicological effects. Therefore, ADME studies of alpha-cypermethrin have been evaluated together with data from cypermethrin studies and metabolic pathways were elucidated in rats following oral administration (List of studies is presented under CA 5.1.1). The Annex I endpoints were fixed in the Review report of alpha-cypermethrin (SANCO/4335/2000 final from 13 February 2004) as follows:

Absorption, distribution, excretion and metabolism in mammals

Rate and extent of absorption:	46% within 24 h
Distribution:	Widely distributed, highest residue in fat and skin
Potential for accumulation:	High accumulation in fat
Rate and extent of excretion:	76-78% within 24 h (43-46% in urine; 30-35% in faeces)
Toxicologically significant compounds:	Parent compound
Metabolism in animals:	± 50 % metabolised via hydrolytic cleavage of ester bound

The different formulations used for absorption, distribution, metabolism and excretion studies are summarized in table below:

Table 5.1.1-1: Characteristics of the formulations of alpha-cypermethrin and cypermethrin used in the metabolism studies

Substance	Position of label, Batch No.	Vehicle	Specific activity	Radiochemical purity (%)	Reference
Alpha-cypermethrin, oral	^{14}C-phenoxy benzyl ring labelling, 1, OCD 594	corn oil	29.7 µCi/mg	99.6	[redacted] 1982 Cyanamid
Alpha-cypermethrin, oral	^{14}C-phenoxy benzyl ring labelling, 30, OCD 594	corn oil	29.7 µCi/mg	99	[redacted] 1983 Cyanamid
Alpha-cypermethrin, oral	^{14}C-CN, B121	?	55.3 mCi/mmol or 825 µCi/mg	97.8	[redacted] nknown Gharda
Cypermethrin (50:50), oral	^{14}C-benzyl	corn oil	9.6 µCi/mg	>99	[redacted] 1977 Cyanamid
WL43481 cis cypermethrin, oral	^{14}C-benzyl	corn oil	34 µCi/mg	>99.5	[redacted] [redacted] [redacted] 1977a

Table 5.1.1-1: Characteristics of the formulations of alpha-cypermethrin and cypermethrin used in the metabolism studies

Substance	Position of label, Batch No.	Vehicle	Specific activity	Radiochemical purity (%)	Reference
WL42641 trans cypermethrin, oral	^{14}C-benzyl	corn oil	35 µCi/mg	>99.5	Cyanamid
Cypermethrin (50:50), oral	^{14}C-benzyl	corn oil	low dose kinetics: 39.8 µCi/mg high dose kinetics: 1.0 µCi/mg low dose metabolites: 39.7 µCi/mg high dose metabolites: 1.0 µCi/mg	99.5 – 99.9	[redacted] 1980 Cyanamid
	^{14}C-cyclopropyl	corn oil	low dose kinetics: 40 µCi/mg high dose kinetics: 1.0 µCi/mg low dose metabolites: 48 µCi/mg high dose metabolites: 1.64 µCi/mg	99.3 – 99.5	
Cypermethrin (50:50), oral	^{14}C-benzyl, Batch No. 981-26	corn oil	0.5 µCi/mg	>99	[redacted] 1980 Cyanamid
	^{14}C-cyclopropyl	corn oil	0.49 µCi/mg	99.6	
Cypermethrin (50:50), oral	^{14}C-benzyl	corn oil	1 µCi/mg	>99	[redacted] 1981 Cyanamid
Cypermethrin (50:50), oral	^{14}C-cyclopropyl	corn oil	1.11 µCi/mg	>99	[redacted] 1978 Cyanamid
Cypermethrin (46:54), iv	^{14}C-phenyl	PEG 300	152.57 µCi/mg	>98.1	[redacted] 1993 Mitchell Cotts
3-phenoxybenzoic acid, oral	^{14}C-benzoyl ring	aqueous NH_4OH	13.9 µCi/mg	>99.9	[redacted] 1978 Cyanamid
3-phenoxybenzoic acid, oral	^{14}C-benzoyl ring	aqueous NH_4OH	13.9 µCi/mg	not stated	[redacted] 1978 Cyanamid

Submission of not yet peer-reviewed studies in this AIRIII-Dossier: A study on oral adsorption kinetics, distribution and excretion of **alpha-cypermethrin** is supplementing the already available data and is discussed in this chapter under point KCA 5.1.1/1 AL-440-017. A verification of the **metabolite identification** in rats is discussed under KCA 5.1.1/2 2013/1086630. Additionally, the dermal absorption of alpha-cypermethrin has been investigated in the representative formulation in vitro in human skin and revealed a maximum absorption of 3% for the concentrate (see KCP 7.3).

According to the new data requirements for active ingredients of plant protection products as set out in Commission Regulation (EU) No. 283/2013 (1 March 2013, OJ L93, 1ff, 3.4.2013), "comparative *in vitro* metabolism studies shall be performed on animal species ... and on human material ...in order to determine the relevance of the toxicological animal data and to guide in the interpretation of findings and in further definition of the testing strategy..." (Section 5, Toxicological and metabolism studies, point 5.1.1., page 22). In the absence of validated test method or guidance documents, this data requirement is waive in accordance to SANCO Guidance Document SANCO/10181/2013-rev 2.1 (13 May 2013).

Table 5.1.1-2: Characteristics of the formulations of alpha-cypermethrin used in the ADME studies newly submitted

Substance	Position of label, Batch No.	Vehicle	Radiochemical purity (%)	Reference
Alpha-cypermethrin, oral	^{14}C-phenoxy benzyl ring labelling, CFQ11156, CP-2386	corn oil	CFQ11156:92%, CP-2386: 98%	[redacted] 2000 KCA 5.1.1/1 AL-440-017
Alpha-cypermethrin, oral	benzyl-U-^{14}C: 775-0401 phenoxy-1,2,3,4,5,6-^{13}C:1025-1034 cyclopropane-1-^{14}C: 986-1046 cyclopropane-1-^{13}C: 990-1024	corn oil	775-0401: 96.1% 1025-1034: 95.1% 986-1046 : 98.2% 990-1024: 97.7%	KCA 5.1.1/2 2013/1086630

A new rat study on absorption, distribution and excretion of alpha-cypermethrin as well as a detailed metabolism is available which extends and augments the previous studies (List of studies is presented under CA 5.1.1). Additionally, the dermal absorption of alpha-cypermethrin has been investigated in the representative formulation in vitro in human skin and revealed a maximum absorption of 3% for the concentrate (see KCP 7.3).

When information from the older and new studies is reviewed in total, the following endpoints are proposed as an update without changing the relevant endpoints.

Proposed endpoints for Absorption, distribution, excretion and metabolism in mammals

Rate and extent of absorption:	Relatively rapid; Tmax 6-9 hours; Oral absorption : 46 %, considering urinary and biliary excretion (within 48 h), and carcass residuals.
Distribution:	Widely distributed, mainly in well perfused organs (mesenteric lymphnode, liver, kidney) and lipophilic organs (fat, brown fat).
Potential for accumulation:	Accumulation in fat (T½ (fat) > 24h),
Rate and extent of excretion:	24 h: 79-87% (urine: 31-39%; faeces: 39-55%) 48 h: 92-94% (urine: 34-46%; faeces: 46-60%)
Toxicologically significant compounds:	Parent compound
Metabolism in animals:	± 50 % metabolised via hydrolytic cleavage of ester bound

The summary of these ADME studies is as follows:

Absorption: Oral absorption of alpha-cypermethrin is relatively rapid with Tmax reached within 6-9 hours. After single oral application approximately 46 % of the dose is absorbed within 48 hours [redacted] 1982 and confirmed in KCA 5.1.1/1 AL-440-017). After 168 hours up to 50 % is considered bioavailable based on urinary excretion, bile excretion and residual carcass radioactivity [see KCA 5.1.1/1 AL-440-017]. The presence of large unchanged portions of the dose in faeces may be interpreted as a sign of limited absorption in the gastro-intestinal (GI) tract and may be considered as potentially non-absorbed [redacted] 1978; [redacted] 1980).

Distribution: Alphacypermethrin is widely distributed, mainly in well perfused organs (mesenteric lymphnode, liver, kidney) and lipophilic organs (fat, brown fat). Distribution and elimination from tissues is rapid, for example the half-life in plasma is biphasic with 4 and 6-8 hours for low and high dose (rapid initial depletion) and around 24 h in a slower second phase (AL-440-017), half-life in liver and kidney is ca. 2 days ([redacted] 1983). The depletion of alpha-cypermethrin from adipose tissue is biphasic with half-lives of ca. 2 days (rapid initial depletion) and 17-26 days (slower second phase), compared to a second phase of 18.9 days for cis-cypermethrin (WHO, 1992, [see KCA 5/10 1992/1005337]).

Metabolism: Alphacypermethrin, like Cypermethrin, is extensively metabolised by ester cleavage [redacted] 1982) and subsequent elimination of the cyclopropanecarboxylic acid moiety largely as its glucuronic acid conjugate [redacted] 1977). The 3-phenoxybenzoic acid portion of the compound undergoes hydroxylation and sulphate conjugation (major route) prior to excretion [redacted] 1982). The major metabolite determined in urine was 3-(4- hydroxyphenoxy) benzoic acid-O-sulphate conjugate (34–40 % of the dose). In faeces, mainly the unchanged parent compound was found (20 % of administered dose).
The present Annex I renewal dossier provides new metabolism studies using alpha-cypermethrin in goat, hen, fish and rat. The new studies mainly confirm the findings and degradation reactions known so far and complement additional details due to more sensitive technology.
The proposed metabolic pathway of Alphacypermethrin in rats is presented in Figure **5.1.1-1**.

Excretion: More than 92% of the radioactivity is excreted within 48 hours of administration, equally partitioned amongst feces and urine in females (both 46%) and slightly less in urine in males (34.2% via urine and 60% via feces). Excretion via bile is considered to be limited to around 3% in the first 24 hours. No radioactivity was excreted in the expired air.

General aspects: Apparent sex differences in the pattern of excretion and identified metabolites were not observed.

Human volunteer study: A human volunteer study was conducted in 1984 to determine oral absorption, and to develop a human biomonitoring procedure based on urinary metabolite analysis. There was no evidence of any accumulation after repeated oral administration of Alphacypermethrin to human volunteers. On average, 49 % of the dose was excreted as ciscyclopropane carboxylic acid, which is also a major metabolite in rats (van Sittert, 1984).

In the following already peer reviewed data taken from the different DARs as indicated are summarized and new data are attached at the end.

Absorption (taken from DAR alpha-cypermethrin, 1999, except where mentioned otherwise)

Oral absorption:

No animal studies measuring oral absorption kinetics of **alpha-cypermethrin** are available. On the basis of distribution, metabolism and excretion, it seems that after low or high dose of alpha-cypermethrin ±45% of the dose was excreted in urine; faecal excretion representing ±33% for the same period. Just over 20% of the compound is not absorbed and is eliminated unchanged in the feces.

As far as **cypermethrin** is concerned, the absorption process itself proceeds quite rapidly. The maximal concentration of radioactivity in the blood after oral administration of a low dose, reaches a maximum within 3-4 h and this for both sexes and for both labeling positions. Increasing the dose by a factor of 100 was followed by a 10-30 times increase in blood concentrations suggesting a saturable mechanism ([redacted] 1980).

After a low dose ±50% of cypermethrin administered was excreted in urine within a 24 h period (32-62% in urine, 16-35% in feces) ([redacted] 1977). Over a 3-7 day period 26-60% of radioactivity is eliminated in urine ([redacted] 1980).
Absorption of cypermethrin is incomplete representing ±50% of a low dose within the first 24 h after oral administration.

Human dermal absorption:

After a four hour dermal application of a formulation containing cypermethrin (49.6 mg) on the underside of the forearm, the formulation was removed and contained 33.4 mg cypermethrin; the dermal retained dose was therefore, 14.2 mg (±28%) by substraction. The cypermethrin metabolite dichlorocyclopropanecarboxylic acid (DCVA), and its free and conjugated forms, was not detected, i.e. <0.3 mg, in urine samples up to 96 h after dermal application.
Little is known about the fate of the remainder of the dose, which may have diffused into and be stored in a nurnber of skin layers and as such be, in part, available for further removal using the washing procedure (Coveney and Eadsforth, 1982).

Human data from the open literature:

Oral absorption:

Cypermethrin (cis/trans ratio = 1:1; 98.9-99.4% purity) was administered orally to 6 male volunteers as a single oral dose of 3.3 mg. Urine samples were collected for up to 5 days. Twice as much trans DCVA was excreted in urine compared to cis DCVA. From the urinary metabolite ratios found, it is apparent from DCVA/(3PBA+4HO-3PBA) ratio that more of the DCVA part of the molecule was accounted for compared to phenoxybenzyl moiety, indicating a greater proportion of the phenoxybenzyl moiety is converted to other metabolites. Peak excretion rates of metabolites occurred between 8 and 24 h after dosing. Based on metabolites measurements, the amount of absorbed dose was estimated between 27 and 57% (mean 36%) of the administered dose.

Dermal absorption:

Cypermethrin (cis/trans ratio = 1:1; 98.9-99.4% purity) was administered dermally to 6 volunteers at a dose range of 31 mg/800 cm^2 as a soya oil-based formulation. A substantial proportion of the dose (mean 41%) was recovered in the mild detergent wash after 8 h. The amounts of cyclopropane acid metabolites are 4 times lower than the amounts derived from the phenoxybenzyl moiety. This indicates that the cyclopropane acid part of the molecule may either not be effectively absorbed, or more probably is converted to other metabolites which are not measured using the assay procedure. The estimate of absorption of cypermethrin, based on cyclopropane acids is of 0.3% and 1.2% for phenoxybenzoic acid metabolites. 24% was recovered from a T-shirt worn overnight. Peak urinary excretion rates of metabolites occurred between 12 and 16 h after dermal dosing. A recovery of 66.55% can be calculated.
These results demonstrate two important differences in the excretion of urinary metabolites when cypermethrin is dosed orally and dermally. First the trans/cis DCVA ratio differs (oral 2:1; dermal 1:1.2) and secondly the ratio of total DCVA/phenoxybenzoic acid metabolites differs (oral 1:0.8; dermal 1:4). For the purpose of biological monitoring total DCVA could substantially underestimate absorption when dermal is the main route and the amount of total phenoxybenzoic acid metabolites will best reflect absorption of cypermethrin by all routes (Woollen et al., 1992).

Distribution (taken from DAR alpha-cypermethrin, 1999, except where mentioned otherwise)

Single dose studies

Maximal tissue concentration is reached 24 h after oral administration of a low or high dose of **alpha-cypermethrin**. At that time, appreciable quantities of test material were present in fat. For each of the fatty tissues there appears to be an initial rapid depletion of residues, with a half-life of about 2 days, followed by a second slower phase. $T_{1/2}$ blood was calculated as 6.4 h for a high dose and 8 h for a low dose ([redacted] unknown date).
Four days after single oral dose of **alpha-cypermethrin**, radioactivity detected in tissues did not exceed 0.01% of the administered dose. Some tissues retained higher amounts, liver (0.1%), skin (0.25%), carcass (0.9%) and intestines (0.3%). The highest concentration was found in adipose tissue (0.42%) ([redacted] 1982a).

Fourteen days after exposure to **alpha-cypermethrin** the concentration of radioactivity was at the detection limit in the majority of tissues except in fat, where an appreciable amount of radioactivity was still present [redacted] unknown date).

Some analysis of the peri-renal and parovarian fat was carried out in an attempt to identify the nature of the residue. It was found that more than 98% of the radioactivity chromatographed as alpha-cypermethrin, indicating that there was little or no isomerization in fat over an extended period of time. Lipophilic metabolite(s) of **alpha-cypermethrin**, with longer half-lives than that of the parent compound were, however, also present [redacted] 982a).

At 1 h after i.v. administration of **cypermethrin**, highest radioactivity levels were found in heart, liver, kidney, ovaries and blood, fat and skin region. Radioactivity was rapidly eliminated from blood with a half-life of 5.9 h. Similar half-lives were determined for most organs/tissues. Only in fat (>24 h) and skin backregion (12.8 h) longer half-lives were found. At 120 h, except for fat and skin region, all radioactivity levels were at or below 0.1 µg/g [redacted] 1993).
3 days after a single oral low dose of **cypermethrin** similar low concentrations as for alpha-cypermethrin were found for both sexes in kidney, muscle, brain and blood (<0.1%); male rat liver contained 3 times the amount found in the female liver. The residue in the fat of the female rats was 2-3 fold higher than that found in the male rats (0.31 for male and 0.72% for female) [redacted] 1977).
A comparison of the data at 3 days for the 2 groups of isomers revealed trans isomer was lesser retained in the tissue than the cis isomer. Between day 1 and day 8 after dosing the cypermethrin residues in liver, kidney, muscle, brain and blood decreased by a factor of 10. The highest and most persistent residual radioactivity was in fat. There was a trend to have lower concentrations of trans isomer than cis isomer; this was most noticeable in the fat of males (5 times lower trans than cis). There was a sex difference for the trans isomer in the fat with a 2.5 times higher level in females than in males ([redacted] 1977).
Seven days after dosing with 200 mg/kg **cypermethrin** very little radioactivity was retained by the animals (<1% of the administered dose). The concentration of radiolabelled material in the tissues were generally low, being less than 1 µg/g with the exception of tissues which were still actively engaged in the metabolism or which possessed a high fat content. The highest residues were found in fat, they were not 100 times higher than after 2 mg/kg [redacted] 980).

Distribution of cyclopropane- or phenoxybenzyl moiety among the different tissues appear to be comparable except in fat were the levels of benzyl moiety were somewhat higher. The extraction of label from tissues with solvent was well above 50% for most tissues (e.g. fat 99%) except for liver (27-48%) [redacted] 980).

Repeated dose study

The following information was taken from the DAR of zeta-Cypermethrin:
The concentration of radioactivity found in tissues of male and female rats after the last of 28 daily oral doses of ^{14}C labeled on the benzyl ring **cypermethrin** showed a similar distribution in male and female rats with the exception of gonads. The highest concentrations of radioactivity were found in fat and tissues with a high fat content i.e. adrenal glands, skin and ovaries. High concentrations were also found in organs associated with absorption (GI tract), metabolism (liver) and excretion (kidney), while lower concentrations were found in whole blood and plasma. All other tissues studied, notably the brain, contained little or no radioactivity. The residual carcass also contained a considerable quantity of radioactivity due at least in part to the remaining adipose tissue and skin [redacted] 1980).

Further information was taken from the DAR of alpha-cypermethrin, without Tables:
When given as a low daily dose over 70 days, **cypermethrin** related radioactivity at day 70 was found predominantly in lipid sites, particularly adipose tissue and skin. In fat the cis isomer represented 88% of the residue, the trans isomer 12%. The cis and trans isomers were eliminated with elimination half-lives of 18.24 and 3.43 days, respectively, to reach background levels 7 weeks after stop of dosing [redacted] 1981).

From the open literature it appears that more rapid elimination of trans **cypermethrin** is not due to a difference in lipophilic character, but is more probably related to a lipase mediated hydrolysis of the ester bond which would be expected to proceed much more rapidly with the trans isomer than with the cis isomer.

Metabolism (taken from DAR alpha-cypermethrin, 1999, except where mentioned otherwise)
The metabolic fate of **alpha-cypermethrin** is similar to that of cypermethrin. Hydrolytic cleavage of the ester bound and elimination of the cis and trans cyclopropanecarboxylic acid moieties in the free and conjugated form is a major route of metabolism of cypermethrin in rats and in man.
Metabolism of **cypermethrin** and elimination of metabolites derived from the cyclopropane carboxylate moiety via urine in man was rapid (Eadsforth, 1981). From open literature it seems that the much less toxicity of the trans isomer as compared to the cis isomer of cypermethrin is largely owing to the facile metabolism of the trans-compounds by carboxylesterase-catalysed hydrolysis [redacted] 1991).

The cis and trans cyclopropanecarboxylic acid moiety were almost exclusively conjugated and rapidly eliminated as the ester glucuronide (30% of the dose in male, 47% of the dose in females; trans representing 51.4% and cis 32.8% of the administered dose) with some free acid (twice as much trans as cis) being found in the urine (4%) together with traces of the trans-hydroxymethyl cyclopropyl acids (2%).

Minimal hydroxylation occurred at the methyl groups attached to the cyclopropane ring, giving a complex mixture of partially unstable metabolites. The cis isomer, somewhat more resistant to hydrolysis than the trans isomer, gave more hydroxylated metabolites than did the trans isomer. Only small amounts of hydroxylated metabolites with the ester bound intact (e.g. 4-HO-cis-cypermethrin) were excreted in the feces.

The **3-phenoxybenzyl product** was mainly converted into 3-phenoxybenzoic acid. 3-Phenoxybenzoic acid was excreted free but aromatic hydroxylation at the 4´-position occurred also before excretion followed by sulphation of 3-(4-hydroxyphenoxy)benzoic acid.
The 4´-hydroxy sulphate forms the major aryl metabolite (16% of the dose) with 3-phenoxybenzoic acid as the second most important (5%). The other identified aryl metabolites are 3-(4´-hydroxyphenoxy)benzoic acid (1%) and the glycine conjugate. N-(3-phenoxybenzoyl)glycine (1%).

There was no evidence of any major sex differences in biotransformation at the higher toxic dose, or of any dose-dependent major change in the route or extent of biotransformation, but a number of uncharacterized minor metabolites (generally <3% of the dose) were detected [redacted] 1980).

The **metabolic fate of 3-phenoxybenzoic acid (3PBA)** and one of its major plant metabolites, the glucoside conjugate, was studied in rats. Rats hydrolyse 3PBA-glucoside and further metabolise it as 3PBA. The metabolism of 3PBA administered as such to rats appears to be virtually identical to that of 3PBA liberated during the metabolism of cypermethrin ([redacted] 1978).
In the skins of rats receiving 3-phenoxybenzoic acid (after 7 daily oral doses giving a total of 750 mg/kg) unchanged 3-phenoxybenzoic acid and a mixture of 3-phenoxybenzoyl-dipalmitins were identified. The components were present in the approximate ratio 3:7. They were also present in the carcass, but in the ratio 9:1 [redacted] 1979).

Only small amounts of hydroxylated metabolites of cypermethrin with the ester bound intact (e.g. 4´-hydroxy-cis-cypermethrin) were excreted in the feces. The very low yield of radioactivity found in the bile was probably comprised of these compounds, and suggests that the cypermethrin found in the feces was there because it was not absorbed during passage through the gut. There is, as yet, no evidence for the metabolism at the 2,2.dichlorovinyl group of cypermethrin [redacted] 1978).

In mice, metabolism of **cypermethrin** was similar as in rats [redacted] 1981).

In vitro studies using liver microsomal preparations from rats and mice showed that ester cleavage is more extensive for trans than for cis pyrethroids, while the relative extent of oxidative metabolism of the 2 isomers depends on the enzyme source. The low toxicity of trans cypermethrin to mice to the corresponding cis isomers is consistent with their greater ease of biodegradation in the mouse microsomal system (Shono et al., 1979).

Human data

The following information on human data was taken from the DAR of zeta-Cypermethrin:

Following a single oral dose 3.3 mg **cypermethrin**, peak excretion rates of the hydrolysis products *cis* and *trans* DCVA are generally seen within the first 4 h. Peak excretion rates of oxidized metabolites 3PBA and 4OH3PBA were seen later between 4 and 24 h after dosing. An average of 93% of the metabolites recovered was excreted within 72 h of dosing. In the majority of individuals all or some of the metabolites were still detectable in urine 5 days after dosing. The mean elimination half-life for total metabolites was 16.5 h. The average trans/cis DCVA ratio was 2:1 and approximately equivalent amounts of total DCVA and total phenoxybenzoic acid metabolites were recovered in urine. Based on the recovery of *trans* DCVA the proportion of the administered dose absorbed is estimated to be between 27% and 57% (mean 36%).

Following a single dermal dose of 31 mg, metabolites were detectable in the majority of the urine samples collected for the period 0-4 h, but peak excretion rates were not seen until between 12 and 36 h after dosing. The mean elimination half-live for total metabolites was 13 h. The average *trans/cis* DCVA ratio was 1:1.2 and the amounts of cyclopropane acid metabolites were, on average, 4 times lower than the amounts of metabolites derived from the phenoxybenzyl moiety. Based on urinary excreted metabolites, dermal absorption reaches approximately 1.88% of the dose. These results demonstrate 2 important differences in the excretion of urinary metabolites when cypermethrin is dosed orally and dermally. First, the *trans/cis* DCVA ratio differs (oral 2:1; dermal 1:1.2) and secondly the ratio of total DCVA/phenoxybenzoic acid metabolites differs (oral 1:0.8; dermal 1:4). Therefore, monitoring of total DCVA could substantially underestimate absorption when dermal is the main route and the amount of 3PBA + 4OH3PBA will best reflect absorption of cypermethrin by all routes (Woollen et al., 1992; published study).

After a single oral dose cypermethrin (0.25-1.5 mg) to 4 human subjects, urine was monitored for the free and conjugated cyclopropane carboxylic acid. Urinary excretion during the first 24 h after dosing showed an average 78% of the *trans* isomer dose and 49% of the *cis* isomer dose in the form of metabolites. These results suggest that, as in other mammals, ester cleavage and elimination of the *cis* and *trans* cyclopropane carboxylic acid moieties is a major route of metabolism of cypermethrin in man (Eadsforth and Baldwin, 1983).

Excretion (taken from DAR alpha-cypermethrin, 1999, except where mentioned otherwise)

After oral administration of a single low or high dose of **alpha-cypermethrin**, 76-78% of the dose was excreted within 24 h (43-46% in urine and 30-35% in feces) reaching 93.1-95.6% in 4 days. Alpha-cypermethrin is eliminated similarly by both sexes [redacted] 1982 DocID AL-905-066).

After oral administration of a single low dose of **cypermethrin** excretion of radioactivity within 3 days was 90.8-100.8% (54.7-66.5% in urine, 28.7-27% in feces) ([redacted] 1977). Urinary excretion of the trans isomer appears to be faster while cis isomer was more excreted via faeces. Total recovery was 100% for male and 97% for female rats for the cis and 103% for males and 101% for females receiving the trans isomer in 3 days [redacted] 1977).

After oral administration of a high dose of cypermethrin, 73-79% of the dose for males and 80-87% for females was eliminated within 3 days. The excretion was generally greater in the feces than in the urine (except females dosed with ^{14}C-cyclopropyl) and suggests that an appreciable portion of the dose was not absorbed. Cyclopropyl label elimination was somewhat slower than that seen with the benzyl label. The benzyl label is more excreted in the feces than the cyclopropyl label ([redacted] 1980).

Detection of some radioactivity in expired air (0.1%) indicates some metabolic breakdown of the cyclopropyl ring.

Biliary excretion of metabolites was investigated and only 1-1.6% of an oral dose was excreted via this route within 4-5 h as conjugated metabolites in a ratio cis:trans of 5:3.

After intravenous administration of cypermethrin to female rats urinary excretion was somewhat lower than after oral administration.

Additional information taken from the DAR of zeta-cypermethrin (2008):
Cumulative radioactivity excreted in the urine amounted to 0.8%, 11.6%, 44.5% and 61.3% after 1, 4, 24 and 120 hours, respectively. Significant amounts of radioactivity (7.9%) were excreted via bile to feces from 4 to 24 hours, reaching 28.4% after 120 hr with the major amount (24.3%) excreted already within the first 48 hours. Total excreted radioactivity amounted, on average, to 89.7% [redacted] 1993).

Human data **(Taken from the DAR of alpha-cypermethrin)**
After single or repeated oral administration of **alpha-cypermethrin** (0.25-0.75 mg), people excreted 43 or 49% respectively of the dose in urine within 24 h after treatment in the form of free and conjugated cis cyclopropane carboxylic acid (van Sittert et al., 1985, DocID AL-445-003).
After a single oral dose of **cypermethrin**, urinary excretion was rapid and confined to the first 24 h sample collection period: 78% of the trans isomer and 49% of the cis isomer as free and conjugated cyclopropanecarboxylic acid (Eadsforth, 1981).

Excretion of plant metabolites in rats
After 3-phenoxybenzoic acid administration to rats the major route of excretion for both sexes was the urine (81.5% for males and 79.8% for females). After 4 days very little radioactivity was observed in the carcass and skin of either sex (0.7% males; 0.65% females).
Following oral dosing to rats of the glucoside derivative, the elimination of radioactivity was very similar to that found with 3PBA with the exception that only 0.3% of the dose was found in the skin [redacted] 1978).

Metabolism in livestock (Taken from the DAR of alpha-cypermethrin)
Alpha-cypermethrin:
^{14}C-labelled alpha-cypermethrin was administered orally to a cow (14 mg/kg) via twice daily doses added to the diet. Whereas muscle, fat and milk mainly contained a single compound with similar chromatographic properties to alpha-cypermethrin (muscle 85%, fat 91%, milk 97%), the liver extract contained at least 8 metabolites with a broad range of polarities, one component (±16%) had similar chromatographic properties to those of alpha-cypermethrin. In urine, the 2 major components were probably 3PBA (3%) [redacted] 1994).

Excretion of ^{14}C-labelled alpha-cypermethrin was rapid in lactating cows after a single oral administration at 0.1, 0.2, or 0.4 mg/kg bw reaching 48% in urine and 28% in stools in the first 24 h. The amount present in milk was 0.08 µg/mL on the 4th day ([redacted] 1990).

Cypermethrin:

Lactating cows were fed with cypermethrin; the insecticide is rapidly metabolized by cows. Urinary metabolites were tentatively identified as 3-(4-hydroxyphenoxy)benzoic acid O-sulphate (minor) and N-(3-phenoxybenzoyl)glutamic acid, the major metabolite. Part of the ingested cypermethrin (36%) was eliminated unchanged in the feces. The major residue in liver and kidney was N-(3-phenoxybenzoyl)glutamic acid. Unchanged cypermethrin was found in the lipophilic component of milk. Unlike rats, cows afford only small amounts of phenyl-hydroxylated metabolites in urine.Metabolites in the urine were the result of hydrolysis and the formation of 3-phenoxybenzoic acid, which was conjugated mostly with glutamic acid and to a lesser extent with glycine. The cows therefore differs from the other species studies in respect of the amino-acid selected for conjugation ([redacted] 1980; [redacted] 1976).

After dermal application to sheeps, absorption was slow and less than 0.5% of the dose was excreted in the urine in 24 h and only 0.5% was excreted in feces within 6 days. Conversely, absorption and elimination after oral treatment was rapid over 48 h (61%). Radioactivity in fat samples was identified as cypermethrin ([redacted] 1977).

The fate of cypermethrin was assessed in laying chickens. Unchanged compound was essentially recovered in yolk; minor metabolites were trans hydroxyl(methyl)-cypermethrin and 3-phenoxybenzoic acid. In fat unchanged cypermethrin was present. In liver beside unchanged cypermethrin polar metabolites which could not be converted chemically to 3PBA or 4HO-3PBA were detected ([redacted] 1982).

Metabolism in plants (Taken from the DAR of alpha-cypermethrin)

Under glasshouse conditions:

Metabolism of cypermethrin was studies in lettuce and cotton foliage under glasshouse conditions after treatment with separate cis- and trans-isomers. The detected metabolites (3-phenoxybenzoic acid, the amide and a hydroxylated derivative of the parent compound, 4´-hydroxy derivative?) suggests a metabolism by conversion of the cyano group to amide and by hydrolysis at the ester bond leading to the formation of 3-phenoxybenzoic acid. There was also evidence for aryl hydroxylation of the intact molecule (Gilham, 1976).

In cotton plants only minor variations in the rate of degradation of cypermethrin were observed: 3-phenoxybenzoic acid and 3-phenoxybenzyl alcohol were identified. The presence of polar metabolites formed by conjugation of the cyclopropane carboxylic acid was suggested. No evidence was provided to suggest that ether cleavage had occurred (Sherren and Dutton, 1981).

Soya bean plants has been treated by spraying of the plant; the major degradation pathway of cypermethrin appears to be hydrolysis of the ester bond of the parent molecule followed by conjugation of the metabolites produced. Small concentrations of metabolites with the ester bond intact were also observed. The concentrations of free metabolites observed were considerably less in soya compared with cotton and lettuce (Edwards,1979).

Using soybean callus tissue cultures the metabolites which were detected are consecutive to the hydrolysis at the ester linkage and oxidation of one of the methyl groups attached to the cyclopropyl ring (Standen, 1980).

Under outdoor conditions
The major metabolite detected in lettuce foliage was the cyclopropane carboxylic acid, which occurred mainly as conjugates with sugars including glucose. Relatively few components have been detected apart from cypermethrin and material of quite high polarity and contrasted to the previous indoor study. The major metabolites of cypermethrin in lettuce under outdoor conditions resulted from ester cleavage and further metabolism or conjugation of the acid and alcohol moieties (Wright, 1977).
Cis and trans isomers of cypermethrin were applied as overall sprays to cotton plants. The major metabolites present in boll cares and leaves were cis- and trans-cyclopropanecarboxylic acids and 3-phenoxybenzoic acid, which all occurred in both free and conjugated forms. In the kernel small radioactive residues occurred as metabolites, which it was not possible to identify (Hitchings and Wright, 1979).
The leaves of apple trees contained unchanged insecticide (36-42%) and the apple peel contained 50-77% of unchanged insecticide. In leaves and fruits treated with cis isomer there had been considerable conversion into trans isomer. There was no conversion into the cis isomer. Metabolites detected in leaves and fruits were cis, trans cypermethrin, 3-phenoxybenzaldehyde, 3-phenoxybenzoic acid, the amide, the hydroxylated amide, 3-phenoxyalcohol and conjugated material (Dutton and Roberts, 1978).

Conclusion: (Taken from the DAR of alpha-cypermethrin)
Absorption of **alpha-cypermethrin** in rats within the first 24 h after oral administration represents approximately 45% of the dose (43-46% in urine and 30-35% in feces) 76-78% of the dose was excreted within 24 h reaching 93.1-95.6% in 4 days. Increasing the dose by a factor of 10 did not modify the excretion.

The absorption of cypermethrin after oral low dose represents approximately 50% of the dose (32-62% in urine; 16-35% in feces) within the first 24 h after administration; 2-3 days later 90-100% of the dose was recovered in excreta (54.7-66.5% in urine as unchanged compound and metabolites and 28.7% in feces mainly as unchanged compound). After high dose administration, absorption of cypermethrin was reduced to 8% (7-19% of the dose is excreted in urine while 7-31% is recovered in feces) suggesting a saturation effect at high dose.
After dermal application to human skin 28% of cypermethrin was retained into the skin within 4h.

Alpha-cypermethrin reached maximal tissue concentrations within 24 h after administration decreasing rapidly within 4 days after administration (<0.01%). Appreciable quantities of test material were present in fat (3.64 µg/g tissue at 24 h), decreasing slowly as a function of time (0.83 µg/g tissue after 7 days).
One hour after i.v. administration of cypermethrin radioactivity was found in heart, liver, kidney, ovaries and blood, fat and skin region. Low tissue concentrations were found (<0.1%) 3 days after oral administration of cypermethrin. Trans isomer was less retained (5 times lower trans than cis) and there was also a sex difference for the trans isomer in the fat 2.5 times higher level in females than in males.
The extraction of label from tissues with solvent was well above 50% for most tissues (e.g. fat 99%) except for the liver (27-48%). Tissue retention of the phenoxybenzyl moiety in fat is slightly higher than cyclopropyl moiety.

The half-life values for elimination of cis and trans isomers were calculated to be 18.24 and 3.43 days showing that cis and trans isomers of cypermethrin accumulated in fat. In contrast, the reduction of radioactivity in the liver and kidney with time is very rapid and after 2 weeks is at or below the limit of detection.

The metabolic fate of alpha-cypermethrin is similar to that of cypermethrin; the ester bound is preferentially cleaved yielding a cyclopropane carboxylate moiety and a dibenzyl (3-phenoxybenzoic acid) moiety. A more facile metabolism of the trans compounds by carboxylesterase-catalysed hydrolysis was reported in mammals. The phenoxybenzoic acid compounds is further hydroxylated and half of the administered dose of cypermethrin or alpha-cypermethrin is recovered as 4-OH-3-phenoxybenzoic acid of which 65% is the sulfate conjugate and to a minor percentage the glycine conjugate. The unchanged compound is also found as glucuronide and sulfate conjugate. In feces, most of the radioactivity is unchaged compound; radioactivity as metabolites represents only 1 to 3% of the administered dose. There is, as yet, no evidence for the metabolism at the 2,2-dichlorovinyl group of cypermethrin.

In humans, urinary **excretion of alpha-cypermethrin** reached 43 to 49% within 24 h after a single or repeated oral administration. After a single oral dose of cypermethrin urinary excretion reached 49% for the cis isomer and 78% for the trans isomer in 24 h after dosing. These data suggest that human absorption of cypermethrin is comparable to rats. Metabolism was also similar to rats. The presence of the trans isomer does not influence the metabolism and excretion of its cis congener.
Metabolites of cypermethrin were identified in mice and cows. Overall, metabolism is similar to rats. Differences that occur are related to the rate of metabolite formation rather than to the nature of the metabolites formed. The only major differences between species relate to conjugation reactions. While the fate of the cyclopropanecarboxylic acid has bennn shown to be largely independent of species, the fate of the 3-phenoxybenzyl moiety, however, is species dependent, with marked differences either in the extent of aromatic hydroxylation or in the amino acid utilized for conjugation of the carboxylic acid.
Ester cleavage also takes place in plants. The phenoxybenzyl and cyclopropyl moieties are readily converted into glucoside conjugates.

Report: CA 5.1.1/1
[redacted] 2000a
Alphacypermethrin (CL 900049): Absorption, distribution, excretion, and pharmacokinetics of orally administered (benzyl-ring-14C)CL 900049 in the rat
AL-440-017
Guidelines: <none>
GLP: yes (It is stated in the report that the study was conducted in compliance with GLP Standard. Quality assurance statement is attached. GLP certificate of the competent aurhority is missing.)
Remark: This study is investigating oral absorption kinetics and bile excretion of alpha-cypermethrin. Furthermore the optical conversion of alpha-cypermethrin in vivo

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alphacypermethrin (BAS 310 I)

Description:	white-light yellow crystalline solid		
Batch/purity #:	Radiolabeled (^{14}C):	CP-2386	98%
	Radiolabeled (^{14}C):	CFQ11156	92%
	Nonlabeled:	351	purity not given
Stability of test compound:	To demonstrate the stability of the test item in the application formulation, HPLC analysis of the application formulation was performed.		

2. Vehicle and/or positive control: corn oil

3. Test animals:

Species:	Rat
Strain:	SD (Crj; CD) (Charles River Japan, Inc.)
Age:	7 weeks
Sex:	males and females
Number of animals:	61 males and 32 females
Weight at dosing:	males: 182.3 – 237.3 g Females: 142.3 – 175.9 g
Acclimation period:	at least 5 days
Diet:	diet for rodent (MF, Oriental Yeast), *ad libitum* Prior to administration rats were fasted about 16 hours before administration and fed at 4 hours after administration.
Water:	Tap water *ad libitum*
Housing:	During acclimatization four animals per cage (designed by DPC), then individually in restraining Bollman cage (designed by DPC)

Environmental conditions:

Temperature:	21 – 25°C
Humidity:	Animal experimental facility: 45 – 65% Experiment facility IV: 40 – 70%
Photoperiod:	Alternating 12-hour light and dark cycles

4. Preparation of dosing solution

Unpurified ^{14}C-alphacypermethrin (batch: CFQ11156) and unlabeled alphacypermethrin were weight out and dissolved in designated amount of corn oil to prepare 0.4 mg alphacypermethrin/mL dosing formulation.

The purified 14C-alphacypermethrin (batch: CP-2386) in ethyl acetate solution was collected, weighed unlabeled alphacypermethrin was added and ethyl acetate was removed by nitrogen gas. The residue was dissolved in the designated amount of corn oil to prepare 0.4, 4 and 7 mg alphacypermethrin/mL dosing formulation.

After preparation, the dosing formulations were stored at approximately 4°C in the dark until use.

In order to demonstrate the stability of the test item in the application formulations, confirm the identity and determine the isotope pattern, HPLC analysis was performed. For all dose groups, diluted application solutions were LSC (liquid scintillation counter) measured and the measured values were taken for calculation. The radiochemical purity of 14C-alphacypermethrin (batch CFQ11156) was as low as 92% before the first use. Therefore, the test substance was purified after administration in the two preliminary studies. The radiochemical purity was 92% after completion of the two preliminary studies. The radiochemical purity of 14C-alphacypermethrin (batch CP-2386) was 98% or more throughout the study period, indicating no decrease in the purity. The total radiochemical purity of cis 1 and cis2 was 98% or more.

Aliquots of the formulated test item were administered to the rats by gavage. The actual applied doses were 2, 20 and 35 mg/ 5 mL/kg bw.

B. STUDY DESIGN AND METHODS

1. Dates of work: November 16, 1999 – March 27, 2000

Two preliminary studies were conducted in order to determine the radioactivity concentrations in plasma, the excretion of radioactivity in urine, feces and expired air and residual radioactivity in carcass.

In four experiments of the definitive study the radioactivity concentrations in plasma and tissues, excretion of radioactivity in urine, feces and bile and the residual radioactivity in carcass was determined.

Each group consisted of four animals excluding preliminary study (one animal).

Determination of radioactivity concentrations in plasma:
After single oral administration of ^{14}C-alphacypermethrin to male and female rats at doses of 2 and 20 mg/mg bw, blood was withdrawn at the designated time from the tail vein using a heparinized micropipette into a microtube containing heparin. The sampling times were 30 minutes, 1, 2, 4, 6, 8, 10, 12, 24, 48, 72, 120 and 168 hours after administration. The radioactivity in the sample was measured in the LSC.

Determination of excretion of radioactivity in urine and feces, and residual radioactivity in carcass:
Male and female rats were housed in metabolic cages after single oral administration of ^{14}C-alphacypermetrin at doses of 2 and 35 mg/kg bw. Urine and feces were collected at the defined times and the excretion ratios of radioactivity in the urine and feces were determined. The sampling times were 0-4, 0-8, 0-24 hours and every 24 hours up to 168 hour for urine, and were every 24 hours up to 168 hours for feces. The expired air was not collected because the excretion of radioactivity in the expired air was not found in the preliminary study. The excretion of radioactivity in the urine and feces and the residual radioactivity in the carcass were determined by LSC. Urine samples up to 48 hours after administration were collected on an ice bath in the dark and stored at -20°C for investigation of quantitative analysis of isomers in urine except the samples of radioactivity. The feces recovered at every sampling time were brought to 300 mL with 50% methanol and homogenized and the radioactivity was counted in LSC. After the urine and feces collections at 168 hours after administration, the rat was sacrificed by ether overdose. The carcass was solubilized in 500 mL of 0.5 mol/L sodium hydroxide and 30 mL of toluene by heating with reflux. The radioactivity was counted in LSC.

Determination of excretion of radioactivity in bile:
Male rats were anesthetized with ether and their common bile ducts were cannulated with polyethylene tubes for bile collection. The animals were dosed orally with ^{14}C-alphacypermetrin at doses of 2 and 20 mg/kg bw after recovery from anesthesia. Bile, urine and feces were collected at the defined times, and excretion ratios of radioactivity in bile, urine and feces were determined. The sampling times were 0-2, 0-4, 0-6, 0-8, 0-24 and 0-48 hours after administration for bile. The sampling times of urine were 0-4, 0-8, 0-24 and 0-48 hours after administration. The sampling times of feces were 0-24 and 0-48 hours after administration. The final sampling time for the group 11 was limited to 24 hours after administration.
After dilution of the obtained bile, urine and feces the radioactivity was counted in the LSC. The gastro-intestinal tract and contents were collected at 24 hours or 48 hours after administration. The radioactivity of the carcass and the gastro-intestinal tract was determined after solubilization by LSC.

Determination of radioactivity concentrations in tissues:
After single oral administration of ^{14}C-alphacypermetrin at doses of 2 and 20 mg/kg bw male and female rats were housed in animal cages. The male rats (2 and 20 mg/kg bw) at 6, 24 and 168 hours after administration and the female rats (2 mg/kg bw) at 8, 24 and 168 hours after administration were sacrificed by exsanguination from the abdominal aorta under ether anesthesia using vacuum tubes containing heparin. The following tissues were excised: plasma, blood, cerebrum, cerebellum, pituitary gland, spinal cord, eyeball, Harderian gland, thyroid gland, trachea, mandibular gland, thymus, heart, lung, liver, kidney, adrenal gland, spleen, pancreas, fat (around the kidney), brown fat, skeletal muscle (from the femoral region), skin (from the axillary region), bone marrow, aorta, mesenteric lymph node, testis (male), epididymis (male), prostate gland (male), ovary (female), uterus (female), stomach, small intestine, large intestine and urinary bladder.
The radioactivity in each sample after preparation was counted in LSC in order to determine the concentrations and distribution of the radioactivity in the tissues.

Quantitative Analysis of Isomers in Urine:
The relative amounts of optical isomers *(cis 1, cis 2, trans* 3 and *trans* 4) in 0-48 hr urine collected from the 2 and 20 mg/kg male rat groups, and the 2 mg/kg female rat group were analyzed by LSC to determine the quantitative extraction ratio of radioactivity in an ethyl acetate and the water layer separately. The obtained layers were then investigated for the different optical isomers via HPLC-RAD in each sample. The collected urine, which was not analyzed soon, was stored at -20°C.

II. RESULTS AND DISCUSSION

Stability and homogeneity control analysis of the test substance preparation
The analytical investigations of 14C-Alphacypermethrin demonstrated the stability and homogeneity of the test substance in the preparation during the period of administration.

Radioactivity concentrations in plasma (see Table 5.1.1-3):
After oral administration of a 2 mg/kg bw dose to male rats, the radioactivity concentration in the plasma reached a maximum of 1268 ppb at 6 hours and then declined biphasically with a half-life of 3.8 and 21 hours. The AUC (0-∞) was 13398 ng eq. hr mL^{-1}, respectively. The C_{max} and AUC (0-∞) of the 20 mg/kg bw dose group were 5.1 and 8.5 times those in the 2 mg/kg bw dose group, respectively. The C_{max} and AUC (0-∞) both increased but the increase was smaller than the increase in the dose level. The half-lives were longer than those in the 2 mg/kg bw dose group.
After single oral administration of ^{14}C-alphacypermetrin to female rats at dose of 2 mg/kg bw, the radioactivity concentration in the plasma reached C_{max} of 1491 ppb at 7 hours, and then declined with a half-life of 3.9 hours from 8 to 24 hours and a half-life of 24 hours after 24 hours. The AUC(0-∞) was 18336 ng eq. hr mL^{-1}. The AUC(0-∞) was larger than in male rats at the same dose level but there were no large differences in the t_{max}, C_{max} and half-lives. The C_{max} and AUC(0-∞) of the 20 mg/kg bw dose group were 5.0 and 6.0 times those in the 2 mg/kg bw dose group, respectively. The C_{max} and AUC(0-∞) both increased but the increase was smaller than the increase in the dose level.

Table 5.1.1-3: Radioactivity concentration in plasma after single oral administration of ^{14}C-alphacypermethrin

	Radioactivity concentration (ppb)			
	2 mg/kg bw		20 mg/kg bw	
Time	male	female	male	female
30 min	48 ±16	53±13	363±30	520±72
1 h	103 ±23	98±13	904±480	1198±244
2 h	256 ±21	198±57	1951±592	2556±1032
4 h	723 ±100	664±121	4462±1715	6303±1235
6 h	1268 ±549	1390±180	6074±1456	7009±2067
8 h	1077 ±485	1442±323	5203±577	6755±1387
10 h	756 ±237	1136±236	5088±704	5631±1186
12 h	526 ±149	922±156	4817±798	4563±1251
24 h	60 ±49	97±19	1459±539	946±301
48 h	11 ±5	11±4	150±45	103±39
72 h	5 ±4	4±3	90±8	35±24
120 h	N.D.	N.D.	22±25	N.D.
168 h	N.D.	N.D.	N.D.	N.D.
Detection limit	4	4	36	35
t_{max} (h)	6±0	7±1	9±3	6±2
C_{max} (ppb)	1268±549	1491±248	6530±919	7528±1686
$t_{1/2}$ (h)	3.8±0.7 (8-24h) 21±10 (24-last)	3.9±0.2 (8-24h) 24±23 (24-last)	7.8±2.5 (10-24h) 31±14 (24-last)	5.6±0.8 (8-24h) 13±4 (24-last)
AUC (ng eq.*h*mL^{-1})				
(0-last) (0-∞)	13264±4590 13398±4619	18193±2388 18336±2450	111678±11026 114175±10528	108212±23587 109132±23719

Data are expressed as the mean values ± S.D. of four animals
N.D.: Not detected

At tmax the variability of radioactivity in the plasma in the low dose group differed by a factor of 2.6 and 1.5, and in the high dose by a factor of 1.2 and 1.7 in male and female rats, respectively, indicative for the range of inter-individual variability of alpha-cypermethrin toxicity in studies with bolus dosing.

Excretion of radioactivity in urine, feces, and residual radioactivity in carcass (see Table 5.1.1-4):
After single oral administration of ^{14}C-alphacypermetrin to male rats at a dose of 2 mg/kg bw, the excretion of radioactivity in the urine was 31.8%, 34.9% and 36.1% of the dose up to 24, 72 and 168 hours, respectively. The excretion of radioactivity in the feces was 55.6%, 61.6% and 62.5% of the dose up to 24, 72 and 168 hours after administration, respectively. The total excretion of radioactivity in the urine and feces was 98.5% of the dose up to 168 hours after administration. The residual radioactivity in the carcass was 1.3% of the dose at 168 hours after administration. In the 35 mg/kg bw dose group the total excretion of radioactivity in the urine and feces was 96.5% of the dose up to 168 hours after administration. The residual radioactivity in the carcass was 1.5% of the dose at 168 hours after administration. As compared with the results in the 2 mg/kg bw dose group, the excretion of radioactivity in the urine was lower by 7.5% of the dose and the excretion of radioactivity in the feces was higher by 5.4% of the dose.

Comparing these results with the female rats at the 2 mg/kg bw dose level, the excretion of radioactivity in the urine was higher by 12.0% of the dose and the excretion radioactivity in the feces was lower by 14.2% of the dose. Comparing the 2 mg/kg bw dose group with the 35 mg/kg bw dose group of the female rats, the excretion of radioactivity in the urine was lower by 13.8% of the dose and the excretion of radioactivity in the feces was higher by 11.9% of the dose.

Table 5.1.1-4: Cumulative excretion of radioactivity in urine and feces

	Excretion of radioactivity (% of dose) – Dose: **2 mg/kg bw**					
	Male			Female		
Time (h)	Urine	Feces	Total	Urine	Feces	Total
0-4	2.0 ± 1.1	-	-	2.2±1.4	-	-
0-8	11.2 ± 2.2	-	-	10.0±6.0	-	-
0-24	31.8 ± 7.0	55.6 ± 6.8	87.4 ± 3.8	39.3±8.3	39.2±7.4	78.6±3.7
0-48	34.2 ± 6.4	60.6 ± 8.6	94.7 ± 4.1	46.1±7.1	46.4±7.6	92.4±2.5
0-72	34.9 ± 6.4	61.6 ± 8.6	96.4 ± 3.7	47.0±7.2	47.5±8.0	94.5±1.9
0-96	35.3 ± 6.4	61.9 ± 8.6	97.3 ± 3.4	47.4±7.3	47.8±7.9	95.2±1.6
0-120	35.6 ± 6.5	62.1 ± 8.5	97.7 ± 3.3	47.7±7.4	48.0±7.9	95.7±1.6
0-144	35.9 ± 6.5	62.2 ± 8.5	98.1 ± 3.1	47.9±7.4	48.2±7.9	96.0±1.5
0-168	36.1 ± 6.5	62.5 ± 8.5	98.5 ± 2.9	48.1±7.4	48.3±7.8	96.3±1.5
Carcass (168h)			1.3 ± 0.5			1.0±0.2
	Excretion of radioactivity (% of dose) – Dose: **35 mg/kg bw**					
0-4	1.2±0.3	-	-	1.0±0.2	-	-
0-8	3.3±1.5	-	-	2.3±0.9	-	-
0-24	15.9±5.2	38.6±29.2	54.5±25.1	15.1±4.4	32.0±14.8	47.0±18.9
0-48	26.4±11.3	65.7±11.2	92.1±1.3	29.7±3.1	54.7±4.8	84.4±3.8
0-72	27.5±11.3	66.9±10.7	64.4±1.9	32.8±3.3	58.7±5.1	91.5±3.2
0-96	27.9±11.3	67.2±10.7	95.1±2.0	33.4±3.4	59.4±5.4	92.8±3.0
0-120	28.2±11.3	67.5±10.7	95.6±2.1	33.7±3.4	59.7±5.5	93.4±2.8
0-144	28.4±11.3	67.6±10.7	96.0±2.2	34.0±3.4	59.8±5.4	93.8±2.7
0-168	**28.6±11.4**	67.9±10.8	96.5±2.2	**34.3±3.4**	60.2±5.4	94.4±2.7
Carcass (168h)			**1.5±0.6**			**3.1±1.6**

Data are expressed as the mean values ± S.D. of four animals
-: Not determined

The single animal data after oral administration of alpha-cypermethrin at a dose of 2 mg/kg bw shows significant variability: after 168 hours the excretion via urine ranged in the low dose males between 29.4 % and 44% of the administered dose, whereas in females the excretion ranged from 41.8% up to 57%. The amount considered bioavailable (excretion via urine plus carcass) range between 30.3-45.4% of the dose administered in males and 42.7 – 58.2% of the dose in females [see Table 5.1.1-5].

Table 5.1.1-5: Individual bioavailability of alpha-cypermethrin in the low dose group based on excretion via urine and amount in carcass [amount of radioactivity in % of administered dose]

Dose	**Low dose males (2 mg/kg)**				**Low dose females (2 mg/kg)**			
Animal	**No.19**	**No.20**	**No.21**	**No.22**	**No.23**	**No.24**	**No.25**	**No.26**
Urine 0-168h	29.4	32.4	38.4	44.0	57.0	51.4	42.1	41.8
Carcass (168 h)	0.9	0.8	1.9	1.4	1.2	0.9	0.9	0.9
Sum	30.3	33.2	40.3	45.4	58.2	52.3	43.0	42.7
Mean	37.3 ± 6.84				49.05 ± 7.55			

Excretion of radioactivity in bile of male rats (see Table 5.1.1-6):
After single oral administration of ^{14}C-alphacypermetrin to male rats at a dose of 2 mg/kg bw, the excretion of radioactivity in the bile was 1.6%. 2.6% and 3.0% of the dose up to 8, 24 and 48 hours, respectively. The excretion of radioactivity in the urine was 1.7%, 3.0% and 3.4% of the dose up to 8, 24 and 48 hours after administration, respectively. The excretion of radioactivity in the feces was 65.0% and 71.5% of the dose up to 24 and 48 hours after administration, respectively. At 48 hours after administration, the ratios of radioactivity in the gastro-intestinal tract and contents were 0.4% and 3.2% of the dose, respectively. The residual radioactivity in the carcass after removing the gastro-intestinal contents was 15.3% of the dose. Looking at the 20 mg/kg bw dose group with male rats, the excretion of radioactivity in the bile and urine was comparable with that in the 2 mg/kg bw dose group. The excretion of radioactivity in the feces was 46.8% and 56.3% of the dose up to 24 and 48 hours after administration, respectively. At 48 hours after administration, the ratios of radioactivity in the gastro-intestinal tract and contents were 0.6% and 12.4% of the dose, respectively. The residual radioactivity in the carcass after removing the gastro-intestinal contents was 21.7% of the dose.
After single oral administration of ^{14}C-alphacypermetrin to male rats at a dose of 2 mg/kg bw, the excretion of radioactivity in the bile and urine collected up to the final sampling time of 24 hours was comparable with that in the group determined up to 48 hours after dosing. The excretion of radioactivity in the feces was 72.3% of the dose up to 24 hours after administration. At 24 hours after administration, the ratios of radioactivity in the gastro-intestinal tract and contents were 0.5% and 9.9% of the dose, respectively. The residual radioactivity in the carcass after removing the gastro-intestinal contents was 8.8% of the dose.

Table 5.1.1-6: Cumulative excretion of radioactivity in bile. urine and feces in male rats

	Excretion of radioactivity (% of dose) **- 48 hours**					
	2 mg/kg bw			20 mg/kg bw		
Time (h)	Bile	Urine	Feces	Bile	Urine	Feces
0-2	0.2±0.1	-	-	0.2±0.1	-	-
0-4	0.8±0.4	0.5±0.4	-	0.6±0.2	0.4±0.2	-
0-6	1.2±0.5	-	-	1.0±0.3	-	-
0-8	1.6±0.6	1.7±0.7	-	1.3±0.4	1.2±0.3	-
0-24	2.6±0.7	3.0±0.8	65.0±9.1	2.8±1.0	2.9±0.1	46.8±9.2
0-48	3.0±0.7	3.4±0.8	71.5±9.9	3.6±1.2	3.7±0.6	56.3±8.5
Gastro-intestinal contents (48h)			3.2±2.4			12.4±8.0
Gastro-intestinal tract (48h)			0.4±0.5			0.6±0.5
Carcass (48h)			15.3±9.1			21.7±4.9
	Excretion of radioactivity (% of dose) **– 24 hours**					
0-2	0.1±0.1	-	-			
0-4	0.6±0.4	0.5±0.5	-			
0-6	1.0±0.5	-	-			
0-8	1.5±0.6	1.7±0.7	-			
0-24	2.5±0.9	3.6±1.0	72.3±14.6			
Gastro-intestinal contents (24h)			9.9±11.3			
Gastro-intestinal tract (24h)			0.5±0.4			
Carcass (24h)			8.8±4.2			

Data are expressed as the mean values ± S.D. of four animals
-: Not determined

Bile excretion accounts for around 3% within the first 24 hours of excretion.

Radioactivity concentration in tissue (see Table 5.1.1-7):
At 6 hours after administration of 2 mg/kg bw test substance to male rats, the radioactivity concentration in the mesenteric lymph node was the highest, being 1.78 times that in the plasma (1255 ppb). The radioactivity concentrations in the other tissues were comparable with or lower than that in the plasma. indicating low tissue distribution at an early time after administration. The radioactivity concentrations in the eyeball, cerebellum, spinal cord and cerebrum were 4-3% of that in the plasma, indicating that the distribution is especially low in these tissues. At 24 hours after administration, the radioactivity concentrations in the brown fat and fat showing a maximum concentration were the highest, being 9.85 and 8.89 times that in the plasma (81 ppb), respectively. The radioactivity concentrations in the tissues except the fat decreased. At 168 hours after administration, the radioactivity concentration in the fat was the highest, being 157 ppb. The radioactivity concentration in the brown fat (27 ppb), mesenteric lymph node (23 ppb), skin (12 ppb) and epididymis (10 ppb) were next high in that order. The radioactivity concentration in the fat was relatively high but decreased to 22% of the 24-hour value, suggesting that the elimination of radioactivity was slightly slower than that from the other tissues. The results from the 20 mg/kg bw dose group showed similar results.
The radioactivity concentration in the tissues in female rats showed a similar tendency to those in male rats as the radioactivity concentration in the mesenteric lymph node was the highest at an early time and the radioactivity concentration in the fat was high at 168 hours after administration. These results suggest no definite sex difference.

Quantitative Analysis of Isomers in Urine:
As the result of quantification of the isomers in the urine after administration of a 20 mg/kg bw dose, a radioactivity peak was observed only at the retention time of cis 2, suggesting that the optical conversion did not occur or occurred a little in vivo. Dosing with 2 mg/kg bw no radioactivity peaks were observed at retention times of isomers.

III. CONCLUSION

Absorption: After oral administration the radioactivity concentration in the plasma reached a maximum at 6-9 hours after administration and then declined biphasically with a half-time of 3.8 and 21 hours. After single oral application in rat between 34% and 46% of the dose is excreted within 48 hours via urine in male and female rats. After 168 hours 50 % is considered an adequate value for absorbed dose based on urinary excretion (29.4-57%), bile excretion (around 3%) and residual carcass radioactivity (around 1%).

Excretion: More than 92% of the radioactivity is excreted within 48 hours of administration, equally partitioned amongst feces and urine in females (both 46%) and slightly less in urine in males (34.2% via urine and 60% via feces). Excretion via bile is considered to be limited to around 3% in the first 24 hours. No radioactivity was excreted in the expired air. The new data are considered to confirm and supplement the originally presented data in the DAR.

Distribution: The highest radioactivity concentrations are found in well perfused organs (mesenteric lymphnode, kidney and liver) as well as in fat with the slowest elimination rate compared to the other tissues.

Quantification of isomers in urine: In the high dose group a radioactivity peak was observed only at the retention time of cis 2, therefore it is assumed that optical conversion did not occur or occurred only a little in vivo.

Table 5.1.1-7: Radioactivity concentration in tissue in male rats

	Radioactivity concentration (ppb)					
	2 mg/kg bw			20 mg/kg bw		
Tissue	6 h	24 h	168 h	6 h	24 h	168 h
Plasma	1255±365 (1.00)	81±40 (1.00)	N.D.	5363±496 (1.00)	1159±1038 (1.00)	N.D.
Blood	754±206 (0.6)	49±25 (0.60)	N.D.	2936±326 (0.55)	649±575 (0.56)	N.D.
Cerebrum	43±11 (0.03)	3±1 (0.04)	N.D.	217±8 (0.04)	31±33 (0.03)	N.D.
Cerebellum	45±12 (0.04)	4±1 (0.05)	N.D.	243±31 (0.05)	41±32 (0.04)	N.D.
Pituitary gland	273±136 (0.22)	N.D.	N.D.	1233±293 (0.23)	N.D.	N.D.
Spinal cord	44±8 (0.04)	7±2 (0.09)	N.D.	206±24 (0.04)	45±27 (0.04)	N.D.
Eyeball	46±8 (0.04)	6±2 (0.07)	N.D.	210±25 (0.04)	65±50 (0.06)	N.D.
Harderian gland	272±79 (0.22)	105±25 (1.30)	1±1	1472±109 (0.27)	694±354 (0.60)	N.D.
Thyroid gland	318±141 (0.25)	72±51 (0.89)	N.D.	1423±224 (0.27)	659±578 (0.57)	N.D.
Trachea	173±36 (0.14)	48±13 (0.59)	N.D.	1086±127 (0.20)	449±205 (0.39)	N.D.
Mandibular gl.	218±72 (0.17)	14±3 (0.17)	1±1	912±55 (0.17)	174±114 (0.15)	N.D.
Thymus	93±21 (0.07)	17±6 (0.21)	1±2	467±93 (0.09)	166±138 (0.14)	N.D.
Heart	346±77 (0.28)	14±5 (0.17)	N.D.	1395±398 (0.26)	196±158 (0.17)	N.D.
Lung	313±93 (0.25)	26±7 (0.32)	2±1	1486±258 (0.28)	315±252 (0.27)	N.D.
Liver	**1172±260 (0.93)**	232±74 (2.86)	9±1	5006±869 (0.93)	1158±715 (1.00)	88±18
Kidney	**845±208 (0.67)**	107±32 (1.32)	3±1	3058±108 (0.57)	1103±604 (0.95)	40±18
Adrenal gland	709±211 (0.56)	58±36 (0.72)	4±5	2441±412 (0.46)	520±417 (0.45)	N.D.
Spleen	246±137 (0.20)	10±3 (0.12)	N.D.	755±206 (0.14)	113±89 (0.10)	N.D.
Pancreas	240±64 (0.19)	36±17 (0.44)	3±1	982±74 (0.18)	280±200 (0.24)	28±10
Fat	**393±50 (0.31)**	**720±118 (8.89)**	**157±51**	**2748±626 (0.51)**	**3382±616 (2.92)**	**1490±584**
Brown fat	**1228±233 (0.98)**	**798±220 (9.85)**	**27±3**	**5873±1347 (1.10)**	**5273±2037 (4.55)**	**234±104**
Skeletal muscle	115±26 (0.09)	8±4 (0.10)	N.D.	508±46 (0.09)	100±86 (0.09)	N.D.
Skin	246±67 (0.20)	170±31 (2.1)	12±4	1324±138 (0.25)	1146±434 (0.99)	105±61
Bone marrow	227±81 (0.18)	15±5 (0.19)	N.D.	945±200 (0.18)	226±157 (0.19)	N.D.
Aorta	246±60 (0.20)	53±21 (0.65)	5±6	1423±426 (0.27)	403±252 (0.35)	N.D.
Mesenteric lymph node	2237±683 (1.78)	259±99 (3.20)	23±12	13373±4861 (2.49)	2021±1342 (1.74)	226±134
Testis	128±36 (0.10)	15±4 (0.19)	N.D.	620±107 (0.19)	173±134 (0.15)	N.D.
Epididymis	188±52 (0.15)	64±22 (0.79)	10±7	1007±73 (0.19)	431±91 (0.37)	121±108
Prostate gland	137±47 (0.11)	44±14 (0.54)	N.D.	806±210 (0.15)	495±232 (0.43)	14±16
Stomach	219±57 (0.17)	59±35 (0.73)	3±2	1259±311 (0.23)	505±265 (0.44)	10±11
Small intestine	374±74 (0.30)	27±6 (0.33)	2±2	1662±465 (0.31)	381±271 (0.33)	14±10
Large intestine	193±47 (0.15)	84±42 (1.04)	4±3	1226±153 (0.23)	657±424 (0.57)	69±45
Urinary bladder	1244±550 (0.99)	211±139 (2.60)	N.D.	6198±1963 (1.16)	2436±3033 (2.10)	67±112

Data are expressed as the mean values ± S.D. of four animals
Figures in parentheses are expressed as the ratio of concentration in tissue relative to plasma.
N.D.: Not detected

Report: CA 5.1.1/2
[redacted] 2013a
Metabolism of 14C-alpha-Cypermethrin (BAS 310 I) after oral administration in rats
2013/1086630
Guidelines: EPA 870.7485, EPA 860.1000: EPA Residue Chemistry Test Guidelines, MAFF Testing Guidelines for Toxicology Studies: Metabolism Animals (Japan), OECD 417, EEC 87/302 B
GLP: yes
(certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: Alpha-cypermethrin (BAS 310 I)
Lot/Batch #: 775-0401 (benzyl-U-^{14}C)
1025-1034 (phenoxy-1,2,3,4,5,6-^{13}C)
986-1046 (cyclopropane-1-^{14}C)
990-1024 (cyclopropane-1-^{13}C)
L80-24 (unlabeled)
Purity: 96.1% (benzyl-U-^{14}C)
95.1% (phenoxy-1,2,3,4,5,6-^{13}C)
98.2% (cyclopropane-1-^{14}C)
97.7% (cyclopropane-1-^{13}C)
99.4% (unlabeled)
CAS#: 67375-30-8
Development code: 4078193
Stability of test compound: Stable during dosing period

2. Vehicle and/or positive control: The vehicle used was corn oil

3. Test animals

Species: Rat
Strain: Wistar (Crl:WI(Han), Charles River Laboratories, Germany)
Age: 7 weeks (arrival) / 8-9 weeks (administration)
Sex: Male and female
Number of animals: 40 (10 males + 10 females for each label)
Weight at dosing: 154-293 g
Acclimation period: 7-14 days
Diet: Kliba lab diet for mouse and rat, *ad libitum*
Water: Tap water *ad libitum*

Husbandry:	In accordance with the German Animal Welfare Act
Housing:	During acclimatization in groups in Macrolon Cages, then individually in Plexiglas metabolism cages
Environmental conditions:	
Temperature:	19-23°C
Humidity:	45-60%
Air changes:	>30 changes/h
Photoperiod:	Alternating 12-hour light and dark cycles

4. Preparation of dosing solutions

For oral administration of the test item, mixtures of ^{14}C labeled, ^{13}C labeled and unlabeled BAS 310 I were weighed in (in ratios of approximately 1:1:1) and suspended in corn oil. Aliquots of the resulting application formulations were subjected to MS analysis for identity check and for determination of the isotope pattern.

The test items used during the present metabolism study were ^{14}C-labeled either in the benzyl ring (benzyl label) or in the cyclopropane moiety (cyclopropane label). The benzyl-U-^{14}C-radiolabeled test item was mixed with phenoxy-1,2,3,4,5,6-^{13}C-BAS 310 I and unlabeled ^{12}C-BAS 310 I, while the cyclopropane-1-^{14}C radiolabeled test item was mixed with cyclopropane-1-^{13}C-BAS 310 I and unlabeled BAS 310 I prior to application in order to facilitate analysis by radio-HPLC and mass spectrometry.

B. STUDY DESIGN AND METHODS

1. Dates of work: October 24, 2011 – September 5, 2014

The treated rats consisted of an oral single dose group (10 rats/sex/label, about 20 mg/kg bw). All animals received the oral dose administered via gavage.

Six hours after administration of the test item, blood samples were taken and centrifuged to separate plasma from blood cells. In the case of female dose group (cyclopropane label), no sufficient blood samples could be obtained.

Urine of all dose groups was collected after 6, 12, 24, 48, 72, 96, 120, 144, 168 and 192 hours (female dose group, benzyl label: 190 h). Feces were collected in intervals of 24 hours up to eight days. After sampling, urine and feces were combined per sampling interval (several animals per dose group) and stored at -18°C or below.

In the case of male dose group (cyclopropane label), two animals had to be sacrificed prior to the first sampling. The other animals were killed 192 hours after administration (female dose group, benzyl label: 190 h). Blood was collected from the sacrificed animals and centrifuged to separate plasma from blood cells. Combined plasma samples (several animals per dose group) as well as combined samples of the cage wash and of liver, kidney and carcass were stored in a freezer at -18°C or below.

Samples were analyzed for total ^{14}C-radioactivity by samples combustion and/or liquid scintillation counting (LSC). Metabolite identification was performed by mass spectrometry and quantitative analysis using HPLC.
The radioactive residues were determined in urine and feces in time intervals of 24 hours for a total of eight days after administration (for urine samples additionally after 6 h and 12 h), as well as in cage wash. Subsamples of homogenized feces were extracted with acetonitrile and water. Urine samples of all time intervals and extracts from feces collected in the time intervals of 0-24 h and 24-48 h were analyzed by HPLC with radio detection.

II. RESULTS AND DISCUSSION

Storage stability
The analytical investigations performed within the present study demonstrated the specific radioactivity, purity, stability and homogeneity of ^{14}C-BAS 310 I in the vehicle for the performed experiments.

Balance and Absorption
The mean total recovery of radioactivity was 104.9% and 93.5% (benzyl label) or 87.6% and 91.0% (cyclopropane label) of the administered dose for male and female rats, respectively.
Approximately half of the administered dose was found as unchanged parent compound in feces extracts for each gender and both labels. Thus, alpha-cypermethrin was either only partly absorbed or only partly metabolized in the rat (sum of biotransformation products in urine and feces: approximately 31-39% of the dose). The unchanged parent molecule BAS 310 I was only detected in feces samples (portions of 39-57% of the dose).

Distribution
No tissues were examined during this study.
The cage wash contained 2.8% and 2.7% (benzyl label) or 2.4% and 2.3% (cyclopropane label) of the radioactivity administered for male and female rats, respectively.

Excretion
Excretion via urine and feces was nearly complete within 72-96 h after administration for both dose groups and both labels, with higher portions of radioactive residues in feces. Within the investigation period of eight days, total excretion of radioactive residues via feces amounted to 58-77% of the dose. Portions of approximately 25% of the administered dose were excreted via urine within eight days for each gender and both radiolabels. In the cage wash, portions of 2-3% of the dose were recovered. No significant differences in the excretion pattern related to gender or label were observed.

Table 5.1.1-8: Excretion of radioactivity via urine and feces after single oral administration of ^{14}C-BAS 310 I to rats at a dose level of nominally 20 mg/kg bw (group mean values)

Matrix	[% of the administered radioactivity]			
	Benzyl label		Cyclopropane label	
	Male	Female	Male	Female
Urine (h)				
0-6	2.447	2.085	2.728	1.868
6-12	3.548	4.433	2.867	5.188
12-24	9.320	8.643	6.667	6.256
24-48	6.128	4.885	8.470	6.015
48-72	1.544	1.659	2.623	1.732
72-96	0.895	1.704	1.093	0.643
96-120	0.486	0.662	0.567	0.384
120-144	0.251	0.548	0.636	0.296
144-168	0.210	0.386	1.036	0.398
168-192[1]	0.269	0.244	0.085	0.237
Subtotal Urine (0-192)	**25.099**	**25.248**	**26.770**	**23.016**
Feces (h)				
0-24	27.765	22.835	23.521[3]	32.912
24-48	44.328	36.240	30.788	27.664
48-72	3.232	3.830	2.803	3.533
72-96	0.772	2.015	0.608	0.349
96-120	0.581	0.175	0.312	1.123
120-144	0.106	0.135	0.178	0.068
144-168	0.079	0.100	0.053	0.056
168-192[1]	0.075	0.073	0.105	0.045
Subtotal feces (0-48)	**72.093**	**59.075**	**54.309[3]**	**60.576**
Subtotal Feces (0-192[1])	**76.939**	**65.404**	**58.369[3]**	**65.749**
Other sources				
Cage wash (after 8-9 days)	2.773	2.711	2.437	2.254
Plasma (6 h)	0.042	0.088	0.014	no sample
Plasma (192 h[1])	0.001	0.002	0.001	0.001
Subtotal plasma (6 and 192 h[1])	0.044	0.089	0.015	n.r.
Liver	not measured	not measured	not measured	not measured
Kidney	not measured	not measured	not measured	not measured
Carcass	not measured	not measured	not measured	not measured
Total (0-192 h[1])	**104.855**	**93.451**	**87.591[2,3]**	**91.020**

1 168 – 190 h or 0 – 190 h or 6 h and 190 h, respectively, in the case of female dose group (benzyl label)

2 The values for this dose group represent portions compared to the test item administered to the eight animals from which the samples were obtained (animals M04 and M10 of dose group DXM, cyclopropane label, had to be sacrificed prior to the first sampling)

3 The % dose value for feces of male dose group, cyclopropane label, time interval of 0 to 24 h, and in consequence the subtotal feces and the total residues, are probably underestimated since the weight of the homogenized workup sample was significantly lower compared to the sample weight documented for the respective storage sample, and no unusually high loss was apparent during homogenization, suggesting that the actual sample weight of the storage sample was lower and the % dose value was actually higher, correspondingly

n.r. Not reported

Extractability

Extractability of radioactive residues of BAS 310 I in feces of male and female rats was high to very high, with 77.5-96.9% of the total radioactive residues extracted. The main portions of radioactive residues were extracted with acetonitrile (46.5-95.0% TRR), and lower portions were subsequently extracted with water (1.9-31.1% TRR). No significant differences in the extraction efficiency related to gender or label were observed.

Table 5.1.1-9: Extractability of feces samples with solvents (acetonitrile, water) after dosing of rats with [benzyl-U-^{14}C]- or [cyclopropane-1-^{14}C]-alpha-cypermethrin

Matrix	TRR (calculated) mg/kg	Solvent extract (ERR)		RRR	
		mg/kg	% TRR	mg/kg	% TRR
Benzyl-U-^{14}C label					
Male rats (0-24 h)	302.95	290.55	95.9	12.40	4.1
Male rats (24-48 h)	277.11	252.12	91.0	25.00	9.0
Female rats (0-24 h)	351.00	338.79	95.5	12.21	3.5
Female rats (24-48 h)	233.84	219.32	93.8	14.52	6.2
Female rats (48-72 h)	22.65	19.97	88.2	2.68	11.8
Female rats (72-96 h)	11.05	10.21	92.3	0.85	7.7
Female rats (96-120 h)	1.14	0.94	81.8	0.21	18.2
Female rats (120-144 h)	0.85	0.66	77.5	0.19	22.5
Female rats (144-168 h)	n.r.	n.r.	n.r.	n.r.	n.r.
Female rats (168-190 h)	n.r.	n.r.	n.r.	n.r.	n.r.
Cyclopropane-1-^{14}C label					
Male rats (0-24 h)	146.00	141.40	96.8	4.61	3.2
Male rats (24-48 h)	163.24	150.67	92.3	12.57	7.7
Female rats (0-24 h)	361.72	350.62	96.9	11.10	3.1
Female rats (24-48 h)	180.19	170.82	94.8	9.37	5.2

TRR: Total radioactive residue (sum of ERR + RRR)
ERR Extractable radioactive residue (solvents: acetonitrile, isohexane, water)
RRR: Residual radioactive residue after solvent extraction (solvents: acetonitrile, water)
n.r. Not reported

Metabolism

Transformation mainly proceeds via cleavage of the ester, loss of the nitrile group and oxidation to form two carboxylic acid cleavage products, metabolites M310I001 and M310I011. These cleavage products and their derivatives are only detectable with the respective radiolabel. In urine, only label-specific cleavage products were detected which is probably due to the high molecular mass of the uncleaved molecule. Metabolite M310I001 can be conjugated with glucuronic acid to form the main component detected in urine (cyclopropane label), metabolite M310I004 (approximately 17% of the dose in sum of the time intervals for each gender). Hydroxylation of metabolite M310I001 at one of the methyl groups at the cyclopropane ring and conjugation with glucuronic acid produce the metabolites M310I003 and M310I002 found in urine at minor concentrations. Metabolite M310I011 can be hydroxylated to form metabolite M310I013 and then conjugated to the sulphate metabolite M310I014, the main component detected in urine (benzyl label, 16-17% of the dose). Conjugation of the carboxyl group of metabolite M310I011 and M310I013 with glycine forms the metabolites M310I010 and M310I012, respectively, which were only detected in urine at minor concentrations. The metabolites M310I001, M310I003, M310I011 and M310I013 were detected in urine and in feces extracts.

A further metabolic reaction is hydroxylation of the phenoxy ring of the molecule, forming metabolite M310I017, the main conversion product detected in feces extracts (1.3-2.5% of the dose in sum of the period of 0-48 h). Additional hydroxylation of one of the methyl groups at the cyclopropane ring produces metabolite M310I015. Cleavage of the ether bridge (loss of the phenyl ring) forms metabolite M310I016. Metabolites M310I017, M310I015 and M310I016 still contain each of the radiolabels. The label-specific metabolites M310I001, M310I003 and M310I013 may also be formed by analogous cleavage of the ester bond in metabolite M310I017 and M310I015. Up to 19 further peaks (per sample) in the chromatograms were characterized by their chromatographic properties (each below 1% dose for the analyzed time periods), including two assigned peaks in feces extracts for which molecular masses were determined.

Table 5.1.1-10: Summary of metabolites identified in urine and feces

Designation	Females			Males		
	Urine (0-192 h) [% dose]	**Feces (0-48 h) [% dose]**	**Sum [% dose]**	**Urine (0-192 h) [% dose]**	**Feces (0-48 h) [% dose]**	**Sum [% dose]**
Benzyl label						
BAS 310 I	n.r.	48.634	**48.634**	n.r.	56.998	**56.998**
M310I010	0.432	n.r.	**0.432**	0.983	n.r.	**0.983**
M310I011	2.658	0.341	**2.999**	3.722	0.990	**4.712**
M310I012	0.322	n.r.	**0.322**	0.379	n.r.	**0.379**
M310I013	2.786	1.183	**3.969**	0.850	1.183	**2.033**
M310I014	16.723	n.r.	**16.723**	16.080	n.r.	**16.080**
M310I015	n.r.	1.071	**1.071**	n.r.	1.098	**1.098**
M310I016	n.r.	0.470	**0.470**	n.r.	0.648	**0.648**
M310I017	n.r.	1.691	**1.691**	n.r.	2.409	**2.409**
M=404 u	n.r.	0.420	**0.420**	n.r.	0.267	**0.267**
M=461 u	n.r.	0.542	**0.542**	n.r.	0.643	**0.643**
Total identified	**22.922**	**53.390**	**76.312**	**22.015**	**63.326**	**85.341**
Sum conversion products	**25.248**	**7.933**	**33.181**	**25.099**	**9.960**	**35.059**

Table 5.1.1-10: Summary of metabolites identified in urine and feces

Designation	Females			Males		
	Urine (0-192 h) [% dose]	Feces (0-48 h) [% dose]	Sum [% dose]	Urine (0-192 h) [% dose]	Feces (0-48 h) [% dose]	Sum [% dose]
Cyclopropane label						
BAS 310 I	n.r.	50.603	**50.603**	n.r.	39.198	**39.198**
M310I001	4.526	1.234	**5.760**	5.482	2.043	**7.525**
M310I002	0.305	n.r.	**0.305**	0.565	n.r.	**0.565**
M310I003	0.990	n.r.	**0.990**	1.245	n.r.[3]	**1.245**
M310I004	16.528	n.r.	**16.528**	17.338	n.r.	**17.338**
M310I015	n.r.	0.606	**0.606**	n.r.	0.847	**0.847**
M310I016	n.r.	0.360	**0.360**	n.r.	0.498	**0.498**
M310I017	n.r.	1.328	**1.328**	n.r.	2.468	**2.468**
M=404 u	n.r.	n.r.	**n.d.**	n.r.	0.245	**0.245**
M=461 u	n.r.	0.547	**0.547**	n.r.	0.606	**0.606**
Total identified	**22.349**	**54.130**	**76.479**	**24.630**	**45.053**	**69.683**
Sum conversion products	**23.016**	**7.525**	**30.541**	**26.770**	**11.998**	**38.768**

1 Component characterized by HPLC-MS with a nominal mass of 404 u: a molecular formula of $C_{21}H_{18}Cl_2O_4$ was proposed according to the measured accurate mass
2 Component characterized by HPLC-MS with a nominal mass of 461 u: a molecular formula of $C_{23}H_{21}Cl_2NO_5$ was proposed according to the measured accurate mass
3 Only detected in the metabolite fraction used for identification and not quantified in feces extracts
n.r. Not reported

Table 5.1.1-11: Structures of metabolites identified in urine and feces

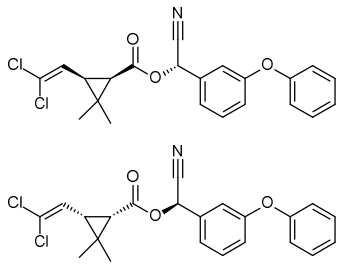
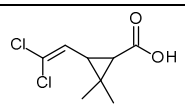
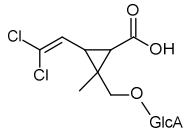
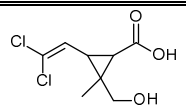
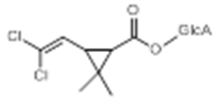
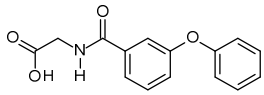
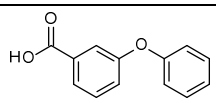
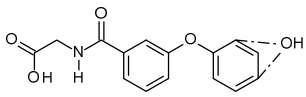
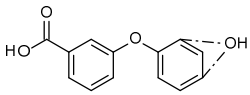
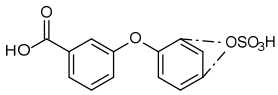
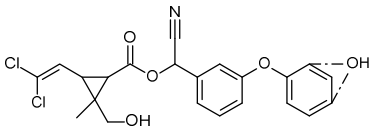
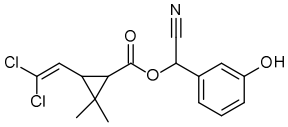
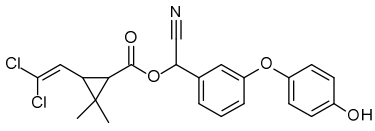
Metabolite designation				Structure/Name
BASF code/ Synonym (mol. weight)	Reg. No	Metabolite code	CAS-No	
BAS 310 I (415)	**4078193**	**-**	**67375-30-8**	
M310I001 DCVA (208)	**4080830**	**M310I001**	**n.r.**	
M310I002 (400)	**-**	**M310I002**	**n.r.**	

Table 5.1.1-11: Structures of metabolites identified in urine and feces

Metabolite designation				Structure/Name
BASF code/ Synonym (mol. weight)	Reg. No	Metabolite code	CAS-No	
M310I003 (224)	-	M310I003	n.r.	
M310I004 (384)	-	M310I004	n.r.	
M310I010 (271)	4108084	M310I010	n.r.	
M310I011 3PBA (214)	130213	M310I011	n.r.	
M310I012 (287)	-	M310I012	n.r.	
M310I013 (230)	-	M310I013	n.r.	
M310I014 (310)	-	M310I014	n.r.	
M310I015 (447)	-	M310I015	n.r.	
M310I016 (339)	6004475	M310I016	n.r.	
M310I017 (431)	6002320	M310I017	n.r.	

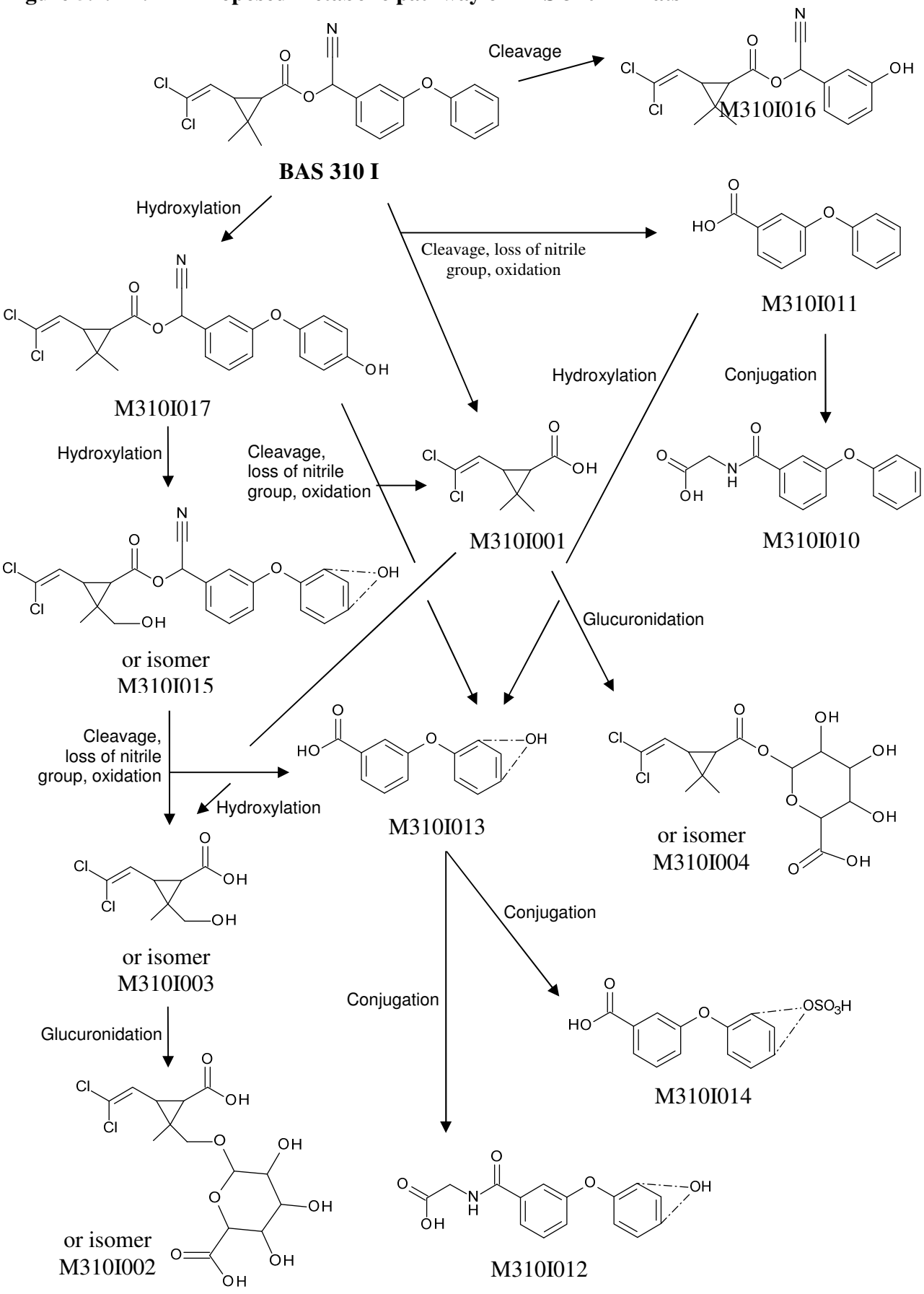
n.r. Not reported

Enantiomer-specific HPLC analysis (chiral column) of the parent fraction isolated from an exemplary acetonitrile extract of feces from male rats (cyclopropane label, 24-48 h) revealed no noticeable change in the ratio of the two enantiomers of alpha-cypermethrin compared to the administered racemic mixture and thus no indication that one enantiomer of alpha-cypermethrin was preferably metabolized in rats.

III. CONCLUSION

Excretion via urine and feces was nearly complete within 72-96 h after administration for both dose groups and both labels, with higher portions of radioactive residues in feces. In the cage wash, portions of 2-3% of the dose were recovered. No significant differences in the excretion pattern related to gender or label were observed.
In comparison, urine and feces extracts from male and female rats showed qualitatively and quantitatively similar metabolite patterns. The experiments with the two radiolabels indicate that only cleavage products of BAS 310 I are excreted via urine. Hydroxylation and / or conjugation (with sulphate or glycine in the case of the benzyl label, with glucuronic acid in the case of the cyclopropane label) occur with both parts of the molecule. The quantitative distribution of the uncleaved components in feces (hydroxylated metabolites and derivative with the phenyl ring lost) was very similar for both labels.

Figure 5.1.1-1: Proposed metabolic pathway of BAS 310 I in rats



CA 5.1.2 CA 5.1.2 Absorption. distribution. metabolism and excretion by other routes

According to the new data requirements for active ingredients of plant protection products as set out in Commission Regulation (EU) No. 283/2013 (1 March 2013, OJ L93, 1ff, 3.4.2013), "comparative *in vitro* metabolism studies shall be performed on animal species ... and on human material ...in order to determine the relevance of the toxicological animal data and to guide in the interpretation of findings and in further definition of the testing strategy..." (Section 5, Toxicological and metabolism studies, point 5.1.1., page 22). In the absence of validated test method or guidance documents, this data requirement is waived in accordance to SANCO Guidance Document SANCO/10181/2013-rev 2.1 (13 May 2013).

This entry is taken from public literature and was not included in the study list of the application.

Report: CA 5.1.2/1
Takaku T. et al., 2011a
In vitro metabolism of trans-Permethrin and its major metabolites, PBalc and PBacid, in humans
2011/1297011
Guidelines: none
GLP: no
(certified by none)

Executive Summary

In the present study, to compare the metabolic profiles (route and rate) between rats and humans, the in vitro metabolism of trans-permethrin by hepatic microsomes from rats and humans was investigated. Trans-[Phenoxyphenyl-^{14}C]permethrin or [phenoxyphenyl-^{14}C] PBalc was incubated at 2.7-98 µM with or without 3 mM NADPH in the presence of male rat, female rat or human liver microsomes. The incubations were carries out for 60 min at 37 °C and stopped by addition of acetonitrile. Control experiments were performed without liver microsomes.
In addition, the P450 isoforms responsible for the 40-hydroxylation of 3-phenoxybenzyl alcohol (PBalc) and the UGT isoforms responsible for the glucuronidation of PBacid were identified. Furthermore, glucuronidation of 3-phenoxybenzoic acid (PBacid) in humans was examined.
In the following, only the results of the in-vitro metabolism of trans-permethrin by hepatic microsoms are described.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description:	Trans-[Phenoxyphenyl-^{14}C]permethrin and [phenoxyphenyl-^{14}C]PBalc; specific activity: 4.44 GBq/mmol
Lot/Batch #:	not applicable
Purity:	100.0% radiochemical purity
CAS#:	Trans-Permethrin: 61949-77-7 PBalc: 13826-35-2
Development code:	not applicable
Spiking levels:	Trans-[Phenoxyphenyl-^{14}C]permethrin: 2.7-98 µM and [phenoxyphenyl-^{14}C] PBalc: 2.7-98 µM
Stability of the test Compound:	not reported

2. Test Commodity:

Cell cultures: Liver microsomes from human (purchased from KAC Co., Ltd); liver microsomes from male and female Crj:CD(SD) rats (purchased from Charles River Japan Inc., Kanagawa, Japan)

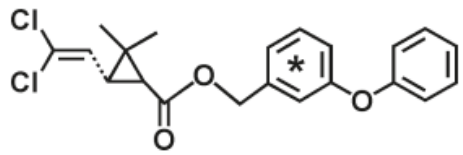
In vitro assay: In Vitro Metabolism of trans-Permethrin and PBalc with Human, Male Rat, and Female Rat Liver Microsomes
Trans-[Phenoxyphenyl-^{14}C]permethrin (2.7-98 µM) or [phenoxyphenyl-^{14}C] PBalc (2.7-98 µM) was incubated with or without 3 mM NADPH in the presence of human, male rat, and female rat liver microsomes (1 mg/mL) for 60 min at 37 °C. All incubations were carried out in 100 mM phosphate buffer (pH 7.4). Control experiments were performed without liver microsomes. After incubation, the reaction was terminated by addition of acetonitrile and the mixtures were stored on ice for 10 min and centrifuged at 10000 g for 5 min. The supernatant was analyzed by HPLC.

B. STUDY DESIGN AND METHODS

1. Test procedure

In the present study, to compare the metabolic profiles (route and rate) between rats and humans, the in vitro metabolism of trans-permethrin by hepatic microsomes from rats and humans was investigated. Trans-[Phenoxyphenyl-^{14}C]permethrin or [phenoxyphenyl-^{14}C] PBalc was incubated at 2.7-98 µM with or without 3 mM NADPH in the presence of male rat, female rat or human liver microsomes. The incubations were carries out for 60 min at 37 °C and stopped by addition of acetonitrile. Control experiments were performed without liver microsomes.
In addition, the P450 isoforms responsible for the 40-hydroxylation of 3-phenoxybenzyl alcohol (PBalc) and the UGT isoforms responsible for the glucuronidation of PBacid were identified. Furthermore, glucuronidation of 3-phenoxybenzoic acid (PBacid) in humans was examined. In the following, only the results of the in-vitro metabolism of trans-permethrin by hepatic microsoms are described.

Figure 5.1.2-1: Structural formula of trans-[phenoxyphenyl-^{14}C]permethrin



2. Description of analytical procedures

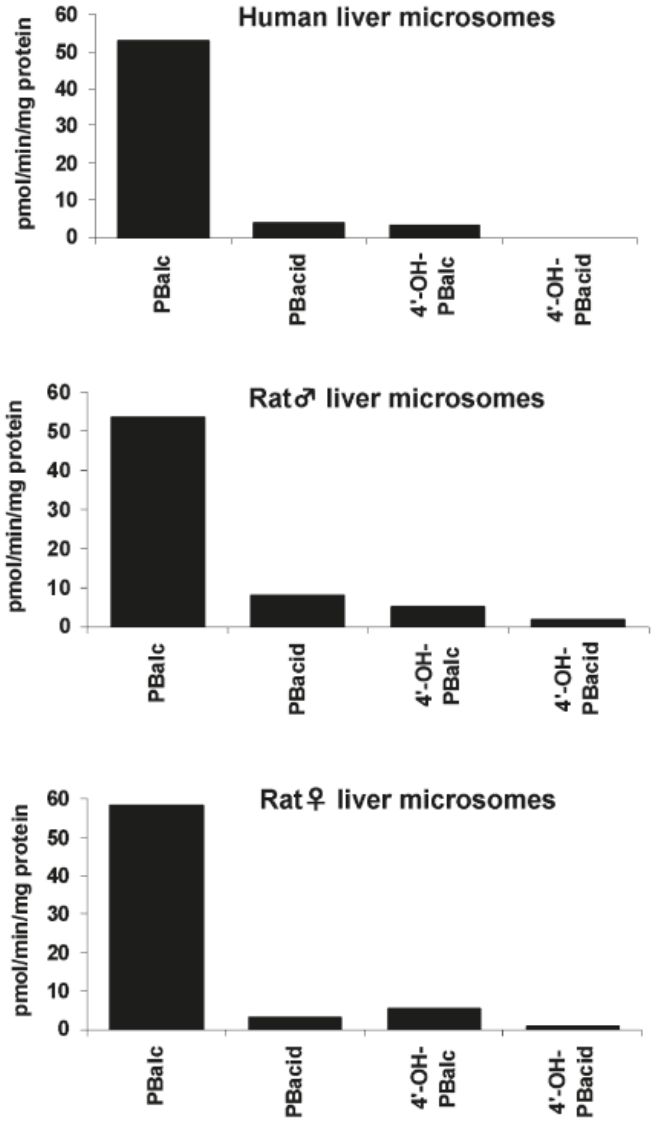
HPLC Analysis
HPLC was carried out on a system consisting of an L-6200 HPLC intelligent pump, an L-4000 UV detector and a Radiomatic 610TR RI detector fitted with an Atlantis dC18 column. The mobile phase consisted of acetonitrile and 0.1% (v/v) acetic acid in water. A flow rate of 1 mL/min was used. For the separation, a gradient program was used.

II. RESULTS AND DISCUSSION

1. Comparative in Vitro Metabolism of trans-[Phenoxyphenyl-^{14}C] permethrin

Trans-[Phenoxyphenyl-^{14}C]permethrin was incubated with human, male rat, and female rat liver microsomes each at concentrations of 2.7, 4.5, 17, 46, and 98 µM. In each incubation solution, PBalc, PBacid, and 4'-OH-PBalc were detected. PBalc (resulting from the hydrolysis of trans-permethrin) was the major metabolite in both humans and rats. The metabolites produced by human microsomes were nearly identical to those produced by rat liver microsomes, although 4'-OH-PBacid was not detected in a human liver microsomal incubation. The rates of metabolic formation were almost similar between humans and rats (see Figure 5.1.2-2). In this study, human and rat liver microsomes metabolized trans-permethrin mainly through hydrolysis, not oxidation, suggesting that the rapid hydrolysis is the major metabolic route of trans-permethrin in both humans and rats.

Figure 5.1.2-2: Rates of metabolite formation of trans-permethrin with human, male rat, and female rat microsomes.



III. CONCLUSION

The main metabolic pathway of trans-permethrin was the hydrolysis of the ester linkage to give PBalc in humans and rats. The metabolites produced by both rat and human microsomes (PBalc, PBacid, and 4'-OH-PBalc) were nearly identical except for 4'-OH-PBacid, which was not detected in a human liver microsomal incubation.

CA 5.2 Acute Toxicity

Studies evaluated in the draft monograph of alpha-cypermethrin by the rapporteur member state Belgium (1999): Alpha-cypermethrin (BAS 310 I) has been tested in various species and via different routes of administration. All studies are scientifically valid; however, partially the studies have been conducted before the release of study guidelines and are without GLP according to the usual practice in those days. These studies have been evaluated by European authorities and Belgium as Rapporteur Member State (European Commission Peer Review Program) and are, for the convenience of the reviewer, listed in Table 5.2-1**:**. Brief summaries of the respective studies were extracted from the monograph of alpha-cypermethrin and are provided under the respective chapters.

Table 5.2-1: Summary of already peer-reviewed acute toxicity studies with alpha-cypermethrin as available in the DAR (1999)

Route	**Purity;** cis1:cis2 ratio: **Species/Sex**	**Dose range (vehicle)**	**Result Classification**	**Reference (BASF Doc ID)**
Oral	95.6%, cis1:cis2 ratio: 3:97; Rat, Crl:CD:BR, m/f	33, 46, 64, 90 and 126 mg/kg bw (2% in corn oil)	LD_{50} (m): 57 mg/kg bw LD_{50} (f): 71 mg/kg bw LD_{50} (f&m): 64 mg/kg bw	AL-410-003
	95% - 99.5%; cis1:cis2 ratio: not stated Rat, Sprague-Dawley, m/f	180, 280, 440 and 700 mg/kg bw (corn oil)	LD_{50} (m): 310 mg/kg bw LD_{50} (f): 430 mg/kg bw	[redacted] 1993[1]
	99.4%; cis1:cis2 ratio: not stated Rat	10, 20, 40, 80, 160 and 320 mg/kg bw (10% in corn oil)	40-80 mg/kg bw	KCA 5.2/1 AL-411-005
		99, 158, 251, 400, 635 and 800 mg/kg bw (20% in corn oil)	368 mg/kg bw	
	96.6%, cis1:cis2 ratio: 3.3:96.7; Rat, Wistar, m/f	394, 626, 995, 1582, 2515 and 4000 mg/kg bw (40% in DMSO)	LD_{50} (m): 4000 mg/kg bw LD_{50} (f): >4000 mg/kg bw	AL-411-004
		492, 782, 1244, 1978, 3144 and 5000 mg/kg bw (50% in CMC)	LD_{50} (m/f) >5000 mg/kg bw	
Dermal	95.6%, cis1:cis2 ratio: 3:97; Rat, Crl:CD:BR, m/f	2000 mg/kg bw (undiluted)	LD_{50} (m/f) >2000 mg/kg bw	AL-410-003
	96%; cis1:cis2 ratio: not stated Rat, Sprague-Dawley, m/f	2000 mg/kg bw	LD_{50} (m/f) >2000 mg/kg bw	[redacted] 1992[1]
Inhalation	95.6%; cis1:cis2 ratio: not stated Rat, Sprague Dawley, m/f	0.98 and 1.59 mg/L (4 h)	LC_{50} (m/f): > 1.59 mg/L	AL-413-001
	95 - 99.5%; cis1:cis2 ratio: not stated Rat, Sprague-Dawley m/f	LC_{50} (mf): > 0.593 mg/L	LC_{50} (m/f): > 0.593 mg/L	[redacted] 1994[1]
Skin irritation	95.6%, cis1:cis2 ratio: 3:97 ; Rabbit, NZW	0.5 g/animal	Not irritating	AL-410-003
	95% - 99.5%; cis1:cis2 ratio: not stated Rabbit, NZW	0.5 g/animal	Not irritating	[redacted] 1992[1]
Eye irritation	95.6%, cis1:cis2 ratio: 3:97 ; Rabbit, NZW	0.1 mL/animal	Not irritating	AL-410-003
	95% - 99.5%: cis1:cis2 ratio: not stated Rabbit, NZW	0.1 g/animal	Not irritating	[redacted] 1992[1]

Table 5.2-1: Summary of already peer-reviewed acute toxicity studies with alpha-cypermethrin as available in the DAR (1999)

Route	**Purity;** cis1:cis2 ratio: **Species/Sex**	**Dose range (vehicle)**	**Result Classification**	**Reference (BASF Doc ID)**
Skin sensitization Magnusson-Kligman test	95.6%, cis1:cis2 ratio: 3:97: Guinea Pig, Dunkin Hartley	Intradermal induction: 2% Epidermal induction: 50% Challenge: 50%	Not sensitizing	AL-410-003
	95% - 99.5%; cis1:cis2 ratio: not stated Guinea pig, Dunkin Hartley	Intradermal induction: 5% Epidermal induction: 50% Challenge: 50%	Not sensitizing	[redacted] 1992[1]

[1] These studies are not BASF proprietary, but owned by Gharda, Chemical Ltd. Bombay, a further notifier for Annex I listing of alpha-cypermethrin in 1999.
NZW: New Zealand White;

Additional information given by the notifier:

Table 5.2-2: Summary of acute toxicity of alpha-cypermethrin in mice:

Route	Purity; cis 1 : cis 2 ratio	Vehicle	LD50 (mg/kg bw)	Reference
oral	96.6%; cis1:cis2 ratio: 3.3/96.7	as a 40% solution in DMSO	762	AL-411-004
	96.6%; cis1:cis2 ratio: 3.3/96.7	as a 50% solution in water	798	AL-411-004
	96.6%; cis1:cis2 ratio: 3.3/96.7	in corn oil	35	AL-411-004
	95-99.5%; cis1:cis2 ratio: not stated	in corn oil	50	[redacted] 1994[1]
dermal	96.6; cis1:cis2 ratio: 3.3/96.7	in corn oil	> 100	AL-411-004

[1] This study is not BASF proprietary, but owned by Gharda, Chemical Ltd. Bombay, a further notifier for Annex I listing of alpha-cypermethrin in 1999.

Furthermore, based on human observations and indications in animal studies alpha-cypermethrin is considered to be irritating to the respiratory tract after single exposure (decision by ECB, 2001). Based on the data available at that time, the following EU agreed endpoints were given in the Review Report (SANCO/4335/2000 final; Doc ID 2004/1040515) from 2004:

Acute toxicity

Rat LD_{50} oral:	57 mg/kg bw (corn oil) T, R 25
Rat LD_{50} dermal:	> 2000 mg/kg bw
Rat LC_{50} inhalation:	> 0.593 mg/L (4-h, nose/mouth) (max. attainable concentration) R 37
Skin irritation:	Non-irritant
Eye irritation:	Non-irritant
Skin sensitization (M&K):	Not skin sensitizing

Submission of not yet peer-reviewed studies in this AIRIII-Dossier:
New oral, dermal and inhalation acute toxicity, skin and eye as well as sensitization studies according to current criteria have been performed with alpha-cypermethrin for global registration and for production site comparison.
In accordance with the data requirements of Commission Regulation SANCO/11802/2010 an in vitro NRU-Phototoxicity study in Balb/c 3T3 cells has been performed and is given in detail in chapter CA 5.2.7. All new studies are submitted within the AIR III process and are tabulated inTable 5.2-3.

Table 5.2-3: Summary of newly available acute toxicity studies with alpha-cypermethrin

Type of study	Species/Sex	purity / vehicle	Result	Reference (BASF DocID)
Oral	Rat, Wistar HanRcc:WIST (STP)/f	99.3% / 0.5% CMC	LD_{50} (f): > 2000 mg/kg bw	KCA 5.2.1/1 2005/1011568
		98.8% / 0.5% CMC	LD_{50} (f): > 2000 mg/kg bw	KCA 5.2.1/2 2005/1011604
Dermal	Rat, Wistar HanRcc:WIST (STP)/m+f	99.3% / 0.5% CMC	LD_{50} (m+f): > 2000 mg/kg bw	KCA 5.2.2/1 2005/1011569
		98.8% / 0.5% CMC	LD_{50} (m+f): > 2000 mg/kg bw	KCA 5.2.2/2 2005/1011605
Inhalation	Rat,Wistar HanRcc:WIST (STP)/ m+f	98.8% / none	LC_{50} (f): 2.29mg/L LC_{50} (m): 2.79 mg/L LC_{50} (m+f): 2.5 mg/L	KCA 5.2.3/1 2005/1013246
		99.3% / none	LC_{50} (f): 1.21 mg/L LC_{50} (m): > 1.33 mg/L LC_{50} (m+f): 1.33 mg/L	KCA 5.2.3/2 2005/1015687
Skin irritation	Rabbit, NZW m+f	99.3% / none	Not irritating	KCA 5.2.4/1 2005/1011570
		98.8% / none	Not irritaing	KCA 5.2.4/2 2005/1011606
Eye irritation	Rabbit, NZW m+f	99.3% / none	Not irritating	KCA 5.2.5/1 2005/1011571
		98.8% / none	Not irritating	KCA 5.2.5/2 2005/1011607
Sensitisation – (M&K)	Guinea pig, Dunkin Hartley (Crl:(HA)BR), f	98.8% / 1% CMC	Not sensitising	KCA 5.2.6/1 2005/1011608
		99.3% / 1% CMC	Not sensitising	KCA 5.2.6/2 2005/1011572
in vitro NRU-Phototoxicity study	Balb/c 3T3 cells	99.4% / DMSO 1% in PBS	Not phototoxic	KCA 5.2.7/1 2013/1389105

The two batches tested from the different production sites showed similar results and were tested according to current guidelines in CMC or pure. Under these conditions alpha-cypermethrin has low acute toxicity by the oral and dermal route. The toxicity by the inhalation route is moderate with an LC_{50} of 1.33 mg/L and 2.5 mg/L. Alpha-Cypermethrin does not induce skin or eye irritation and is neither sensitizing nor shows phototoxic potential in Balb/c 3T3 cells.

The solutions of alpha-cypermethrin in corn oil are substantially more toxic than those in water, in DMSO or in suspension. Therefore, the new studies for acute oral toxicity performed with aqueous suspensions do not affect the LD_{50} relevant for classification. The new studies were only taken into consideration for the classification proposal when they indicated higher toxicity than seen in the original data set. This is only the case for acute inhalation toxicity.

Considering all available studies alpha-cypermethrin is acutely toxic by the oral route and harmful via inhalation. Based on human and animal data alpha-cypermethrin is regarded as irritating to the respiratory tract after single exposure. Very low toxicity is induced by the percutaneous route. Alpha-Cypermethrin is no skin or eye irritant, not a skin sensitizer and not phototoxic.

The proposed endpoints based on all available studies are shown below in Table 5.2-4

Table 5.2-4: Proposed acute toxicity endpoints of alpha-cypermethrin*

Study type/species	Results	Classification
		Reg. EC 1272/2008 (CLP)
Acute oral toxicity, rat	LD_{50} = 57 mg/kg bw	Acute tox 3 (H301)
Acute dermal toxicity, rat	LD_{50} > 2000 mg/kg bw	-
Acute inhalation toxicity, rat	**LC_{50} (f) = 1.21 mg/L**	Acute tox 4 (H332) STOT SE 3 (H335)
Dermal irritation, rabbit	Slightly Irritating	-
Eye irritation, rabbit	Not irritating	-
Maximisation test, guinea pig	Not sensitising (M & K maximisation test)	-
In vitro NRU Phototoxicity Test, Balb/c 3T3 cells	**Not phototoxic**	-

* new endpoints or values differing from the current agreed EU endpoints are marked in bold

Under consideration of all available data the harmonized classification of alpha-cypermethrin will differ under Reg. EC 1272/2008 (CLP) with regard to the additional classification as Acute tox 4 (H332). The List of endpoints will be adapted accordingly as shown below:

Acute toxicity (SANCO/11802 data point 5.2)

Rat LD_{50} oral	LD_{50} = 57 mg/kg bw Acute tox 3 (H301)
Rat LD_{50} dermal	LD_{50} > 2000 mg/kg bw No classification required
Rat LC_{50} inhalation	LC_{50} (f) = 1.21 mg/L **Acute tox 4 (H332),** STOT SE 3 (H335)
Skin irritation	Mildly irritating No classification required
Eye irritation	Not irritating No classification required
Skin sensitization (test method used and result)	Not sensitising (M & K maximisation test) No classification required

Acute toxicity (SANCO/11802 data point 5.2)

Phototoxicity	Not phototoxic No classification required

For convenience of the reviewer brief summaries of the respective studies as extracted from the monograph of alpha-cypermethrin are provided below together with the summaries of recently conducted studies.

CA 5.2.1 Oral

[redacted] 1993 (AL-410-003)

Guidelines: In compliance with the test method B.1 of directive 92/69/EEC
GLP: Yes
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 Crl:CD:BR rats/sex received by gavage a single dose of alpha-cypermethrin (95.6%, cis1:cis2 ratio: 3:97) in corn oil at dose levels of 33, 46, 64, 90 and 126 mg/kg bw.
Mortalities occurred between four hours after dosing and day 3. There were deaths among rats dosed at 46 mg/kg bw (3♂, 1♀), at 64 mg/kg bw (3♂) at 90 mg/kg bw (3♂, 5♀) and at 126 mg/kg bw (5♂, 5♀). Clinical signs comprised hunched posture, piloerection, diarrhoea, an unkempt appearance and yellow staining of the anogenital fur at all dose levels. In animals that died, the following clinical signs were observed in addition: splayed gait, thrashing and bloody discharge from the mouth and nose, twitching, tremor, fasciculations, convulsions, vasodilatation, salivation and in isolated cases lacrimation, chromodacryorrhoea, lethargy, abasia vocalisation (distressed noises), hypothermia, cyanosis, bradypnoea and prostration. Surviving animals gained body weight as expected over the 14 day observation period. At pathology, pallor or a darkened appearance of the liver, kidneys and spleen, lung congestion and abnormal content of the stomach and small intestines was observed.

Conclusion
Under the conditions of this study, the LD_{50} for male and female animals was considered to be 57 and 71 mg/kg bw, respectively.

[redacted] 1993

Guidelines: In compliance with the test method B.1 of directive 92/69/EEC
GLP: Yes
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 Sprague Dawley rats/sex received alpha- Cypermethrin (95% - 99.5%) in corn oil by gavage at dose levels of 180, 280, 440 and 700 mg/kg bw.
Mortalities occurred within 2 days after dosing. There were deaths among rats dosed at 280 mg/kg bw (2♂, 2♀), at 440 mg/kg bw (4♂, 2♀) and at 700 mg/kg bw (5♂, 4♀). Clinical signs comprised nasal secretion, lacrimation, salivation, polyuria, ataxia, drowsiness, tremors and convulsions. Surviving animals were free of symptoms on 1 to 4 days after treatment. Body weight development was unaffected. At pathology, hermorrhage and congestion in lung and liver of the animals that died during the study was observed.

Conclusion
Under the conditions of this study, the LD_{50} for male and female animals was considered to be 310 and 430 mg/kg bw, respectively.

[redacted] 1982a (AL-411-004)

Guidelines: Not fully in compliance with the test method B.1 of directive 92/69/EEC
Deviations: Variability in test volume was not minimized by adjusting the concentration to ensure a constant volume at all dose levels
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

6 Wistar rats/sex received by intubation a single dose of alpha-cypermethrin (96.6%, cis1:cis2 ratio: 3.3:96.7) as 40% solution in DMSO at dose levels of 394, 626, 995, 1582, 2515 and 4000 mg/kg bw (group 1) and as 50% aqueous suspension in carboxy-methylcellulose at dose levels of 492, 782, 1244, 1978, 3144 and 5000 mg/kg bw (group 2). In group 1 all deaths occurred within 2 days of dosing at 995 mg/kg bw (1♂), 1582 mg/kg bw (1♂, 1♀), 2516 mg/kg bw (2♂) and at 4000 mg/kg bw (4♂). In group 2, one female animal of the 5000 mg/kg bw dose group died. Clinical signs in group 1 were observed over all dose levels until study day 2 and comprised salivation, lethargy, diarrhea, piloerection, clonic convulsions, blood around nose (some animals), tip-toe hind leg walk and lacrimation. In group 2, clonic convulsions, salivation, lethargy, aggressive, abasia, piloerection, diarrhea, hypersensitivity to sensory stimuli, blood around the nose and chromodacryorrhoea were observed until day 4 over all dose levels. In group 1, all surviving animals recovered from an initial weight loss until the end of the 14 day observation period. In group 2, body weight decrease was observed at the highest dose but reversed by the end of the 14-day observation period except for two males. Description of necropsy findings are missing.

Conclusion
Under the conditions of this study, the LD_{50} for male and female animals was considered to be 4000 and >4000 mg/kg bw, respectively, for the solution in DMSO (group 1) and >5000 mg/kg bw for the aqueous suspension (group 2).

[redacted] 1982b (AL-411-005)

Guidelines: Not fully in compliance with the test method B.1 of directive 92/69/EEC
Deviations: Variability in test volume was not minimized by adjusting the concentration to ensure a constant volume at all dose levels
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

6 Wistar rats/sex received by intubation a single dose of alpha-cypermethrin (99.4%, b,n° OCD/7) as 10% solution in corn oil at dose levels of 10, 20, 40, 80, 160 and 320 mg/kg bw (group 1) and as 20% solution in corn oil at dose levels of 99, 158, 251, 400, 635 and 800 mg/kg bw (group 2). In group 1, all deaths occurred within 2 days of dosing at 80 mg/kg bw (4♂, 4♀) and at 160 mg/kg bw (6♂, 6♀). In group 2, all deaths occurred within 3 days of dosing at 158 mg/kg bw (1♀), 251 mg/kg bw (2♂, 3♀), 400 mg/kg bw (2♂, 5♀), 635 mg/kg bw (6♂, 4♀) and 800 mg/kg bw (6♂, 3♀). Clinical signs in group 1 comprised piloerection, abasia, salivation, splayed hind leg gait, lethargy, diarrhea, clonic convulsion, tremor, blood around mouth and abnormal respiratory and were observed over all groups. In group 2, clonic convulsion, abasia, piloerection, diarrhea, lethargy, salivation, tremors, ataxia, splayed hind leg gait, chromodacryorrhoea, blood around nose was observed over all dose levels. All surviving animals recovered from an initial weight loss until the end of the 14 day observation period. Description of necropsy findings are missing.

Conclusion
Under the conditions of this study, the LD_{50} for male and female animals was considered to range from 40 to 80 mg/kg bw for the 10% solution (group 1) and from 282 to 487 mg/kg bw for 20% suspension (368 mg/kg bw, group 2).

Report:	CA 5.2.1/1 [redacted] 2005a BAS 310 I (Alpha-Cypermethrin) - Acute oral toxicity study in rats 2005/1011568
Guidelines:	OECD 423, EEC 2004/73 B.1 tris, EPA 870.1100
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

Single doses of 2000, 300 and 50 mg/kg bw of BAS 310 I (batch: COD-000166; purity: 99.3%) preparations in 0.5% CMC-solution in doubly distilled water were given to 4 administration groups of three fasted female animals, each (2000 mg/kg in 6 females, 300 and 50 mg/kg in 3 females each) by gavage in a sequential manner. Animals were observed for 14 days. No mortality occurred in the administration groups. Accordingly, the oral LD_{50} was found to be greater than 2000 mg/kg bw:

Rat, oral: LD_{50} > 2000 mg/kg bw

No clinical signs and findings were observed. The mean body weights of the administration groups increased throughout the study period. No macroscopic pathologic abnormalities were noted in the animals examined at the end of the observation period.
Under the conditions of this study the median lethal dose of BAS 310 I after oral administration was found to be greater than 2000 mg/kg bw in rats. DocID (2005/1011568)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I (alpha-cypermethrin)
Description:	Solid; powder/white
Lot/Batch #:	COD-000166
Purity/content:	99.3%
Stability of test compound:	The stability of the test substance in the vehicle was determined indirectly by the concentration control analysis.
2. Vehicle:	0.5% CMC-solution (cleaned sodium carboxymethylcellulose) in doubly distilled water

3. Test animals:

Species:	Rat
Strain:	Wistar / HanRcc:WIST(SPF)
Sex:	female
Age:	approximately 8 -12 weeks
Weight at dosing (mean):	172 - 188 g
Source:	RCC Ltd Laboratory Animal Services, Wölferstrasse 4, CH-4414 Füllinsdorf, Switzerland
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät (Maus / Ratte Haltung "GLP"), Provimi Kliba SA, Kaiseraugst, Basel, Switzerland, ad libitum
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages, type DK-III (Becker & Co., Castrop-Rauxel, FRG)

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 15-Feb-2005 - 12-Apr-2005

2. Animal assignment and treatment:

Single doses of 2000, 300 and 50 mg/kg bw of test material preparations in 0.5% CMC-solution in doubly distilled water were given to 4 administration groups of three fasted animals each (2000 mg/kg in 6 females, 300 and 50 mg/kg in 3 females) by gavage in a sequential manner. Clinical signs and symptoms were recorded several times on the day of administration and afterwards at least once each workday for the individual animals up to 14 days post-administration. A check for any dead or moribund animal was made twice each workday and once on Saturdays, Sundays and public holiday. Individual body weights were determined shortly before administration, weekly thereafter and at the end of the study. The animals were sacrificed by CO_2-inhalation and subjected to necropsy including gross pathological examination on the last day of the observation period or as soon as possible after death in case of animals that died before.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality occurred in the administration groups.

B. CLINICAL OBSERVATIONS

No clinical signs and findings were observed.

C. BODY WEIGHT

The mean body weights of the administration groups increased throughout the study period.

D. NECROPSY

No abnormalities were observed at gross necropsy.

III. CONCLUSION

Under the conditions of this study, the oral LD_{50} in rats for BAS 310 I was determined to be greater than 2000 mg/kg bw.

Report:	CA 5.2.1/2 [redacted] 2005b BAS 310 I (Alpha-Cypermethrin) - Acute oral toxicity study in rats 2005/1011604
Guidelines:	OECD 423, EEC 2004/73 B.1 tris, EPA 870.1100
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

Single doses of 2000, 300 and 50 mg/kg bw of BAS 310 I (batch: COD-000165; purity: 98.8%) preparations in 0.5% CMC-solution in doubly distilled water were given to 4 administration groups of three fasted female animals, each (2000 mg/kg in 6 females, 300 and 50 mg/kg in 3 females each) by gavage in a sequential manner. Animals were observed for 14 days. No mortality occurred in the administration groups. Accordingly, the oral LD_{50} was found to be greater than 2000 mg/kg bw:

Rat, oral: LD_{50} > 2000 mg/kg bw

No clinical signs and findings were observed. The mean body weights of the administration groups increased throughout the study period. No macroscopic pathologic abnormalities were noted in the animals examined at the end of the observation period.
Under the conditions of this study the median lethal dose of BAS 310 I after oral administration was found to be greater than 2000 mg/kg bw in rats. DocID (2005/1011604)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I (alpha-cypermethrin)
Description:	Solid; powder/white
Lot/Batch #:	COD-000165
Purity/content:	98.8%
Stability of test compound:	The stability of the test substance in the vehicle was determined indirectly by the concentration control analysis.
2. Vehicle:	0.5% CMC-solution (cleaned sodium carboxymethylcellulose) in doubly distilled water

3. Test animals:

Species:	Rat
Strain:	Wistar / HanRcc:WIST(SPF)
Sex:	female
Age:	approximately 8 -12 weeks
Weight at dosing (mean):	178 - 185 g
Source:	RCC Ltd Laboratory Animal Services, Wölferstrasse 4, CH-4414 Füllinsdorf, Switzerland
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät (Maus / Ratte Haltung "GLP"), Provimi Kliba SA, Kaiseraugst, Basel, Switzerland, ad libitum
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages, type DK-III (Becker & Co., Castrop-Rauxel, FRG)

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 15-Feb-2005 - 14-Apr-2005

2. Animal assignment and treatment:

Single doses of 2000, 300 and 50 mg/kg bw of test material preparations in 0.5% CMC-solution in doubly distilled water were given to 4 administration groups of three fasted animals each (2000 mg/kg in 6 females, 300 and 50 mg/kg in 3 females) by gavage in a sequential manner. Clinical signs and symptoms were recorded several times on the day of administration and afterwards at least once each workday for the individual animals up to 14 days post-administration. A check for any dead or moribund animal was made twice each workday and once on Saturdays, Sundays and public holiday. Individual body weights were determined shortly before administration, weekly thereafter and at the end of the study. The animals were sacrificed by CO_2-inhalation and subjected to necropsy including gross pathological examination on the last day of the observation period or as soon as possible after death in case of animals that died before.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality occurred in the administration groups.

B. CLINICAL OBSERVATIONS

No clinical signs and findings were observed.

C. BODY WEIGHT

The mean body weights of the administration groups increased throughout the study period.

D. NECROPSY

No abnormalities were observed at gross necropsy.

III. CONCLUSION

Under the conditions of this study, the oral LD_{50} in rats for BAS 310 I was determined to be greater than 2000 mg/kg bw.

CA 5.2.2 Dermal

[redacted] **1993 (AL-410-003)**

Guidelines: Not fully in compliance with the test method B.3 of directive 92/69/EEC
Deviations: Occlusive dressing was used
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 Crl:CD:BR rats/sex were exposed to undiluted alpha-cypermethrin (95.6%, cis1:cis2 ratio: 3:97) at a dose level of 2000 mg/kg bw, by dermal occlusive application for 24 h.
Mortality was not observed. Clinical signs comprised salivation, hypersensitivity to stimuli, staining of anogenital fur and hyperactivity from day 2 but until day 4. Sites of application of the test material were discoloured (white) after removal of the dressings on day 2. This effect disappeared on day 5. At pathology, limited to slight vascular congestion of the sites of application of the test material was observed.

Conclusion
Under the conditions of this study, the LD50 for male and female animals was >2000 mg/kg bw.

Pore, 1992

Guidelines: In compliance with the test method B.3 of directive 92/69/EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 albino Sprague-Dawley rats/sex were exposed to solid alpha-cypermethrin (96%) at a dose level of 2000 mg/kg bw, by dermal semi-occlusive application for 24 h.
Mortality, clinical signs and local effects were not observed.

Conclusion
Under the conditions of this study, the LD_{50} for male and female animals was >2000 mg/kg bw.

Report: CA 5.2.2/1
[redacted] 2005c
BAS 310 I (Alpha-Cypermethrin) - Acute dermal toxicity study in rats
2005/1011569
Guidelines: OECD 402, EEC 92/69 B 3, EPA 870.1200
GLP: yes
(certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an acute dermal toxicity study groups of 5 male and 5 female Wistar rats were exposed to 2000 mg/kg bw of a test material (batch COD-000166; purity: 99.3%) preparation in 0.5% CMC solution in doubly distilled water. The preparation was applied to the clipped skin under semi-occlusive conditions for 24 hours. The animals were observed for 14 days after administration. No mortality occurred in the male dose group. One animal of the female dose group was found dead on study day 13. However, this was considered to be due to an abszess of the lower jaw and not test substance related. Accordingly, the LD_{50} was determined to be greater than 2000 mg/kg bw:

Rat, dermal: LD_{50} > 2000 mg/kg bw

No systemic clinical observations or skin effects were noted in the animals. The mean body weights of the animals increased throughout the study period. During necropsy the animal that died showed a green to yellow pasty abszess (1 mL) of the right lower jaw. No further macroscopic pathologic abnormalities were noted in the surviving animals at the end of the study.
DocID (2005/1011569)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I (alpha-cypermethrin)
Description:	Solid; powder/white
Lot/Batch #:	COD-000166
Purity/content:	99.3%
Stability of test compound:	The stability of the test substance in the vehicle was determined indirectly by the concentration control analysis.
2. Vehicle:	0.5% CMC-solution (cleaned sodium carboxymethylcellulose) in doubly distilled water

3. Test animals:

Species:	Rat
Strain:	Wistar / HanRcc:WIST(SPF)
Sex:	male / female
Age:	male animals approx. 8 – 10 weeks, female animals approx. 12 - 14 weeks
Weight at dosing (mean):	males: 258 g, females: 222 g
Source:	RCC Ltd Laboratory Animal Services, Wölferstrasse 4, CH-4414 Füllinsdorf, Switzerland
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät (Maus / Ratte Haltung "GLP"), Provimi Kliba SA, Kaiseraugst, Basel, Switzerland, ad libitum
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages, type DK-III (Becker & Co., Castrop-Rauxel, FRG)
Environmental conditions:	
Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 01-Mar-2005 - 12-Apr-2005

2. Animal assignment and treatment:

Five male and five female rats were given a single application of test material preparation in 0.5% CMC solution in doubly distilled water to the clipped epidermis on the dorsal and dorsolateral parts of the trunk (about 40 cm^2, corresponding to at least 10% of the body surface) for 24 hours. The fur was clipped one day before application of the test substance.
A bandage consisting of four layers absorbent gauze (Ph. Eur. Lohmann GmbH & co. KG) with the calculated amount of the test substance was applied to the test site. The application site was covered with a semi-occlusive dressing (Fixomull Stretch (adhesive fleece), Beiersdorf AG). After the exposure period, the dressing and the bandage were removed and residual test substance was rinsed with warm water.
Rats were observed for signs and symptoms several times on the day of application and afterwards at least once each workday for a total of 2 weeks. Individual body weights were determined shortly before application, weekly thereafter and at the end of the study. Necropsy with gross-pathology examination on the last day of the observation period was done after killing the animals with CO_2.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality occurred in the male dose group.
One animal of the female dose group was found dead on study day 13. However, this was considered to be due to an abszess of the lower jaw and not test substance related.

B. CLINICAL OBSERVATIONS

No systemic clinical observations or skin effects were noted in the animals.

C. BODY WEIGHT

The mean body weights of the animals increased throughout the study period.

D. NECROPSY

During necropsy the animal that died showed a green to yellow pasty abszess (1 mL) of the right lower jaw. No macroscopic pathologic abnormalities were noted in the surviving animals examined at the end of the study.

III. CONCLUSION

Under the conditions of this study, the dermal LD_{50} in rats for BAS 310 I was determined to be greater than 2000 mg/kg bw.

Report:	CA 5.2.2/2 [redacted] 2005d BAS 310 I (Alpha-Cypermethrin) - Acute dermal toxicity study in rats 2005/1011605
Guidelines:	OECD 402, EEC 92/69 B 3, EPA 870.1200
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an acute dermal toxicity study groups of 5 male and 5 female Wistar rats were exposed to 2000 mg/kg bw of a test material (batch COD-000165; purity: 98.8%) preparation in 0.5% CMC solution in doubly distilled water. The preparation was applied to the clipped skin under semi-occlusive conditions for 24 hours. The animals were observed for 14 days after administration. Based on the absence of mortality in this study the acute dermal LD_{50} was determined to be greater than 2000 mg/kg bw:

Rat, dermal: LD_{50} > 2000 mg/kg bw

No systemic clinical observations or skin effects were noted in the animals. The mean body weights of the animals increased throughout the study period. No macroscopic pathologic abnormalities were noted in the animals examined at the end of the study.

DocID (2005/1011605)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I (alpha-cypermethrin)
Description:	Solid; powder/white
Lot/Batch #:	COD-000165
Purity/content:	98.8%
Stability of test compound:	The stability of the test substance in the vehicle was determined indirectly by the concentration control analysis.
2. Vehicle:	0.5% CMC-solution (cleaned sodium carboxymethylcellulose) in doubly distilled water

3. Test animals:

Species:	Rat
Strain:	Wistar / HanRcc:WIST(SPF)
Sex:	male / female
Age:	male animals approx. 8 – 10 weeks, female animals approx. 12 - 14 weeks
Weight at dosing (mean):	males: 264 g, females: 217 g
Source:	RCC Ltd Laboratory Animal Services, Wölferstrasse 4, CH-4414 Füllinsdorf, Switzerland
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät (Maus / Ratte Haltung "GLP"), Provimi Kliba SA, Kaiseraugst, Basel, Switzerland, ad libitum
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages, type DK-III (Becker & Co., Castrop-Rauxel, FRG)
Environmental conditions:	
Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 01-Mar-2005 - 14-Apr-2005

2. Animal assignment and treatment:

Five male and five female rats were given a single application of test material preparation in 0.5% CMC solution in doubly distilled water to the clipped epidermis on the dorsal and dorsolateral parts of the trunk (about 40 cm^2, corresponding to at least 10% of the body surface) for 24 hours. The fur was clipped one day before application of the test substance.
A bandage consisting of four layers absorbent gauze (Ph. Eur. Lohmann GmbH & co. KG) with the calculated amount of the test substance was applied to the test site. The application site was covered with a semi-occlusive dressing (Fixomull Stretch (adhesive fleece), Beiersdorf AG). After the exposure period, the dressing and the bandage were removed and residual test substance was rinsed with warm water.
Rats were observed for signs and symptoms several times on the day of application and afterwards at least once each workday for a total of 2 weeks. Individual body weights were determined shortly before application, weekly thereafter and at the end of the study. Necropsy with gross-pathology examination on the last day of the observation period was done after killing the animals with CO_2.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality occurred.

B. CLINICAL OBSERVATIONS

No systemic clinical observations or skin effects were noted in the animals.

C. BODY WEIGHT

The mean body weights of the animals increased throughout the study period.

D. NECROPSY

No macroscopic pathologic abnormalities were noted in the animals examined at the end of the study.

III. CONCLUSION

Under the conditions of this study, the dermal LD_{50} in rats for BAS 310 I was determined to be greater than 2000 mg/kg bw.

CA 5.2.3 Inhalation

[redacted] **993 (AL-413-001)**

Guidelines: In compliance with the test method B.2 of directive 92/69/EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 Sprague-Dawley rats/sex were exposed for 4 hours by inhalation exposure to an atmosphere contacting 0.98 and 1.59 mg/L of alpha-cypermethrin (95.6%) produced by a dust generator (MMAD: 6.1 – 9 mm, 49.3 and 34.8% respirable).
One female animal of the 0.98 mg/L dose group died. Clinical signs comprised exaggerated or irregular respiratory movements during exposure and exaggerated respiratory movements, staggering, fascicular tremors, writhing, ataxia, hunched posture, poorly groomed appearance and yellow staining around the urogenital region post-exposure until day 6 of the observation period. Body weight gain was reduced for up to 2 days following exposure. At pathology, lungs of the rat that died were moderately congested and stomach distended with gas. At the high dose, surviving rats showed dark subpleural foci on the lung in two animals. No abnormalities were observed at 0.98 mg/L.

Conclusion
Under the conditions of this study, the LC_{50} for male and female animals was >1.59 mg/L.

[redacted] **1994**

Guidelines: In compliance with the test method B.2 of directive 92/69/EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 Sprague-Dawley rats/sex were exposed for 4 hours by nose and mouth route to an atmosphere containing 0.593 mg/L (max. attainable concentration) of alpha-cypermethrin (95 - 99.5%) produced by a dust generator (MMAD: 5.1 mm, respirable range of 0-7 mm).
No mortality was observed. Clinical signs comprised nasal secretion, lacrimation, salivation, polyurea and wet fur at the termination of the exposure period. Nasal secretion was observed further on in females until study day 8 and in males until day 12. No effects on body weight development were observed. At pathology, congestion of the lung was observed in one male and one female animal each.

Conclusion
Under the conditions of this study, the LC_{50} for male and female animals was >0.593 mg/L.

Report:	CA 5.2.3/1 [redacted] 2005e BAS 310 I (Alpha-Cypermethrin) - Acute inhalation toxicity study in Wistar rats - 4-hour dust exposure 2005/1013246
Guidelines:	OECD 403, EEC 92/69, EPA 870.1300, EEC 67/548 Part B.2 L383A, EPA 712-C-98-193
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an acute inhalation toxicity study, groups of 5 male and 5 female Wistar rats were exposed to alpha-cypermethrin as dust (Batch COD-000165; Purity: 98.8%) at concentrations of 1.07 and 2.47 mg/L for 4 hours. The animals were observed for 14 days after exposure.
No mortality occurred at 1.07 mg/L. At 2.47 mg/L, one of five male and four of five female animals died. Accordingly, the acute inhalation LC_{50} for alpha-cypermethrin after dust inhalation exposure was determined to be:

LC50 (females) : 2.29mg/L
LC50 (males) : 2.79 mg/L
LC_{50} (male and female rats): 2.5 mg/L

Clinical signs of toxicity in animals exposed to 1.07 mg/L comprised visually accelerated respiration, eyelid closure, apathy, squatting posture, tremor, ataxia, startle reflex and smeared fur. Clinical signs of toxicity in animals exposed to 2.47 mg/L comprised some additional unspecific symptoms like gasping, discharged nose and salivation, indicative for respiratory distress, local irritant action and systemic toxicity. Findings were observed from hour 0 of exposure until study day 7. In the 1.07 mg/L dose group the mean body weight of the male animals did not increase adequately during the first post exposure observation week, but increased during the second week. The mean body weight of the female animals did not increase adequately throughout the whole study period. This effect is observed at times in the rat strain used, because in the required age range the female animals have already reached the phase of slow growth. No gross pathological abnormalities were noted in the animals necropsied at termination of the post exposure observation period. Regarding the 2.47 mg/L dose group, the mean body weight of the male animals decreased during the first post exposure observation week, but increased during the second week. The body weight of the only surviving female animal increased slightly throughout the whole study period. During necropsy, dark red discoloration of all lung lobes were noted in two female animals that died one day after exposure. Additionally, contaminated and wet fur around the snout were noted in all three female animals that died one day after exposure. No gross pathological abnormalities were noted in the one male and the one female animal that died 1 hour after exposure. No gross pathological abnormalities were noted in all animals that were necropsied at termination of the study. Cascade impactor measurements resulted in particle size distributions with a mass median aerodynamic diameters (MMAD) of 2.7 - 2.8 µm and a geometric standard deviation (GSD) of 2.7 – 2.8.

(DocID 2005/1013246)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I
Description:	Solid; powder/white
Lot/Batch #:	COD-000165
Purity/content:	98.8%
Stability of test compound:	Stable (Expiry date: 01-Nov-2006)
2. Vehicle:	Test substance was applied unchanged.
3. Test animals:	
Species:	Rat
Strain:	Wistar / RccHan™:WIST
Sex:	male and female
Age:	Males: approx. 8 - 10 weeks; females: approx. 8 - 12 weeks
Weight at dosing (mean):	Males: 264.0 g; females: 201.6 g
Source:	RCC Ltd Laboratory Animal Services; Wölfersraße 4, CH-4414 Füllinsdorf
Acclimation period:	at least 1 week
Diet:	KLIBA mouse / rat laboratory diet 10 mm pellets "GLP", Provimi Kliba SA, Kaiseraugst, Basel Switzerland, ad libitum
Water:	Drinking water, ad libitum
Housing:	Singly in cages type DK III (Becker, Germany) without bedding.
Environmental conditions:	
Temperature:	20 - 24 °C
Humidity:	30 - 70%
Air changes:	No data. The animals were kept in fully air-conditioned rooms.
Photo period:	Alternating 12-hour light and dark cycles

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: 27-Apr-2005 - 14-Jul-2015

2. Animal assignment and treatment:

For determination of the acute inhalation toxicity (head-nose inhalation, 4-hour-exposure) groups of five male and five female rats were exposed to 1.07 and 2.47 mg/L of the test substance. The test substance was desagglomerated in a mixer under addition of 2 % (w/w) of Aerosil® R972 before introduction into the dust generator, in order to improve dust formation. After exposure, animals were observed for 14 days. Individual body weights were recorded on arrival, shortly before exposure (day 0) and on days 7 and 14. Additionally, body weight was measured in animals that died from study day 1 onward. A check for overt clinical signs of toxicity or mortality as well as a check for the presence of feed and drinking water was made twice a day on workdays and once daily on weekends and public holidays. Detailed clinical observations were recorded for each animal separately several times during exposure and at least once on each workday of the observation period. Additionally, on one Saturday details clinical observation was carried out in both test groups. At the end of the observation period the surviving animals that were sacrificed with CO_2 were subjected to gross-pathological examination like all other animals which had died before.

3. Statistics/calculations:

The LC_{50} was calculated by Probit analysis by means of a computer program. For results of the type ”LC_{50} greater than”, ”LC_{50} approx.”, or ”LC_{50} smaller than”, the binomial test was used for statistical evaluation. The calculation of the particle size distribution was carried out in the inhalation laboratory on the basis of mathematical methods for evaluating particle measurements.

4. Generation of the test atmosphere and exposure:

The test substance was desagglomerated in a mixer under addition of 2 % (w/w) of Aerosil® R972 before introduction into the dust generator, in order to improve dust formation. For each test group the dust was produced inside the dust pre-chamber with a dosing-wheel dust generator (Gericke/BASF) and compressed air and passed into the inhalation system. The concentrations of the dust in the atmosphere were adjusted by varying the apertural width and rotation of the dosing wheel. The exposure systems were located inside an exhaust cabin in an air-conditioned laboratory. A supply air flows (compressed air) of 1.5 m³/h were used for the exposures. The exhaust air flows were set to 1.35 m³/h. An air change of about 27 times per hour can be calculated by dividing the supply air flows by the volume of the inhalation systems. The lower amounts of exhaust air, which were adjusted by means of a separate exhaust air systems, achieved positive pressure inside the exposure systems. This ensured that the mixtures of test substance and air were not diluted with laboratory air in the breathing zones of the animals. The animals were exposed to the inhalation atmospheres for 4 hours plus equilibration time of the inhalation systems (t_{99} about 10 min).

5. Analytical investigation:
The flows of supply and exhaust air were adjusted and continuously measured with flowmeters (Rota). They were recorded four times in about 1-hour intervals. The temperature in the inhalation systems were measured continuously with a digital thermometer and recorded four times in about 1-hour intervals. The humidities in the inhalation systems were measured with a dielectric probe four times in about 1-hour intervals. No surveillance of the oxygen content in the inhalation systems was performed. The air change was judged to be sufficient to prevent oxygen depletion by the breathing of the animals, and the concentrations of the test substance used could not have a substantial influence on oxygen partial pressure. The nominal concentration was calculated from the amounts of substance dosed and the supply air flows.
Gravimetric determination of the inhalation atmosphere concentration was performed with a balance Mettler AT 250. Preweighed filters were placed into the filtration equipment. By means of the vacuum pump metered volumes of the dust aerosol were drawn through the filter. For each sample the dust aerosol concentration in mg/L was calculated from the difference between the preweight of the filter and the weight of the filter after sampling, with reference to the sample volume of the inhalation atmospheres. Mean and standard deviation were calculated for the concentration from the results of the individual measurements. The mean concentration were corrected for the amount of additive used

6. Particle Size Analysis:
Before sampling, the impactor was assembled with preweighed glass-fiber collecting discs, and a backup particle filter. The impactor was connected to the vacuum pump and one sample per exposure was taken from the breathing zone of the animals starting not earlier than 30 minutes after the beginning of the exposure. The sample volumes were 6 and 15 L. After sampling the impactor was taken apart. The collecting discs and the backup particle filter were re-weighed. The amounts of material adsorbed to the walls of the impactor and in the sampling probe (wall losses) were also determined quantitatively. The results from the particle size analysis were not corrected for the additive.

II. RESULTS AND DISCUSSION

A. MORTALITY

No lethality occurred at the tested concentration of 1.07 mg/L during the study period of 14 days. At 2.47 mg/L one male and 4 female animals died.

Table 5.2.3-1: Lethality in rats exposed for 4 hours to BAS 310 I as dust

Test group (mg/L)	Cumulated lethality		Time interval of lethality
	Males	Females	
1.07	0/5	0/5	-
2.47	1/5	4/5	d0 – d1

B. CLINICAL OBSERVATIONS

The nature and duration of the observations are indicated in Table 5.2.3-2. All animals recovered within the 14-day observation period.

Table 5.2.3-2: Nature and duration of clinical signs observed in rats exposed for 4 hours to BAS 310 I as dust

	Males		Females	
Test group (mg/L)	**1.07**	**2.47**	**1.07**	**2.47**
Total number of animals	5	5	5	5
Respiration, visually accelerated	h0 – d7	h0 – d7	h0 – d7	h0 – d7
Gasping	-	-	-	d0
Nose, discharge	-	-	-	d0
Eyelid closure	d0	-	d0	-
Salivation	-	-	-	d0
Apathy	d0 – d7	d0 – d5	d0 – d7	d0 – d5
Unconsciousness	-	-	-	d0
Squatting posture	d0 – d7	d1 – d5	d0 – d7	d1 – d5
Tremor	d0 – d2	-	d0 – d2	-
Ataxie	d1 – d3	d0 – d5	d1 – d3	d1
Startle reflex	d1 – d3	-	d1 – d3	-
Convulsions	-	-	-	d0
Piloerection	-	d0 – d7	-	d0 – d1
Fur, smeared	d0 – d2	d0 – d1	d0 – d2	d0 – d7
Reduced general state	-	d0	-	d0

hn: hour n of exposure; d0: post-exposure on the day of exposure; dn: day n after exposure

C. BODY WEIGHT

Regarding the 1.07 mg/L dose group, the mean body weight of the male animals did not increase adequately during the first post exposure observation week, but increased during the second week. The mean body weight of the female animals did not increase adequately throughout the whole study period. This effect is observed at times in the rat strain used, because in the required age range the female animals have already reached the phase of slow growth.

In the 2.47 mg/L dose group, the mean body weight of the male animals decreased during the first post exposure observation week, but increased during the second week. The body weight of the only surviving female animal increased slightly throughout the whole study period.

D. NECROPSY

Regarding the 1.07 mg/L dose group, no gross pathological abnormalities were detected the animals that were necropsied at termination of the study.
In the 2.47 mg/L dose group, dark red discoloration of all lung lobes were noted in two female animals that died one day after exposure. Additionally, contaminated and wet fur around the snout were noted in all three female animals that died one day after exposure. No gross pathological abnormalities were noted in the one male and the one female animal that died 1 hour after exposure.
No gross pathological abnormalities were noted in all animals that were necropsied at termination of the study.

E. ANALYTICAL MEASUREMENTS

The exposure conditions are summarized in Table 5.2.3-3.

Table 5.2.3-3: Exposure conditions

Test group (mg/L)	Supply air (m^3/h)	Exhaust air (m^3/h)	Temperature (°C)	Relative humidity (%)	Substance f (g/h)[4]
1.07	1.5	1.35	21.5±0.3	55.5±0.6	20.9
2.47	1.5	1.35	21.4±0.1	51.7±0.6	53.8

[4] corrected for 2 % (w/w) Aerosil.

Test atmosphere concentrations are presented in Table 5.2.3-4.

Table 5.2.3-4: Atmosphere concentrations

Mean achieved (mg/L)	Standard deviation	Nominal (mg/L)
1.07	0.21	13.9
2.47	0.49	35.9

The measurements of particle-size distribution revealed mass median aerodynamic diameters (MMAD) of 2.7 – 2.8µm with a geometric standard deviation of 2.7 - 2.8 respectively (see Table 5.2.3-5).

Table 5.2.3-5: Particle size distribution

Mean achieved (analytical) atmosphere concentration (mg/L)	Mean mass median aerodynamic diameter (µm)	Inhalable fraction (% <3 µm)	Standard deviation
1.07	2.7	54.6	2.7
2.47	2.8	53.1	2.8

III. CONCLUSION

Under the conditions of this study the 4 hour inhalation LC_{50} of alpha-cypermethrin for male and female rats was estimated to be 2.5 mg/L.

Report:	CA 5.2.3/2 [redacted] 2005a BAS 310 I (Alpha-Cypermethrin) - Acute inhalation study in Wistar rats - 4-hour dust exposure 2005/1015687
Guidelines:	OECD 403, EPA 870.1300, EEC 92/69 B 2, EEC 67/548, EPA 712-C-98-193
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an acute inhalation toxicity study, groups of 5 male and 5 female Wistar rats were exposed to alpha-cypermethrin as dust (Batch COD-000166; Purity: 99.3%) at concentrations of 0.42 and 1.16 mg/L for 4 hours. The animals were observed for 14 days after exposure.
No mortality occurred at 0.42 mg/L. Two of five females but no males died at 1.16 mg/L. Accordingly, the acute inhalation LC_{50} for alpha-cypermethrin after dust inhalation exposure was determined to be:

LC_{50} (female rats): 1.21 mg/L
LC_{50} (male rats): >1.16 mg/L
LC_{50} (male and female rats): 1.33 mg/L

Clinical signs of toxicity in animals exposed to 0.42 mg/L comprised visually accelerated respiration, squatting posture, piloerection and smeared fur. Findings were observed from hour 0 of exposure until including study day 2. Clinical signs of toxicity in animals exposed to 1.16 mg/L comprised visually accelerated respiration, squatting posture, tremor, abdominal position, staggering, high-stepping gait, startle reflex, piloerection and smeared and contaminated fur. Moreover, reddish discoloration in the anogenital region and around the snout was observed. Findings were observed from hour 0 of exposure until including study day 6. In the 0.42 mg/L dose group, the mean body weight of the male animals increased throughout the study period. The mean body weights of the female animals decreased slightly during the first post exposure observation week and increased slightly during the second week. This effect is observed at times in the rat strain used, because in the required age range the female animals have already reached the phase of slow growth. Regarding the 1.16 mg/L dose group, the mean body weight of the surviving animals increased throughout the study period. No gross pathological abnormalities were noted at pathology. Cascade impactor measurements resulted in particle size distributions with a mass median aerodynamic diameters (MMAD) of 2.6 - 2.8 µm and a geometric standard deviation (GSD) of 2.8 – 3.7.

(DocID 2005/1015687)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I
Description:	Solid; powder/white
Lot/Batch #:	COD-000166
Purity/content:	99.3%
Stability of test compound:	Stable (Expiry date: 01-Nov-2006)
2. Vehicle:	Test substance was applied unchanged.
3. Test animals:	
Species:	Rat
Strain:	Wistar / RccHan™:WIST
Sex:	male and female
Age:	Males: approx. 8 - 9 weeks; females: approx. 11 - 12 weeks
Weight at dosing (mean):	Males: 287.5 g; females: 202.0 g
Source:	RCC Ltd Laboratory Animal Services; Wölfersraße 4, CH-4414 Füllinsdorf
Acclimation period:	at least 1 week
Diet:	KLIBA mouse / rat laboratory diet 10 mm pellets "GLP", Provimi Kliba SA, Kaiseraugst, Basel Switzerland, ad libitum
Water:	Drinking water, ad libitum
Housing:	Singly in cages type DK III (Becker, Germany) without bedding.
Environmental conditions:	
Temperature:	20 - 24 °C
Humidity:	30 - 70%
Air changes:	No data. The animals were kept in fully air-conditioned rooms.
Photo period:	Alternating 12-hour light and dark cycles

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: 17-May-2005 - 08-Jul-2005

2. Animal assignment and treatment:

For determination of the acute inhalation toxicity (head-nose inhalation, 4-hour-exposure) groups of five male and five female rats were exposed to 0.42 and 1.16 mg/L of the test substance. The test substance was desagglomerated in a mixer under addition of 1 % (w/w) of Aerosil® 200 before introduction into the dust generator, in order to improve dust formation. After exposure, animals were observed for 14 days. Individual body weights were recorded on arrival, shortly before exposure (day 0) and on days 7 and 14. Additionally, body weight was measured in animals that died from study day 1 onward. A check for overt clinical signs of toxicity or mortality as well as a check for the presence of feed and drinking water was made twice a day on workdays and once daily on weekends and public holidays. Detailed clinical observations were recorded for each animal separately several times during exposure and at least once on each workday of the observation period. Additionally, on one weekend details clinical observation was carried out in both test groups. At the end of the observation period the surviving animals that were sacrificed with CO_2 were subjected to gross-pathological examination like all other animals which had died before.

3. Statistics/calculations:

The LC_{50} was calculated by Probit analysis by means of a computer program. For results of the type "LC_{50} greater than", "LC_{50} approx.", or "LC_{50} smaller than", the binomial test was used for statistical evaluation. The calculation of the particle size distribution was carried out in the inhalation laboratory on the basis of mathematical methods for evaluating particle measurements.

4. Generation of the test atmosphere and exposure:

The test substance was desagglomerated in a mixer under addition of 1 % (w/w) of Aerosil® 200 before introduction into the dust generator, in order to improve dust formation. For each test group the dust was produced inside the dust pre-chamber with a dosing-wheel dust generator (Gericke/BASF) and compressed air. The concentrations of the dust in the atmosphere were adjusted by varying the aperture width and rotation of the dosing wheel. The exposure systems were located inside an exhaust cabin in an air-conditioned laboratory. A supply air flows (compressed air) of 1.5 m³/h were used for the exposures. The exhaust air flows were set to 1.35 m³/h. An air change of about 27 times per hour can be calculated by dividing the supply air flows by the volume of the inhalation systems. The lower amounts of exhaust air, which were adjusted by means of a separate exhaust air systems, achieved positive pressure inside the exposure systems. This ensured that the mixtures of test substance and air were not diluted with laboratory air in the breathing zones of the animals. The animals were exposed to the inhalation atmospheres for 4 hours plus equilibration time of the inhalation systems (t_{99} about 10 min).

5. Analytical investigation:
The flows of supply and exhaust air were adjusted and continuously measured with flowmeters (Rota). They were recorded four times in about 1-hour intervals. The temperature in the inhalation systems were measured continuously with a digital thermometer and recorded four times in about 1-hour intervals. The humidities in the inhalation systems were measured with a dielectric probe four times in about 1-hour intervals. No surveillance of the oxygen content in the inhalation systems was performed. The air change was judged to be sufficient to prevent oxygen depletion by the breathing of the animals, and the concentrations of the test substance used could not have a substantial influence on oxygen partial pressure. The nominal concentration was calculated from the amounts of substance dosed and the supply air flows.
Gravimetric determination of the inhalation atmosphere concentration was performed with a balance Mettler AT 250. Preweighed filters were placed into the filtration equipment. By means of the vacuum pump metered volumes of the dust aerosol were drawn through the filter. For each sample the dust aerosol concentration in mg/L was calculated from the difference between the preweight of the filter and the weight of the filter after sampling, with reference to the sample volume of the inhalation atmospheres. Mean and standard deviation were calculated for the concentration from the results of the individual measurements. The mean concentration were corrected for the amount of additive used

6. Particle Size Analysis:
Before sampling, the impactor was assembled with preweighed glass-fiber collecting discs, and a backup particle filter. The impactor was connected to the vacuum pump and one sample per exposure was taken from the breathing zone of the animals starting not earlier than 30 minutes after the beginning of the exposure. The sample volume were 30 and 15 L. After sampling the impactor was taken apart. The collecting discs and the backup particlevfilter were re-weighed. The amounts of material adsorbed to the walls of the impactor and in the sampling probe (wall losses) were also determined quantitatively. The results from the particle size analysis were not corrected for the additive.

II. RESULTS AND DISCUSSION

A. MORTALITY

No lethality occurred at the tested concentration of 0.42 mg/L during the study period of 14 days. At 1.16 mg/L 2 female animals died.

Table 5.2.3-6: Lethality in rats exposed for 4 hours to BAS 310 I as dust

Test group (mg/L)	Cumulated lethality		Time interval of lethality
	Males	Females	
0.42	0/5	0/5	-
1.16	0/5	2/5	d0 – d1

B. CLINICAL OBSERVATIONS

The nature and duration of the observations are indicated in Table 5.2.3-7. All animals recovered within the 14-day observation period.

Table 5.2.3-7: Nature and duration of clinical signs observed in rats exposed for 4 hours to BAS 310 I as dust

	Males		Females	
Test group (mg/L)	**0.42**	**1.16**	**0.42**	**1.16**
Total number of animals	5	5	5	5
Respiration, visually accelerated	h0 – d1	h0 – d3	h0 – d1	h0 – d3
Squatting posture	d0	d0 – d3	d0	d0 – d3
Tremor	-	d0	-	d0
Adominal position	-	d0	-	d0
Staggering	-	d0	-	d0
High-stepping gait	-	d1 – d3	-	d1 – d2
Startle reflex	-	d1 – d3	-	d1 – d3
Reddish discoloration in the anogenital region and the region of the snout	-	-	-	d0
Piloerection	d0 – d2	d0 – d6	d0 – d2	d0 – d3
Fur, smeared	d0	d0	d0	d0

hn: hour n of exposure; d0: post-exposure on the day of exposure; dn: day n after exposure

C. BODY WEIGHT

Regarding the 0.42 mg/L dose group, the mean body weight of the male animals increased throughout the study period. The mean body weights of the female animals decreased slightly during the first post exposure observation week and increased only slightly during the second week. This effect is observed at times in the rat strain used, because in the required age range the female animals have already reached the phase of slow growth.
In the 1.16 mg/L dose group, the mean body weight of the surviving male and female animals increased throughout the study period.

D. NECROPSY

No gross pathological abnormalities were noted in all animals.

E. ANALYTICAL MEASUREMENTS

The exposure conditions are summarized in Table 5.2.3-8.

Table 5.2.3-8: Exposure conditions

Test group (mg/L)	Supply air (m^3/h)	Exhaust air (m^3/h)	Temperature (°C)	Relative humidity (%)	Substance flow (g/h)[4]
0.42	1.5	1.35	21.6±0.5	45.5±2.6	7.4
1.16	1.5	1.35	21.3±0.8	48.2±3.4	11.6

[4] corrected for 1 % (w/w) Aerosil.

Test atmosphere concentrations are presented in Table 5.2.3-9.

Table 5.2.3-9: Atmosphere concentrations

Mean achieved (mg/L)	Standard deviation	Nominal (mg/L)
0.42	0.03	4.9
1.16	0.24	7.7

The measurements of particle-size distribution revealed mass median aerodynamic diameters (MMAD) of 2.6 – 2.8µm with a geometric standard deviation of 2.8 -3.7 respectively (see Table 5.2.3-10).

Table 5.2.3-10: Particle size distribution

Mean achieved (analytical) atmosphere concentration (mg/L)	Mean mass median aerodynamic diameter (µm)	Inhalable fraction (% <3 µm)	Standard deviation
0.42	2.6	54.3	3.7
1.16 (sample 1)	2.8	52.8	2.9
1.16 (sample 2)	2.6	55.1	2.8

III. CONCLUSION

Under the conditions of this study the 4 hour inhalation LC_{50} of alpha-cypermethrin for male and female rats was estimated to be 1.33 mg/L.

CA 5.2.4 Skin irritation

[redacted] 1993 (AL-410-003)

Guidelines: In compliance with the test method B.4 of directive 92/69/EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

6 New Zealand white rabbits were exposed to 0.5 g alpha-cypermethrin (95.6%, cis1:cis2 ratio: 3:97) applied to the intact skin and covered with a semi-occlusive dressing during 4 h.
An overall mean erythema and edema score of 0.222 and 0, respectively, were determined over the 24, 48 and 72 h reading time points.

Conclusion
Under the conditions of this study, alpha-cypermethrin was considered not to be skin-irritating.

[redacted] 1994a

Guidelines: Not fully in compliance with the test method B.4 of directive 92/69/EEC
Deviations: Substance was applied on abraded skin
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

3 New Zealand white rabbits were exposed to 0.5 g alpha-cypermethrin (95% - 99.5%) applied to the shorn skin and covered with a semi-occlusive dressing during 4 h.
For both, erythema and edema, scores of 0/0/0 were determined for the three animals over the 24, 48 and 72 h reading time points.

Conclusion
Under the conditions of this study, alpha-cypermethrin was considered not to be skin-irritating.

Report:	CA 5.2.4/1 [redacted] 2005a BAS 310 I (Alpha-Cypermethrin) - Acute dermal irritation/corrosion in rabbits 2005/1011570
Guidelines:	OECD 404, EEC 2004/73 B.4, EPA 870.2500, JMAFF No 12 Nosan No 8147
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an acute dermal irritation study, the skin irritation/corrosion potential of BAS 310 I (batch COD-000166, purity: 99.3%) was tested. The intact clipped skin of 3 New Zealand White rabbits was exposed to 0.5 g of the unchanged test substance for 4 hours covered with a semi-occlusive dressing.
The cutaneous reactions were assessed immediately after removal of the patch, approximately 1, 24, 48 and 72 hours after removal of the patch and then in weekly intervals until day 14 after treatment.
Slight erythema, observed in all animals immediately after removal of the patch, increased to moderate in all animals within 1 hour. Moderate erythema persisted in one animal up to day 7 and decreased to slight erythema in two animals at the 24- hour reading. Slight erythema persisted in one of these animals until 72 hours. In one animal severe scaling was noticed on day 7. The cutaneous reactions were reversible in one animal within 48 hours, in another animal within 7 days and in the third animal within 14 days after removal of the patch.
Individual mean skin irritation scores (24 to 72 hours) for each of the three animals were:

- Erythema: 1.0-0.3-2.0
- Edema: 0.0 each

- Mean scores for erythema and edema: 1.1-0.0

Consequently, alpha-cypermethrin **does not show a relevant skin irritation potential** under the test conditions chosen.

(DocID 2005/1011570)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BAS 310 I

Description:	Solid; powder/white
Lot/Batch #:	COD-000166
Purity:	99.3 %
Stability of test compound:	Stable

2. Vehicle: The test substance was administered unchanged.

3. Test animals:

Species:	Rabbit
Strain:	New Zealand white A 1077 INRA (SPF)
Sex:	2 males / 1 female
Age:	ca. 6 months
Weight at dosing:	males: 3.60 and 3.70 kg, female: 3.80 kg
Source:	Centre Lago S. A., 01540 Vonnas, France
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät (Kaninchen & Meerschweinchenhaltung "GLP"), Provimi Kliba SA, Kaiseraugst, Basel, Switzerland (about 130 g/animal per day)
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages with grating, floor area: 3000 cm²

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 22-Feb-2005 - 12-Apr-2005

2. In-vitro pre-test: No *in vitro* pre-test was conducted. *In vivo* data already existed.

3. Animal assignment and treatment:
The potential of BAS 310 I to cause acute dermal irritation or corrosion was assessed by a single topical application of 0.5 g of the unchanged test substance for 4 hours to the intact untreated skin of three New Zealand White rabbits using a patch of 2.5 cm x 2.5 cm. The test substance was covered with a test patch (Idealbinde, Pfaelzische Verbandstoff-Fabrik, Kaiserslautern) and Fixomull stretch (adhesive fleece, Beiersdorf AG). The test substance was removed at the end of the exposure period with Lutrol and Lutrol/water (1:1).
At least 24 hours before treatment, the dorsolateral part of the trunk of the animals was clipped. The cutaneous reactions were assessed immediately after removal of the patch, approximately 1, 24, 48 and 72 hours, on day 7 and maximally up to day 14 after removal of the patch.
Body weights were measured before application of the test substance and after the last reading. The animals were checked for mortality and morbidity twice on working days and once daily at weekends and on public holidays.

II. RESULTS AND DISCUSSION

Slight erythema (grade 1), observed in all animals immediately after removal of the patch, increased to moderate (grade 2) in all animals within 1 hour. Moderate erythema persisted in one animal up to day 7 and decreased to slight erythema (grade 1) in two animals at the 24-hour reading. Slight erythma persisted in one of these animals until 72 hours. In one animal severe scaling was noticed on day 7. The cutaneous reactions were reversible in one animal within 48 hours, in another animal within 7 days and in the third animal within 14 days after removal of the patch. Mean scores over 24, 48 and 72 hours for each animal were 1.0, 0.3 and 2.0 for erythema and 0.0 for edema. The overall 24 to 72 hour skin irritation scores were 1.1 for erythema and 0.0 for edema. Individual and mean irritation scores after 4 hour dermal application of BAS 310 I are presented in Table 5.2.4-1.

Table 5.2.4-1: Individual and mean skin irritation scores after 4 hour dermal application of BAS 310 I

Readings	Animal	Erythema	Edema	Additional findings
0 h	01	1	0	
	02	1	0	
	03	1	0	
1 h	01	2	0	
	02	2	0	
	03	2	0	
24 h	01	1	0	
	02	1	0	
	03	2	0	
48 h	01	1	0	
	02	0	0	
	03	2	0	
72 h	01	1	0	
	02	0	0	SD
	03	2	0	
7 d	01	0	0	SD
	03	2	0	Severe scaling
14 d	03	0	0	
Mean 24 - 72 h	01	1.0	0.0	
	02	0.3	0.0	
	03	2.0	0.0	
Mean		1.1	0.0	

SD: Study discontinued because the animal was free of symptoms

III. CONCLUSION

Based on the findings of this study, the BAS 310 I showed no relevant skin irritation potential to rabbits under the test conditions chosen.

Report:	CA 5.2.4/2 [redacted] 2005b BAS 310 I (Alpha-Cypermethrin) - Acute dermal irritation / corrosion in rabbits 2005/1011606
Guidelines:	OECD 404, EEC 2004/73 B.4, EPA 870.2500, JMAFF No 12 Nosan No 8147
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an acute dermal irritation study, the skin irritation/corrosion potential of BAS 310 I (batch COD-000165, purity: 98.8%) was tested. The intact clipped skin of 3 New Zealand White rabbits was exposed to 0.5 g of the unchanged test substance for 4 hours covered with a semi-occlusive dressing.
The cutaneous reactions were assessed immediately after removal of the patch, approximately 1, 24, 48 and 72 hours after removal of the patch and then in weekly intervals until day 14 after treatment.
Slight erythema was observed in all animals immediately after removal of the patch and persisted in one animal up to 24 hours. Moderate erythema was noted in two animals from 1 hour up to 48 hours and in one animal up to 72 hours. Moderate erythema increased to marked in one animal after 72 hours and decreased to moderate again on day 7. In this animal moderate edema was observed after 72 hours, too. Additionally scaling or severe scaling was noted in two animals on day 7. The cutaneous reactions were reversible within the 14 day observation period. Mean scores over 24, 48 and 72 hours for each animal were 2.0, 2.3 and 0.3 for erythema and 0.0, 0.7 and 0.0 for edema. The overall 24 to 72 hour skin irritation scores were 1.6 for erythema and 0.2 for edema. Individual mean skin irritation scores (24 to 72 hours) for each of the three animals were:

- Erythema: 2.0-2.3-0.3
- Edema: 0.0-0.7-0.0

- Mean scores for erythema and edema: 1.6 and 0.2, respectively.

Consequently, alpha-cypermethrin **does not show a relevant skin irritation potential** under the test conditions chosen.

(DocID 2005/1011606)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BAS 310 I

Description:	Solid; powder/white
Lot/Batch #:	COD-000165
Purity:	98.8 %
Stability of test compound:	Stable

2. Vehicle: The test substance was administered unchanged.

3. Test animals:

Species:	Rabbit
Strain:	New Zealand white A 1077 INRA (SPF)
Sex:	2 males / 1 female
Age:	ca. 8-9 months
Weight at dosing:	males: 4.01 and 3.73 kg, female: 4.14 kg
Source:	Centre Lago S. A., 01540 Vonnas, France
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät (Kaninchen & Meerschweinchenhaltung "GLP"), Provimi Kliba SA, Kaiseraugst, Basel, Switzerland (about 130 g/animal per day)
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages with grating, floor area: 3000 cm²

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 22-Feb-2005 - 20-Apr-2005

2. In-vitro pre-test: No *in vitro* pre-test was conducted. *In vivo* data already available.

3. Animal assignment and treatment:
The potential of BAS 310 I to cause acute dermal irritation or corrosion was assessed by a single topical application of 0.5 g of the unchanged test substance for 4 hours to the intact untreated skin of three New Zealand White rabbits using a patch of 2.5 cm x 2.5 cm. The test substance was covered with a test patch (Idealbinde, Pfaelzische Verbandstoff-Fabrik, Kaiserslautern) and Fixomull stretch (adhesive fleece, Beiersdorf AG). The test substance was removed at the end of the exposure period with Lutrol and Lutrol/water (1:1).
At least 24 hours before treatment, the dorsolateral part of the trunk of the animals was clipped. The cutaneous reactions were assessed immediately after removal of the patch, approximately 1, 24, 48 and 72 hours, on day 7 and maximally up to day 14 after removal of the patch.
Body weights were measured before application of the test substance and after the last reading. The animals were checked for mortality and morbidity twice on working days and once daily at weekends and on public holidays.

II. RESULTS AND DISCUSSION

Slight erythema (grade 1) was observed in all animals immediately after removal of the patch and persisted in one animal up to 24 hours. Moderate erythema (grade 2) was noted in two animals from 1 hour up to 48 hours and in one animal up to 72 hours. Moderate erythema increased to marked (grade 3) in one animal after 72 hours and decreased to moderate again on day 7. In this animal moderate edema (grade 2) was observed after 72 hours, too. Additionally scaling or severe scaling was noted in two animals on day 7. The cutaneous reactions were reversible within 48 hours, 7 days or 14 days after removal of the patch in one animal each. Mean scores over 24, 48 and 72 hours for each animal were 2.0, 2.3 and 0.3 for erythema and 0.0, 0.7 and 0.0 for edema. The overall 24 to 72 hour skin irritation scores were 1.6 for erythema and 0.2 for edema. Individual and mean irritation scores after 4 hour dermal application of BAS 310 I are presented in Table 5.2.4-2.

Table 5.2.4-2: Individual and mean skin irritation scores after 4 hour dermal application of BAS 310 I

Readings	Animal	Erythema	Edema	Additional findings
0 h	01	1	0	
	02	1	0	
	03	1	0	
1 h	01	2	0	
	02	2	0	
	03	1	0	
24 h	01	2	0	
	02	2	0	
	03	1	0	
48 h	01	2	0	
	02	2	0	
	03	0	0	
72 h	01	2	0	
	02	3	2	
	03	0	0	SD
7 d	01	0	0	Scaling, SD
	02	2	0	Severe scaling
14 d	02	0	0	
Mean 24 - 72 h	01	2.0	0.0	
	02	2.3	0.7	
	03	0.3	0.0	
Mean		1.6	0.2	

SD: Study discontinued because the animal was free of symptoms

III. CONCLUSION

Based on the findings of this study, the BAS 310 I showed no classification relevant skin irritation potential to rabbits under the test conditions chosen.

CA 5.2.5 Eye irritation

[redacted] **1993 (AL-410-003)**

Guidelines: In compliance with the test method B.5 of directive 92/69/EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

0.1 mL (45 mg) of alpha-cypermethrin (95.6%, cis1:cis2 ratio: 3:97) was instilled into the conjunctival sac of the eye of 6 New Zealand white rabbits.
All rabbits developed injection of the conjunctival blood vasculature and an ocular discharge within 1 hours of treatment. One animal showed injection of the conjunctival blood vasculature, chemosis sufficient to cause partial eversion of the eyelids and a slight ocular discharge. These effects reversed within 72 h. For corneal opacity and iris and a mean overall score of 0 was determined for the unwashed eye over the 24, 48 and 72 h reading time points. For erythema and chemosis values of 0.1 and 0.2 were determined.

Conclusion
Under the conditions of this study, alpha-cypermethrin was considered not to be eye-irritating.

[redacted] **1994b**

Guidelines: In compliance with the test method B.5 of directive 92/69/EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

0.1 g of alpha-cypermethrin (95 - 99.5%) was instilled into the conjunctival sac of the right eye of 3 New Zealand white rabbits. The eyes were rinsed 24 h after treatment.
For corneal opacity and iris an overall score of 0/0/0 was determined over the 24, 48 and 72 h reading time points. For conjunctival redness and chemosis values of 0.3/1.3/0 and 2/2.3/0.66 were determined.

Conclusion
Under the conditions of this study, alpha-cypermethrin was considered not to be eye-irritating.

Remark: The scores for chemosis would justify a classification for eye irritation. Similar effects were neither produced in the study of [redacted] 1993 nor in the studies presented newly in the AIR III process and therefore a classification for eye irritation is not considered justified for the nowadays produced material.

Report:	CA 5.2.5/1 [redacted] 2005c BAS 310 I (Alpha-Cypermethrin) - Acute eye irritation in rabbits 2005/1011571
Guidelines:	OECD 405, EEC 2004/73 B.5, EPA 870.2400, JMAFF No 12 Nosan No 8147
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an eye irritation study, the eye irritation/corrosion potential of BAS 310 I (batch COD-000166, purity: 99.3%) was determined by instillation of 0.1 mL bulk volume (about 40 mg) of the unchanged test substance into the conjunctival sac of the right eye of three New Zealand White rabbits. The application of the test substance was performed in a stepwise procedure starting with one animal and supplementing two additional animals. About 1 hour after application the eye was rinsed with tap water.
The ocular reactions were assessed approximately 1, 24, 48 and 72 hours after the administration of the test substance.
Moderate conjunctival redness, observed in all animals 1 and 24 hours after application, decreased to slight in all animals at the 48-hour reading. Moderate conjunctival chemosis was noted in all animals at the 1-hour reading. Slight conjunctival chemosis was observed in one animal after 24 hours. Slight discharge was seen in all animals after 1 hour and in one animal after 24 hours. The ocular reactions were reversible in all animals within 72 hours.
Individual mean eye irritation scores (24 to 72 hours) for each of the three animals were:

- Corneal opacity: 0.0 each
- Iritis: 0.0 each
- Redness: 1.0 each
- Chemosis 0.3-0.0-0.0

- Mean scores for corneal opacity, iritis, redness and chemosis: 0-0-1-0.1

Consequently, alpha-cypermethrin is **not an eye irritant** under the conditions of this study.
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DocID (2005/1011571)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BAS 310 I

Description:	Solid; powder/white
Lot/Batch #:	COD-000166
Purity:	99.3%
Stability of test compound:	Stable

2. Vehicle: The test substance was administered undiluted.

3. Test animals:

Species:	Rabbit
Strain:	New Zealand white A 1077 INRA (SPF)
Sex:	one male / two females
Age:	about 3 months
Weight at dosing:	2.41 - 2.77 kg
Source:	Centre Lago S.A., 01540 Vonnas, France
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät, Provimi Kliba SA, Kaiseraugst, Basel, Switzerland (about 130 g/animal per day)
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages with grating, floor area: 3000 cm²

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 22-Feb-2005 - 12-Apr-2005

2. In-vitro pre-test: No *in vitro* pre-test was conducted. *In vivo* data already existed.

3. Animal assignment and treatment:
The potential of BAS 310 I to cause acute eye irritation/corrosion was assessed by instillation of 0.1 mL of the undiluted test substance (about 40 mg of the comminuted test substance) into the conjunctival sac of the right eye. The left eye, which remained untreated, served as the negative control. About 1 hours after application of the test substance, the treated eye was rinsed with 3 to 6 mL of hand warm tap water for 1 to 2 minutes using a syringe with a blunt probe. The ocular reactions were assessed approximately 1, 24, 48 and 72 hours and then in weekly intervals maximally up to day 28 after the administration of the test substance.
Body weights were determined shortly prior to application. The animals were checked for mortality and morbidity twice on working days and once daily at weekends and on public holidays.

II. RESULTS AND DISCUSSION

Moderate conjunctival redness (grade 2), observed in all animals 1 and 24 hours after application, decreased to slight (grade 1) in all animals at the 48-hour reading. Moderate conjunctival chemosis (grade 2) was noted in all animals at the 1-hour reading. Slight conjunctival chemosis (grade 1) was observed in one animal after 24 hours. Slight discharge (grade 1) was seen in all animals after 1 hour and in one animal after 24 hours. The ocular reactions were reversible in all animals within 72 hours. Mean scores calculated for each animal over 24, 48 and 72 hours were 0.0 for corneal opacity and for iris lesions, 1.0 for redness of the conjunctiva and 0.3, 0.0 and 0.0 for chemosis.
For details regarding the individual and mean scores as well as additional findings see Table 5.2.5-1.

Table 5.2.5-1: Individual and mean eye irritation scores after ocular application of BAS 310 I

Readings	Animal	Cornea		Iris	Conjunctiva			Additional findings
		Opacity	Area involved		Redness	Chemosis	Discharge	
1 h	01	0	0	0	2	2	1	-
	02	0	0	0	2	2	1	-
	03	0	0	0	2	2	1	-
24 h	01	0	0	0	2	1	1	-
	02	0	0	0	2	0	0	-
	03	0	0	0	2	0	0	-
48 h	01	0	0	0	1	0	0	-
	02	0	0	0	1	0	0	-
	03	0	0	0	1	0	0	-
72 h	01	0	0	0	0	0	0	-
	02	0	0	0	0	0	0	-
	03	0	0	0	0	0	0	-
Mean 24 - 72 h	01	0.0		0.0	1.0	0.3		
	02	0.0		0.0	1.0	0.0		
	03	0.0		0.0	1.0	0.0		
Mean		0.0		0.0	1.0	0.1		

III. CONCLUSION

Based on the findings of this study, BAS 310 I does not show an eye irritation potential to rabbits under the test conditions chosen.

Report:	CA 5.2.5/2 [redacted] 2005d BAS 310 I (Alpha-Cypermethrin) - Acute eye irritation in rabbits 2005/1011607
Guidelines:	OECD 405, EEC 2004/73 B.5, EPA 870.2400, JMAFF No 12 Nosan No 8147
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In an eye irritation study, the eye irritation/corrosion potential of BAS 310 I (batch COD-000165, purity: 98.8%) was determined by instillation of 0.1 mL bulk volume (about 40 mg) of the unchanged test substance into the conjunctival sac of the right eye of three New Zealand White rabbits. The application of the test substance was performed in a stepwise procedure starting with one animal and supplementing two additional animals. About 1 hour after application the eye was rinsed with tap water.
The ocular reactions were assessed approximately 1, 24, 48 and 72 hours after the administration of the test substance.
Moderate conjunctival redness was observed in all animals 1 hour after application and persisted in one animal up to 24 hours. Moderate conjunctival redness decreased to slight in two animals after 24 hours and in one animal at the 48-hour reading. Slight or moderate conjunctival chemosis was noted in all animals 1 hour after application. Moderate conjunctival chemosis decreased to slight in one animal after 24 hours. Slight discharge was observed in all animals after 1 hour and persisted in one animal up to 24 hours. The ocular reactions were reversible in two animals within 48 hours and in one animal within 72 hours after application.

- Corneal opacity: 0.0 each
- Iritis: 0.0 each
- Redness: 1.0-0.3-0.3
- Chemosis 0.3-0.0-0.0

- Mean scores for corneal opacity, iritis, redness and chemosis: 0-0-0.6-0.1

Consequently, alpha-cypermethrin is **not an eye irritant** under the conditions of this study
.

DocID (2005/1011607)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BAS 310 I

Description:	Solid; powder/white
Lot/Batch #:	COD-000165
Purity:	98.8%
Stability of test compound:	Stable

2. Vehicle: The test substance was administered undiluted.

3. Test animals:

Species:	Rabbit
Strain:	New Zealand white A 1077 INRA (SPF)
Sex:	one male / two females
Age:	about 3 months
Weight at dosing:	2.60 - 2.77 kg
Source:	Centre Lago S.A., 01540 Vonnas, France
Acclimation period:	At least 5 days
Diet:	Kliba-Labordiät, Provimi Kliba SA, Kaiseraugst, Basel, Switzerland (about 130 g/animal per day)
Water:	Tap water, ad libitum
Housing:	Single housing in stainless steel wire mesh cages with grating, floor area: 3000 cm^2

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 22-Feb-2005 - 21-Apr-2005

2. In-vitro pre-test: No *in vitro* pre-test was conducted.

3. Animal assignment and treatment:
The potential of BAS 310 I to cause acute eye irritation/corrosion was assessed by instillation of 0.1 mL of the undiluted test substance (about 38 mg of the comminuted test substance) into the conjunctival sac of the right eye. The left eye, which remained untreated, served as the negative control. About 1 hours after application of the test substance, the treated eye was rinsed with 3 to 6 mL of hand warm tap water for 1 to 2 minutes using a syringe with a blunt probe. The ocular reactions were assessed approximately 1, 24, 48 and 72 hours and then in weekly intervals maximally up to day 28 after the administration of the test substance.
Body weights were determined shortly prior to application and after the last reading. The animals were checked for mortality and morbidity twice on working days and once daily at weekends and on public holidays.

II. RESULTS AND DISCUSSION

Moderate conjunctival redness (grade 2) was observed in all animals 1 hour after application and persisted in one animal up to 24 hours. Moderate conjunctival redness decreased to slight (grade 1) in two animals after 24 hours and in one animal at the 48-hour reading. Slight or moderate conjunctival chemosis (grade 1 or 2) was noted in all animals 1 hour after application. Moderate conjunctival chemosis decreased to slight in one animal after 24 hours. Slight discharge (grade 1) was observed in all animals after 1 hour and persisted in one animal up to 24 hours. The ocular reactions were reversible in two animals within 48 hours and in one animal within 72 hours after application.
Mean scores calculated for each animal over 24, 48 and 72 hours were 0.0 for corneal opacity and for iris lesions, 1.0, 0.3 and 0.3 for redness of the conjunctiva and 0.3, 0.0 and 0.0 for chemosis. For details regarding the individual and mean scores as well as additional findings see Table 5.2.5-2.

Table 5.2.5-2: Individual and mean eye irritation scores after ocular application of BAS 310 I

Readings	Animal	Cornea		Iris	Conjunctiva			Additional findings
		Opacity	Area involved		Redness	Chemosis	Discharge	
1 h	01	0	0	0	2	2	1	-
	02	0	0	0	2	2	1	-
	03	0	0	0	2	1	1	-
24 h	01	0	0	0	2	1	1	-
	02	0	0	0	1	0	0	-
	03	0	0	0	1	0	0	-
48 h	01	0	0	0	1	0	0	-
	02	0	0	0	0	0	0	-
	03	0	0	0	0	0	0	-
72 h	01	0	0	0	0	0	0	-
	02	0	0	0	0	0	0	-
	03	0	0	0	0	0	0	-
Mean 24 - 72 h	01	0.0		0.0	1.0	0.3		
	02	0.0		0.0	0.3	0.0		
	03	0.0		0.0	0.3	0.0		
Mean		0.0		0.0	0.6	0.1		

III. CONCLUSION

Based on the findings of this study, BAS 310 I **does not show an eye irritation potential** to rabbits under the test conditions chosen.

CA 5.2.6 Skin sensitisation

[redacted] 1993 (AL-410-003)

Guidelines: In compliance with the test method B.6 of directive 84/449//EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

20 Dunkin Hartley guinea pigs received an intradermal induction of alpha-cypermethrin (batch 02156, drum 1085, 95.6%, cis1:cis2 ratio: 3:97) as 2% solution in corn oil:FCA. Percutaneous induction and challenge were performed with a 50% test substance solution in corn oil. A group of 10 animals served as control group.
None of the 20 test animals showed any positive response at either 24 or 48 h after removal of the challenge patches.

Conclusion
Under the conditions of this study, alpha-cypermethrin was considered not to be skin-sensitizing.

[redacted] 1994c

Guidelines: In compliance with the test method B.6 of directive 84/449//EEC
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.
20 Dunkin Hartley guinea pigs received an intradermal induction of alpha-cypermethrin (95% - 99.5%) 5% in propylene glycol. Percutaneous induction and challenge were performed with 50% test substance in petrolatum. A group of 20 animals served as control group.
An allergenicity score of 25% was reported resulting from erythema observed at 24 h after challenge in 4 animals (very slight erythema) and 1 animal (slight erythema).

Conclusion
Under the conditions of this study, alpha-cypermethrin was considered not to be skin-sensitizing.

Report:	CA 5.2.6/1 [redacted] 2005f BAS 310 I (Alpha-Cypermethrin) - Maximization test in guinea pigs 2005/1011608
Guidelines:	EEC 96/54 B 6, OECD 406, EPA 870.2600, JMAFF No 12 Nosan No 8147
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

BAS 310 I (batch: COD-000165, purity: 98.8%) was tested in a maximization test in female Dunkin Hartley guinea pigs (HsdPoc: DH). Based on the results of a pre-test, the intradermal induction was performed with a 5% test item preparation in 1% CMC-solution into the neck region of the animals. The epicutaneous induction (7 days after intradermal induction) and the challenge exposure (14 days after epicutaneous induction) were performed with a 50 % test item preparation. The study was performed in 5 control and 10 test group animals. Readings were performed 24 hours after the intradermal injection and 24 hours after removal of the patch with regard to epicutaneous induction. The challenge was carried out with the test substance preparation applied for 24 hours to the intact skin of the flank under occlusive conditions. 24 and 48 hours after removal of the patch, skin readings were performed. A positive control with a known sensitizer was not included into the study. However, studies with Alpha-Hexylcinnamaldehyde (techn. 85%) are regularly performed as reliability check in the laboratory and showed that the test system is able to detect sensitizing compounds under the laboratory conditions by showing a sensitization rate of 100% in the guinea pig strain.

The intradermal induction caused moderate and confluent to intense erythema and swelling at the injection sites of the test substance preparation in all test group animals. After the epicutaneous induction, incrustation, partially open (caused by the intradermal induction) could be observed in addition to moderate and confluent erythema and swelling in all test group animals. The challenge resulted in skin reactions in 0/5 control and 0/10 test group animals.

Based on the results of this study alpha-cypermethrin **does not have a sensitizing effect** on the skin of the guinea pig in the Maximization Test under the test conditions chosen.

(DocID 2005/1011608)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I
Description:	Solid; powder/white
Lot/Batch #:	COD-000165
Purity:	98.8%
Stability of test compound:	Stable

2. Vehicle / Positive control:

Vehicles:
1% CMC (cleaned sodium carboxymethylcellulose) in doubly distilled water
Positive control: Alpha-Hexylcinnamaldehyde

3. Test animals:

Species:	Guinea Pig
Strain:	Dunkin Hartley, HsdPoc: DH
Sex:	female
Age:	6 - 8 weeks
Weight at dosing (mean):	424 - 507 g
Source:	Harlan Winkelmann, Gartenstr. 27, 33178 Borchen, Germany
Acclimation period:	14 days
Diet:	Kliba Labordiät (Kaninchen/ Meerschweinchen-Haltungsdiät) Provimi Kliba SA, Kaiseraugst, Switzerland, ad libitum
Water:	Tap water, ad libitum
Housing:	Groups of 5 animals were housed in stainless steel wire mesh cages with plastic-coated grating, minimum floor area: 2000 cm²

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 11-Mar-2015 - 26-Apr-2005

2. Animal assignment and treatment:

The skin sensitizing potential of BAS 310 I was assessed using the Maximization Test based on the method of Magnusson and Kligman. For this, female guinea pigs were randomly allocated to groups. Five animals were used as control group animals and 10 animals in the test group. Based on the results of a pre-test, animals were intradermally induced with 5% test substance preparations. Epidermal induction and challenge were conducted with 50% test substance preparations. The homogeneity and the stability of the test substance in the vehicle were determined indirectly by the concentration control analysis. The fur was clipped at least 2 hours before each test item application at the appropriate application sites. If necessary, the fur was additionally clipped at least 2 hours before evaluation of the skin reactions.

3. Clinical observation:
Mortality was checked once each day. No detailed clinical examination of the individual animals was performed but any obvious signs of systemic toxicity and/or local inflammation at the application sites were recorded.

4. Body weights:
Individual body weights were determined on day 0 on the last day of observation.

5. Pre-test:
A 5% test substance preparation (TS in 1% CMC-solution in doubly distilled water) was intradermally injected to two animals. 6 intradermal injections were applied at the neck region of each animal: front row: 2 injections each of 0.1 mL Freund´s complete adjuvant without test item emulsified with 0.9 % aqueous NaCl-solution in a ratio of 1:1; middle row: 2 injections each of 0.1 mL of a test item preparation in vehicle at the selected concentration; back row: 2 injections each of 0.1 mL Freund's complete adjuvant / vehicle (1 : 1) with test item at the selected concentration. Skin reactions were assessed 24 hours after the beginning of the application. For detecting a possible influence on irritating effects of previous intradermal treatment with Freund's complete adjuvant, animals pretreated with Freund's complete adjuvant / 0.9% aqueous NaCl-solution (1 : 1) each, in the same manner as intradermal pretest referring front row and back row without test substance about 3 weeks prior to the application of the test substance were used. 2 x 2 cm gauze patches (6 layers surgical gauze Ph. Eur. from Lohmann GmbH & Co. KG) containing 0.5 mL of test substance preparation (25 and 50% in 1% CMC-solution in doubly distilled water) were applied to the skin of the flanks under an occlusive dressing. The dressing consisted of rubberized linen patches (4 x 4 cm from Russka), patches of Idealbinde (5 x 5 cm from Pfälzische Verbandstoff-Fabrik) and Fixomull® Stretch (adhesive fleece) from Beiersdorf AG. The animals were exposed for 24 hours and skin readings were performed 1, 24 and 48 h after removal of the patch.

6. Main study – intradermal induction:
Based on the results of the pretest, test group animals received intradermal injections of 5% test substance preparations analogously to the intradermal pretest (see above). Control group animals received the same injections but with the test substance preparation being replaced by the vehicle.

7. Main study – epicutaneous induction:
One week after intradermal induction, 1 mL of the 50% test item preparation was applied to each test group animal under the same conditions as described in the epidermal pretest. The control animals were not treated since the 1% CMC-solution in doubly distilled water used as formulating agent was not expected to influence the result of the study.

8. Main study - challenge:
The challenge was carried out 14 days after the epicutaneous induction. 0.5 mL of the 50% test item preparation was applied to the test and control group animal. The animals were exposed under occlusive conditions as described above for 24 hours and skin readings were performed 24 and 48 h after removal of the patch.

9. Evaluation of results
The number of animals with skin findings at 24 and/or 48 hours after the removal of the patch was taken into account for the determination of the sensitization rate. The evaluation "sensitizing" results if at least 30% of the test animals exhibit skin reactions.

10. Positive controls
A positive control (reliability check) with a known sensitizer was not included in this study. However, a separate study with the positive control Alpha-Hexylcinnamaldehyde (techn. 85%) is regularly performed in the laboratory.

II. RESULTS AND DISCUSSION

A. PRE-TEST

Injections of a 5% test substance preparation in 1% CMC-solution in doubly distilled water caused moderate and confluent erythema and swelling. At the injection sites of a 5% test substance preparation in Freund's complete adjuvant/0.9% aqueous NaCl-solution (1 : 1) intense erythema and swelling were seen. After epidermal induction and challenge no skin findings were observed in the animals treated with 50% and 25% test substance preparations 24 and 48 hours after removal of the patch.

B. OBSERVATIONS

No abnormalities were observed during general observation.

C. BODY WEIGHTS

Body weight gain was not adversely affected during the course of the study.

D. SKIN REACTIONS AFTER INTRADERMAL INDUCTION

Injections of a 5% test item preparation in 1% CMC-solution in doubly distilled water caused moderate and confluent to intense erythema and swelling at the injection sites of the test substance preparation in all test group animals (see Table 5.2.6-1**:**).

Table 5.2.6-1: BAS 310 I - Skin reactions after intradermal induction

Position of injection: Neck region				
Form of application:				
Findings 24 hours after the beginning of application				
Animal #	**Application site**	**A) Freund's complete adjuvant / 0.9% aqueous NaCl Solution (1 : 1)**	**B) Test item 5 % in 1% CMC-solution in doubly distilled water**	**C) Test item 5 % in A)**
411	right	3	2 E	3
	left	3	2 E	3
412	right	3	2 E	3
	left	3	2 E	3
413	right	3	2 E	3
	left	3	2 E	3
414	right	3	2 E	3
	left	3	2 E	3
415	right	3	2 E	3
	left	3	2 E	3
416	right	3	2 E	3
	left	3	2 E	3
417	right	3	2 E	3
	left	3	2 E	3
418	right	3	2 E	3
	left	3	2 E	3
419	right	3	2 E	3
	left	3	2 E	3
420	right	3	2 E	3
	left	3	2 E	3

E: Swelling

E. SKIN REACTIONS AFTER EPICUTANEOUS INDUCTION

The epicutaneous induction with a 50% test substance preparation in 1% CMC-solution in doubly distilled water led to incrustation, partially open (caused by the intradermal induction) and moderate and confluent erythema in all test group animals (see Table 5.2.6-2).

Table 5.2.6-2: BAS 310 I - Skin reactions after epicutaneous induction

Animal #	Test item 50 % in 1% CMC-solution in doubly distilled water
411	2 E K
412	2 E K
413	2 E K
414	2 E K
415	2 E K
416	2 E K
417	2 E K
418	2 E K
419	2 E K
420	2 E K

E: Swelling; K: incrustation, partially open

F. SKIN REACTIONS AFTER CHALLENGE

The challenge with a 50% test substance preparation in 1% CMC-solution in doubly distilled water did not cause any skin reactions in animals of the control group and test group 24 and 48 hours after removal of the patch (see Table 5.2.6-3). Since no borderline results were observed, a 2nd challenge was not performed.

Table 5.2.6-3: BAS 310 I - Skin reactions after challenge

	Challenge			
Skin findings	**Control group**		**Test group**	
	24 h	**48 h**	**24 h**	**48 h**
Grade 0	5/5#	5/5	10/10	10/10

[#] x/y = number of findings / number of animals tested

G. POSITIVE CONTROL

The positive control Alpha-Hexylcinnamaldehyde showed a sensitization rate of 100% in the guinea pig strain. The results of the latest study conducted with the positive control are presented in Table 5.2.6-4.

Table 5.2.6-4: Skin reactions after challenge with the positive control

	Challenge			
Skin findings	**Alpha-Hexylcinnamaldehyde (techn. 85%) 5% in Lutrol® E 400**		**Vehicle Control: Lutrol® E 400**	
	24 h	**48 h**	**24 h**	**48 h**
Control group	0/5#	0/5	0/5	0/5
Test group	10/10	9/10	0/10	0/10

[#] x/y = number of positive reactions/number of animals tested (reading at 24 h and/or 48 h after the removal of the patch)

III. CONCLUSION

Based on the results of this study it is concluded that BAS 310 I does not have sensitizing properties in the guinea pig maximization test under the test conditions chosen. 0% of the animals were considered positive after challenge application.

Report:	CA 5.2.6/2 [redacted] 2005g BAS 310 I (Alpha-Cypermethrin) - Maximization test in guinea pigs 2005/1011572
Guidelines:	EEC 96/54 B 6, OECD 406, EPA 870.2600, JMAFF No 12 Nosan No 8147
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

BAS 310 I (batch: COD-000166, purity: 99.3%) was tested in a maximization test in female Dunkin Hartley guinea pigs (HsdPoc: DH). Based on the results of a pre-test, the intradermal induction was performed with a 5% test item preparation in 1% CMC-solution into the neck region of the animals. The epicutaneous induction (7 days after intradermal induction) and the challenge exposure (14 days after epicutaneous induction) were performed with a 50 % test item preparation. The study was performed in 5 control and 10 test group animals. Readings were performed 24 hours after the intradermal injection and 24 hours after removal of the patch with regard to epicutaneous induction. The challenge was carried out with the test substance preparation applied for 24 hours to the intact skin of the flank under occlusive conditions. 24 and 48 hours after removal of the patch, skin readings were performed. A positive control with a known sensitizer was not included into the study. However, studies with Alpha-Hexylcinnamaldehyde (techn. 85%) are regularly performed as reliability check in the laboratory and showed that the test system is able to detect sensitizing compounds under the laboratory conditions by showing a sensitization rate of 100% in the guinea pig strain.

The intradermal induction caused moderate and confluent to intense erythema and swelling at the injection sites of the test substance preparation in all test group animals. After the epicutaneous induction, incrustation, partially open (caused by the intradermal induction) could be observed in addition to moderate and confluent erythema and swelling in all test group animals. The challenge resulted in skin reactions in 0/5 control and 0/10 test group animals.

Based on the results of this study alpha-cypermethrin **does not have a sensitizing effect** on the skin of the guinea pig in the Maximization Test under the test conditions chosen.

(DocID 2005/1011572)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BAS 310 I

Description:	Solid; powder/white
Lot/Batch #:	COD-000166
Purity:	99.3%
Stability of test compound:	Stable

2. Vehicle / Positive control:

Vehicles:
1% CMC (cleaned sodium carboxymethylcellulose) in doubly distilled water
Positive control: Alpha-Hexylcinnamaldehyde

3. Test animals:

Species:	Guinea Pig
Strain:	Dunkin Hartley, HsdPoc: DH
Sex:	female
Age:	6 - 8 weeks
Weight at dosing (mean):	415 - 510 g
Source:	Harlan Winkelmann, Gartenstr. 27, 33178 Borchen, Germany (For the intradermal pretest animals of the strain/quality "Dunkin Hartley, Crl:HA" of the supplier Charles River Deutschland GmbH, 88353 Kisslegg were used.)
Acclimation period:	14 days
Diet:	Kliba Labordiät (Kaninchen/ Meerschweinchen-Haltungsdiät) Provimi Kliba SA, Kaiseraugst, Switzerland, ad libitum
Water:	Tap water, ad libitum
Housing:	Groups of 5 animals were housed in stainless steel wire mesh cages with plastic-coated grating, minimum floor area: 2000 cm²

Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	30 - 70 %
Air changes:	Central air-conditioning
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 11-Mar-2015 - 28-Apr-2005

2. Animal assignment and treatment:
The skin sensitizing potential of BAS 310 I was assessed using the Maximization Test based on the method of Magnusson and Kligman. For this, female guinea pigs were randomly allocated to groups. Five animals were used as control group animals and 10 animals in the test group. Based on the results of a pre-test, animals were intradermally induced with 5% test substance preparations. Epidermal induction and challenge were conducted with 50% test substance preparations. The homogeneity and the stability of the test substance in the vehicle were determined indirectly by the concentration control analysis. The fur was clipped at least 2 hours before each test item application at the appropriate application sites. If necessary, the fur was additionally clipped at least 2 hours before evaluation of the skin reactions.

3. Clinical observation:
Mortality was checked twice each workday and once on Saturdays, Sundays and on public holidays. No detailed clinical examination of the individual animals was performed but any obvious signs of systemic toxicity and/or local inflammation at the application sites were recorded.

4. Body weights:
Individual body weights were determined on day 0 on the last day of observation.

5. Pre-test:
A 5% test substance preparation (TS in 1% CMC-solution in doubly distilled water) was intradermally injected to two animals. 6 intradermal injections were applied at the neck region of each animal: front row: 2 injections each of 0.1 mL Freund´s complete adjuvant without test item emulsified with 0.9 % aqueous NaCl-solution in a ratio of 1:1; middle row: 2 injections each of 0.1 mL of a test item preparation in vehicle at the selected concentration; back row: 2 injections each of 0.1 mL Freund's complete adjuvant / vehicle (1 : 1) with test item at the selected concentration. Skin reactions were assessed 24 hours after the beginning of the application. For detecting a possible influence on irritating effects of previous intradermal treatment with Freund's complete adjuvant, animals pretreated with Freund's complete adjuvant / 0.9% aqueous NaCl-solution (1 : 1) each, in the same manner as intradermal pretest referring front row and back row without test substance about 3 weeks prior to the application of the test substance were used. 2 x 2 cm gauze patches (6 layers surgical gauze Ph. Eur. from Lohmann GmbH & Co. KG) containing 0.5 mL of test substance preparation (25 and 50% in 1% CMC-solution in doubly distilled water) were applied to the skin of the flanks under an occlusive dressing. The dressing consisted of rubberized linen patches (4 x 4 cm from Russka), patches of Idealbinde (5 x 5 cm from Pfälzische Verbandstoff-Fabrik) and Fixomull® Stretch (adhesive fleece) from Beiersdorf AG. The animals were exposed for 24 hours and skin readings were performed 1, 24 and 48 h after removal of the patch.

6. Main study – intradermal induction:
Based on the results of the pretest, test group animals received intradermal injections of 5% test substance preparations analogously to the intradermal pretest (see above). Control group animals received the same injections but with the test substance preparation being replaced by the vehicle.

7. Main study – epicutaneous induction:
One week after intradermal induction, 1 mL of the 50% test item preparation was applied to each test group animal under the same conditions as described in the epidermal pretest. The control animals were not treated since the 1% CMC-solution in doubly distilled water used as formulating agent was not expected to influence the result of the study.

8. Main study - challenge:
The challenge was carried out 14 days after the epicutaneous induction. 0.5 mL of the 50% test item preparation was applied to the test and control group animal. The animals were exposed under occlusive conditions as described above for 24 hours and skin readings were performed 24 and 48 h after removal of the patch.

9. Evaluation of results
The number of animals with skin findings at 24 and/or 48 hours after the removal of the patch was taken into account for the determination of the sensitization rate. The evaluation "sensitizing" results if at least 30% of the test animals exhibit skin reactions.

10. Positive controls
A positive control (reliability check) with a known sensitizer was not included in this study. However, a separate study with the positive control Alpha-Hexylcinnamaldehyde (techn. 85%) is regularly performed in the laboratory.

II. RESULTS AND DISCUSSION

A. PRE-TEST

Injections of a 5% test substance preparation in 1% CMC-solution in doubly distilled water caused moderate and confluent erythema and swelling. At the injection sites of a 5% test substance preparation in Freund's adjuvant/0.9% aqueous NaCl-solution (1 : 1) intense erythema and swelling were seen. No skin findings were observed in the animals treated with a 50% and 25% test substance preparation 24 and 48 hours after removel of the patch.

B. OBSERVATIONS

No abnormalities were observed during general observation.

C. BODY WEIGHTS

Body weight gain was not adversely affected during the course of the study.

D. SKIN REACTIONS AFTER INTRADERMAL INDUCTION

Injections of a 5% test item preparation in 1% CMC-solution in doubly distilled water caused moderate and confluent to intense erythema and swelling at the injection sites of the test substance preparation in all test group animals (see Table 5.2.6-5).

Table 5.2.6-5: BAS 310 I - Skin reactions after intradermal induction

Position of injection: Neck region				
Form of application:				
Findings 24 hours after the beginning of application				
Animal #	**Application site**	**A) Freund's complete adjuvant / 0.9% aqueous NaCl Solution (1 : 1)**	**B) Test item 5 % in 1% CMC-solution in doubly distilled water**	**C) Test item 5 % in A)**
711	right	3	2 E	3
	left	3	2 E	3
712	right	3	2 E	3
	left	3	2 E	3
713	right	3	2 E	3
	left	3	2 E	3
714	right	3	2 E	3
	left	3	2 E	3
715	right	3	2 E	3
	left	3	2 E	3
716	right	3	2 E	3
	left	3	2 E	3
717	right	3	2 E	3
	left	3	2 E	3
718	right	3	2 E	3
	left	3	2 E	3
719	right	3	2 E	3
	left	3	2 E	3
720	right	3	2 E	3
	left	3	2 E	3

E: Swelling

E. SKIN REACTIONS AFTER EPICUTANEOUS INDUCTION

The epicutaneous induction with a 50% test substance preparation in 1% CMC-solution in doubly distilled water led to incrustation, partially open (caused by the intradermal induction) and moderate and confluent erythema in all test group animals (see Table 5.2.6-6).

Table 5.2.6-6: BAS 310 I - Skin reactions after epicutaneous induction

Animal #	Test item 50 % in 1% CMC-solution in doubly distilled water
711	2 E K
712	2 E K
713	2 E K
714	2 E K
715	2 E K
716	2 E K
717	2 E K
718	2 E K
719	2 E K
720	2 E K

E: Swelling; K: incrustation, partially open

F. SKIN REACTIONS AFTER CHALLENGE

The challenge with a 50% test substance preparation in 1% CMC-solution in doubly distilled water did not cause any skin reactions in animals of the control group and test group 24 and 48 hours after removal of the patch (see Table 5.2.6-7). Since no borderline results were observed, a 2nd challenge was not performed.

Table 5.2.6-7: BAS 310 I - Skin reactions after challenge

	Challenge			
Skin findings	**Control group**		**Test group**	
	24 h	**48 h**	**24 h**	**48 h**
Grade 0	5/5#	5/5	10/10	10/10

x/y = number of findings / number of animals tested

G. POSITIVE CONTROL

The positive control Alpha-Hexylcinnamaldehyde showed a sensitization rate of 100% in the guinea pig strain. The results of the latest study conducted with the positive control are presented in Table 5.2.6-8.

Table 5.2.6-8: Skin reactions after challenge with the positive control

	Challenge			
Skin findings	**Alpha-Hexylcinnamaldehyde (techn. 85%) 5% in Lutrol® E 400**		**Vehicle Control: Lutrol® E 400**	
	24 h	**48 h**	**24 h**	**48 h**
Control group	0/5#	0/5	0/5	0/5
Test group	10/10	9/10	0/10	0/10

x/y = number of positive reactions/number of animals tested (reading at 24 h and/or 48 h after the removal of the patch)

III. CONCLUSION

Based on the results of this study it is concluded that BAS 310 I does not have sensitizing properties in the guinea pig maximization test under the test conditions chosen. 0% of the animals were considered positive after challenge application.

CA 5.2.7 Phototoxicity

Report: CA 5.2.7/1
Cetto V.,Landsiedel R., 2013a
BAS 310 I (Alphacypermethrin) - In vitro 3T3 NRU phototoxicity test
2013/1389105

Guidelines: OECD 432 (2004) In vitro 3T3 NRU Phototoxicity test, (EC) No 440/2008 of 30 May 2008 laying down test methods pursuant to (EC) No 1907/2006 of European Parliament and of Council on the REACH - Part B No. B.41 No. L 142

GLP: yes
(certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

Alpha-Cypermethrin (Batch L80-24; purity 99.4%) was tested for its ability to induce phototoxic effects in Balb/c 3T3 cells in vitro. The photo-cytotoxicity was estimated by the means of the Neutral Red Uptake (NRU) method. A single experiment was carried out with and without UVA irradiation. Vehicle and positive controls were included into the study. Based on the range-finding phototoxicity test the same concentrations were used in the main experiment with and without Irradiation: 4.6 - 10 - 21.5 - 46.4 - 100 - 215.4 - 464.2 - 1000 µg/mL.
Test substance precipitation in culture medium at the end of treatment was observed at 215.4 µg/mL and above in the absence and presence of UVA irradiation. In addition, changes in cell morphology were observed at the end of exposure period at 215.4 µg/mL and above without and with irradiation. After treatment with the test substance, no cytotoxic effects indicated by Neutral Red absorbance values of below 50% of control were observed in the Main Experiment in the absence and the presence of UVA irradiation. Therefore, no EC_{50} values could be calculated. Based on these observations a formal PIF = *1 has to be used to characterize the result. The positive control chlorpromazine led to the expected cytotoxicity both with and without UVA irradiation (PIF: 60.1).

Thus, under the experimental conditions of this study, alpha-cypermethrin is considered **not to be a phototoxic** substance in the in vitro 3T3 NRU Phototoxicity Test using Balb/c 3T3 cells.

(DocID 2013/1389105)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material BAS 310 I

Description: Solid; white

Lot/Batch #: L80-24

Purity: 99.4% (tolerance ± 1.0%) (see Certificate of Analysis, study code ASAP13_123)

Stability of test compound: The stability of the test substance under storage conditions over the test period was guaranteed until 01 Sep 2019 by the sponsor, and the sponsor holds this responsibility. The homogeneity of the test substance was ensured by the high purity and by mixing prior to preparation of test substance solutions.
The stability of the test substance in the vehicle DMSO was not determined analytically.

Solvent used: DMSO

2. Control Materials:

Vehicle control: The vehicle control cultures with and without irradiation only contained the vehicle used for the test substance/positive control at the same concentration and volume as used for the test substance and the positive control.

Solvent/final concentration: DMSO 1% (v/v) in PBS

Positive control compounds: Chlorpromazine (CPU) was dissolved in DMSO

A complete 96-well plate containing 8 concentrations was performed in parallel to demonstrate sensitivity of the test method.

Without irradiation	1.9-3.8-7.5-15-30-60-90-180 µg/mL
With irradiation	0.03-0.05-0.1-0.2-0.4-0.8-1.6-3.2 µg/mL

3. Test organisms: The BaIb/c 3T3, clone A31, cell line was isolated from the muscle tissue of mouse embryo. This fibroblast cell line has a high proliferation rate (doubling time 16 - 20 hours) and a high plating efficiency (>70%)) of untreated cells both necessary for the appropriate performance of the study. The Balb/c 3T3 cell line which was used in this experiment was obtained from the “European Collection of Cell Cultures” Salisbury, Wiltshire SP4 OJG, UK (date 09 Aug 2006) and is stored at -196°C (liquid nitrogen).

4. Culture media and reagents:

Culture medium:	Dulbecco's Modified Eagle's Medium (DMEM) supplemented with - 10% (v/v) newborn calf serum (NCBS) - 4 mM L-glutamine - 100 IU penicillin - 100 µg/mL streptomycin
Neutral Red solution:	- 0.4 g Neutral Red powder (NR; Sigma N4638) - 100 mL deionized water
Neutral Red medium:	- 1 mL Neutral Red solution - 79 mL culture medium (DMEM incl. supplements) Incubated overnight at 37° C with 5% CO_2 and filtered with a 0.22 µm filter prior to use.
Other solutions and reagents:	- phosphate buffered saline (PBS) without Ca/Mg - trypsin/EDTA solution (0.05%; 0.02%) - Neutral Red desorb solution (1 mL acetic acid, 50 mL ethanol, 49 mL deionized water)

5. Irradiation source: The Sol 500 solar simulator (Dr. Hönle AG, 82166 Gräfelfing, Germany) used with filter H1 produced wavelength > 320 nm. The exposure rates were determined with UV-meter RM-21 (Dr. Gröbel GmbH, 76275 Ettlingen, Germany).

6. Test concentrations:

Pretest:	Up to 1000 µg/mL with and without irradiation.
NRU test conditions:	An appropriate amount of test article substance was taken up in the vehicle, shaken thoroughly and diluted in accordance with the planned doses under light protection conditions immediately before administration. The experiment was performed in 96 well plates in one experiments (6 replicates per concentration with and without irradiation; two plates per substance (test substance or positive control) were prepared.) The test substance concentrations were: Without: 4.6; 10.00; 21.5; 46.4; 100.0, 215.4, 464.2, 1000.0 µg/mL With: 4.6; 10.00; 21.5; 46.4; 100.0, 215.4, 464.2, 1000.0 µg/mL

B. TEST PERFORMANCE:

1. Dates of experimental work: 05-Nov-2013 - 29-Nov-2013

2. Treatment and NRU Phototoxicity test:

Two 96 well-plates per substance (test substance or positive control) were used for cultivation of cells (1.5 x 10^5 cells/well). After an attachment period of about 24 hours the cells were washed once with 100 µL PBS and subsequently treated with the respective substance (8 concentrations each with 6 replicates of the test substance or the positive control) and the vehicle control in parallel for 1 hour in the dark (5% (v/v) CO_2, ≥90% humidity; 37° C). Then, one microtiterplate per substance was irradiated for 50 minutes with UVA (UV intensity underneath the lid 1.5 - 2.1 mW/cm^2 = 5 J/cm^2) whereas the respective reference plate was kept in the dark for the same period. After test substance removal and washing step (100 µL PBS) the cells were incubated in culture medium overnight. The medium was removed after 24 hours, the cells washed again, 100 µL medium containing 50 µg/mL Neutral red was added and the plates were incubated for another 3 hours. Each step was performed under light protected conditions in the lab to prevent uncontrolled photo activation. Afterwards, the cells were washed and the dye was extracted by Neutral Red desorb solution. Cytotoxicity was determined by measuring the Neutral Red Uptake using a microplate reader (Perkin Elmer, Waltham, Massachusetts, US; Wallac 1420 multilabel counter) equipped with a 550 nm filter to read the absorption of the extracted dye. The absorption shows a linear relationship with the number of surviving cells.

3. Evaluation/Assessment

For the assessment of the phototoxic potential of a compound two prediction models are currently available:

- The Photo-Irritancy-Factor Prediction model for substances which allow the comparison of two equi-effective concentrations (EC_{50}) in the concurrently performed experiments in the presence and absence of light. This model includes the special case of absence of cytotoxicity in the presence and absence of light for substances obviously showing no phototoxic potential (see below).
- The Mean Photo Effect prediction model which is used if no equi-effective concentrations (EC_{50}) are obtained in the absence and presence of UV light. This special cases do not apply to this study. Even though described in the report this case is not described in this summary.

3.1 Cytotoxicity

The mean absorbance values obtained for each test group of every plate were used to calculate the percentage of cell viability relative to the respective vehicle control, which is arbitrariliy set at 100 %.

$$Cytotoxicity\,[\%] = \frac{\text{Absorbance}_{\text{mean of the test group}}}{\text{Absorbance}_{\text{mean of the vehiclecontrol}}} \times 100$$

In case of cytotoxicity, an EC_{50} value (Inhibition concentration 50% relative to the respective vehicle control) was calculated by a linear interpolation method (linear dose-response curve).

3.2 Photo-Irritancy-Factor

If no cytotoxicity occurs to determine an EC_{50} value in the concurrently performed experiments in the absence and presence of UV light up to the highest applied test concentration it has to be considered that the test substance has no phototoxic potential.

In this case, a formal PIF = *1 is used to characterize the result:

$$PIF = *1 = \frac{C\max(-UVA)}{C\max(+UVA)}$$ resulting in the following classification rule:

PIF = *1	no phototoxic potential predicted

3.3 Other parameters

pH:
The pH was measured at least for the two top doses and for the vehicle controls with and without irradiation.

Osmolarity:
Osmolarity was measured at least for the two top doses and for the vehicle controls with and without irradiation.

Solubility:
Test substance precipitation was checked immediately after treatment and at the end of treatment.

Cell morphology:
Test cultures of all test groups were examined microscopically before staining with NRU, which allows conclusions to be drawn about attachment of the cells.

4. Statistics:

No special statistical tests were performed.
Mean absorbance values and standard deviations were calculated from the single values using calculation software (e.g. MS Excel). The calculations were made using the unedited values. For the report the values were rounded, therefore there may be deviations in the given relative values. If technical errors occurred in single wells (outlier) at least 4 single values per test group were sufficient for calculating reliable mean values. Outliers are defined as values that have half or double the value of the respective mean.

5. Acceptance criteria:

The assay has to be considered valid if the following criteria are met:

- The vehicle control needs to fulfill the following criteria:
  - The mean OD550 value (with and without UVA irradiation) should be ≥ 0.3.
  - Cell viability after irradiation should be at least 80% of the concurrent non-irradiated vehicle control.
  - The standard deviation of the mean values of both vehicle control rows should not exceed ± 15%.
- The positive control chlorpromazine needs to fulfill the following criteria:
  - the EC50 value should be in the ranges:
    With irradiation (+UVA): 0.1 - 2.0 µg/mL
    Without irradiation (-UVA): 7.0 - 90.0 µg/mL
  - and the PIF ≥ 6.

II. RESULTS AND DISCUSSION

A. ANALYTICAL DETERMINATIONS

Osmolarity and pH values were not influenced by test substance treatment. In this study, in the absence and the presence of UVA irradiation precipitation in culture medium was observed at test substance concentrations of 215.4 µg/mL and above. In addition, changes in cell morphology were observed at the end of exposure period at 215.4 µg/mL and above without and with irradiation.

B. CYTOTOXICITY OF THE TEST SUBSTANCE

After treatment with the test substance, no cytotoxic effects indicated by Neutral Red absorbance values of below 50% of control were observed in the main experiment in the absence and the presence of UVA irradiation. Therefore, no EC50 values could be calculated. Based on these observations a formal PIF = *1 has to be used to characterize the result.

Table 5.2.7-1: Mean relative cytotoxicity of BAS 310 I with and without UVA irradiation in Balb 3T3 cells

Test group	UVA irradiation*	Precipitation**	Mean $OD_{corr.}$ ***	Cytotoxicity [% of control]
Vehicle control (1% DMSO)	-	-	0.359	100.0
BAS 310 I				
4.6 µg/mL	-	-	0.327	90.9
10.0 µg/mL	-	-	0.334	93.0
21.5 µg/mL	-	-	0.347	96.6
46.4 µg/mL	-	-	0.301	83.7
100.0 µg/mL	-	-	0.275	76.5
215.4 µg/mL	-	+	0.213	59.2
464.2 µg/mL	-	+	0.198	55.0
1000.0 µg/mL	-	+	0.201	56.0
Vehicle control (1% DMSO)	+	-	0.346	100.0
BAS 310 I				
4.6 µg/mL	+	-	0.336	97.4
10.0 µg/mL	+	-	0.329	95.2
21.5 µg/mL	+	-	0.326	94.2
46.4 µg/mL	+	-	0.314	90.9
100.0 µg/mL	+	-	0.292	84.5
215.4 µg/mL	+	+	0.257	74.3
464.2 µg/mL	+	+	0.224	64.9
1000.0 µg/mL	+	+	0.239	69.1

*: Irradiation with Sol 500 solar simulator for 50 minutes (approx.. 5 J/cm²)
**: Precipitation in PBS at the end of exposure period
***: Mean OD corrected: mean absorbance (test group) minus mean absorbance (blank)

C. CYTOTOXICITY OF THE POSITIVE CONTROL

After treatment with the positive control chlorpromazine clear cytotoxic effects indicated by Neutral Red absorbance values of below 50% of control were observed in the absence and the presence of UVA irradiation at least in the highest applied concentrations.
Without UVA irradiation, there was a decrease in the cell number from 60.0 µg/mL (EC_{50}: 35.4 µg/mL) onward. With UVA irradiation, there was a decrease in the cell number at 0.8 µg/mL (EC_{50}: 0.6 µg/mL) and above. Based on the EC_{50} values a PIF of 60.1 (phototoxic potential) was obtained (see Table 5.2.7-2).

Table 5.2.7-2: Mean relative cytotoxicity of Chlorpromazine with and without UVA irradiation in Balb/c 3T3 cells

Test group	UVA irradiation	Mean OD *	Mean $OD_{corr.}$ **	Relative Cytotoxicity [% of control]	Standard deviation [%]
Blank	-	0.036	-	-	-
Vehicle control 1	-	0.419	0.384	-	4.1
Vehicle control 2	-	0.424	0.389	-	1.6
Vehicle control mean (1% DMSO)	-	0.422	0.386	100.0	3.0
Chlorpromazine					
1.9 µg/mL	-	0.433	0.397	102.8	3.7
3.8 µg/mL	-	0.456	0.421	108.9	3.3
7.5 µg/mL	-	0.455	0.419	108.6	3.0
15.0 µg/mL	-	0.436	0.400	103.6	5.6
30.0 µg/mL	-	0.270	0.234	60.7	35.8
60.0 µg/mL	-	0.040	0.004	1.1	2.8
90.0 µg/mL	-	0.037	0.001	0.2	0.6
180.0 µg/mL	-	0.037	0.001	0.2	0.2
Blank	+	0.035	-	-	-
Vehicle control 1	+	0.376	0.341	-	1.4
Vehicle control 2	+	0.384	0.348	-	1.7
Vehicle control mean (1% DMSO)	+	0.380	0.345	100.0	1.9
Chlorpromazine					
0.03 µg/mL	+	0.365	0.329	95.6	2.3
0.05 µg/mL	+	0.380	0.345	100.0	2.7
0.10 µg/mL	+	0.372	0.337	97.8	2.7
0.20 µg/mL	+	0.366	0.331	96.0	3.6
0.40 µg/mL	+	0.311	0.276	80.1	5.9
0.80 µg/mL	+	0.092	0.056	16.4	8.6
1.60 µg/mL	+	0.035	0.000	0.1	0.1
3.20 µg/mL	+	0.039	0.004	1.1	0.2

*: Mean absorbance at 550 nm of 6 wells, in general
**: Mean absorbance (test group) minus mean absorbance (blank)

III. CONCLUSIONS

According to the results of the present study, alpha-cypermethrin is considered **not to be a phototoxic** substance in the in vitro 3T3 NRU Phototoxicity Test.

CA 5.3 Short-Term Toxicity

Studies evaluated in the draft monograph of rapporteur member state Belgium of Sep. 1999:
Short-term toxicity of alpha-cypermethrin was evaluated in subacute and subchronic studies (rat, mouse, dog) and all relevant data have been reviewed during the last Annex I inclusion process by European authorities and Belgium as Rapporteur Member State (European Commission Peer Review Program). All studies are scientifically valid; however, partially the studies have been conducted before the release of study guidelines and are without GLP according to the usual practice in those days. The studies are, for the convenience of the reviewer, listed in Table 5.3-1., and brief summaries were extracted from the monograph of alpha-cypermethrin and are provided under the respective chapters.

For several studies the achieved dose was newly calculated on the basis of data given in the respective report whereas in the former evaluation process the dose was estimated based on simple conversion factors. For the convenience of the reviewer the NOAEL is given in the Table as former estimated value and as newly calculated value.

The summary of the short-term toxicity studies from the DAR of alpha-cypermethrin (1999) is still valid, except for the respective achieved dose, which is amended in square brackets: “The short-term repeated dose exposure tests allow the hazard assessment, identification and dose-response, of alpha-cypermethrin.
Oral administration of alpha-cypermethrin primarily causes neurotoxicity in rats, mice and dogs, producing symptoms similar to those observed in the acute experiment. Doses that produce CNS effects also influence the general well-being of the animal. At the MTD, axonal degeneration has been seen in the rat. Above the MTD, hepatotoxicity is observed in the mouse.
The lowest NOAEL was 2.3 mg/kg bw/d [newly calculated: 3.5 mg/kg bw] (dog, 90 day) and 1.5 mg/kg bw/day [newly calculated: 2.0 mg/kg bw] (dog, 1 year).
Short term, repeated dose administration via inhalation (14 days), mainly produced local inflammatory reactions in the airways, compatible with the airway irritation seen in the acute inhalation experiment. These already appeared at exposure concentrations of 0.0028 mg/l. The absence of systemic effects is compatible with the low dose, 2.44 mg/kg bw/day, generated by the highest exposure concentration. Short term, repeated dose, dermal administration (21 days) to rabbits, did not show adverse effects.”

Table 5.3-1: Summary of already peer-reviewed alpha-cypermethrin short-term toxicity studies as available in the DAR (1999)

Study	Dosages (mg/kg bw/ day)	NOAEL (mg/kg bw/day)	Main adverse effect	Reference
5-week feeding Wistar rat 0, 25, 100, 200, 400 and 800 ppm	M&F: 2.5, 10, 20, 40 and 80 (estimated)	200 ppm Estimated: 20 Calculated: M: 17.9 F: 20.0	Reduced food intake Reduced body weight	[redacted] 1982 (AL-420-003) (Dossier Cyanamid)
6-week feeding* Crl:CD(SD) BR rat 0, 50, 200, 800 and 1200 ppm	M&F: 5, 20, 80 and 120 (estimated)	200 ppm Estimated: 20 Calculated: M: 17.6 F: 20.2	neurotoxicity	[redacted] 1984 (AL-420-004)
2-7 day oral MTD study Beagle dog	M&F: 200, 250, 300, 400 ppm	MTD: >250 - <300 ppm	Ataxia, tremors	[redacted] 1993 (AL-420-006) (Dossier Cyanamid)
14-day oral administration Mongrel dog*	M&F: 0, 1.25, 2.5, 5, 10, 20 or 40	10	Vomiting	[redacted] 1988a[1] (Dossier Gharda)
29-day feeding CD-1 mice 0, 200, 400, 800, 1200 and 1600 ppm	M: 27.4, 55.9, 121.4, 165.9 and 241.3; F: 33.6, 73.1, 145.6, 211.5 and 294.1	400 ppm M: 55.9 F: 73.1	Decreased body weight and body weight gain, Increased relative kidney weight	[redacted] 1993 (AL-420-005) (Dossier Cyanamid)
90-day feeding Wistar rat 0, 20, 60, 180, and 540 ppm	M&F: 0, 1, 3, 9 and 27	180 ppm Estimated: 9 Calculated: M: 13.1 F: 15.1	Decreased body weight and food intake, liver enlargement, small peripheral nerve toxicity	[redacted] 1982 (AL-425-003) (Dossier Cyanamid)
90-day feeding CD-1 mice 0, 50, 250 and 1000 ppm	M: 0, 6.3, 33.2 and 169.8 F: 0, 7.4, 36.3 and 184.4	50 ppm M: 6.3 F: 7.4	Altered haematological parameters, increased AST, increased liver weight	[redacted] 1994 (AL-425-006) (Dossier Cyanamid)
90-day feeding Beagle dog 30, 90, 270 ppm	0, 0.75, 2.25 and 6.75 (estimated) Calculated	90 ppm Estimated 2.3 Calculated: M: 3.5 F: 3.8	neurotoxicity	[redacted] 1984 (AL-425-005) (Dossier Cyanamid)
90-day oral administration Mongrel Dog*	0, 1, 3 and 10	3	Decreased food intake Decreased body weight	[redacted] 1988b[1] [2] (Dossier Gharda)
52-week feeding Beagle dog 0, 60, 120 and 240 ppm	0, 1.5, 3 and 6 (estimated)	60 ppm Estimated 1.5 Calculated: M: 2.0 F: 2.2	Skin irritation	[redacted] 1995 (AL-427-001) (Dossier Cyanamid)
14-day inhalation* Wistar rat	0.0028, 0.012, 0.029 mg/L	< 0.0028 mg/L	Local adverse effects	[redacted] 1988c[1] (Dossier Gharda)
21-day dermal* Albino rabbit	0, 500, 1000 and 2000	> 2000	none	[redacted] 1988c[1] (Dossier Gharda)

[1] These studies are not BASF proprietary, but owned by Gharda, Chemical Ltd. Bombay, a further notifier for Annex I listing of alpha-cypermethrin in 1999.
[2] This studies formally shows the lowest NOAEL value but is considered less accurate than the 90-day feeding study in Beagle dogs AL-425-005. Therefore this NOAEL was not considered in the endpoint definition.
* No GLP study;

Studies submitted in this AIR 3 dossier (not yet peer-reviewed):
There is one 90-day study performed in 1994 that was not submitted in the former registration process under the aegis of Cyanamid. This feeding study contains a complete neurotoxic evaluation and is therefore described in detail in KCA 5.7. Furthermore a 15 day adult male rat assay was performed to address the endocrine potential of alpha-cypermethrin and potentially effects on sperm parameters. This study is described in detail in KCA 5.8.3.

Table 5.3-2: Summary of not yet reviewed alpha-cypermethrin short-term toxicity studies

Study	Dosages (mg/kg bw/ day)	NOAEL (mg/kg bw/day)	Main adverse effect	Reference
15-day adult male rat assay Wistar rat Vehicle: Corn oil	M: 2.0, 3.5, 6.6	M: 6.6	none (Range finder showed BW loss and neurotoxic symptoms at 10 mg/kg bw)	BASF Doc ID KCA 5.8.3/50 & 51 2014/1275120 2004/1289319
90-day feeding study Crl:CD (SD) BR rat 0, 50, 250, 500 ppm	M: 0, 3.7, 17.9 and 36.1 F: 0, 4.2, 21 and 42	50 ppm M: 3.7 F: 4.2	Transient reduced food intake, altered hematology parameters	1994 KCA 5.7.1/4 AL-425-007
Literature data: 31-day gavage study Wistar rat Vehicle: 1ml DMSO	0, 14.5 mg/kg bw	n.a.	Changed biochemical parameters in blood and liver, Microscopic pathology	Manna et al., 2004 KCA 5.3.1/1 2004/1040514 Not further considered
28-day gavage study Swiss albino mice Vehicle: arachis oil	0, 250 mg/kg bw Alpha-cypermethrin or Cypermethrin, unclear	n.a.	Changed biochemical parameters in blood and liver, Microscopic pathology	[redacted] 2010 KCA 5.3.1/2 2010/1232197 Not further considered
28-day dermal study rabbit	M: 0, 40, 200, 1000 mg/kg bw	n.a.	Dose-dependent increase of abnormal sperms, TRIG, histopath. changes in testes	[redacted] 2009 KCA 5.3.3/1 2009/1130982 Not further considered

n.a.: not applicable- These studies do not mention a NOAEL level.

However, under consideration of the general toxicity parameters these studies do not change the overall risk assessment, which is still valid to be performed based on the NOAEL from the 1-year dog study, as the species which showed overall the lowest NOAEL.

Based on the available studies, the following endpoints were determined in the Annex I listing of alpha-cypermethrin in 2004.

Short term toxicity

Target / critical effect:	Irritation secondary to systemic toxicity (dogs)
Lowest relevant oral NOAEL/NOEL	1 year dog study: 60 ppm (1.5 mg/kg bw/day); R48/22 90 day dog study: 90 ppm (2.3 mg/kg bw/day)
Lowest relevant dermal NOAEL/NOEL	No data – not required
Lowest relevant inhalation NOAEL/NOEL	6h/day; 5 d/week, 3 week rat: >0.029 mg/L (no systemic effects, only local inflammatory effects)

Based on the newly calculated achieved dose at the respective NOAELs in the 90 day and 52-week feeding dog studies the following endpoints are proposed in the Annex renewal process:

Short term toxicity

Target / critical effect:	Irritation secondary to systemic toxicity (dogs)
Lowest relevant oral NOAEL/NOEL	1 year dog study: 60 ppm (**2.0** mg/kg bw/day); STOT RE 2 (H373) 90 day dog study: 90 ppm (**3.5** mg/kg bw/day)
Lowest relevant dermal NOAEL/NOEL	No data – not required
Lowest relevant inhalation NOAEL/NOEL	6h/day; 5 d/week, 3 week rat: >0.029 mg/L (no systemic effects, only local inflammatory effects)

CA 5.3.1 Oral 28-day study

Rat, 5 week feeding study, 0, 25, 100, 200, 400 and 800 ppm (AL-420-003)

Guidelines: Not fully in compliance with the test method B.7 of directive 92/69/EEC. This study is a Range finder
Deviations: The duration was longer than 28 days
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

Groups of 5 male and 5 female Wistar rats per dose group received alpha-cypermethrin (batch: OCD/7: ST 81/002; purity 96.5%) at dietary doses of 0, 25, 100, 200, 400 and 800 ppm for 5 weeks.

Rats administered 25, 100 and 200 ppm alpha-cypermethrin did not show treatment-related effects.
Feeding of 800 ppm alpha-cypermethrin to rats of both sexes for five weeks produced abnormal gait and increased sensitivity to noise. Abnormal gait was recorded only in one male at 400 ppm. Decreases of food intake and bodyweight were observed in rats of both sexes fed with 400 and 800 ppm alpha-cypermethrin.
Some haematological findings were seen in males at 400 and 800 ppm and females at 800 ppm. Changes in blood chemistry were considered to be a consequence of alterations in hepatic protein metabolism associated with the reduced nutritional status of the animals. Urinalysis did not reveal significant differences.
Increases in adjusted organ weights at 400 and 800 ppm were considered to be an adaptive response to increased functional demands associated with treatment. No significant treatment-related macroscopic abnormalities were identified at necropsy. Histological examination revealed no significant treatment induced effects other than axonal degeneration in the sciatic nerves of one male of the 800 ppm group killed due to severe neurological disturbance. There was no evidence of axonal degeneration in the sciatic nerves in either sex fed 400 ppm test substance.

Conclusion
Based on the described findings at 400 and 800 ppm the NOAEL in this study was identified at 200 ppm.

<u>New calculation of substance intake at NOAEL:</u>
No conversion from Concentration [ppm] to Dose [mg/kg bw/day] was done in the study report although food consumption and body weight were determined. In the monograph the dose was estimated based on a conversion factor of 10 resulting in 2.5, 10, 20, 40 and 80 mg/kg bw/day substance intake for 25, 100, 200, 400 and 800 ppm, respectively. Since the relevant values are available and well-documented in the report of the study, the group substance intake in “mg/kg bw/day” is newly calculated based on weekly values according the following equation:
Mean Substance intake [mg/kg bw/day] = Concentration (ppm) x (Mean (weekly group food consumption / weekly group mean body weight)/7 (see Table 5.3.1-1). The last week was not included due to food restrictions. The resulting values were already submitted in the biocide registration process for alpha-cypermethrin (Rapporteur: Belgium) and were considered acceptable.

Table 5.3.1-1: Substance Intake values at the NOAEL taken from the DAR (1999) and newly calculated based on data in the report

Dose (ppm)	estimated Substance Intake (mg/kg bw/day)	calculated Substance Intake (mg/kg bw/day)
200 (NOAEL)	Male: 20 Female: 20	Male:17.9 Female: 20.0

Rat, 6 week feeding study, 0, 50, 200, 800 and 1200 ppm (1993, AL-420-006)

Guidelines: Not fully in compliance with the test method B.7 of directive 92/69/EEC. This study is a Range finder.
Deviations: The duration was longer than 28 days
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999.

Groups of 5 male and 5 female Crl:CD BR rats per dose group received alpha-cypermethrin (batch: 02156; drum 1085; purity 95.6%) at dietary doses of 0, 50, 200, 800 and 1200 ppm for 6 weeks.

Rats of both sexes fed 1200 ppm and males fed 800 ppm alpha-cypermethrin were sacrificed during weeks 2 to 4 because of severe treatment related clinical signs characteristic for pyrethroid neurotoxicity. The signs were characterized by changes in gait, hunched posture, abasia and hypersensitivity. Signs of gait changes, limb position, hypersensitivity and increased reactivity to stimuli were also observed in the functional observational battery (FOB) during week 2 in rats of both sexes fed 1200 ppm and males fed 800 ppm. Gait changes were also observed in the females received 800 ppm test substance during week 6. There were no statistically significant changes in fore or hind limb grip strength or hind limb landing foot splay.
Throughout the study rats of both sexes fed 1200 and 800 ppm test substance had lower mean body weight and food intake compared to controls.

There were only minor changes in haematology and clinical chemistry parameters in the males receiving 200 ppm and females fed with 800 ppm alpha-cypermethrin.

Increased absolute organ weights were not seen when adjusted for terminal body weight and considered to reflect terminal body weight reduction. There were no macroscopic treatment-related findings. Stress related histopathological findings were confined to lymphocyte depletion of the thymus in the decedent rats in the 800 and 1200 ppm treated groups and two surviving females fed with 800 ppm test substance. There were no significant microscopic findings in the nervous system or muscle tissues.

Conclusion
Based on the described findings at 800 and 1200 ppm the NOAEL in this study was identified at 200 ppm.

New calculation of substance intake at NOAEL:
No conversion from Concentration [ppm] to Dose [mg/kg bw/day] was done in the study report although food consumption and body weight were determined. In the monograph the dose was estimated based on a conversion factor. This estimated intake at the NOAEL was replaced by calculated substance intake values (see description under study AL-420-003). Thereby the following substance intake values at the NOAEL were calculated (see Table 5.3.1-2). The last week was not included due to food restrictions.

Table 5.3.1-2: Substance Intake values at the NOAEL taken from the DAR (1999) and newly calculated based on data in the report

Dose (ppm)	estimated Substance Intake (mg/kg bw/day)	calculated Substance Intake (mg/kg bw/day)
200	Male:20 Female: 20	Male:17.6 Female: 20.2

Mouse, 29-day feeding study, 0, 200, 400, 800, 1200, 1600ppm (1993, AL-420-005)

Guidelines: In compliance with the test method B.7 of directive 92/69/EEC. This study is a Range finder
GLP: Yes
Acceptance: The study was considered acceptable in the EU registration process 1999.

Groups of eight male and eight female CD-1 mice received alpha cypermethrin via the diet, at concentrations of 200, 400, 800, 1200, or 1600 ppm for 29 days. This study was conducted to assess the toxicity of alpha cypermethrin and to determine suitable dose levels to be used in longer-term studies. A similarly constituted control group received untreated diet.

One female receiving 1200 ppm and one male receiving 1600 ppm were sacrificed in extremis during the treatment period following signs of neurologic disturbances; these deaths were considered to be related to treatment. One male receiving 1200 ppm died without ante mortem signs on day 16; however its death could not readily be attributed to treatment.

Clinical signs of toxicity including ungroomed coats, ataxia/abnormal gait, overactivity and/or hunched posture were observed in animals receiving 1200 or 1600 ppm. At 800 ppm, ungroomed coats were observed in four males and three females and abnormal gait was observed in one female.
Significantly reduced overall body weight gains, when compared to the controls, were seen in males and females receiving 1600 and 1200 ppm, and in females receiving 800 ppm. The effects on overall weight gains were generally a consequence of weight loss or significantly reduced weight gains during the first one or two weeks of treatment, however gains during weeks 3 and 4 in males and during week 4 in females receiving 800 ppm or more tended to be lower than the controls.

When compared with the controls, food intake was reduced during the first two weeks in males receiving 1200 or 1600 ppm and during the first week in females receiving 1200 ppm. Overall, food consumption was comparable between treated and control animals. Reduced food conversion efficiency, when compared to controls, was observed only in animals exhibiting decreased weight gains.
Significantly reduced lymphocyte counts, in comparison with those of the controls after adjustment for outlying control values, were seen after 24 days in males receiving 1600 ppm. No significant hematologic effects were seen in treated females. After 29 days of treatment, increased plasma alanine and aspartate aminotransferase activities were seen in males receiving 1200 or 1600 ppm. Small, but statistically significant, reductions in total protein and albumin concentrations, with a concomitant reduction in albumin to globulin ratios, were seen in males receiving 1600 ppm.
Terminal body weights were significantly lower for males and females treated at 800, 1200 and 1600 ppm. Kidney-to-body weight ratios for males fed 800, 1200 and 1600 ppm and for females fed 800 and 1200 ppm were increased compared to those of controls. It is likely that these findings resulted from decreased body weights observed for the animals in these dose groups. In addition, there were no changes in absolute kidney weights for either males or females at any dose level.
There were no macroscopic or microscopic findings which could be attributed to treatment with alpha cypermethrin.

Conclusion:
Based on the described findings at 1600, 1200 and 800 ppm the NOAEL in this study was identified at 400 ppm, corresponding to 55.9 mg/kg bw/d for male and 73.1 mg/kg bw/d for female mice as given in the report.

Dog, 14-day oral administration, 0, 1.25, 2.5, 5, 10, 20, 40 mg/kg bw (1988a, [redacted]

Guidelines: Not fully in compliance with the test method B.7 of directive 92/69/EEC. This study is a Range finder
Deviations: Rat is the preferred species: low number of animals, duration shorter than 28 day, incomplete description of experimental protocol and results.
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999.

One female and one male Mongrel dog per dose group received alpha-cypermethrin (batch: not given; purity 95 – 99.5%) received orally 1.25, 2.5, 5, 10, 20 and 40 mg/kg bw for 14 days.

No significant toxic symptoms were exhibited by control and treated animals except vomiting reported at the two highest doses. No mortality, normal body weight gain, relative values of organ weights of treated animals were normal and no significant gross pathological effect were noted. Administration of test material was discontinued from 3rd day at doses of 20 and 40 mg/kg bw as the animals vomited.

Conclusion:
Based on the described findings at 20 and 40 mg/kg bw/day the NOAEL in this study was identified at 10 mg/kg bw/day.

Dog, increasing dose feeding study (1984, AL-420-004)

Guidelines: Not fully in compliance with the test method B.7 of directive 92/69/EEC. This study is a Range finder
Deviations: low number of animals, the duration of treatment is normally 28 day with the same dose
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

The objective of this study was to determine the oral maximum tolerated dose of alpha-cypermethrin in beagle dogs in a two phase range-finding study.
In phase 1, one male and one female dog were fed alpha-cypermethrin in the diet at concentrations of 200 ppm for 7 days, 400 ppm for 2 days (followed by ground diet for 5 days), and 300 ppm for 7 days. Animals were sacrificed after the third week.
In phase 2, one male and one female dog were fed diets containing 300 ppm alpha-cypermethrin for 3 or 4 consecutive days (followed by 4 or 3 days ground diet) in the first week and 250 ppm daily for 7 days in the second week. Animals were sacrificed after the second week.
No clinical manifestations in male and female dogs were seen at concentration of 200 ppm alpha-cypermethrin. At concentrations of 300 and 400 ppm in the diet, dogs of both sex showed ataxia, body tremors, subdued behaviour, head nodding, food regurgitation and inflammation of the gums and tongue. At 250 ppm alpha-cypermethrin only the female dog exhibited these symptoms.
Body weight losses were recorded in both parts of the study only in animals receiving 300 ppm alpha-cypermethrin, but recovery was evident in animals subsequently treated with 250 ppm in phase 2.
No treatment-related haematological or clinical chemistry findings, significant changes in absolute or relative organ weights, gross pathological findings, remarkable effects on urine parameters or faeces were noted that could be ascribed to treatment with alpha-cypermethrin.

Conclusion:
It was concluded that the maximum tolerated dose (MTD) of alpha-cypermethrin when administered to dogs in their diet was greater than 250 ppm but less than 300 ppm. Therefore, it was recommended that the maximum dose level in any subsequent 13 week dietary study should not exceed 300 ppm.

Additional information from the open literature presented in the DAR (1999):

Rat, gavage, 28-days, 0, 4, 8, 12mg/kg bw in soy bean oil [redacted] 1996)

16 F344 male rats received by gavage 0, 4, 8 or 12 mg/kg bw alpha-cypermethrin (99.1%) in soy bean oil during 28 days.

The main subject of this publication was a possible effect of alpha-cypermethrin on immunotoxicity. No effects on the clinical appearance, weight gain, haematological parameters and bone marrow cellularity were found. There were no effects on the relative or absolute spleen weights. The relative testes and brain weights were significantly increased in the high dose group. Histopathological examination revealed no dose-related alterations.

Conclusion:
Based on the increased relative adrenal weight at 12 mg/kg bw/d the NOAEL in this study was identified at 8 mg/kg bw/day. Alpha-cypermethrin had no effect on the immune system.

New information from the open literature presented in the AIR process:

Report: CA 5.3.1/1
Manna S. et al., 2004a
Repeated dose toxicity of alpha-Cypermethrin in rats
2004/1040514
Guidelines: none
GLP: no
(certified by none)

Short term toxicity rat gavage study; 14.5 mg/kg bw (Manna et al., 2004; Doc ID 2004/1040514)

The LD_{50} of alpha-cypermethrin (99%, Gharda Chemicals, Bombay) administered in DMSO to Wistar rats was determined by 6 dose levels ranging from 100 mg/kg bw up to 225 mg/kg bw in 25mg steps. Vehicle in each dose group was 1ml DMSO. Based on this outcome 5 rats/sex were dosed daily by gavage with 1/10 LD50 (14.5 mg/kg bw) in 1 ml DMSO for 30 days compared to a control group receiving only DMSO. After sacrifice on day 31 the activities of various hepatic enzymes and tissue residue concentrations, hematology parameters and pathological changes were investigated.
Findings: Serum AST, ALT, LDH, ALP and blood glucose were significantly elevated, RBC, PCV and HB levels were reduced. Cytochrome P450, Catalase (CAT), superoxide dismutase (SOD) and glycogen level in liver were reduced whereas Malondialdehyde (MDA) was increased. GSH levels were not affected. Residue levels were determined in brain, lungs, liver, heart, kidney and testes. Macroscopic and histological alterations were observed in lungs, liver, stomach, kidneys, testes and cerebellum.

The following tables were copied from the report:

Table 2. Effects of α-CP on certain biochemical parameters in serum and blood of rats after daily oral administration at 14.5 mg/kg for 30 days (Values are mean ± SE, n = 10)

Parameters	Control	α-CP treated
ALP activity (IU/L)	78.03 ± 2.58	161.53 ± 6.60*
AST activity (IU/L)	59.45 ± 3.52	72.00 ± 4.97*
ALT activity (IU/L)	12.00 ± 1.43	26.50 ± 1.67*
LDH activity (IU/L)	49.41 ± 2.58	64.80 ± 2.01*
TP (gm/dl)	8.12 ± .022	6.41 ± 0.17*
ALB (gm/dl)	4.53 ± 0.29	4.50 ± 0.26
GLB (gm/dl)	3.81 ± 0.21	2.16 ± 0.49*
Blood Glucose mmol/L	3.70 ± 0.48	6.22 ± 0.85*

*$p < 0.05$ in comparison with control
ALP: Alkaline Phosphatase, AST: Aspartate transaminase, ALT: Alanine transaminase; LDH: Lactate dehydrogenase, TP: Total protein; ALB: Albumin; GLB: Globulin.

Table 3. Effects of α-CP on certain biochemical parameters in liver of rats after daily oral administration at 14.5 mg/kg for 30 days (Values are mean ± SE, n = 10)

Parameters	Control	α-CP treated
CAT activity (U/mg protein)	0.39 ± 0.04	0.07 ± 0.01*
SOD (U/mg protein)	0.48 ± 0.02	0.13 ± 0.01*
MDA (nmol/mg protein)	0.24 ± 0.02	2.85 ± 0.18*
GSH (μmol/mg protein)	1.41 ± 0.16	1.30 ± 0.05
Glycogen (mg%)	7.94 ± 0.24	5.15 ± 0.34*
P450 (nmol/mg microsomal protein)	2.91 ± 0.02	2.74 ± 0.04*
b5 (nmol/ mg microsomal protein)	1.16 ± 0.07	1.28 ± 0.05

*$p < 0.05$ in comparison with control
CAT: Catalase, SOD: Superoxide dismutase, MDA: Malondialdehyde, GSH: Reduced glutathione.

Residue level of α-CP

The levels of α-CP following daily oral administration for 30 days were 0.07 ± 0.01, 0.08 ± 0.02, 0.12 ± 0.10, 0.58 ± 0.11, 1.02 ± 0.21 and 0.21 ± 0.01 ppm in liver, brain, testis, kidney, lung, and heart, respectively. Concentration of α-CP was maximum in the lungs.

Conclusion from the author: Repeated oral doses of alpha-cypermethrin to Wistar rats altered the biochemical parameters, decreased cytochrome P450 content and antioxidant status, which correlated with histopathological changes of tissues.

Comment from the applicant:
General toxicity in this study is not documented in the main study over the 30 day period.No body weights, food consumption or neurotoxicity is reported, but severe stomach and intestine hemorrhages with desquamation and necrosis of the stomach epithelium, as well as congestions in lungs, liver and kidneys are mentioned. These strong local effects in the intestinal tract are normally associated with subsequent food intake reduction. It cannot be excluded that these effects are vehicle related as DMSO can cause significant local toxic effects and data are not given to distinguish between control and treated groups. Care needs to be taken when tissue *pieces* are used for histopathology based on squeezing artefacts of tissues. In addition, testes fixation in bouins fixative leads to tubular shrinkage and condensation of chromatin which can interfere with the correct interpretation of treatment related findings. The shrinkage is also well documented in Fig.3 of the publication (see copy below left side).

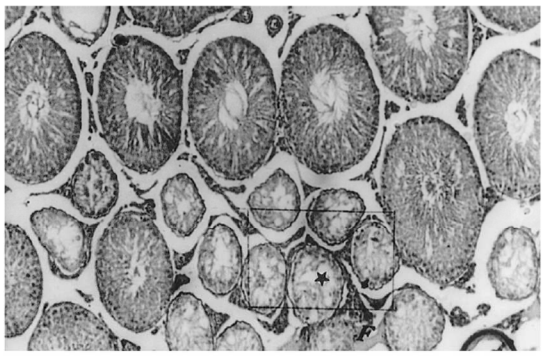


Fig. 3. Photomicrograph of rat testis showing edematous fluid accumulation between the tubules (F) and vacuolation (*) within the tubule after daily oral administration of α-CP at 14.5 mg/kg for 30 days, (H & E, 100×).

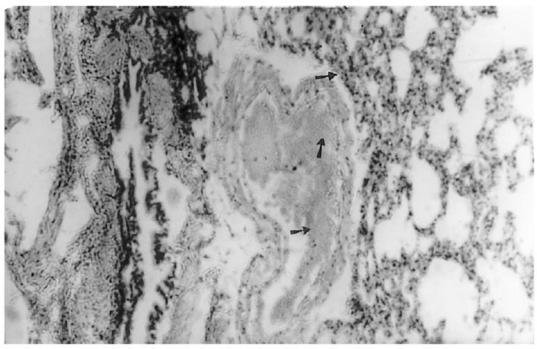


Fig. 1. Photomicrograph of rat lungs showing hemorrhages, and thickened inter-alveolar septae with infiltration of mononuclear cells (arrows) after daily oral administration of α-CP at 14.5 mg/kg for 30 days, (H & E, 450×).

This shrinkage might have been interpreted as a fluid accumulation between the tubules, which is seen only at one point (marked with F) at the border of figure 3 and would be expected to be evenly distributed between the tubules if treatment related.

Figure 1 (see copy above on the right side) in the literature shows a picture of a suboptimal preserved lung, and the arrows indicating mononuclear cell infiltration are pointing towards a blood vessel which is not appropriately recognized. Several enzyme parameters are determined which are, based on the strong intestinal effects with questionable test item-relation, not clearly attributable to alpha-cypermethrin.
In conclusion, the presented data are not convincing and the effects described might rather be attributed to administration of the vehicle or might be secondary effects to general impaired health status. Therefore, the study was not considered relevant to be taken into account in the overall conclusion

Classification of study: not further considered

Report:	CA 5.3.1/2 Prakash N. et al., 2010a Evaluation of testicular toxicity following short-term exposure to Cypermethrin in albino mice 2010/1232197
Guidelines:	none
GLP:	no (certified by none)

Mouse 28-day gavage study, 250 mg/kg bw (Prakash, 2010, Doc ID 2010/1232197)

Groups of 12 male Swiss albino mice were gavaged once daily with 250 mg/kg bw alpha-cypermethrin (1:9 in arachis oil) or vehicle alone. Six animals in each group were sacrificed on day 14 of administration of cypermethrin and remaining 12 animals in both the groups were sacrificed on day 28 of the experiment. Increased testicular wet weight but no differences in body weights were observed. Increased activities of testicular AST, ALT, and decreased epididymal ALP levels were observed. Increased cholesterol levels in the testes and reduced plasma testosterone levels were observed. The section of testis of treated group studied under Transmission electron microscope revealed changes in spermatogonic cells like rupture of cell membrane, shrinkage in the nucleus, stages of apoptosis, and condensation of chromatin decreased or absence of cytoplasmic organelles. The histological studies also revealed the pathological damage induced by the alpha-CYP on testicular tissue, which ranges from cell membrane damage of the seminiferous tubules to shrinkage in the nucleus, stages of apoptosis, condensation of chromatin, and decreased cytoplasmic organelles.

Comment from applicant:
Title, abstract and materials and methods are referring to cypermethrin dissolved in arachis oil. Otherwise alpha-cypermethrin is mentioned in the material part but without stereospecific information, so the used test substance is unclear. In the methods part it is described on the one hand that testes were frozen immediately in liquid nitrogen, but elsewhere there is the statement that testes were freshly trimmed using chilled saline solution for further investigations-so there is an unclear statement with regard to the methods used and anyway both methods are known to produce artefacts preventing evaluation of normal tissue structure. This is also documented in Fig.2 of the publication (copied below) which shows the suboptimal preservation of structures of a control seminiferous tubule.

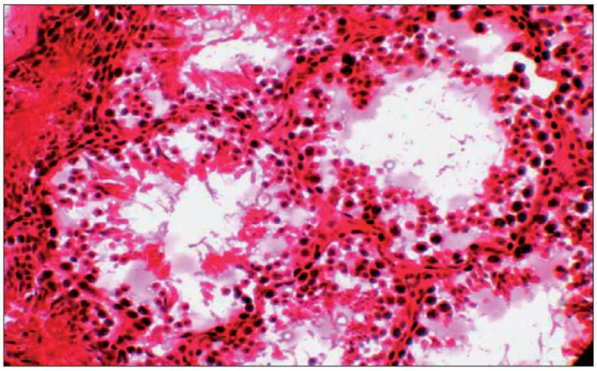


Figure 2: Transverse section of testes showing different spermatogonic cells in the semineferous tubules in control group (HandE, ×10) on day 28

A figure for comparison showing treatment-related changes is not presented. Bouins is also known to induce artefacts like tubular shrinkage and condensation of chromatin. Furthermore the paraffin sections (up to 8 µm) are too thick for microscopy. Transmission electron microscopy shown in Figure 3 and 4 of the publication presents only treatment-related slides, but no control. The described findings in Fig. 3 (not presented), like rupture of cell membrans, increased lipochrome pigment and shrinkage in the nucleus, and in Fig. 4 (not presented), like decreased cytoplasmic organelles are not seen in the images. There is no indication that the histopathological evaluation of spermatogonic cells was performed in a way which would allow to distinguish the different stages and therefore it is questionable if tubuli of the same stage were compared. Is the author aware of the spermatogenic cell turnover which includes apoptosis in some stages? The captions of the Figures 3 and 4 each do indicate 3 pictures a) to c) but there is only one picture displayed. In addition to that, there are no indices a) to c) given in these pictures. Taken everything together the inconsistency of the presentation of results makes the accuracy of the investigation rather questionable. The weight increase of testes and accessory glands combined with testosterone decrease are not a convincing finding. Furthermore, there is no information regarding general systemic toxicity. Irrespective of the used test substance (cypermethrin or alpha-cypermethrin) a high dose (250 mg/kg bw) was tested. Thus, effects in this peer-reviewed literature are likely to be secondary to impaired health status and are not relevant for risk assessment. Overall, the reliability is considered questionable and the study is therefore not further considered.

Classification of study: not further considered

CA 5.3.2 Oral 90-day study

Rat, 90-day feeding study, 0, 20, 60, 180, 540 ppm (1982, AL-425-003)

Guidelines: In compliance with the test method B of directive 87/302/EEC or OECD 408.
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

Groups of 30 male and 30 female Wistar rats received diets containing 20, 60, 180 and 540 ppm of alpha-cypermethrin (batch: OCD/7, purity: 96.5%) for up to 13 weeks. 60 male and 60 female animals served as control. After 6 weeks, one third of the animals were killed for interim haematological, clinical chemistry and gross post mortem examination.

Three males fed 540 ppm had an abnormal gait with splayed hind limbs after 14–28 days of treatment, but the histopathological examination showed no lesions in the sciatic nerves. Several instances of transient skin sores and fur loss were seen, particularly in the animals receiving 540 ppm. Other effects were generally unremarkable and females showed a lower incidence of clinical signs than males. Treatment of males and females with 540 ppm alpha-cypermethrin caused significantly decreased body weights and food consumption throughout the 13-week study period, which resulted in significantly reduced terminal body weights. Haematological and clinical chemistry findings at 540 ppm were not treatment-related, but a reflection of reduced food intake. Increased specific gravity and decreased urinary volumes at 540 ppm were attributed to decreased food and water intake.

Increased organ-to-liver weight ratios at 180 and 540 ppm were considered to represent an adaptive response of the liver to treatment with xenobiotics.
Gross and histopathological examination did not indicate any biologically significant lesions in any organ or other effects that were consistent with treatment.

No conversion from ppm to mg/kg bw/day was done in the study although food consumption and body weight were determined. In the monograph an estimation for males and females together was performed which resulted in 1, 3, 9 and 27 mg/kg bw/day for 20, 60, 180 and 540 ppm, respectively.

Conclusion:
Based on decreased body weights and clinical signs of neurotoxicity at 540 ppm, the NOAEL was determined to be 180 ppm.

New calculation of substance intake at NOAEL:
No conversion from Concentration [ppm] to Dose [mg/kg bw/day] was done in the study report although food consumption and body weight were determined. In the monograph the dose was estimated based on a conversion factor. This estimated intake at the NOAEL was replaced by calculated substance intake values (see description under study AL-420-003). Thereby the following Substance intake values at the NOAEL were calculated (see Table 5.3.2-1). The last week was not included due to food restrictions.

Table 5.3.2-1: Substance Intake values at the NOAEL taken from the DAR (1999) and newly calculated based on data in the report

NOAEL (ppm)	estimated NOAEL (mg/kg bw/day)	calculated NOAEL (mg/kg bw/day)
180	Male:9 Female: 9	Male: 13.1 Female: 15.1

Mouse, 90-day feeding study, 0, 50, 250, 1000 ppm (1994, AL-425-006)

Guideline: The study is not compulsory
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

12 CD-1 mice /sex/dose received via the diet 0, 50, 250 or 1000 ppm alpha-cypermethrin (batch: 02156, purity: 95.4%) for 13 weeks. The converted doses correspond to 6.3, 33.2 and 169.8 mg/kg bw/d for males and 7.4, 36.3 or 184.8 mg/kg bw/d for females.

At 1000 ppm 4 male mice died and 1 male mouse died from the 250 ppm dose group. These animals showed a slightly higher kidney weight. The clinical signs observed in the treated animals were thin build, ungroomed coats, hair loss (also seen at 50 ppm dose group) and encrustrations (also seen at 250 ppm) of dorsal body surface. These signs were probably due to a poor general condition.
Body weight gains were markedly lower at 1000 ppm. The overall food conversion efficiency of animals receiving 1000 ppm or 250 ppm was lower compared to the controls.

Relative kidney and liver weight were increased at 1000 ppm. An increased liver weight was also noted in females at 250 ppm. AST was increased and glucose values were decreased in males at 250 ppm and 1000 ppm. Furthermore, at the high dose an increased urinary specific gravity and a reduced urinary volume were observed.
Microscopically examination of the livers found a decreased incidence of centrilobular vacuolation/pallor and of inflammatory foci with single cell necrosis in males at 1000 ppm. These effects are considered to represent an altered hepatic metabolism which was further suggested from the increased plasma AST activity and reduced plasma glucose.

Conclusion:
Based on the effects found at 250 and 1000 ppm in this study the NOAEL was determined to be 50 ppm (6.3 mg/kg bw/day for males and 7.4 mg/kg bw/day for females).

Dog, 90-day feeding study, 0, 30, 90, 270 ppm (1984, AL-425-005)

Guidelines: In compliance with the test method B of directive 87/302/EEC or OECD 408 (1981).
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

Groups of 4 male and 4 female Beagle dogs received diets containing 0, 30 and 90 ppm of alpha-cypermethrin (batch F830047B, purity: 95.8%) while a fourth group comprised of 6 males and 6 females received 270 ppm test substance for up to 13 weeks. Dose levels were based on the results of a short-term repeated dose study in dogs ("Increasing dose feeding study in dogs").
One female animal receiving 270 ppm alpha-cypermethrin was sacrificed for human reasons in week 5. All animals receiving 270 ppm alpha-cypermethrin exhibited marked clinical signs from day 2 or 3 onwards within 3-6 hours after feeding. The signs comprised body tremors together with variable degrees of head nodding, lip licking, subduedness, ataxia or agitation. In week 3 all these dogs developed a high stepping gait and this observation, together with the instances of body tremors, head nodding, lip licking and ataxia increased gradually as the study progressed. There were no ophthalmoscopic findings.
Body weight gains and food consumption for all groups of animals were within expectation for animals of this type and age. No treatment related changes on haematology, clinical chemistry and urine parameters were observed. No treatment-related effects on absolute or relative organ weights, macroscopic examination and histopathology were seen.

Conclusion:
Based on the severity of clinical signs at 270 ppm the NOAEL was determined to be 90 ppm.

New calculation of substance intake at NOAEL:
No conversion from Concentration [ppm] to Dose [mg/kg bw/day] was done in the study report although food consumption and body weight were determined. In the monograph the dose was estimated based on a conversion factor. This estimated intake at the NOAEL is replaced by calculated substance intake values (more detailed description is given under BASF Doc ID 2013/1089655). Thereby the following Substance intake values at the NOAEL were calculated (see Table 5.3.2-2).

Table 5.3.2-2: Substance Intake values at the NOAEL taken from the DAR (1999) and newly calculated based on data in the report

Dose group	Intake estimation (mg/kg bw/day as given in the DAR (1999)	Intake (mg/kg bw/day) as calculated in BASF DocID 2013/1089655	
		male	female
90 ppm	2.25	3.5	3.8

Remark: As shown above the estimation of the intake value was lower than the calculated value. That means that the intake value given in the PPP monograph was underestimated. Since this study is pivotal and has been used for the setting of the reference value AOEL for the PPP registration it becomes absolutely necessary to base the reference value on the exact calculated value and not on mere estimation. The resulting values were already submitted in the biocide registration process for alpha-cypermethrin (Rapporteur: Belgium) and were considered acceptable.

Dog, 90-day oral administration study, 0, 1, 3, 10, and 3 mg/kg bw (1988b, [redacted])

Guidelines: Not fully in compliance with the test method B of directive 87/302/EEC or OECD 408(1981).
Deviations: Low number of animals
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999.
5 groups of 3 Mongrel dogs per sex were fed (mixed soup) with alpha-cypermethrin (purity: 95 – 99.5%) at doses of 0, 1, 3, 10 and 3 mg/kg bw/day for 90 days consecutively. One group dosed with 3 mg/kg bw/day served as recovery group. The effect of withdrawal of alpha-cypermethrin was measured 30 days after stopping oral feeding.

No deaths occurred during the study. Some of treated animals suffered from emesis and diarrhea. Food consumption was reduced in the high dose group. The animals of the 10 mg dose group showed reduction of body weight gain. Haematological parameters were normal and BUN, serum AP, SGPT and blood sugar were comparable to the control animals. Urinanalysis was normal. Relative organ weights were normal.

Conclusion:
Based on effects found at 10 mg/kg bw/day the NOAEL was determined to be 3 mg/kg bw/day.

Remark: This study is considered to be less accurate than the 90 day dog study AL-425-005 with regard to GLP status and Deviation from Guideline. The lower NOAEL value is a result of different dose spacings but not due to higher sensitivity in this study. The 90day dog study AL-425-005 indicates that the NOAEL is slightly higher. Therefore this study is not considered for endpoint definition.

Dog, 52-week feeding study, 0, 6, 120, 240 ppm (1995, AL-427-001)

Guidelines: Not fully in compliance with the test method B of directive 87/302/EEC or OECD 452 (1981)
Deviations: 2 liver function tests, GGT and ornithine decarboxylase, were omitted. The MTD was not reached.
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

Three groups of 4 male and 4 female Beagle dogs were dosed orally via the diet, 7 days per week, for 52 consecutive weeks at concentrations of 0, 60, 120 or 240 ppm (estimated intake: 1.5, 3 and 6 mg/kg bw/day) alpha-cypermethrin (batch: 02156, purity: 95.4%).

Findings:
Daily dietary administration of alpha-cypermethrin to Beagle dogs at a concentration of 240 ppm resulted in two males showing skin reddening on the tail leading to ulceration, necrosis and amputation in one male. In addition, treatment-related abdominal skin reddening and hair loss were seen in one male receiving 240 ppm and one female receiving 120 ppm alpha-cypermethrin. It was considered possible that there were some transient effect of the test material on the skin of susceptible animals. No consistent effects that were attributable to administration of alpha-cypermethrin were observed on food consumption, body weight gain, organ weights, haematology, clinical chemistry, urinalysis, ophthalmoscopic examinations, macroscopic and microscopic examinations.

Conclusion:
Based on clinical signs of skin irritation in one female receiving 120 ppm alpha-cypermethrin the NOAEL was determined to be 60 ppm.

New calculation of substance intake at NOAEL:
No conversion from Concentration [ppm] to Dose [mg/kg bw/day] was done in the study report although food consumption and body weight were determined. In the monograph the dose was estimated based on a conversion factor. This estimated intake at the NOAEL is replaced by calculated substance intake values (the calculation is given in KCA 5.3.2/1 2013/1089655). Thereby the following Substance intake values at the NOAEL were calculated (see Table 5.3.2-3).

Table 5.3.2-3: Substance Intake values at the NOAEL taken from the DAR (1999) and newly calculated based on data in the report

Dose group	Intake estimation (mg/kg bw/day as given in the PPP monograph	Intake (mg/kg bw/day) as calculated in the statement	
		male	female
60 ppm	1.5	2.0	2.2

Remark: As shown above the estimation of the intake value was lower than the calculated value. That means that the intake value given in the PPP monograph (1999) was underestimated. Since this study is pivotal and has been used for the setting of the reference value AOEL for the PPP registration it becomes absolutely necessary to base the reference value on the exact calculated value and not on mere estimation. The resulting values were already submitted in the biocide registration process for alpha-cypermethrin (Rapporteur: Belgium) and were considered acceptable.

CA 5.3.3 Other routes

Rat, 6h/day, 5day/week, 14-day inhalation study, 0, 0.0028, 0.012, 0.029mg/l (1988c, [redacted])

Guidelines: Not fully in compliance with the test method B.9 of directive 92/69/EEC (or 84/449/EEC).
Deviations: The duration of 2 weeks is too short (28d)
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999.

5 Wistar rats per sex and dose were exposed for 6 hours, 5 days per week for 2 weeks by nose and mouth route to an atmosphere containing 0.0028, 0.012 and 0.029 mg/L of alpha-cypermethrin (purity: 95-99.5%) produced by nebulisation. A control group was exposed to the vehicle (kerosene, 0.3 mg/L). An additional group was exposed to 0.012 mg/L and observed for 14 days after the last exposure. The particle size distribution yielded a mass median aerodynamic diameter of 3.7 µm which was within the respirable range of 0 to 7 µm.

The highest exposure concentration tested (0.029 mg/L) would be approximately equal to a dose of 3.65 mg/kg bw/day.
No mortality was observed during the study. During exposure all animals showed salivation, lacrimation and redness to nose. Food consumption and body weight gain was decreased in animals exposed to the highest dose. No treatment related findings were observed in hematology, clinical chemistry, urinanalysis, organ weights and necropsy. At the histopathological examination emphysema, perivascular and peribronchiolar lymphoid aggregation in lungs, an increased subcellularity in trachea and larynx, moderate amount of exudate containing polymorphs and desquamated cells in nose were observed in some animals exposed at different dose levels and were not dose-related. These findings were not seen in the control animals. The histopathological findings, however, not seen in the control group, point to a potential local adverse effect on the airway already produced at the lowest exposure concentration of 0.0028 mg/L.

Conclusion:
Based on the histopathological findings up to the lowest dose the NOAEL was determined to be lower than 0.0028 g/L (< 0.756 mg/kg bw/day).

Rabbit, 5days/week, 21-day dermal study, 0, 500, 1000, 2000 mg/kg bw (1988d, [redacted])

Guidelines: Not fully in compliance with the test method B.9 of directive 92/69/EEC (or 84/449/eEC),or OECD 410 (1981).
Deviations: The duration was 21 days instead of 28 days. Low number of animals
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999.
Three rabbits per sex and dose were exposed 5 times per week with alpha-cypermethrin technical (purity: 95-99.5%) to the clipped intact dorsal skin under semi-occluded conditions for 3 weeks with a dose of 0, 500, 1000 or 2000 mg/kg bw/day. At the end of 35 days all animals were killed.

No animal died during the study. No test substance related findings were observed concerning clinical signs, skin reactions, body weight and food consumption, hematology, blood chemistry, urinanalysis and organ weights. During histopathlogical examination small tiny foci of lymphocytic infiltration in the parenchyma, occasional degenerating hepatocyte in the liver, emphysematous lung, less colloid at some places in a few follicles in thyroid were observed. These changes were found in some of the control and treated animals.

Conclusion:
Since no treatment related findings were observed up to the highest dose the NOAEL is considered to be > 2000 mg/kg bw/day.

New information from the open literature presented in the AIR process

Report: CA 5.3.3/1
Anonymous, 2009a
Adverse drug reactions /risk assessment
2009/1130982
Guidelines: none
GLP: no
(certified by none)

Alpha-cypermethrin, rabbit 28-day dermal study (Aksoy et al., 2009), Doc ID 2009/1130982

Groups of 7 adult male rabbits were treated with alpha-cypermetrhin at doses of 40, 200, and 1000 mg/kg bw. DMSO was used as a vehicle.
Findings: None of the animals died during the study. From day 18 on slight hind limb paralysis was observed in four animals of the 1000 mg/kg bw dose group. No other clinical signs were observed. There were no significant changes in body weight or food consumption during the 28-day study. There were no statistically significant changes in semen volume, concentration or motility between treatment and control groups. However, rates of abnormal spermatozoal abnormalities (head acrosome, central and caudal parts) increased significantly in a dose-dependent manner. Serum AST, ALT, GGT, ALP and cholesterol remained unchanged. However, triglyceride concentrations decreased significantly in the treated groups. There were no gross pathological changes observed in brain, liver, spleen, kidney, stomach, intestines or gonads. Histopathological changes were limited to the testes of treated animals. Many tubules had severe testicular degeneration including vacuolation between spermatogonial cells, collapsed/shrunken tubules, decreased spermatogonial cell count and intratubular giant cell formation. The severity of the testicular findings was dose-related. Blood and semen drug concentrations were dose linear.

Comment from the applicant:
The publication was only available as a short-summary of a poster presented at a conference. Thus, all findings were only presented in text form and no data were given to make an evaluation or derivation of LOAEL or NOAEL values. Material purity and supplier were not stated. Methods were not described. The histopathology findings were not described according to accepted nomenclature. The weights of the rabbits used were not in the range stated in the guideline (OECD 410) where 2-3 kg is suggested and the variation was quite high (2.36 – 3.91 kg). Based on the limited data presented the study is considered to be insufficient for assessment and was therefore not further taken into account. A regulatory study addressing this type of study was already presented in the first Annex I inclusion which did not find any substance related effects on biochemical parameters in blood despite a dosing up to 2000 mg/kg bw.

Classification of study: not considered

Remark: To address testes and sperm toxicity a 15-day gavage study in adult males has been conducted which is described in KCA 5.8.3 (see Doc ID 2014/1275120). This study with oral administration of alpha-cypermethrin to Wistar rats in corn oil did not show any effect on testes and sperm parameters

CA 5.4 Genotoxicity Testing

Studies evaluated in the draft monograph of the rapporteur member state Belgium of Sep. 1999: A sufficient data-package of in vitro genotoxicity studies in bacterial, yeast and mammalian cell systems and of in vivo genotoxicity studies conducted with alpha-cypermethrin is available. These studies were performed guideline-compliant at that time and have been evaluated by European authorities and Belgium as rapporteur member state (European Commission Peer Review Program) and were considered to be acceptable. For the convenience of the reviewer, these studies are summarized below in table form as extracted from the monograph in Table 5.4.1-1, Table 5.4.2-1 and Table 5.4.3-1 below.

Submission of not yet peer-reviewed studies in this AIR3-Dossier: No additional data on genotoxicity of alpha-cypermethrin was generated. Only additional literature data is **newly** included and discussed in chapter 5.4.1, but does not lead to a change of endpoints. The following literature data was considered relevant for discussion but was disclosed from further consideration based on the used test substance:

Table 5.4-1: *In vitro* mutagenicity study with alpha-cypermethrin (literature data)

Study type	Test System	Exposure	Result	Reference
In vitro MNT with fluorescent in situ hybridization and ethidium bromide / acridine orange fluorescence staining	*human peripheral blood lymphocyte culture* 10% EC formulation (Super Takimethrin 100 EC) Concentration: 1-30 µM	24 and 48 h without metabolic activation	Equivocal	Muranli, 2013 Doc ID 2013/1417284 Not futher considered

The summary of genotoxicity, as given in the monograph of alpha-cypermethrin, is still valid and was also supported in the biocide registration process by the respective rapporteur (Belgium).

"Alpha-cypermethrin has been tested in *in vitro* test systems using bacteria, yeast and mammalian cells to assess gene mutations, and human peripheral lymphocytes to assess chromosomal damage. Alpha-cypermethrin was not mutagenic or genotoxic in these in vitro assays. The genotoxic potential of alpha-cypermethrin was assessed *in vivo* using three test systems to evaluate potential to induce chromosomal damage or DNA damage. The results obtained show that alpha-cypermethrin was not mutagenic or genotoxic in these in vivo assays." It is therefore concluded, that alpha-cypermethrin has no mutagenic or genotoxic properties both *in vitro* and *in vivo*.

Thus, the conclusion for relevant endpoints for the current re-registration remains as follows:

Genotoxicity	
In vitro studies	No genotoxic concern
In vivo studies in somatic cells	No genotoxic concern
In vivo studies in germ cells	No genotoxic concern

CA 5.4.1 In vitro studies

A summary of the in vitro genotoxicity studies evaluated for Annex I inclusion of alpha-cypermethrin is summarized in Table 5.4.1-1 below.

Table 5.4.1-1: *In vitro* mutagenicity studies with alpha-cypermethrin

Study type	Test System	With S-9 mix	Result	Reference
In vitro Mutagenicity in bacterial cells (Ames test) OECD 471 (1983)	*Salmonella thyphimurium* (TA 98, 100, 1535, 1537, 1538); *Escherichia coli* (WP2 uvrA); Concentration up to 5000 µg/plate	No Yes	Negative Negative	1993, AL-435-005 (Cyanamid Dossier)
In vitro Mutagenicity in bacterial cells (Ames test)	*Salmonella thyphimurium* (TA 98, 100, 1535, 1537, 1538); Concentration up to 5000 µg/plate	No Yes	Negative Negative	Jones et al., 1989[1] (Gharda Dossier)
In vitro Mutagenicity in mammalian cells OECD 476 (1984)	L5178Y Mouse lymphoma cells; Concentrations up to 50 µg/mL	No Yes	Negative Negative	1993, AL-435-007 (Cyanamid Dossier)
In vitro Cytogenicity OECD 473 (1983)	Chromosome aberration in human lymphocytes; Concentrations up to 1000 µg/mL, precipitation from 625 µg/ml onwards	No Yes	Negative Negative	1993, AL-435-006 (Cyanamid Dossier)
Additional information: *In vitro* Mutagenicity in yeast	*Saccharomyces cerevisiae*; Concentrations up to 4000 µg/mL	No Yes	Negative Negative	1984, AL-435-002 (Cyanamid Dossier)

[1] This study is not BASF proprietary, but owned by Gharda, Chemical Ltd. Bombay, a further notifier for Annex I listing of alpha-cypermethrin in 1999.

Further information from the literature submitted in the AIR 3 dossier:

Report:	CA 5.4.1/1 Muranli F.D.G., 2012a Genotoxic and cytotoxic evaluation of pyrethroid insecticides Gamma-Cyhalothrin and Alpha-Cypermethrin on human blood lymphocyte culture 2013/1417284
Guidelines:	none
GLP:	no (certified by none)

Executive Summary of Literature

The authors investigated the genotoxic, cytotoxic and aneugenic potential of alpha-cypermethrin and lambda-cyhalothrin on human peripheral blood lymphocyte culture using micronucleus and fluorescence in situ hybridization (FISH) methods. For the determination of micronuclei and apoptotic effects concentrations of 1, 2, 3.75, 7.5, 15, and 30 µM were tested. For FISH analysis only 7.5 and 15 µM cypermethrin were used. Cytotoxicity was also observed using trypan blue and acridine orange/ethidium bromide fluorescence staining methods to measure the apoptotic effect. The test substance was a commercial EC formulation containing 10% active substance (for alpha-cypermethrin).

Conclusion of the author: The results of the MN assay revealed that alpha-cypermethrin showed a weak genotoxic effect on human peripheral blood lymphocyte culture. Alpha-cypermethrin has aneugenic effects shown by the FISH assay. Alpha-cypermethrin might be a spindle poison or caused damaged to centromere/kinetochore function. The results of the cell viability method indicated that alpha-cypermethrin induced cell death, at lower concentrations via apoptosis.

Conclusion of the applicant: The study shows some minor methodic deficiencies with regard to mitogenic stimuli (not over 48 h before test substance incubation as requested for human lymphocytes according to OECD guideline 487), cell proliferation index not calculated according to OECD guideline 487 (Nuclear devision index is not mentioned in OECD guideline 487), no presentation of historic control range. Besides this, the results are considered borderline and not relevant: First the viability of the lymphocytes was significantly reduced at all concentrations, and reached a cytotoxic level of 60% at 30µM and 45% at 15 µM. Significant induction of cytotoxic effects were also confirmed with the ethidium bromide/acridine orange stain at concentrations as low as 2 µM. Secondary, the MN frequency was not significantly increased at any concentration tested. FISH staining was performed to distinguish between MN induced by aneugenic or clastogenic effect. Since the increase in MN was not significant, this staining is of limited use. In addition, the validity of this staining method is of doubt based on the occurrence of only 82% C+ MN cells with the aneugenic positive control.

In conclusion, the borderline effect in this in vitro MNT is considered to be not relevant in vivo, especially based on the results of an in vivo MNT in mouse bone marrow (AL-435-008).

Furthermore, this study was performed with a 10% EC formulation of unknown composition, so that the result is not directly assignable to the active ingredient alpha-cypermethrin. Genotoxicity testing of formulations is in addition no data requirement within the EU.

Classification of study: not further considered

CA 5.4.2 In vivo studies in somatic cells

A summary of the *in vivo* genotoxicity studies evaluated for Annex I inclusion of alpha-cypermethrin is summarized in Table 5.4.2-1 below.

Table 5.4.2-1: *In vivo* mutagenicity studies with alpha-cypermethrin

Study type	Test System	With S-9 mix	Result	Reference
In vivo Cytogenicity OECD 474 (1983)	Mouse Micronucleus test (0, 1, 5, 10 mg/kg bw); Oral gavage	Not applicable	Negative	1995, AL-435-008 (Cyanamid Dossier)
In vivo Cytogenicity (B11-92/69/EEC)	Chromosome aberrations in rat bone marrow (0, 2, 4, 8 mg/kg bw); Oral gavage	Not applicable	negative	1984, AL-435-003 (Cyanamid Dossier)
In vivo Alkaline elution of rat DNA	DNA single strand breaks in rat liver/ hepatectomied rats (40 mg/kg bw); Oral gavage	Not applicable	Negative	1982, AL-435-004 (Cyanamid Dossier)

CA 5.4.3 In vivo studies in germ cells

A summary of the *in vivo* genotoxicity studies in germ cells evaluated for Annex I inclusion of alpha-cypermethrin is summarized in Table 5.4.3-1 below.

Table 5.4.3-1: In vivo mutagenicity studies with alpha-cypermethrin in germ cells

Study type	Test System	With S-9 mix	Result	Reference
In vivo Germ cell mutation	Rodent dominant lethal test Concentrations: 5, 10, 15 mg/kg bw, 5 consecutive days	Not applicable	Negative	Sengupta, 1989a, (Gharda Dossier)[1]

[1] These studies are not BASF proprietary, but owned by Gharda, Chemical Ltd. Bombay, a further notifier for Annex I listing of alpha-cypermethrin in 1999.

CA 5.5 Long-Term Toxicity and Carcinogenicity

Studies evaluated in the draft monograph of the rapporteur member state Belgium of Sep. 1999: Studies evaluated in the alpha-cypermethrin (BAS 310 I) draft monograph of rapporteur member state Belgium of September 1, 1999 consisted of: one chronic toxicity study in rats performed with cypermethrin and one carcinogenicity studies in mice performed with alpha-cypermethrin. These studies have been evaluated by European authorities and Belgium as the rapporteur member state and were considered to be acceptable. For the convenience of the reviewer, these studies are summarized below as extracted from the monograph. The results are summarized in Table 5.5-1.

Table 5.5-1: Summary of already peer-reviewed long-term toxicity/carcinogenicity studies as available in the monograph (1999)

Study	Dosages (mg/kg bw/day)	NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Main adverse effect	Reference and year
Cypermethrin 2 year chronic toxicity study in Wistar rats (diet administration of 0, 1, 10, 100,1000 ppm)	0, 0.05, 0.5, 5 and 50	5 [100 ppm]	50 [1000 ppm]	Systemic toxicity: Reduced body weight and food consumption; increased blood urea level Oncogenicity: no increase in tumor incidences	[redacted] 1978 (Dossier Cyanamid); CY-427-001 *CY-427-002* *CY-427-003* *CY-427-004*
Alpha-cypermethrin 78 week carcinogenicity study inCD1 mice (diet administration of 0, 30, 100, 300 ppm)	M: 3.0, 10.6 and 35.2 F: 3.5, 11.5 and 37.7	3 [30 ppm]	10.6 [100 ppm]	Systemic toxicity: Reduced body weight gain and food conversion efficiency; clinical signs of toxicity Oncogenicity: no evidence of carcinogenicity	[redacted] 1996 (Dossier Cyanamid) AL-428-001 AL-428-002# AL-428-003#

These studies were not listed in the former reference list but have been reported in the monograph of 1999

The results of the 2-yr chronic/carcinogenicity studies in rats with cypermethrin indicated that a maximum tolerated dose was clearly met at the high dose of 1000 ppm in males and females (corresponding to 50 mg/kg bw/day). This is demonstrated by a decrease of body weight (at nearly all weeks in males, and from weeks 1 to 75 in females) and a decrease of food consumption and an increased blood urea level. No further substance related adverse effects were observed. There was no evidence of a treatment-related increase in neoplasms. Due to the composition and toxicological similarities of cypermethrin and alpha-cypermethrin, it is unlikely that there would be significant differences between these two compounds with regard to carcinogenicity. These findings are supported by the absence of genotoxic activity for alpha-cypermethrin.

A carcinogenicity study in mice was conducted up to 300 ppm with alpha-cypermethrin. The maximum tolerated dose was found to be at 100 ppm as evidenced by body weight gain depression, reduced food conversion efficiency and clinical signs of toxicity. No further substance related effects were observed in this study. There was no evidence of a treatment-related increase in neoplasms.

Based on the available data, the following endpoint was determined during the last Annex I listing of alpha-cypermethrin concerning:

Long term toxicity and carcinogenicity

Target/critical effect	Clinical signs of toxicity, body weight reduction
Lowest relevant NOAEL / NOEL	78-wks, mouse: 3 mg/kg bw/day
Carcinogenicity	No evidence of carcinogenicity

Supporting evidence from peer-reviewed studies from other DARs: Furthermore, one chronic toxicity study in rats performed with cypermethrin is available which has been evaluated in the zeta-cypermethrin draft monograph of November 2006 by the rapporteur member state Belgium. In addition beta-cypermethrin was investigated in a chronic toxicity study in wistar rats and the study has been evaluated in the beta-cypermethrin DAR by the rapporteur member state United Kingdom (2013).

Table 5.5-2: Summary of other peer reviewed long-term toxicity/carcinogenicity studies in rats published in related DARs

Study	Dosages (mg/kg bw/day)	NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Main adverse effect	Reference and year
Cypermethrin 2 year chronic toxicity study in Wistar rats (diet administration 0, 20, 150, 1500 ppm)	0, 1, 7.5 and 75	7.5 [150 ppm]	75 [1500 ppm]	Systemic toxicity: Reduced body weight gain, food consumption and efficient food utilization; clinical signs of toxicity Oncogenicity: no increase in tumor incidences	From the DAR of zeta- and cypermethrin: [redacted] 1982;
Beta-cypermethrin 2 year chronic toxicity study in Wistar rats (diet administration 0, 50, 250, 1000/500 ppm	0, 2.3/3.0, 12.3/16.5, 25/31.8 mg/kg bw males / females	12.3 [250 ppm]	25 [500 ppm]	Systemic toxicity: Clinical signs of toxicity, reduced body weight gain, food consumption and liver histopathology Oncogenicity: no increase in tumor incidences	From the DAR of beta-cypermethrin; (2009)

In these studies with cypermethrin and beta-cypermethrin in Wistar rats (summarized in the respective DARs) findings were broadly similar to those described in the original alpha-cypermethrin DAR for chronic toxicity studies in Wistar rats, with no evidence of carcinogenic potential and the primary findings being related to food consumption and body weight.

Studies submitted in this AIR 3 dossier:
There are no new studies available with alpha-cypermethrin that affects the overall evaluation for long-term toxicity and carcinogenicity. Thus, the conclusion for relevant endpoints for the current re-registration remains as follows:

Long term toxicity and carcinogenicity

Target/critical effect	Clinical signs of toxicity, body weight reduction **No classification required**
Lowest relevant NOAEL / NOEL	78-week, mouse: 30 ppm (3 mg/kg bw/day)
Carcinogenicity	No evidence of carcinogenicity **No classification required**

For convenience of the reviewer brief summaries of the studies listed in the DAR of the draft monograph of alpha-cypermethrin are provided below.

Cypermethrin, Rat 2-year oral study (1978, CY-427-001, CY-427-002, CY-427-003, CY-427-004)

Guidelines: Not fully in compliance with the test method B of directive 87/302/EEC.
Deviations: Low number of rats, blood albumin, glucose, GGT, and ornithine decarboxylase were not measured. Urinalysis was not performed.
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999.

The chronic toxicity and carcinogenicity of cypermethrin (WL 43467, batch 30, purity: 98%) was tested in 24 Wistar rats per sex fed the test substance at dietary concentrations of 1, 10, 100, and 1000 ppm. A control group of 48 animals per sex received untreated diet. Groups of rats were scheduled for necropsy after 6 months (6 per sex treated, 12 per sex control), 12 months (6 per sex treated, 12 per sex control), 18 months (12 per sex treated, 24 per sex control) and 2 years (24 per sex treated, 48 per sex control) of treatment.

Male and female rats exposed to 1000 ppm cypermethrin had reduced body weights throughout the study. The trend was consistent but not always statistically significant. During the first 13 weeks, small, sometimes statistically significant, reductions in food consumption were seen in the 1000 ppm group males and females. Thereafter, only minor fluctuations in food intake were observed.
No significant differences in survival were seen between treatment and control groups. Clinical chemistry revealed induction of Cyp 2B1 at high dose. There were no treatment-related effects on haematology and organ weights. Macroscopic and histopathological examination did not indicate any biologically significant lesions in any organ or other effects that were considered to be related to treatment. Sciatic nerves showed age dependent nerve fiber exhibiting the changes of Wallerian degeneration, but the incidence of this was not dose related.
A wide spectrum of tumors and degenerative and other lesions were seen at post-mortem and microscopically. None appeared to be compound-related.
Conclusion: Based on the described findings for both sexes the NOAEL (chronic effects) was determined to be 100 ppm corresponding to 5 mg/kg bw/day. Since no treatment related tumors were observed a NOAEL (for oncogenic effects) was determined to be 1000 ppm corresponding to 50 mg/kg bw/day for both sexes.

Alpha-cypermethrin, mouse 78-week oral study [redacted] 1996)

Guidelines: Fully in compliance with the test method B of Directive 87/302/EEC.
Deviations: No
GLP: Yes
Acceptance: The study was considered acceptable in the EU registration process 1999.

Groups of 72 male and 72 female CD-1 mice received diets containing 0, 30, 100 and 300 ppm of alpha-cypermethrin, equivalent to 3/3.5, 10.6/11.5 and 35.2/37.7 mg/kg bw for males /females, respectively. 20 males and 20 females of each group were sacrificed for interim examination, after 52 weeks, the remainders were sacrificed after 78 weeks of treatment.

Administration of alpha-cypermethrin (batch: 02156, purity: 95.4%; cis1/cis2 isomer ration: 1:99) to CD-1 mice for 78 weeks was associated with a number of non-specific effects.
Treatment of animals with 300 ppm alpha-cypermethrin resulted in poor growth performance and changes in appearance. In males, this was evident by a higher incidence of thin build during the in-life phase. Signs of reaction to treatment were, generally, confined to males receiving 300 ppm and included ungroomed coats, hair loss and surface encrustations on the skin and were probably related to the irritant properties of the test material. Other signs of reaction included occasional hunched posture in males and over-activity in females receiving 300 ppm.
Overall weight gains of males and females in the 300 ppm group were 26 and 24% lower than those of the controls and were associated with a reduced efficiency of food conversion. This is considered indicative of a non-specific toxicity rather than an influence of food intake. A similar, less marked effect on weight gain and food conversion efficiency was noted for males receiving 100 ppm.
No treatment-related effects on hematology or organ weight changes were seen, and macroscopic and histopathological examinations revealed no changes attributable to treatment with the test substance. There was no evidence that alpha-cypermethrin had any oncogenic potential at concentrations up to 300 ppm.
Conclusion: Based on the poor growth performance and changes in appearance the NOAEL was determined to be 30 ppm corresponding to 3.0 mg/kg bw/day for males and 3.5 mg/kg bw/day for female mice. Alpha-cypermethrin showed no oncogenic potential in mice up to the highest dose tested.

Alpha-cypermethrin, mouse 18-month oral study (disregarded; [redacted] 1990; Dosssier Gharda)

Guidelines: Not fully in compliance with test method B of Directive 87/302/EEC.
Deviations: Number of animals not given. Blood sampling not performed at 12 months. Animals were not weighed 1 time/week and food consumption is not measured 1 time/week. The dose is given in ppm in food and in some tables expressed in mg/kg of ?. Incomplete histopathology and individual data are missing.
GLP: No data
Acceptance: The study was not used for final evaluation in the EU registration process 1999.

Albino mice (Swiss strain) were expose to alpha-cypermethrin (99%) in diet, 6 day/week during 18 months at 6.5, 13 or 65 ppm for males and at 7.8, 15.6 or 78 ppm for females. This study has many qualitative deficits and is quite difficult to interpret. The deficits are: the experimental protocol is not clearly defined; the number of animals used is not given; blood sampling is not performed at 12 months; the animals were not weighed one time/week and food consumption is not measured one time/week; the dose is given in ppm in food and in some tables expressed as mg/kg ?; incomplete histopathology; individual data are missing. Due to these deficits the study was not used for the final evaluation.
Nevertheless, possible treatment related findings were observed: hepatocytic cytoplasm vacuolization with loss of cytoplasm in the high dose, scarring of renal capsule with varying degree of nephritis in the medium and high doses, fatty degeneration and tubular swelling in the high dose animals; dilatation of the spleen with distortions of the white and red pulp in the medium and high dose; spermatic tubes more or less choked with Sertoli cells, and degeneration of Leydig cells in the high dose with only temporary arrest of spermatogenesis in some animals.

CA 5.6 Reproductive Toxicity

Studies evaluated in the draft monograph of the rapporteur member state Belgium of Sep. 1999: The alpha-cypermethrin studies - evaluated in the draft monograph of the rapporteur member state Belgium of September 1999 - consisted of a three-generation study in rats and developmental toxicity studies in rats and rabbits. Furthermore, a three-generation study in rats is available with cypermethrin. These studies have been evaluated by European authorities and Belgium as Rapporteur member state and were considered to be acceptable. For the convenience of the reviewer, these are summarized below as extracted from the monograph and discussed during the ECCO Peer review meeting (2001) - Full Report on alpha-cypermethrin (DocID AL-901-031). For a better overview the endpoints parental toxicity, reproductive toxicity and developmental toxicity were addressed separately.

Table 5.6-1: Summary of already peer-reviewed reproduction toxicity studies as available in the monograph (1999) and supplemented according to discussions during the ECCO Peer review meeting (2001)

Study	Dosages (mg/kg bw/day)	NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Main adverse effect	Reference and year
Alpha-cypermethrin 3-Generation study, oral, Young Charles Foster rats (gavage, vehicle unknown)	M: 0, 2.5, 10 and 25 F: 0, 5, 10 and 20	Parental toxicity: 20 Reproductive toxicity: 20	Parental toxicity: > 20 Reproductive toxicity: > 20	Parental toxicity: no maternal toxicity Reproductive toxicity: no effects	[redacted] 1989b (Gharda Dossier)
Cypermethrin 3-Generation, oral, feed Wistar rats (diet concentrations: 0, 10, 100 and 500 ppm)	0, 1, 10 and 50 (nominal) Calculated: 0, 1, 9.0/10.8, 44.8/51.4	Parental toxicity: 9 [100 ppm] Reproductive toxicity: 9 [100 ppm] Developmental toxicity: 9 [100 ppm]	Parental toxicity: 44.8 [500 ppm] Reproductive toxicity: 44.8 [500 ppm] Developmental toxicity: 44.8 [500 ppm]	Parental toxicity: reduced body weight and food consumption; Reproductive toxicity: reduced litter size No adverse effects on fertility. Developmental toxicity: reduced pup body weight day 21 (F1B and F3B)	[redacted] 978 CY-430-001 (Cyanamid Dossier) Addendum/ Corrigendum CY-430-002 CY-430-003 CY-430-004 CY-430-005
Alpha-cypermethrin Developmental toxicity, gavage (days 6-15), Sprague-Dawley rat	0, 3, 9, 18/15 and 15	Maternal toxicity: 9 Developmental toxicity: 9	Maternal toxicity: 15 Developmental toxicity: 15	Maternal toxicity: Clinical signs (unsteady gait, hind limb splay), reduced body weight and food consumption Developmental toxicity: Reduced fetal weight	[redacted] 1994a AL-432-002 [redacted] 1994a AL-432-001 (Cyanamid Dossier)

Table 5.6-1: Summary of already peer-reviewed reproduction toxicity studies as available in the monograph (1999) and supplemented according to discussions during the ECCO Peer review meeting (2001)

Study	Dosages (mg/kg bw/day)	NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Main adverse effect	Reference and year
Alpha-cypermethrin Developmental toxicity, gavage (days 7-19), New Zealand White rabbits	0, 3, 15 and 30	Maternal toxicity: 15 Developmental toxicity: 30	Maternal toxicity: 30 Developmental toxicity: > 30	Maternal toxicity: Reduced body weight and food consumption Developmental toxicity: No evidence of developmental toxicity.	[redacted] 1994b AL-432-004 [redacted] 1994b AL-432-003 (Cyanamid Dossier)

The summary of reproductive toxicity studies as given in the monograph and amended in accordance to the discussions at the ECCO 102 meeting is as follows: "*There is no good study currently available to assess the multigeneration reproductive toxicity potential of alpha-cypermethrin in the rat. Data from a badly documented study did not show any toxic effect on reproduction or development. This is compatible with the results of an acceptable study with cypermethrin. Cypermethrin did not cause any significant effects on fertility or reproductive performance of rats.*" The delayed pregnancies mentioned in the DAR were stated as being a misinterpretation of the rapporteur, therefore it is concluded that there was in addition no evidence of significant pre- or post-natal effects.

"*In the developmental toxicity studies in rat and rabbit with alpha-cypermethrin, there was no evidence of adverse effects on embryonic development. There was no maternal or developmental toxicity associated with oral administration of alpha-cypermethrin.*

In the dossier on cypermethrin, the RMS reported data from the open literature in which it was suggested that cypermethrin might induce a delayed maturation of cerebral cortex and caused a long-lasting impairment of dopamine receptors. The neurobehavioral findings and biochemical changes published in the open literature are very limited in scope, are not reproduced and no firm conclusions can be drawn. As cypermethrin has been tested in a multigenerational reproduction study in rats, without any indication of persistent effects in the offspring, which were also exposed to the substance neonatally, it is suggested that receptor binding changes are not predictive or causally related to the behavioral changes. Moreover, the vulnerable phase for humans during the brain growth spurt is prenatal and not post-natal as in rodents. Therefore, exposure of the human fetus will be limited by maternal pharmacokinetics as well as maternal toxicity."

Based on the available data, the following endpoints were determined during the last Annex I listing of alpha-cypermethrin.

Reproductive toxicity

Reproduction target / critical effect	No reproductive toxicity
Lowest relevant reproductive NOAEL / NOEL	>20 mg/kg bw/day
Developmental target / critical effect	Reduced fetal weight at maternal toxic doses
Lowest relevant developmental NOAEL / NOEL	Rat: 9 mg/kg bw/day

Supporting evidence from peer-reviewed studies not belonging to BASF from other DARs: According to the DARs of zeta-cypermethrin and beta-cypermethrin, prepared by Belgium (2008) and UK (2013), neither cypermethrin, nor zeta-cypermethrin or beta-cypermethrin are considered to show evidence for reproduction toxicity from scientifically acceptable studies. For the convenience of the reviewer, the reproduction toxicity studies of zeta-cypermethrin, cypermethrin, and beta-cypermethrin are summarized in the Table below as extracted from the DARs.

Table 5.6-2: Summary of already peer-reviewed confirmatory data on cypermethrin, zeta-cypermethrin and beta-cypermethrin (as presented in the DARs)

Study	Dosages (mg/kg bw/day)	NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Main adverse effect	Reference and year
Cypermethrin 3-Generation, oral, feed Wistar rats (diet concentrations: 0, 50, 150 and 750/1000 ppm)	**Conversion factor of 1/10*:** 0, 5, 15, 75	Parental toxicity: 5 [50 ppm] Reproductive toxicity: 75 [750 ppm] Developmental toxicity: 15 [150 ppm]	Parental toxicity: 15 [150 ppm] Reproductive toxicity: - Developmental toxicity: 75 [750 ppm]	Parental toxicity: decreased parental bw gain, food consumption and food utilization Reproductive toxicity: no effects Developmental toxicity: decreased litter body weight gain in F1B, F2B and F3B	[redacted] 1982 (taken from DAR of zeta-cypermethrina nd adapted to 1/10 conversion factor)
Zeta-cypermethrin 2-Generation study, oral, feed, Sprague-Dawley rats (diet concentrations: 0, 7.5, 25, 100, 375, 750 ppm (exceeded MTD)	0.4, 1.5, 5.9, 22 and 43	Parental toxicity: 5.9 [100 ppm] Reproductive toxicity: 22-43 [375-750 ppm] Developmental toxicity: 5.9 [100 ppm]	Parental toxicity: 22 [375 ppm] Reproductive toxicity: - Developmental toxicity: 22 [375 ppm]	Parental toxicity: Clinical signs of neurotoxicity, reduced body weight, body weight gain, food consumption Reproductive toxicity: - Developmental toxicity: reduced pup weight during lactation; pup mortality at 750 ppm probably related to less maternal care	[redacted] 1991 (taken from DAR of zeta-cypermethrin)
Beta-cypermethrin 2-generation study, oral, gavage, Sprague-Dawley rats in 1% CMC	**0, 3, 12, 30/40**	Parental toxicity: 30 Reproductive toxicity: 30 Developmental toxicity: 30	Parental toxicity: >30 Reproductive toxicity: >30 Developmental toxicity: >30	No treatment related effects on reproductive performance, parents or offspring up to the highest dose tested.	2005 (taken from DAR of beta-cypermethrin)

* conversion factor of 1/20 is considered to be not adequate based on the comparison with the study performed by [redacted] 1978 in the same rat strain at the same time

The study performed with cypermethrin was summarized in the DAR of Zeta-cypermethrin in the following passage:

*Parental toxicity was evidenced by the reduced bw gain, food consumption/utilization seen at 150 ppm (*15 mg/kg bw*). Signs of neurotoxicity were reported at the high dose of 1000 ppm (*100 mg/kg bw*). No reproductive toxicity was observed. Body weight gain was reduced in pups of both sexes in some litters from the 750 ppm group where clear maternal toxicity was observed. (taken from the DAR of Zeta-cypermethrin except for the substance intake values).*

The study performed with zeta-cypermethrin was summarized in the DAR of Zeta-cypermethrin in the following passage:
In a 2-generation study, zeta-cypermethrin was investigated at doses ranging from 7.5 to 750 ppm [in Sprague-Dawley rats]. The NOAEL reproduction at 750 ppm (43 mg/kg bw/d)for the P1 generation and 375 ppm (22 mg/kg bw/d) for the F1 generation is proposed because mortality prevented evaluation of the reproductive performance of the 750 ppm group second generation. Decreased body weights, body weight gains, and clinical signs of toxicity were seen at 375 ppm (22 mg/kg bw/d). Reduced pup body weights in both generations during the lactation periods and pup mortality were reported at maternal toxic dose of 375 ppm.

The study performed with beta-cypermethrin was summarized in the DAR of beta-cypermethrin in the following passage:
"In the two-generation-study, using gavage dosing, there were no adverse effects on reproductive performance or on general toxicity in parents up to the highest dose tested of 30 mg/kg bw/day."
This study also included oestrus cycle and sperm analysis data at the highest dose of the F0 and F1 generation and did not find adverse effects.

Studies submitted in this AIR 3 dossier (not yet peer-reviewed):
There are no new studies available with alpha-cypermethrin that could affect the overall evaluation for reproductive toxicity. Thus, the conclusion for relevant endpoints for the current re-registration remains as follows:

Reproductive toxicity (SANCO/11802 data point 5.6)

Reproduction target / critical effect	No reproductive toxicity effects No classification required
Lowest relevant reproductive NOAEL / NOEL	> 20 mg/kg bw/day
Developmental target / critical effect	Reduced fetal weight at maternal toxic doses No classification required
Lowest relevant developmental NOAEL / NOEL	Rat: 9 mg/kg bw/d

No classification for reproductive or developmental toxicity is considered necessary.

CA 5.6.1 Generational studies

Alpha-cypermethrin - Three Generation reproduction study in rats ([redacted] 1989b)

Guidelines: Not fully in compliance with the test method B of directive 87/302/EEC.
Deviations: Daily dosing of males should begin when rats are about 5 to 9 weeks old for 10 weeks prior to the mating period. Administration route is not defined. During premating and mating periods, after parturition and during lactation food consumption was not measured and only female body weights were reported.
GLP: No
Acceptance: The study was considered acceptable in the EU registration process 1999

Eight males and sixteen female Young Charles Foster rats were exposed to alpha-cypermethrin at doses of 0, 2.5 (♂) – 5 (♀), 10 and 25 (♂) – 20 (♀) mg/kg bw/day. Dosing of the test compound started at about 100 days of age. Male rats were treated for at least 60 days (= 8.5 weeks) and females for 15 days prior mating, treatment then continued through mating, gestation and lactation. At weaning the administration of the substance is continued to F1 offspring during their growth into adulthood, mating and production of F2 generation. The administration is continued as for F1 generation till F3 generation is weaned and attains 100 days age. The conduct of the study was consistent to OECD 416 (2001) in all important aspects with the exception that the test procedure was extended to three generations and the daily dosing of males should begin when rats are about 5 to 9 weeks old for 10 weeks prior to the mating period. Administration route and vehicle is not defined. During pre-mating and mating periods, after parturition and during lactation food consumption was not measured and only female body weights were reported.
Findings:
Body weight was normal in pregnant females. Parturition was normal. The various reproductive indices calculated for the test and control animals do not indicate any adverse effect on reproduction and lactation response. There is no significant difference in the total number of pups born to control and treated P0, P1 and P2 mothers. The proportion of live pups at birth and the proportion surviving to 3/4 days post-partum in various treatment groups are similar to the controls and highest and intermediate groups. The number of pregnancies observed in the P0, P1, and P2 generations are comparable in the controls and highest and intermediate groups. The comparative litter size of F1A, F2A and F3A dams is similar in all the treatment and control groups. (In the discussions during the ECCO Meetings the RMS stated that the delayed pregnancy finding was a mis-interpretation. See DocID AL-901-031, page 100 of 222, Point No 4. This is also documented in the List of endpoints in the Review Report.) No parameter of developmental toxicity is discussed in this summary.

Conclusion
Alpha-cypermethrin was not reported to have adverse effects. Therefore the NOAEL for systemic toxicity and reproductive toxicity is considered to be 20 mg/kg bw/day.

Cypermethrin - Three Generation reproduction study in rats ([redacted] 1978) CY-430-001-CY-430-005)

Guidelines: Not fully in compliance with the test method B of directive 92/69/EEC.
Deviations: Males and females were exposed for 5 weeks: normally, in male rats dosing is continued for 10 weeks prior to the mating period. Females must be dosed throughout the 3 week mating period, pregnancy and up to the weaning of the F1 offsprings.
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.
Remark: The terminology used to designate the parental and offspring groups of animals in the study report is different from the currently accepted designators. The report uses "F0, F1,and F2" to describe the original parental and first, second and third generation offspring generations with appropriate subscripts for the first or second litter in that generation. Current terminology employs the descriptors "P0, F1 and F2" to describe the parental generations and F1, F2 and F3, with the added subscript "a" or "b" to describe the offspring. The terminology used here follows the latter scheme and updates that used in the report.

Groups of approx. 30 Wistar rats per sex in the parental generation (and less in other generations due to mating not resulting in pregnancy, single sex litters, or low survival up to weaning, see Table below) received cypermethrin at dietary concentrations of 0, 10, 100 or 500 ppm throughout the entire experimental period up to weaning of F3 litters (pre-mating, mating, gestation and rearing).

		Number of animals			
		Control	10 ppm	100 ppm	500 ppm
Parents P0	Male	30	30	30	30
	Female	30	30	30	30
F1	Male	28	25	28	25
	Female	28	25	28	25
F2	Male	18	20	19	19
	Female	18	20	19	19

Two litters were produced from each parental generation, one was killed at weaning (PND 21) whereas the second litters were randomly selected for mating. The conduct of the study was consistent to OECD 416 (1983) in all important aspects, with the exception that the test procedure was extended to three generations, that males of the P-generation were dosed for less than 10 weeks prior to mating, and that food consumption and body weight were only recorded during the pre-mating period.

Findings in the adults:
General health and behaviour of treated animals were comparable to control animals. P0, F1 and F2 body weights were lower at 500 ppm but not at 10 and 100 ppm. Reduced food intake for the P0 females at 500 ppm (week 3 to 7), sporadically for males at 10, 100 and 500 ppm, was lower than controls and was believed to be related to non-palatability of the diet. Lower food intakes at weeks 6 and 7 were in females were associated with adverse body weight effects and were considered to be related to exposure.
Fertility, gestation, viability and lactation indices were similar for treated and control animal within each generation.

Table 5.6.1-1: Three generation body weight and food consumption data

Dose	0 ppm		10 ppm		100 ppm		500 ppm	
Adult rat P0	♂	♀	♂	♀	♂	♀	♂	♀
Food intake			↓wk 3,4,5 (4%)		↓ wk 3,4 (4%)		↓ wk 3&7 (7%)	↓ wk 3-7 (6-7%)
Body weight							↓	↓ (4-5%)
Adult rat F1, F2								
Food intake			↓ wk 3 (F1: 8%)					↓ wk 3-7 (F1: 6-8%) ↓ wk5&7 (F2: 11-16%)
Body weight							↓ (F1: 4-5%)	↓ (F1: 4-7%) ↓ (F2: 5-6%)

Findings in the litters:
At 500 ppm: Lower litter sizes and weights and lower pup weights, mainly for the F1A. Mean pup weights were significantly lower compared with controls for F1B females pups and for pups of both sexes for F3B pups.
At 100 ppm and at 10 ppm, the number of female pups per litter for F1B litters was higher. No treatment-related adverse effects on litter weights were observed. Mean pup weight was reduced at in F3B at PND 21 male offspring. Using the litter as experimental unit, no statistically significant differences were reported at 10 and 100 ppm.
At 10 ppm, significantly lower litter size for F1A pups and F2B pups were not considered to be toxicologically relevant as similar changes were not observed at 100 ppm. Significantly increased mean pup weights recorded for F1A litters reflected the lower litter sizes at day 7,14 and 21.

Table 5.6.1-2: Three generation reproductive toxicity in the rat – litter data.

Endpoint/dose	0 ppm	10 ppm	100 ppm	500 ppm
Litter survival:				↓ F1A (PND 0, 7, 21)
Litter size:				
F1A				↓ PND 7-21
F1B, F2A, F2B, F3A, F3B	No effect			
Number of female pups / litter				
F1A				↓ PND 1&21
F1B		↑ PND 1&21	↑ PND 1&21	
F2A, F2B, F3A, F3B	No effect			
Mean litter weights:				
F1A				↓ PND 7-21
F1B		↑ PND 7		
F2A, F2B, F3A, F3B	No effect			
Mean pup weight:				
F1A		↑ PND 14 & 21		
F1B				♀: ↓ 10 % PND 21
F2A, F2B, F3A	No effect			
F3B		♂: ↓ 10 % PND 21	♂: ↓ 11 % PND 21	♂: ↓ 9% PND 21 ♀: ↓ 6 % PND 21

Upon necropsy, no compound-related gross or microscopic pathological findings were observed. Beside the tables described in the monograph, more detailed tables are found in full report on alpha-cypermethrin from the ECCO peer review program.
Remark from BASF: The test compound intake has been calculated based on the weekly mean intake values

		Test compound intake (mg/kg bw/d)*	
		100 ppm	500 ppm
Parents P0	Male	9.9	50.3
	Female	11.2	56.4
F1	Male	9.6	48.9
	Female	11.1	57.2
F2	Male	9.0	44.8
	Female	10.8	51.4

* Calculated with weekly mean values (week 2-7);
NOAEL/LOAEL=NOAEC/LOAEC x daily food consumption / body weight

Based on these calculations it seems to be adequate to take a factor of 1/10 for Wistar rats in this typ of study when converting diet concentration in dose. This was already done in the previous monograph and is in compliance with the EFSA Guidance on selected default values (EFSA Journal 2012; 10(3): 2579). This information is of importance when the substance intake in the cypermethrin study of [redacted] 1982 is estimated. In this study a default value of 1/20 was considered but seems to be not adequate with regard to this study in the same strain of rats.

Conclusion
Based on the described maternal toxicity and reproductive effects at 500 ppm (corresponds to approx. 50 mg/kg bw (lowest value 44.8 mg/kg bw/day) the NOAEL for maternal toxicity and reproductive effects was considered to be 100 ppm (corresponding to approx. 10 mg/kg bw (lowest value 9 mg/kg bw/day).

Additional data taken from the open literature:

Report:	CA 5.6.1/1 Anonymous, 2006a Draft Assessment Report (DAR) Zeta-Cypermethrin - Volume 1 to volume 3 - Initial risk assessment provided by the RMS Belgium of the third stage (part A) of the review programme referred to in article 8(2) of council directive 91/414/EEC 2006/7013990
Guidelines:	none
GLP:	no (certified by none)

Remark: [redacted] 1982 ***) (copied from Monograph of zeta-cypermethrin)***
Cypermethrin - Three Generation reproduction study in rats [redacted] 1982)

Remark from BASF:
The conversion of concentration in feed into daily dose in rats was estimated in the study of [redacted] 1982 based on a factor of 1/20. As this study was performed in the same rat strain than that used in the Cypermethrin study performed by [redacted] 1978 (see study before) it is considered more adequate to use a factor of 1/10. This is also in accordance to the EFSA Guidance on default values, where only chronic toxicity studies are converted with a 1/20 factor, whereas subchronic studies are converted with a 1/10 factor. Taking this into account the NOAEL in the cypermethrin study performed by [redacted] 1978 is considered adequate as it is below the LOAEL generated in [redacted] under consideration of a conversion factor of 1/10. Furthermore the study comprises some discrepancies that render the study less valid. On the one hand there was a light cycling problem which is important for the correct mating and fertility parameters and on the other hand 1/3 of the control animals are mentioned to be sub-/or infertile, which indicate further difficulties in the study that might have compromised the integrity of the study or affect the scientific interpretation of the results.

Guidelines:	*Study protocol is not fully conform to EEC 87/302 Annex V B.35 or OECD guideline 416 (1983)*
Deviations:	*Highest dose was reduced at the end of premating period; weanling was postponed. Females, which were not gravid after 10 days, were placed by another male. Histopathology of vagina and coagulating gland was not performed.*
GLP:	*No (not mandatory at that time)*
Acceptance:	*The study was considered acceptable in the EU registration process 2008.*

Material and methods: 15 males and 30 females Wistar rats received in their diet cypermethrin (batch n° P19, P24, P26; purity 91.5, 93.1, 90.6 %) at 0, 50, 150, 750 ppm throughout the entire experimental period up to weaning of F3 litters. The high dose level was 1000 ppm for the first 12 weeks of the F0 parental rats but was reduced to 750 ppm due to neurological disturbance seen in the early part of the period. Stability and homogeneity in food was evaluated and satisfying and food was prepared every one to two weeks.
Findings: The high dose level was 1000 ppm for the first 12 weeks of the P0 parental rats but was reduced to 750 ppm due to neurological disturbance seen in the early part of the period.
Two problems were encountered in the conduct of the study. Firstly the automatic time switch on the lighting in the animal room was faulty for up to one month leaving rats in almost light on each day during mating and gestation to produce F1A litter. During continuous light the diurnal rhythms would have been affected leading to failure of the normal estrus cycle. This was the most probable cause of the overall low fertility during production of the F1A litter.
Secondly, there was difficulty in obtaining the required number of F1B and F2B litters; it was necessary to arrange three pairings before this was achieved. No reason for this difficulty was identified and it was unexpected since the F1 parents had readily produced the F2A litters a few weeks earlier with no difficulties and since the probable reason for low fertility of P0 females producing the F1A litter had been identified and rectified.
The above problems did not compromise the integrity of the study or affect the scientific interpretation of the results.
Clinical signs: During the first three weeks of feeding of the F0 parents at 1000 ppm, neurological disturbance was seen in most rats. The clinical signs were typical of those associated with pyrethroid toxicity. For this reason, the dietary concentration of cypermethrin administered in the top dose group was reduced from 1000 to 750 ppm after feeding to the F0 parents for 12 weeks. In addition separation of pups from mothers took place at 28 instead of 21 days post partum and pups were then kept in litters for a further seven days before selection of the next generation of parents. These alterations in experimental design ensured that there was no difficulty with undue toxicity in pups of subsequent generations.
No treatment related changes were detected in the central or peripheral nervous systems of any animal examined on the study in spite of clinical signs of neurological disturbance at 1000 ppm. This was also true for the F0 male, which died after showing signs of neurological disturbance. A dietary concentration of 750 ppm produced a definite toxic effect.
Body weight gain: a reduction in parental body weight gain throughout the study was reported, generally accompanied by reduced food consumption and less efficient food utilization. The only consistent evidence of a toxic effect in rats fed 150 ppm was a reduction in body weight gain in F2 parents. There was no evidence of any effects at a dietary concentration of 50 ppm.
In pregnant females the only effects seen was reduction in body weight gain in F1 and F2 females producing their respective B litters, at 750 ppm. Similarly the only effect in offspring was a reduction in the body weight gain of B litter offspring at 750 ppm.
Reproductive parameters were not affected.
In all the offsprings of each generation there were no clinical or behavioral abnormalities, which could be related to treatment.
Soft tissue examination of pups revealed no abnormalities, which could be compound-related.

Table 5.6.1-3: three generation rat study with cypermethrin

Dose/endpoints	***0 ppm***		***50 ppm***		***150 ppm***		***750/1000 ppm***	
Parent data	*M*	***F***	***M***	***F***	***M***	***F***	***M***	***F***
Mortality: killed/found dead								
F0	*1*	*1*		*1*	*1*	*3*	*1*	*1*
F1		*1*		*1*			*1*	
F2	*1*	*2*		*2*				
Clinical signs							*Neurological disturbance wk 1-3*	
Body weight gain								
F0						*↓5%*	*↓8% wk1-5*	*↓9% premating*
F1				*↓6% wk3-11*				*↓6% intermittently premating*
F2					*↓7%*	*↓9%*	*↓8%*	*↓10%*
Food consumption								
F0						*↓3%*	*↓3% wk1-2*	*↓7% premating*
F1							*↓12% wk 1-11*	*↓15% premating*
F2							*↓13% wk1-11*	
Food utilization								
F0							*↑16% wk9-12*	*↑5% wk1-4*
F2						*↑15% wk1-4*		
Reproductive failure in parents								
N° F0 females mated for:								
F1A litter		*30*		*30*		*30*		*30*
F1B litter		*29*		*29*		*28*		*29*
Total n° sub-/infertile females		*11*		*19*		*11*		*13*
N° F1 females mated for:								
F2A litter		*29*		*30*		*28*		*30*
F2B litter		*28*		*30*		*28*		*30*
Total n° sub-/infertile females		*10*		*11*		*11*		*6*
N° F2 females mated for:								
F3A litter		*29*		*29*		*30*		*30*
F3B litter		*28*		*29*		*30*		*30*
Total n° sub-/infertile females		*11*		*12*		*13*		*9*
Reproductive performance			*No adverse effects on length of gestation, live born index, survival index, maternal neglect index, male fertility, viable litter size*					
Initial pregnancy weight:								
F0 parent for:								
F1A litter							*↓8%*	
F1B litter							*↓5%*	
F1 parent for:								
F2A							*↓6%*	
F2B					*↓6%*		*↓8%*	
F1 parent for F2B litter day 14							*↓10%*	
***F2 parent for**:*								
F3A litter					*↓7%*		*↓10%*	
F3B litter					*↓5%*		*↓11%*	
F2 parent for F3B litter day 14							*↓10%*	

Table 5.6.1-3: three generation rat study with cypermethrin

Dose/endpoints	***0 ppm***		***50 ppm***		***150 ppm***		***750/1000 ppm***	
Parent data	*M*	***F***	***M***	***F***	***M***	***F***	***M***	***F***
Offspring data								
Litter bw gain								
F1B from day 10							*↓14%*	*↓21%*
F1B from day 21							*↓16%*	*↓16%*
F1B from day 28							*↓12%*	*↓12%*
F2B from day 21							*↓12%*	*↓11%*
F2B from day 28							*↓9%*	*↓8%*
F3B from day 21							*↓17%*	*↓16%*
F3B from day 28							*↓14%*	*↓12%*

↓↑ Statistically significantly different from control

Conclusion*:*
NOAEL reproductive toxicity >750 ppm; NOAEL systemic toxicity = 50 ppm based on the decreased parental body weight gain, food consumption and food utilization seen at 150 ppm; NOAEL developmental toxicity = 150 ppm based on decreased litter body weight gain seen at maternal toxic doses of 750 ppm in F1B, F2B and F3B. A conversion factor of 1/10 is considered adequate for this kind of study in this rat strain.

see KCA 5.6.1/1 2006/7013990 Remark: [redacted]1991*) (copied from Monograph of zeta-cypermethrin)*
Cypermethrin - Three Generation reproduction study in rats [redacted] 1982)

Guidelines: Not fully in compliance with the test method B.35 of directive 87/302/EEC or OECD guideline 416 (1983).
Deviations: Males were changed if mating did not succeed after 2 weeks.
GLP: Yes (only attest of study director)
Acceptance: The study was considered acceptable in the EU registration process 2008.
Material and methods: 30 males and 30 females Sprague Dawley rats received in their diet zeta-cypermethrin (batch n° E-6539-78; purity 89.6 %) at 7.5, 25, 100, 375, 750 ppm for 12 weeks prior to mating and throughout the reproduction phase. Stability and homogeneity in food was evaluated and satisfying and food was prepared every one to two weeks.
Findings:

Table 5.6.1-4: Mean test substance intake (mg/kg bw/day)

Period	***0 ppm***		***7.5 ppm***		***25 ppm***		***100 ppm***		***375 ppm***		***750 ppm***	
P1	***M***	***F***	***M***	***F***	***M***	***F***	***M***	***F***	***M***	***F***	***M***	***F***
Premating	*0*	*0*	*0.4*	*0.6*	*1.5*	*1.9*	*5.9*	*7.4*	*22.1*	*27.6*	*43.4*	*53.*
Gestation		*0*		*0.5*		*1.6*		*6.4*		*24.2*		*47.8*
Lactation		*0*		*0.9*		*2.9*		*11.8*		*40.7*		*67.5*
F1												
Premating	*0*	*0*	*0.4*	*0.6*	*1.8*	*2.1*	*7.2*	*8.6*	*27.8*	*32.9*	*-*	
Gestation		*0*		*0.5*		*1.7*		*6.8*		*25.3*		
Lactation		*0*		*0.9*		*2.9*		*12.1*		*45.7*		

Statistics: Bartlett's test, Kruskall Wallis test, analysis of variance, Fisher's exact test and Dunnett's test were applied.

Parental data: see ***Fehler! Verweisquelle konnte nicht gefunden werden.***

Mortality
P1 generation: in male rats mortality was not substance related. In females, 2 deaths, occurring during lactation, at top dose are considered compound-related. One 100 ppm female rat was found dead on day 28 of lactation period was not considered compound related because there was no clinical observations prior to its death.
F1 generation: At 750 ppm deaths of 24/27 male and of 27/30 females rats was reported.
Clinical signs
P1 males at 750 ppm had soft or liquid feces. Female P1 rats that died at top dose had clinical signs related to substance intake. These effects occurred essentially during lactation.
Necropsy findings in females that were found dead such as urine stained abdominal fur, bedding in the mouth and no feed in the stomach and gastric erosion were considered treatment-related.
F1 generation: at 750 ppm, small pups were hypersensitive to sound, ataxic and had whole body tremors. This group was eliminated from the study at the end of week 3 post weaning. Alopecia occurred at 375 ppm.
In females, the small pups dying at 750 ppm were ataxic, had whole body tremors and clonic convulsions. They had urine-stained abdominal fur, gaseous distended stomach and/or intestines and gastric erosion. This group was eliminated from the study. Localized alopecia was increased in females at 375 ppm during premating, gestation and lactation periods.
Body weights
P1 generation: In male rats, at top dose, bw gains were frequently reduced for the entire premating period resulting in significant reductions in average body weights. Body weight gains were reduced for the 375 ppm group on days 64 to 71. This observation was considered unrelated to treatment because it was a single event followed by a significant increase on days 71 to 79 of the study.
Female rats given the 750 ppm had significant reductions in bw gain during premating and lactation period. Bw was reduced significantly for this group from the completion of the first week of exposure until termination at the end of lactation period. At 375 ppm, females tended to have reduced average bw gains and bw during premating period. Although not affected during gestation, maternal bw gains tended to be reduced for this group during the first 2 weeks of lactation period, and average bw were reduced during the lactation period.
F1 generation: Significant reduced bw occurred at 375 and 750 ppm prior to weaning in males and continued to be reduced through day 99 post weaning and for the 750 ppm through day 22 post weaning (this group was discontinued at this point of the study). Bw gains of males were affected at 375 and 750 ppm. Terminal bw was significantly reduced for the 375 ppm group and this group had significant increase in the relative brain weight.
In females, bw reduction was seen at 750 ppm groups prior to weaning; bw gain was reduced post weaning at 375 and 750 ppm. Average bw tended to be reduced for the 2 top dose groups on day 1-22 through premating period and for surviving 375 ppm group until the end of premating period. Body weight gains were not affected during gestation and lactation at 375 ppm but body weight was reduced during these periods. Terminal bw at 375 ppm was significantly reduced and brain weight was increased.
Food consumption:

P1 generation: was reduced in male rats at 375 and 750 ppm, during the first days of the study. At top dose, reduction was observed throughout the remainder of the premating period.
P1 females had reduced food consumption for the 375-ppm group at the beginning of the premating period. At 750 ppm, food consumption was reduced throughout the premating period. Food consumption was reduced for the 375-ppm group on days 0-6 of gestation period. Food consumption was reduced during the entire gestation
period at 750 ppm. Reduction was also observed during lactation period at 375 and 750 ppm.
F1 generation: food consumption was significantly reduced in males at 375 and 750 ppm during post weaning.
In females, food consumption was reduced at 375 and 750 ppm during the post-weaning period but was not affected during gestation at 375 ppm. Some periods were affected during lactation in this group.
Mating and fertility:
P1 generation: Mating performance of male and female rats of P1 was not affected. Duration of gestation and parturition, gestation indices, implantations, delivered pups, live born and stillborn pups or pup sex ratios was not affected.
F1 generation: at 375 ppm, no effects were seen on the mating performance or fertility of male and female rats. Duration of gestation and parturition, gestation indices, implantations, delivered pups, live born and stillborn pups or pup sex ratios was not affected.

Pup data: see Table 5.6.1-6
F1 generation: The viability and lactation indices were significantly decreased for the 750-ppm. At 750 ppm, litters died before day 28 postpartum. The average number of live pups/litter at weighing was significantly decreased on days 7, 14 and 21 postpartum. The average number of surviving pups/litter was significantly reduced on days 7, 14, 21, and 28 postpartum.
Increased numbers of pups in the 750-ppm group had clinical or necropsy observations related to the compound. Pups were pale, cold to touch, not nursing, weak and/or dehydrated. Necropsy of the 750 ppm pups frequently revealed observations interrelated with their failure to thrive, including evidence of maternal cannibalization, no
milk in the stomach, gaseous distention of GI tract, GI bleeding and incidental urinary tract bleeding.
At 375 and 750 ppm, reduction in pup body weight was observed. These effects first occurred on days 7 and 4-post partum for the 375 and 750-ppm groups respectively.
F2 generation: the 375-ppm group had significantly reduced pup body weights on day 14 and 21 postpartum. Gestation, viability and lactation were unaffected by exposure to 375 ppm. No clinical or necropsy observations for the offsprings were considered effects of the test substance.

Table 5.6.1-5: Two-generation rat study with zeta-cypermethrin: parental data

Endpoint/dose	***0 ppm***		***7.5 ppm***		***25 ppm***		***100 ppm***		***375 ppm***		***750 ppm***	
	M	***F***	***M***	***F***	***M***	***F***	***M***	***F***	***M***	***F***	***M***	***F***
Mortality: P1	*1*							*1*				*2*
F1					*1*		*1*				*24**	*27**
Clinical signs*: n° affected rats*												
Emaciated appearance P1										*2*		*25**
Ataxia P1												*24**
F1											*17**	*23**
Sound sensitivity P1												*21**
F1											*3**	
Clonic convulsions P1												*12**
End of tail cut P1												*6**
Soft feces P1											*3*	
Tremors F1											*17**	*23**
Convulsions F1												*2**
Alopecia F1									*14**	*10**		
Bw changes												
P1 *Day 1-83*											*↓27%*	*↓34%*
P1 lactation Day 1-28 (g)		*-4*		*-8.1*		*-12*		*-7.3*		*-9.7*		*-11.6*
F1 *Day 1-85 (m) Day 1-106 (f)*									*(↓7%)*	*(↓4%)*	-	
Body weight: P1												
Day 8-83											*↓11%*	*↓7-12%*
Gestation: Day 0-20												*↓7-12%*
Lactation: Day 1-28										*(↓5-7%)*		*↓14-25%*
Body weight: P1												
Day 1-50									*↓8-12%*		*↓60%*	*↓48%*
Day 57-106									*↓8%*	*↓7%*	-	-
Terminal									*↓8%*	*↓6%*	-	-
Gestation day 15										*↓6%*	-	-
Lactation day 1-21										*↓6-10%*	-	-
Food consumption												
P1												
Premating											*↓13%*	*↓19%*
Gestation day 0-20												*↓11%*
Lactation d1-21										*↓16%*		*↓40%*
F1												
Premating day 1-8									*↓12%*	*↓10%*	*↓80%*	*↓77%*
Day 8-22									*↓13%*		*↓56%*	*↓32%*
Day 1-85									*↓6%*	*↓7%*		-
Lactation day 1-14										*↓8%*		-

Table 5.6.1-5: Two-generation rat study with zeta-cypermethrin: parental data

Endpoint/dose	*0 ppm*		*7.5 ppm*		*25 ppm*		*100 ppm*		*375 ppm*		*750 ppm*	
	M	*F*	*M*	*F*	*M*	*F*	*M*	*F*	*M*	*F*	*M*	*F*
Fertility index												
N° pregnant/Mated												
P1		*25/29*		*28/30*		*27/30*		*29/30*		*27/30*		*27/30*
F1		*24/30*		*20/30*		*22/29*		*23/29*		*24/29*		*-*
N° dams with pups dying prior d28												
P1		*0*		*1*		*0*		*0*		*0*		*12*
F1		*0*		*0*		*0*		*0*		*0*		*-*
Organ weight												
Brain F1											*↑8%*	*↑8%*

↑↓ Statistically significantly different from control; () not statistically significant*

Table 5.6.1-6: Two-generation rat study with zeta-cypermethrin: litter data

Effect/dose	*0 ppm*	*7.5 ppm*	*25 ppm*	*100 ppm*	*375 ppm*	*750 ppm*
N° Live/Stillborn pups						
F1	*285/4*	*348/3*	*319/3*	*350/3*	*351/0*	*331/1*
F2	*297/5*	*215/7*	*243/6*	*281/1*	*284/3*	*-*
Pup mortality: F1/F2						
Day 1	*0/3*	*2/0*	*0/1*	*0/3*	*0/3*	*1/-*
Day 2-4	*2/4*	*7*/2*	*2/1*	*5/2*	*0/4*	*9*/-*
Day 5-7	*0/1*		*1/0*	*0/2*	*0/1*	*26*/-*
Day 8-14						*44*/-*
Day 15-21						*18*/-*
Day 22-28						*52*/-*
Mean n° surviving pups/litter F/F2						
Day 1	*11.9/12.4*	*12.4/11.9*	*11.8/11*	*12.1/12.2*	*11.7/12.3*	*12.2/-*
Day 4 pre/ post culling	*11.8/7.8 12.1/7.8*	*12.1/7.7 11.8/7.8*	*11.7/7.7 11/7.7*	*11.9/7.8 12/7.8*	*11.7/7.7 12/7.6*	*11.9/7.9*
Day 7	*7.8/7.8*	*7.7/7.8*	*7.6/7.7*	*7.8/7.7*	*7.7/7.6*	*6.9*/-*
Day 14	*7.8/7.8*	*7.7/7.8*	*7.6/7.7*	*7.8/7.7*	*7.7/7.6*	*5.3*/-*
Day 21	*7.8/7.8*	*7.7/7.8*	*7.6/7.7*	*7.8/7.7*	*7.7/7.6*	*4.5*/-*
Day 28	*7.8*	*7.7*	*7.6*	*7.8*	*7.7*	*2.4*/-*
Mean pup weight/litter (g) F1/F2						
Day 4 pre/ post culling						*↓19%/↓19%*
Day 7					*↓10%/-*	*↓32%/-*
Day 14					*↓12%/↓6%*	*↓46%/-*
Day 21					*↓11%/↓10%*	*↓57%/-*
Necropsy: F1						
Stomach w/o milk	*0*	*2*	*0*	*0*	*0*	*24*

Conclusion

NOAEL systemic toxicity parents = 100 ppm (5.9 mg/kg bw/d) based on decreased body weights, body weight gains, clinical signs of toxicity seen at 375 ppm (22 mg/kg bw/day).
The NOAEL reproduction = 750 ppm (43 mg/kg bw/d) for the P1 generation and 350 ppm (22 mg/kg bw/d) for the F1 generation because mortality prevented evaluation of the reproductive performance of the 750 ppm group second generation.
NOAEL developmental toxicity = 100 ppm (5.9 mg/kg bw/d) based on reduced pup body weights in both generations during the lactation periods. Pups mortality was seen at top dose.

CA 5.6.2 Developmental toxicity studies

Alpha-cypermethrin, rat, oral administration by gavage, 0, 3, 9, 15 and 18 mg/kg bw [redacted] 1994a, AL-432-001 and [redacted] 1994a, AL-432-002)

Dose Range Finding Study (AL-432-001)

Guidelines:	Dose range finder for main study. Main study in compliance with the test method B of Directive 87/302/EEC or OECD guideline 414 (1981).
Deviations:	None
GLP:	Yes (no attest of competent authority)
Acceptance:	The study was considered acceptable in the EU registration process 1999.

This study consisted of two parts. The first part was conducted in non-mated females to establish the maximum tolerated dose (MTD) of the test article; the second part was conducted in mated females to investigate the effect of the test article on the pregnant rat and offspring in utero in order to select dose levels for a subsequent developmental toxicity study.
MTD Phase: Groups of three non-mated, sexually mature, female Sprague-Dawley rats were dosed, once daily, orally by gavage, with solutions of alpha-cypermethrin in corn oil vehicle for five consecutive days. Dose levels of 15, 18, 20, 30 and 50 mg/kg were employed.
Mated Phase: Groups of five timed-mated Sprague-Dawley rats were dosed, once daily, by oral gavage, from days 6 through 15 of pregnancy at dose levels of 3, 9, 15, and 18 mg/kg bw/day alpha-cypermethrin in corn oil. A group dosed with the vehicle only served as controls. On day 20 of pregnancy a necropsy was performed. The fetuses were weighed, sexed and subjected to external examination.
Findings:
MTD Phase: Treatment at levels of 20, 30 and 50 mg/kg bw/day was associated with severe changes in clinical condition, including convulsions, ataxia, hypersensitivity to touch and sound, spasms, piloerection, limb splay and hunched posture that were typical pyrethroid-associated effects. The severity of the effects was dose related. Dosing was discontinued prematurely, after the second treatment for these groups, and all animals in the 50 and 30 mg/kg bw/day groups and one animal in the 20 mg/kg bw/day group were sacrificed. In the 18 mg/kg bw/day group, there were similar clinical signs of toxicity as described above, as well as weight loss.
On day 5, prior to dosing, one female displaying severe clinical signs of toxicity was sacrificed. The others were dosed as scheduled. In the 15 mg/kg bw/day group, all three females were dosed for five consecutive days; animals in this group exhibited slight weight loss, and two had observations of piloerection and/or hunched posture. None of the surviving females showed treatment-associated abnormalities at necropsy. The MTD was determined to be 18 mg/kg bw/day.

Mated phase: Clinical signs of treatment at 18 mg/kg bw/day included hind limb splay and unsteady gait. One female treated at 15 mg/kg day also exhibited similar signs. One female in the 9 mg/kg bw/day group in poor clinical condition, was sacrificed on day 16; the condition of this animal was determined not to be treatment associated as similar signs were not seen at higher dose levels. No treatment-related clinical changes were noted in other animals in the 9 mg/kg bw/day group or at lower dose levels. Mean body weight gain in the 18 mg/kg bw/day group was significantly reduced in comparison to the control group over the 6-15 day dosing period. Body weight gains in the groups treated at 9 and 15 mg/kg bw/day were lower during the first half of the dosing period, but the differences were not statistically significant and a compensatory increase was observed in the 15-20 day interval. Body weight gain in the group treated at 3 mg/kg bw/day was similar to that of the control group throughout the dosing period. Similar to body weight gains, mean food consumption values in the 18 mg/kg bw/day group were lower than that of the control group over the entire dosing period, and were significantly lower during days 6-12. Food consumption values in the 15 mg/kg bw/day group were slightly, but not significantly lower between days 6 and 9 only. At the lower dose levels, food consumption values were similar to that of the control group throughout the dosing period. There were no treatment-related abnormalities at maternal necropsy. There were no significant intergroup differences in the mean numbers of corpora lutea, implantations or live fetuses. Pre- and post-implantation losses were increased, as compared to the controls, for all treatment groups and post-implantation losses were statistically significantly increased for the 9 mg/kg bw/day group. These effects were believed, however, to be related to unusually low pre- and post-implantation losses in the control animals and were not considered to be toxicologically significant. There were no effects of treatment on fetal weights or sex ratios.

Conclusion:
Dose levels established for the main study were between 3 and 18 mg/kg bw/day. The highest dose level was selected as a dose expected to elicit minimal maternal toxicity, and the lowest level was selected as a probable "no effect" level with respect to maternal toxicity.

Main study [redacted] 1994a)

Guidelines:	Study in compliance with the test method B of Directive 87/302/EEC or OECD guideline 414 (1981).
Deviations:	None
GLP:	Yes (no attest of competent authority)
Acceptance:	The study was considered acceptable in the EU registration process 1999.

The effects of alpha-cypermethrin on the pregnancy and embryonic or foetal development of the Sprague-Dawley rat were investigated at 0 (control), 3, 9, 18/15 and 15 mg/kg bw/day administered in corn oil by gavage from day 6 to day 15 post mating. The dose level was 18 mg/kg bw/day from day 6 to 9 of pregnancy, and 15 mg/kg bw/day from day 10 to 15 of pregnancy. The study was performed according to US EPA 83-3. The conduct of the study is consistent in all important aspects to OECD 414 (2001) and EC method B.31 (2004/73/EC), with the exception that the test substance was administered solely during the period of organogenesis.

Findings:
Administration of alpha-cypermethrin via gavage at 15 or 18 mg/kg bw/day to pregnant rats during foetal organogenesis elicited maternal toxicity characterised by changes in clinical conditions and reduction in food consumption and body weight gain during the dosing period. Further, a slight reduction in foetal weights was observed at these dose levels most probably being a consequence of the observed maternal toxicity. However, there was no other evidence of embryo- or fetotoxicity or teratogenicity at any tested dose level.

Conclusion:
Based on the described maternal toxicity and embryonal toxicity effects at 15 or 18 mg/kg bw/day the NOAEL for maternal toxicity and embryotoxic/teratogenic effects was considered to be 9 mg/kg bw/day.

Disregarded Developmental toxicity study in rats [redacted] unknown date) (Dossier Gharda)

Guidelines: Study not in compliance with the method B of Directive 87/302/EEC.
Deviations: The route of administration is not defined. Experimental results are not fully described. Sex of foetuses was not determined. Tables describing abnormalities are confusing. No statistical treatment of results.
GLP: No data
Acceptance: The study was not accepted in the EU registration process 1999.

Twenty pregnant Young Charles Foster rats were exposed to alpha-cypermethrin at doses of 0, 5, 10 and 20 mg/kg bw/day on day 6 through day 18 of gestation. Controls were dosed with vehicle only (DMSO). Females were sacrificed on day 20 of gestation. The conduct of the study was not consistent to OECD 414 (2001) and EC method B.31 (2004/73/EC) because the route of administration is not defined, experimental results are not fully described, sex of foetuses was not determined, the tables describing abnormalities are confusing and no statistical evaluation of the results were performed. Therefore, the study is not accepted.
Findings:
The number of litters, body weight of pregnant females, number implants per dam and mean foetal body weight were normal except the number of resorptions increasing with the dose as well as the number of live foetuses which appeared to decrease with the dose. No teratogenic effects were observed.

Conclusion:
This study exhibits severe deficits in the experimental protocol. Therefore, it was not possible to evaluate clearly if alpha-cypermethrin induced abnormalities in foetuses and the study was not accepted for evaluation.

Alpha-cypermethrin, rabbit, oral administration by gavage, 0, 3, 15, 30 mg/kg bw/day ([redacted] 1994b, AL-432-003 and [redacted] 1994b, AL-432-004)

Dose Range Finding Study ([redacted] 1994b)
Guidelines: Dose range finder for main study. Main study was in compliance with the test method B of Directive 87/302/EEC.
Deviations: None
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

This preliminary study in New Zealand white rabbits consisted of two parts. The first part was a study in non-mated females to establish the maximum tolerated dose (MTD). The second part was a study in mated females to investigate the effect of the test article on the pregnant rabbit and offspring in utero in order to select dose levels for a subsequent developmental toxicity study.
MTD Phase: A group of five non-mated, sexually mature, female New Zealand White (NZW) rabbits was dosed, once daily, by oral gavage with solutions of alpha-cypermethrin in corn oil for twenty five days within a thirty day period. The dose levels increased as follows; 10 mg/kg bw/day for days 1 to 5, 15 mg/kg bw/day for days 6 to 10, 25 mg/kg bw/day for days 12 to 16, 35 mg/kg bw/day for days 19 to 23, and 40 mg/kg bw/day for days 26 to 30. On intervening days, the animals were not dosed. Once the MTD was established, a second group of five non-mated females was dosed, once daily, orally by gavage at 35 mg/kg bw/day for seven consecutive days.
Mated Phase: Groups of 5 timed-mated NZW rabbits were dosed, once daily, by oral gavage from days 7 to 19 of pregnancy, inclusive, at dose levels of 5, 15, 25, and 30 mg/kg bw/day alpha-cypermethrin in corn oil. A group dosed with the vehicle only served as controls. On day 28 of pregnancy, animals were sacrificed by caesarean section. The fetuses were weighed, sexed and subjected to external examination.

Findings:
Increasing Dose MTD Phase: During the first two days of dosing at 10 mg/kg bw/day, there were slight reductions in body weight and food consumption values. The animals recovered, however, and there were no further signs of treatment at 10, 15, or 25 mg/kg bw/day. At 35 mg/kg bw/day, one animal appeared ataxic and languid on the first day of dosing, and there was a trend toward a slight decrease in body weight, food consumption and feces production during the five days of treatment at this dosage. These animals recovered in the two off-dose days. At 40 mg/kg bw/day, the animals showed slight fluctuations in body weight and food consumption, similar to the 35 mg/kg bw/day dose group. The MTD was considered to have been reached at 35 mg/kg bw/day. There were no treatment-related abnormalities at necropsy.

Fixed dose MTD Phase: The protocol specified that the animals would be dosed for eight days, however, dosing was discontinued after seven days because of the significant toxicity associated with the treatment. There were no unscheduled deaths, but two animals were sacrificed prematurely (one on day 6 and one on day 7) due to negligible food consumption and marked body weight loss. All animals showed body weight loss and reduced fecal output after one or two doses, which persisted throughout dosing; however, on day 9, two days after cessation of dosing, two of the three surviving females showed a slight body weight gain. There was a marked decrease in food consumption during dosing, as well, with appetite recovery observed for all animals after cessation of dosing. There was a lack of food found in the stomachs of those females sacrificed prematurely, however there were no abnormalities at necropsy that were considered to be related to treatment.
Mated phase: Two females (one in each group at 15 and 25 mg/kg bw/day) were sacrificed prematurely because of low food consumption values and body weight loss. Clinical signs were limited to a reduction in the amount of feces for control and treated animals. Animals in the control, 25 and 30 mg/kg bw/day groups exhibited a mean body weight loss at the onset of dosing. This may have been the result of the low tolerance of the pregnant rabbit to administration of corn oil (vehicle). The weight gains of the animals dosed at 5 and 15 mg/kg bw/day were unaffected by treatment. There was a trend toward further reductions in mean body weights during the latter third of the treatment period for animals dosed with 25 and 30 mg/kg bw/day only. After the conclusion of the dosing period, all affected animals showed a compensatory increase in mean body weight. Food consumption was similarly reduced during the dosing period in animals which exhibited weight loss; a slight reduction in food consumption was also noted during the treatment period for animals dosed at 5 and 15 mg/kg bw/day. Mean food consumption for all groups returned to expected levels at the cessation of treatment. There were no treatment-related abnormalities at maternal necropsy and no apparent adverse effects of treatment on mean numbers of corpora lutea, implantations, live fetuses, or pre- or post-implantation losses. There was no effect of treatment on fetal weight, sex ratio, or the incidence of fetuses with external abnormalities.

Conclusion:
Dose levels established for the main study were 3, 15 and 30 mg/kg bw/day. The highest dose level was selected as a dose expected to elicit minimal maternal toxicity; higher doses were not recommended in light of the toxicity observed at 35 mg/kg bw/day in the preliminary MTD study. The lowest level (3 mg/kg bw/day) was selected as a probable "no effect" level with respect to maternal toxicity.

Main study ([redacted] 1994b)

Guidelines: Study in compliance with the test method B of Directive 87/302/EEC.
Deviations: None
GLP: Yes (no attest of competent authority)
Acceptance: The study was considered acceptable in the EU registration process 1999.

The effects of alpha-cypermethrin on the pregnancy and embryonic or foetal development in the New Zealand White rabbit was investigated at 0 (control), 3, 15 and 30 mg/kg bw/day administered in corn oil from day 7 to day 19 post mating. The study was conducted according to US EPA 83-3. The conduct of the study was consistent in all important aspects to OECD 414 (2001) and EC method B.31 (2004/73/EC), with the exception that less than 20 female animals with implantation sites were investigated, and that the
test substance was administered solely during the period of organogenesis.

Findings:
There was no maternal or developmental toxicity associated with oral administration at 3 or 15 mg/kg bw/day. Administration via gavage of 30 mg/kg bw/d of alpha-cypermethrin to pregnant rabbits during foetal organogenesis caused reduced maternal food consumption and body weights during the latter stages of the dosing period. There was no evidence of embryo or foetal lethality, growth retardation or teratogenicity at any tested dose level.

Conclusion:
Based on the maternal reduced body weight and food consumption the NOAEL for maternal toxic effects is considered to be 15 mg/kg bw/day. Since no embryotoxic and/or teratogenic effects were observed the NOAEL for embryotoxic/teratogenic effects corresponds to 30 mg/kg bw/day which is the highest dose tested.

CA 5.7 Neurotoxicity Studies

EU Dossier update (November 2015) containing an update of data which were not present at the dossier submission in February 2015: The DNT-study under point 5.7.1/3 was not finalized at dossier submission and data on stability, homogeneity and concentration control, pathological examinations of dams and pups and examination of pups brain (weight, length and width) were missing. Furthermore a separate analytical report (Doc ID 2015/1175543) is included which analysed the alphacypermethrin content in the pups, separated into amount in brain and carcass. *The data are combined in chapter CA 5.7.1/3 of this dossier and highlighted in yellow. The methodical details are included in chapter CA 4-Analytical methods.*

Studies evaluated in the draft monograph of rapporteur member state Belgium of September, 1999: Two acute neurotoxicity studies in hens and rats are available for Alpha-Cypermethrin. These studies, in addition with one mechanistic study, have been evaluated by European authorities and Belgium as Rapporteur member state (European Commission Peer Review Program) and were considered to be acceptable. For the convenience of the reviewer, these are summarized below as extracted from the monograph and brief summaries are provided under the respective chapters.

Table 5.7-1: Summary of already peer-reviewed alpha-cypermethrin neurotoxicity studies as available in the DAR (1999)

Study Dose levels	Substance information	NOAEL mg/kg bw/d	LOAEL mg/kg bw/d	Adverse effects at LOAEL	Reference
Neurotoxicity studies					
Acute neurotoxicity study Crl:CD:BR rat, 0, 4, 20 and 40 mg/kg bw/d (gavage, corn oil)	Alpha-cypermethrin (batch No. 021.56)	4	20	neurotoxicity; increased neurologic reactivity	[redacted] 1993, CA 5/13 AL-451-004
Acute neurotoxicity study Hen, 0, 70, 140, 700 mg/kg bw/day (gavage, DMSO)	Alpha-cypermethrin (99%)	-	70	brain inflammation; spinal cord altered cellular pattern; sciatic nerve axonal fibres distension	[redacted] 1989, Gharda[1]
Supplementary studies already reviewed in the monograph of alpha-cypermethrin and at that time discussed under CA 5.8.2					
Subacute neurotoxicity study, rat 10, 20, 40 mg/kg bw/day (gavage, DMSO)	Alpha-cypermethrin (batch No. 7; 96.6%)	10	20	increase of beta-galactosidase activity in sciatic posterior tibial nerve, trigeminal ganglia and nerve	[redacted] 1983, AL-451-002

[1] These studies are not BASF proprietary, but owned by Gharda, Chemical Ltd. Bombay, a further notifier for Annex I listing of alpha-cypermethrin in 1999

The summary of these neurotoxicity studies is still valid:
In hens, alpha-cypermethrin at doses from 70 to 700 mg/kg bw produced inflammatory reaction in brain, disruption of cellular pattern in spinal cord and distension and disruption of axonal fibres in sciatic nerve. Alpha-cypemiethm is toxic for the CNS and peripheral motor nerves.

The acute neurotoxicity of alpha-cypermethrin was studied in rats from 4 to 40 mg/kg bw. Clinical signs of neurotoxicity were apparent from 20 mg/kg bw onwards. Male rats were more susceptible than female rats and presented abnormal splayed gait, thrashing, prostration, vocalization, piloerection, hunched posture, unkempt appearance, soiled/stained body areas and diarrhea. Increased neurologic reactivity was seen in most males. Animals recovered by day 7. Motor activity was not affected. Very slight to slight sporadic fiber degeneration was shown in the proximal part of the sciatic nerve. In this study the NOAEL is 4 mg/kg bw.

Biochemical evidence of damage to the sciatic posterior tibial nerve, trigeminal ganglia and nerve were reported by measuring increases in beta-galactosidase enzyme activity after 4 weeks treatment with alpha-cypermethrin at doses of 20 mg/kg bw/day. The NOAEL in this study is 10 mg/kg bw/d.
Neurodevelopmental toxicity of pyrethroids was comprehensively discussed in the ECCO Peer review meeting - Full Report on alpha-cypermethrin ([see KCA 5.7/1 AL-901-031], page 164-172 of 222, presented as copy in CA 5.7.1) based on Literature data. The discussed literature data are for the convenience of the reviewer summarized below in tabular form (see Table 5.7-2), a short summary of the some points is presented below:

Table 5.7-2: Summary of already peer-reviewed literature data on developmental neurotoxicity as available in the ECCO Peer review meeting - Full Report on alpha-cypermethrin (DocID AL-901-031, page 164-172 of 222)

Authors	Exp.conditions	Endpoints	Results	ECCO Comments
Cantalamessa et al, 1998	Cyp treatment from PND 10-16, Wistar, rats 1 mg/kg bw/d	Dopamine DI and D2 like receptor assay in kidney	↑ density of receptors D1 in 30, 60, 90 day old rats	Dopamine exerts renal actions including diuresis. No effects seen in the 3 generation rat study at 25 mg/kg bw/d
Husain et al, 1992	Cyp in formulation, Rats 15 mg/kg bw/d GD 5-21	Behavioral teratogenicity	Delayed fur Develop., incisor eruption, eye and ear opening, reduced surface righting reflex	Range of days of occurrence and not a mean. Effects obtained from 10 pups/group (1 pup from each of 10 litters/group). Parameters should be determined for litters
Malaviya et al., 1993	Cyp, 15 mg/kg bw/d from GD 5-21. Nursing mothers, day 1 PN up to 3 week	Brain enzymes : MAO, AChE, Na^+,K^+ ATPase	Exposure *in utero:* no effect Lactation exp: ↓ AChE, Na^+,K^+ ATPase and ↑ level dopamine and muscarinic receptor of striatal membrane	Fenvalerat also reported in this study has different effects although it is a type II pyrethroid. No neurobehavioral effects in 3-generation rat study at neurotoxic maternal doses.
Kavlock et al.	Decamethrin (type II) 2.5 or 5 mg/kg bw/d GD 7 to LD15	Neurobehavioral study in litters	Neurotoxicity in dams at 5 mg/kg No effects in neurobehavioral parameters	additional doubt on study by Husain
Eriksson and Fredriksson	NMRI mice pups with bioallethrin 0.7 mg/kg bw/d (type I) or deltamethrin 0.7 mg/kg bw/d (type II) from PND 10-16	Density muscarinic receptors in brain; motor activity in adult	↓Density muscarinic receptors in brain; ↑ motor activity	Results not reproductible for same compounds at same laboratory. Inconsistent results and opposite to the other study

Authors	Exp.conditions	Endpoints	Results	ECCO Comments
Muhammad and Ray	NMRI mice pups with bioallethrin 0.7 and 3.5 mg/kg bw/d (type I) or deltamethrin 0.7 and 3.5 mg/kg bw/d (type II) from PND10-16	Density muscarinic receptors in brain; motor activity in adult	Muscarinic receptors density: inconsistent; Minimal changes in motor activity in some experiments and not in others. No dose related effects	Changes in muscarinic receptor density and motor activity changes found by Eriksson and Fredriksson not confirmed
Bayer AG	Inhalation with several yrethroids in NMRI mice PND10-16	Spontaneous motor activity; muscarinic receptor density at 17d and 4 month	Slight ↑ in muscarinic receptor density on PND17 not correlated with changes in spontaneous motor activity. No effect in adults	Neurobiochemical changes are likely not biologically significant
IVA, 1994		Neurotoxicity of pyrethroids to developing animals: retrospective analysis	Alpha-cyp or cyp are not selectively toxic to developing offsprings : rearing, nursing, behavior of dams, social and sukling behavior of neonates not affected except at toxic parental doses	Majority of NOEL offspring ≥ NOEL parental tox. No functional or morphological changes in nervous system
Sheets, 2000	Rats, cypermethrin 9, 19, 38 mg/kg bw, 21 day old rats, comparison with adult rats	Auditory startle response	38 mg/kg bw: ↓ startle response of 50% comparable to adult 50% reduction	
Conclusion : No additional studies are necessary.				

Sheets (Neurotoxicology, 21(1-2): 57-64, 2000) showed that neonates are more sensitive than adults to acute lethal dose of certain pyrethroids but not to much lower levels that are relevant for dietary risk assessment. This higher sensitivity is not based on differences at the target sites in the brain of the neonates. Studies with Deltamethrin showed that at the respective LD50 dose, the same brain level was observed in adults and weaning rats, which received a seven time lower dose. This indicates that the main age related difference only occur at dose levels that exceed the neonate`s capacity for detoxification.

Furthermore the vulnerable periods during ontogenesis of the CNS were discussed. They can be divided into 2 major courses of events:

1. Early brain development: brain acquires its general adult shape
2. Brain growth spurt: rapid fundamental changes including maturation of axonal and dendritic outgrowth;

In humans, the brain growth spurt occurs maximally at the 3rd trimester of pregnancy up to 2 years of life. In mouse and rat, this period is neonatal spanning the first 3-4 weeks of life. Therefore early postnatal exposure in the rodent (postnatal day (PND) 10-17) encompasses a time span equivalent to peri-neonatal exposure in the human.

Additional support which negates the likelihood of developmental neurotoxic activity is coming from the developmental toxicity studies with alpha-cypermethrin and the developmental and the 3–generation study with cypermethrin. Results indicate that neither alpha-cypermethrin nor cypermethrin is a selective developmental toxicant or a teratogenic agent in either the rat or rabbit. Because neither alpha-cypermethrin nor cypermethrin is a selectively toxicant to the developing fetus or offspring, there is no increased sensitivity of developing offspring following prenatal or postnatal exposure to alpha-cypermethrin or cypermethrin. In the absence of any selectivity or increased sensitivity to the developing fetus or offspring, a developmental neurotoxicity study with alpha-cypermethrin is not warranted.
This was also confirmed by a pyrethroid ad hoc working group sponsored by Industrieverband Agra e.V. which developed a white paper and discussed the potential neurotoxicity of pyrethroids to developing animals. Taken together, the retrospective analysis presented in this white paper provides strong evidence that treatment of lactating and/or pregnant females with pyrethroids is not associated with developmental neurotoxicity.

In the end it was concluded in the Review Report (SANCO/4335/2000 final from Feb. 2004 under the Header “List of studies to be generated:”
“*No further studies were identified at this stage considered necessary in relation to the inclusion of alpha-cypermethrin in Annex I under the current inclusion conditions.*
… As for other pyrethroids, confirmatory data to further address concerns related to potential developmental neurotoxicity should be generated, when internationally agreed testing protocols are available.”
Based on the available data, the following endpoint was determined during the last Annex I listing of Alpha-Cypermethrin concerning neurotoxicity/delayed neurotoxicity:

Neurotoxicity / Delayed neurotoxicity

Effects observed	Alpha-cypermethrin is toxic for CNS and peripheral motor nerves; neurobehavioral changes are reversible within 3 days following single dose
Acute rat study	NOAEL = 4 mg/kg bw (in corn oil)
4 week oral rat study	NOAEL = 10 mg/kg bw/day (DMSO)

Studies submitted in this AIR 3 dossier (Part I: peer-reviewed studies from other DARs):
To address the request of a developmental neurotoxicity study expressed in the Review report of alpha-cypermethrin (2004) when internationally agreed testing protocols are available, it is considered appropriate to bridge to the dietary developmental neurotoxicity study with zeta-cypermethrin. This study has been summarized and evaluated in the DAR of zeta-cypermethrin (2008) by the rapporteur Member state Belgium. This study as well as a dietary placental and lactational transfer study in rats (presented in CA 5.8.2) are not data protected anymore and fulfill the criteria to wave a developmental neurotoxicity study with direct pup dosing because zeta-cypermethrin has been shown to be transferred in utero as well as via milk. Thereby a continuous exposure of the developing offspring is guaranteed and direct pup dosing should be not needed. Based on the previously presented general bridging approach (see CA 05.00) this study is considered to be valid for alpha-cypermethrin, too.

Within the EFSA conclusion of zeta-cypermethrin (EFSA Scientific Report (2008) 196, p. 15) it is stated: *"A developmental neurotoxicity study in rats was summarized in the DAR showing a maternal and developmental toxicity NOAEL of 9 mg/kg bw/day based on reduced bodyweight gain. Zeta-cypermethrin did not show any developmental neurotoxic potential.*

Placental and lactational transfer was investigated after dietary exposure of dams. Zeta-cypermethrin was identified in milk following dietary administration indicating that a transfer occurred from food to milk and that exposure of pups occurred via milk."
Based on this study it is concluded that the related alpha-cypermethrin is not suspected to show any developmental neurotoxic potential.

This conclusion is furthermore supported by evaluations published by different authorities that investigated the developmental neurotoxic potential of pyrethroids and that all came to the conclusion that possible DNT effects induced by pyrethroids are covered by the acute neurotoxicity and medium term studies, since DNT effects from acceptable OECD TG 426 performed studies are taking place at higher LOAELs than acute neurotoxicological effects. Therefore additional DNT study according to OECD TG 426, if such a study is not present, is not necessary.

- EPA "Pyrethroids: Evaluation of Data from Developmental Neurotoxicity Studies and consideration of Comparative Sensitivity" [see KCA 5.7/9 2010/1232203].
- Biocides Technical Meeting (14-18 June 2010) Annex point 6. Survey of DNT studies for pyrethroids. (Conclusion is reflected in a Manual of Technical Agreements and should be available to authorities on CIRCA)
- EFSA Scientific Opinion: Potential developmental neurotoxicity of deltamethrin; EFSA Journal (2009) 921, 1-34; (online available under the following link: http://www.efsa.europa.eu/de/efsajournal/doc/921.pdf)

However, in 2013 the Rapporteur UK published the DAR of Beta-cypermethrin with a developmental neurotoxicity study (Vol. 3, B.6.7 d) split up into 2 administration szenarios:
Szenario I consisted of an oral gavage study of dams from GD 6 to PND 21 and pup exposure via milk (a transfer of beta-cypermethrin into milk was shown in the corresponding 2-generation study). The dams within the developmental neurotoxicity study showed clinical signs and body weight reduction at 12 mg/kg bw whereas pups showed increased motor activity only at the maternal toxic dose of 30 mg/kg bw and only on PND13. The study clearly indicated that although pups are sensitive to the neurotoxic effects of beta-cypermethrin, direct lactational transfer through milk is not at sufficient quantities to induce adverse effects in pups.
Szenario II consisted of an oral gavage study of dams from GD 6 to PND9 and direct pup dosing from GD10 to GD16 at 0.5 mg/kg bw. The direct dosing of pre-weanling pups produced sever clinical signs of toxicity, body weight reduction, clonic convulsions, altered grooming and deaths already at a dose of 0.5 mg/kg bw, but only at the time of direct pup treatment.

Based on the results the rapporteur UK concluded: *"Direct dosing of pre-weanling pups with beta-cypermethrin is a highly unrealistic exposure technique and* ***the findings in this section of the study are not considered relevant to a human risk assessment for infants*** *who will be either breast or bottle fed."*

However, this opinion was changed within the Peer review meetings on beta-cypermethrin where it was argued that relevance for human risk assessment is given because

1. today babies are not only breastfed but receive additional exposure via bottle milk, baby food or air drift as residents.
2. Neurological development of infants and toddlers partly corresponds to early post-natal development in rodents

Taking a look into the Meeting minutes (3717prr public.pdf available online via: http://registerofquestions.efsa.europa.eu/roqFrontend/outputLoader?output=ON-3717) it appears that mainly 2 member states were arguing for the use of these endpoints.

Finally, the EFSA conclusion on beta-cypermethrin (EFSA Journal 2014; 12 (6):3717) reported: *"The majority of the experts agreed that the findings after gavage of pups were relevant for human risk assessment. It is noted that the toxicity of the other cypermethrins might need further evaluation on the basis of these new data of developmental neurotoxicity with beta-cypermethrin."*

In the following Table 5.7-3 the results of developmental neurotoxicity studies with zeta- and beta-cypermethrin are summarized as presented in the respective DARs.

Table 5.7-3: Developmental neurotoxicity endpoints for beta- and zeta-cypermethrin

Study	Test substance information	Endpoints		Reference
		NOAEL (mg/kg bw/day)	**LOAEL (mg/kg bw/day)**	
Developmental neurotoxicity, rat				
Rat developmental neurotoxicity Dams/pups (study I): 3, 12, 30 mg/kg bw/day Pre-weanling pups (study II): 0.5 mg/kg bw/day oral, gavage vehicle: 1% methylcellulose/ 0.1% polysorbate 80	Beta-cypermethrin: batch No. PL9511038; 97.6%	Dams: 3 mg/kg bw/day	12 mg/kg bw/day clinical signs, body weight	Adapted from DAR beta-cypermethrin (2013) and supplemented by EFSA conclusion (2014) data
		Pups: 12 mg/kg bw/day	30 mg/kg bw/day increasing motor activity count	
		Pre-weanling pups direct dosing: <0.5 mg/kg bw/day	0.5 mg/kg bw/day Clinical signs, decreased body weight, tremors, clonic convulsions, altered grooming and death	
Rat developmental neurotoxicity 50, 125, 300 ppm (3.6, 9, 21.1 mg/kg bw/day) (dietary administration)	Zeta-cypermethrin: batch No. PL03-0427; 81.8%	Dams: maternal: 9 mg/kg bw/day neurotoxic: 21.1 mg/kg bw/day	Dams: maternal: 21.1 mg/kg bw/day body weight, body weight changes and food consumption neurotoxic: > 21.1 mg/kg bw/day	Adapted from DAR zeta-cypermethrin (2008) and supplemented by EFSA conclusion (2008) data
		Pups: developmental: 9 mg/kg bw/day neurotoxic: 21.1 mg/kg bw/day	Pups: 21.1 mg/kg bw/day body weight, body weight changes neurotoxic: > 21.1 mg/kg bw/day	

While the exposure via the dam showed for zeta- and for beta-cypermethrin comparable results with no increased sensitivity of developing offspring following prenatal or postnatal exposure, the direct gavage of pups showed drastically higher susceptibility of the pups.

BASF is of the opinion that this higher sensitivity is predominantly due to a substantial double exposure and the artificial test system which is a highly unrealistic scenario for human exposure as it is a bolus administration. There are many reasons why such an exposure leads to artificial findings at low doses that are not relevant under normal conditions and we fully share the scientific opinion of UK that this study is designed for hazard assessment in case of non exposure of pups within the lactation phase and should not be relevant for human risk assessment.

Studies submitted in this AIR 3 dossier (Part II: not peer-reviewed studies)
However, in bilateral discussions with the Rapporteur Belgium it was agreed to perform a targeted DNT study with direct dosing of alpha-cypermethrin to pups focusing on the effects seen in the developmental neurotoxicity studies performed with beta-cypermethrin in order to address the concerns of the Rapporteur. The findings of this study are comparable to those described in the beta-cypermethrin study and demonstrate the drastic increase of toxicity by this artificial form of administration which is not considered relevant for the human risk assessment.

In addition a 90-day study performed in 1994 that was not submitted in the former registration process under the aegis of Cyanamid is now presented for completeness in detail.

Table 5.7-4: Not peer-reviewed neurotoxicity studies with alpha-cypermethrin

Study	Dosages (mg/kg bw/ day) Test substance information	NOAEL (mg/kg bw/day)	Effect at LOAEL (mg/kg bw/day)	Reference
90-day feeding study Crl:CD(SD) BR rat 0, 50, 250, 500 ppm	M: 0, 3.7, 17.9 and 36.1 F: 0, 4.2, 21 and 42	systemic: 3.7 / 4.2 (50 ppm) neurotoxic: 36.1 / 42 (500 ppm)	17/ 21 (250 ppm) Transient reduced food intake, altered hematology parameters neurotoxic: > 36.1 / 42 (500 ppm)	1994 CA 5.7.1/4 AL-425-007 **New data**
Rat developmental neurotoxicity with direct pup dosing PND 10-21 Wistar rats; oral, gavage; vehicle:1% CMC	cohorte I: 0.5, 2, 5 mg/kg bw/day dams and pups; cohorte II-IV: dams: 0.5, 2 mg/kg bw/day pups: 0.25 mg/kg bw each alpha-cypermethrin; 99.2%; PMAM000622	Dams: 5 mg/kg bw	Dams: > 5mg/kg bw No clinical effects	CA 5.7.1/3 ~~2014/1275121~~ 2015/1001621 & Results of the Analytical Report 2015/1175543 (CA 4.1.2/3) **New study**
		Pups (lactational exposure): 5 mg/kg bw	Pups (lactational exposure): >5mg/kg bw No clinical effects (PND 1-9)	
		Pups (direct dosing) Approx.. 0.25 mg/kg bw; (in case dams are dosed < 0.5 mg/kg bw) Calculated: 0.22 mg/kg bw	Pups (direct dosing): 0.25 mg/kg bw; (dose of dams 0.5 mg/kg bw) Calculated: 0.262 mg/kg bw Effect: neurotoxic signs (slight tremors & twitching)	

The **90 day feeding study** in SD rats showed systemic toxicity in form of reduced food intake and altered hematological parameters at the mid dose but no neurotoxicity was observed up to the highest dose of 36 mg/kg bw.

In the rat developmental neurotoxicity study with direct pup dosing the data of beta-cypermethrin were confirmed. Alpha-cypermethrin did not induce neurotoxicity in dams or in pups prior to the onset of direct pup dosing up to a dose of 5 mg/kg bw/day. With the onset of direct pup dosing clinical signs of acute neurotoxicity were induced in pups at a dose level of 0.5 mg/kg bw while higher doses (2 and 5 mg/kg bw) induced mortality after the first bolus application. A high rate of pups that died revealed necropsy observations (gas bubble in the stomach or blood coagulum in renal pelvis). A 50% reduction of the pup dose to 0.25 mg/kg bw/day - while remaining the pre-treatment of dams until PND 8 at 0.5 and 2 mg/kg bw/day - resulted in similar signs of neurotoxicity, however reduced in incidence and severity in a maternal-dose dependent manner. This is indicative for a substantial exposure via milk. Apart from these acute neurotoxic symptoms, pup development was unaffected in all test groups. The transient character of clinical signs with no other indication of developmental changes is furthermore a proof of acute and not developmental neurotoxicity.

The analytical determination of alphacypermethrin in PND 4 and PND 10 pups confirmed the lactational transfer and substantiated the lack of neurotoxic effects prior to direct dosing: it could be demonstrated that the pup body burden is maternal-dose dependent and measureable amounts were still detectable after a 2 day period without maternal treatment. Furthermore, alphacypermethrin was shown to be primarily stored in the carcass with no relevant findings in the brain after lactational transfer. Based on the quantitative determination of the alphacypermethrin in PND 4 and PND 10 pups, under consideration of the bioavailability of 46 %, the LOAEL is calculated to be 0.265 mg/kg bw, while the NOAEL is calculated to be 0.22 mg/kg bw.

A further indication that double exposure via milk is crucial for the reported clinical signs of neurotoxicity comes from the open literature with cypermethrin and is discussed in CA 5.8.2/8 (Doc ID 2007/1070386). This publication reports lack of any neurotoxic clinical signs in pups gavaged with 1.5 mg/kg bw Cypermethrin from PND 10 to PND 16. The main difference in this report is that pups were reared by unexposed dams.

Due to clear indications for substantial double exposure in this study and based on the extreme form of application, BASF does not consider the DNT study endpoint after direct gavage as relevant for risk assessment but will use the endpoint generated in the dietary DNT study with zeta-cypermethrin based on the following reasons:

Reasoning for the appropriateness of dietary DNT studies for cypermethrins

What was the argument for demanding pup exposure via direct gavage with beta-cypermethrin? It was the example of Carbofuran which was mentioned exemplarily within the peer review meetings of beta-cypermethrin that was assessed on the basis of such a study.
Carbofuran shows nearly no accumulation potential in fat and therefore extremely low amounts in milk. Residue levels in fat and milk of ruminants amount for 0.0003 and 0.000083 mg/kg, respectively. So, lactational transfer is highly limited with carbofuran and the investigation of the impact of this substance on newborns within the most vulnerable phase of brain growth sprint is not possible without direct dosing. Here, a DNT study with direct pup dosing is reasonable to investigate the hazard. According to the OECD Guideline 426 for DNT studies it is stated that: "*direct dosing of pups should be considered in those cases where there is a lack of evidence of continued exposure to offspring*."

The case is different for the group of cypermethrins. Both zeta-cypermethrin and beta-cypermethrin are mentioned in the respective EFSA conclusions to have accumulation potential in fat. Residue levels in fat and milk of ruminants amount for ≤0.02 and <0.005 mg/kg for zeta-cypermethrin; and 0.044 and ≤0.002 mg/kg for beta-cypermethrin, respectively. Thereby the residue levels are approximately 66-133 fold higher than for Carbofuran.
In utero and lactational transfer in rats has been shown both for zeta-cypermethrin and beta-cypermethrin, and there is no reasonable argument that a lactational transfer would not also be expected for alpha-cypermethrin.
Indirectly a lactational transfer has been shown with the current DNT study with alpha-cypermethrin because the maternal pre-exposure with 0.5 or 2 mg/kg bw resulted in a dose dependent exacerbation of the clinical symptoms of the pups at the start of direct gavaging, although all pups were dosed at the same level of 0.25 mg/kg bw. This is only explainable under the assumption that the exposure via milk resulted in a dose-dependent pre-exposure of the pups. Further indications of lactational transfer are coming from cattle studies with alpha-cypermethrin (see CA 6.4.2)

As mentioned before, the two major concerns with regard to the beta-cypermethrin developemental neurotoxicity study (direct pup dosing) were:

1. Neurological development of infants and toddlers partly corresponds to early post-natal development in rodents
2. today babies are not only breastfed but receive additional exposure via bottle milk, baby food or air drift as residents.

These concerns are addressed in the following:

Point 1:
A bolus administration in rat at PND 10-17 covers a timespan of human brain development that to a great extent takes place already in utero and that is mainly completed up to birth. For example is the extent of neurogenesis, synaptogenesis or myelination of early postnatal rodents less developed and partly comparable to the respective extent found in humans at birth [see KCA 5.7/2 2006/1051313]. The predominant purpose of direct pup dosing is to ensure pup exposure in those cases where exposure during lactation in animal testing is definitely not given, i.e. exposure of the animal (rat) during the most vulnerable time of brain spurt (PND8 to 14) is not given and therefore effects in humans until birth are not covered. The aim of direct pup dosing is therefore a hazard based approach to get valuable information about substance-related impairment of the brain development during pregnancy and not to simulate the “bolus administration” during breast feeding. In cases where lactational exposure is given, this continous exposure is more relevant to investigate a substance induced effect than the artificial pup dosing with all its secondary consequences.

Point 2:
The concern that babies may be exposed to a relevant extent in a residential situation should be ruled out. Exposure via the inhalation route will be limited. Given the low application rate (maximum of 30 g active substance/ha) in combination with the low vapour pressure of alpha-cypermethrin (3.8 x 10^{-7} Pa at 20°C) inhalation exposure is considered irrelevant. This has also been demonstrated by air concentration measurement in an environmental setting which suggests air concentrations higher than should ever be expected outdoors. Detailed information has been provided in MCP 7.2.2 of the representative preparation BAS 310 55 I.
Exposure via the dermal route is considered negligible too, because babies will have limited activities outdoors in areas that may have experienced contamination via spray drift. Therefore both kinds of exposure scenarios are considered of low relevance which is in full detail addressed in the risk assessment of bystanders and residents (see MCP 7.2.2).

Quite interesting is a comparison of rat exposure vs. human exposure via milk.
The study of dietary lactation transfer of zeta-cypermethrin at 50, 125, 300 ppm in rats from Gestational day (GD) 6 to PND 21 revealed plasma levels half as high as the corresponding dams at PND5 and 21 [see CA 5.7.1/2 2013/1418020]. The measurement of zeta-cypermethrin in milk following dietary administration at 125 and 375 ppm from GD 6- LD17 [see CA 5.7.1/2 2013/1418020] revealed the following mean concentration in milk (see Table 5.7-5). These mean milk concentrations can be used to roughly estimate the mean substance uptake following the equation and parameters used in model simulation for lipophilic substances in rats, published by Lehmann et al., 2014, [see KCA 5.7/3 2014/1320560].

Table 5.7-5: Rough estimation of mean zeta-cypermethrin uptake based on mean concentrations in milk (ppm)

Endpoints/dose	***0***	***125 ppm***	***375 ppm***
Compound ingestion *during gestation Day 6-20 (mg/kg bw/d)*	*0*	*8.59*	*24.38*
Compound ingestion *during lactation Day 1-17 (mg/kg bw/d)*	*0*	*16.71*	*47.73*
Mean zetacypermethrin in milk (ppm)			
Day 4; *Pup ingestion week 1: 3 gr milk/day;* *Mean pup weight week 1: 6,6 gr*		*0.58*	*7.86*
Day 11; *Pup ingestion week 2: 5.4 gr milk/day* *Mean pup weight week 2: 14 gr*		*3.9*	*10.46*
Day 17; *Pup ingestion week 3: 5.9 gr milk/day* *Mean pup weight week 3: 28 gr*		*3.17*	*11.55*
Mean zetacypermethrin uptake (mg/kg bw/day)*			
Day 4		0.26	3.57
Day 11		1.50	4.03
Day 17		0.67	2.43

*Mean zeta-cypermethrin uptake is calculated as mean concentration in milk (ppm) x ingestion rate [kg milk /day]/ pup body weight [kg]

Based on these values, which are in comparison with the data generated in the range finder study for the developmental neurotoxicity study rather underestimations at day 4 and day 17 ([see KCA 5.7/1 AL-901-031], Table 5.7.1-1 the dietary maternal exposure at 375 ppm is considered the NOAEL for developmental neurotoxic effects) resulted in peak levels at around PND 11 of up to 1.5 mg/kg bw/day and 4.03 mg/kg bw. The main developmental neurotoxicity study, which was performed at the same dose levels, showed reduced litter weight and "*occasional statistically significant differences were obtained in some offspring behavioral parameters and in one of 32 brain morphometric measurements of top dose pups, but these apparent alterations did not occur to a degree or in a manner suggesting a compound-related pattern of developmental neurotoxicity" [see KCA 5.7/2 2006/1051313 and Table 5.7.1-2]*. In conclusion the uptake of doses up **to 4.03 mg/kg bw/day** did not induce developmental neurotoxic effects.

In comparison to that, human exposure scenarios via lactation or bottle milk / baby food show a definite lower level of exposure. According Commission Directive 99/39/EC baby food is required to contain no detectable levels of pesticide residues, meaning not more than 0.01 milligrams of pesticide residues per kilogram. Data from the "2011 European Union Report on Pesticide Residues" (EFSA Journal 2014; 12(5):3694, online available: http://www.efsa.europa.eu/de/efsajournal/doc/3694.pdf) indicate that from 1796 surveillance samples of baby food only 3 samples showed cypermethrin residues at a level of 4-5 µg/kg.

Human breast milk concentrations of cypermethrin are available in the open literature from areas where pyrethroids have been widely used for agriculture and disease control. In Table 5.7-6 human breast milk concentrations from Brazil and Colombia (from Corcellas et al., 2012; [see KCA 5.7/4 2012/1367722]) as well as Mozambique (taken from Feo et al., 2012, [see KCA 5.7/5 2012/1367723]) are presented together with concentrations found in baby food. Two of the given milk concentrations, namely Rondonia (2003) and Mosambique (2002), are pool data considered to represent a worst case scenario because the milk samples were taken after epidemic years in which pyrethroids were applied for dengue control at an extensive level. According to the WHO's practitioner's guide (WHO, 2003: Space spray application of insecticides for vector and public health pest control – a practitioner's guide. Geneva, World Health Organization, online available at: http://whqlibdoc.who.int/hq/2003/WHO_CDS_WHOPES_GCDPP_2003.5.pdf) the frequency of indoor treatments can vary widely with the country or geographical area, from annual or occasional treatment to weekly treatments in epidemic situations and disease outbreaks. In addition, the milk concentration from human breast milk samples is given for Columbia and Rondonia in 2006, which were taken in the years after installation of approved guidelines of pesticide use to control dengue in a national program. In all situations, the use and the exposure is considered to be higher than that considered realistic in case of agricultural use.

Table 5.7-6: Lactational exposure levels in humans*

Human situation	Cypermethrin						
data	Baby food	Brazil, Rondonia (2003)*	Brazil, Rondonia (2006)*	Columbia*		Mosambique (2002) **	
		mean	mean	mean	max	mean	max
Concentration of Cypermethrin in milk fat (ng/g fat)		96	4.14	4.25	16.4	54	160
Mean lipid weight in milk [%]		4.0%	4.0%	2.48%	6.43%	4.4%	
Milk levels [µg/kg milk]	5	3.84	0.17	0.11	1.05	2.4	7.0
Estimated daily intake [µg/kg bw]	0.87	0.67	0.02	0.02	0.18	0.42	1.23

*Corcellas et al., 2012; Env. International 47, 17-22, [see KCA 5.7/4 2012/1367722]
**Feo et al., 2012, Env. International 38, 67-72 [see KCA 5.7/5 2012/1367723]
Baby food data: EFSA Journal 2014;12(5):3694; online available; see link in text above

The highest mean cypermethrin residue value was found in the post epidemic Brazilian pool data (10 individuals) from 2003 with a mean concentration in milk fat of 96 ng/g fat (see Table 5.7-6). An estimation of the **maximal value** in this pool sample, considering the highest ratio between mean and max value found in the dataset (Columbia: 16.4 / 4.25 = 3.8), leads to an **estimated maximum value of 370 ng/g fat**. Applying a mean lipid weight in human breast milk of 4% ends up in an **estimated max value of 15 µg/kg milk**.

The estimated daily intake (EDI) for infants of 1 month age can be calculated following WHO recommendations based on the following equation: EDI = C*F*Mb;
with C=concentration in milk fat [ng/g lipid weight]; F =fraction of fat in breast milk [%]; Mb= daily consumption of milk taken as 0.175 kg milk / kg bw / day (WHO parameter).

The EDI is calculated for the pool data and given in Table 5.7-6 in the last line. The worst case exposure for infants of 1 month age based on the estimated max value of 15 µg/kg milk would result in an **EDI_{max} of 2.6 µg/kg bw/day**. This highest estimated daily intake for human infants is by a factor of 1500 below the estimated pup exposure of 4 mg/kg bw realized in the developmental neurotoxicity study with zeta-cypermethrin at 375 ppm that did not induce neurotoxicity.

Further human breast milk pool data from years after installation of approved guidelines of pesticide use to control dengue in a national program demonstrate the drop in cypermethrin concentration in human breast milk when application is done controlled but still with the objective of effective disease control (see Table 5.7-6: Rondonia, 2006 and Columbia). The highest value is represented by the Columbia dataset with a maximal content of 16.4 ng Cypermethrin/g fat. This maximal value is in addition by a factor of 22 lower than that considered as worst case scenario calculated above.

Otherwise, **babies fed with bottle milk will**, even in the hypothetic case of daily exposure to contaminated bottle milk, stay below an **EDI of 1 µg/kg bw/day.** This value is calculated based on the highest residue level found according to the "2011 European Union Report on Pesticide Residues" in 0.2% of probed bottle milk samples. In this respect also here the dietary DNT study can be assumed to be already a worst case scenario that completely covers the human lactational exposure via the different sources.

In conclusion, the concern that babies are exposed not only via breast milk but receive additional exposure via bottle milk, baby food, hand to mouth exposure or air drift as residents is adequately addressed via a normal DNT study in which the exposure of rat pups is at least 1500 times higher than that expected for newborns under worst case scenarios.
Further arguments are discussed in the following that substantiate the limitations of direct pup dosing for the use in human risk assessment of cypermethrins:

- While babies are normally breast- or bottle-fed all 2-4 hours, direct pup dosing is done once a day as bolus administration. This bolus administration leads to artificially high plasma peaks, which is crucial in a case where an effect is related to peak concentrations at the target organ.
- By this bolus administration the complete metabolic system is easily overwhelmed, especially for CYP P450 enzymes which are only little active in the rat at birth and are mainly responsible for cypermethrin detoxification ([see KCA 5.7/7 2009/1130987] Scollon et al., 2008). In humans, 1/3 of the adult enzyme capacity of CYP P450 enzymes is already available at birth [see KCA 5.7/2 2006/1051313] so that low levels of exposure are detoxified.
- The development of the blood brain barrier in humans starts in utero and is considered present at birth, whereas in rats the permeability of the blood brain barrier decreases during the first few weeks after birth to exert full maturation not before PND20.
- The volume of distribution of cypermethrins in neonates is related to the body fat content and rat newborns have less body fat than babies.
- The kind of administration in combination with removal from the dams is highly stressful to the pups and can negatively affect their susceptibility and potentially shows effects on the behavior of the pups [see CA 5.8.2/8 2015/1001752].
- Further critical points with bolus administration of cypermethrins are the volume- and vehicle- dependent toxicity which is also found in adult animals.
- Last but not least there are investigations that indicate that the relevant $Na_v1.3$ sodium channel, which is the principal isoform expressed in the developing brain, is significantly more sensitive in rats than in humans [see Reference in CA 5/2 2012/1367222].

In conclusion, direct pup dosing in rats is prone to produce artificial high plasma concentrations with all its worst case consequences but does not reflect the relevant exposure for human risk assessment.

Consequences of artificial high plasma concentrations with regard to metabolic capacity in rat and a comparison with humans:

The plasma concentration of alpha-cypermethrin is the most important factor for its neurotoxic action because the concentration at the sodium channel relates to the extent of toxicity. Two physiological processes contribute to the concentration: a) distribution and b) metabolic degradation. Distribution within the body mainly happens in fat, and alpha-cypermethrin molecules that are stored in fatty tissue are not available to interact with the sodium channels. Therefore sensitivity to alpha-cypermethrin can be considered to decrease within the first 2 month of child growth due to the steady increase of body fat. Pyrethroid metabolism is mainly based on carboxylesterases and P450 enzymes in general. Age-dependent differences in metabolic capacity are largely responsible for the juvenile susceptibility.

In rats the capacity to detoxify Typ II pyrethroids increases steadily within the first 90 days. This was exemplarily shown with deltamethrin in postnatal day 10, 21, 40, and 90day old Sprague Dawley rats [see KCA 5.7/6 2010/1232432]. Doses of 0.4, 2, 10 mg/kg bw led to dose dependent increase but a age dependent decrease of brain concentrations. The following two figures are taken directly from the literature.

Brain DLT Toxicokinetic Parameter Estimates in Orally-Dosed PND 10, 21, 40 and 90 Rats

	Age (PND)	Dosage (mg/kg)	C_{max} (μg/ml)	T_{max} (h)	Half-life (h)	AUC_0^6 (μg·h/ml)	AUC_0^∞ (μg·h/ml)
Brain	10	0.4	0.04 ± 0.02[a]	2 – 6	28.3	0.2	0.8
		2	0.11 ± 0.04[a]	2 – 6	22.5	0.5	3.6
		10	0.38 ± 0.12[b*]	2 – 6	NA	1.5	NA
	21	0.4	0.05 ± 0.02[a]	2	28.9	0.2	0.5
		2	0.06 ± 0.03[a]	2	32.6	0.3	3.0
		10	0.25 ± 0.04[b]	4 – 6	NA	1.0	NA
	40	0.4	0.02 ± 0.01[a]	2	NA	0.1	0.2
		2	0.05 ± 0.01[a]	2	23.5	0.2	0.9
		10	0.14 ± 0.07[b]	4	22.9	0.6	2.5
	90	0.4	0.02 ± 0.00[a]	2	NA	ND	NA
		2	0.04 ± 0.01[a]	1	12.5	ND	0.8
		10	0.18 ± 0.02[b]	2	19.5	ND	2.2

C_{max} values are means ± SE. All others are means. n = 3 – 5
NA – Sufficient time-course data not available to estimate parameter due lethality or lack of DLT detection.
ND – Not determined.
C_{max} group means for each age-group with different letters are significantly different from one another at $p \leq 0.05$.
* Significantly different from PND 40 and 90 values at the same dosage ($p \leq 0.05$).
Values for PND 90 rats are from Kim *et al.* (2008).

The age dependent decrease of brain concentrations was clearly attributed to the increasing capacity of metabolism as shown in the following table for hepatic CYP and esterase dependent clearance.

Age- and Pathway-Dependent Intrinsic Clearance

Age (PND)	Liver Weight[a] (g)	Hepatic CYP[b] Vmax (nmol/h) per g liver	Hepatic CYP[b] Km (nmol/ml)	Hepatic CYP[b] Intrinsic Clearance (ml/h)	Hepatic Esterase[b] Vmax (nmol/h) per g liver	Hepatic Esterase[b] Km (nmol/ml)	Hepatic Esterase[b] Intrinsic Clearance (ml/h)	Liver Intrinsic Clearance[c] (ml/h)	Liver Blood Flow[d] (ml/h)	Liver Extraction Ratio[e] (0-1)
10	0.55	185.30	37.79	2.7	23.78	73.69	0.18	2.87	125.6	0.02
21	2.08	381.33	23.40	33.9	114.25	74.76	3.2	37.1	282.9	0.11
40	6.98	1231.73	34.15	224	296.65	118.78	17.4	269.2	588.4	0.21
90	13.6	2611.30	74.90	474	1981.80	172.5	156.3	630.4	1093	0.34

[a] Liver weights were are from Mirfazaelian et al. (2007).
[b] Metabolic rate constants are from Anand et al. (2006a). Intrinsic clearance is computed on a whole liver basis.
[c] Total hepatic intrinsic clearance (Cl_h) is sum of CYP and CaE intrinsic clearance.
[d] Liver blood flows (Q_h) were estimated as a fixed fraction (17%) of cardiac flow (QC) (Delp et al., 1991). The QC (L/h) was based on body weight (kg): 14.1 x $kg^{0.75}$ (Delp et al., 1991). Body weights (g) were 19.6, 57.9, 153.7, and 351, respectively, for PND10, 21, 40, and 90 rats.
[e] Calculated according to Gabrielsson and Weiner (2000).

Scollon et al, 2008 [see KCA 5.7/7 2009/1130987] confirmed that rat hepatic microsomes show mainly oxidative (85%) and to a minor extend hydrolytic (15%) cleavage. In contrast human hepatic microsomes show mainly hydrolytic cleavage of cypermethrin. The enzymes relevant for hepatic metabolism in humans are mainly Carboxylesterase 1 (hCE1) and 2 (hCE2) and only to a minor extent CYP 1A2, 2C8, 2C9 and 3A4.

Already in 2005 Pope et al., [see KCA 5.7/8 2005/1043480] investigated the carboxylesterase activity in 5 infant and 5 adult human liver samples. In this study esterase activity of liver S9 fractions from 4 - 8 month old infants and older ones were comparable to those of 20 years old adult ones whereas samples of younger (2-3 month old) showed definitely lower activity. The relative density of carboxylesterase 1 and 2 in human liver samples was reduced only in the sample of 2 month old infants. All other samples starting from 3 month onwards showed no significant changes to samples of adult ones. While this study is limited by the number of samples investigated, the findings suggest that if maturational expression of liver carboxylesterase contributes to age-related sensitivity of alpha-cypermethrin in humans, it may only be important during very early postnatal maturation. A further study (Yang et al., 2009; Doc ID 2009/1131242) with 104 samples (48 fetal probes, 34 0-10 year old, 22 adult) investigated the age-dependent expression of hCE1 and hCE2 mRNA. The data indicates a significant increase of hCE1 and hCE2 mRNA expression in the first and second months after birth and thereafter a less steep increase up to the age of 10 years. The inter-individual variability of mRNA levels for hCE1 in fetus and child samples was fairly high (218 fold for children and 431 fold for fetuses) whereas for hCE2 it was less variable (21 fold). On protein level the data were given as pooled data for the 0-10 years old and the adult ones, thereby a conclusion on the time point of highest increase on protein level within the group of the very young ones (0-2 years) compared to 6 years and older was not possible. However, it was shown that deltamethrin is 3 times faster hydrolyzed in microsomal preparations from adults compared to 0-10 year old.

Although the mRNA expression level increases mainly within the first weeks after birth and thereby it is likely that the found protein level differences between the group of 0-10 year and >18 year old is mainly due to differences found in the early postnatal phase of the first months this was not adequately shown in this study.
More differentiated investigations are expected within the next time under the aegis of the council for the advancement of pyrethroid human risk assessment (CAPHRA) which has initiated research projects to learn more about factors that may contribute to age-dependent sensitivity to pyrethroids but which are not yet published.

In conclusion, based on the already available literature findings, it is evident that a maturational expression of liver carboxylesterases and CYP P450 enzymes contributes to age-related sensitivity of alpha-cypermethrin in humans and rats. While rats develop their full metabolic capacity within 90 days -humans display a significant increase of metabolic capacity within the first few months of life and a full maturation within childhood. Main source of dietary exposure of infants is milk and supplemental baby food products, which are not considered to cause relevant exposure in humans compared to the significant exposure in the dietary DNT study of zeta-cypermethrin. Further non-dietary exposure via drift, vapour, dermal route and hand to mouth exposure are also not considered to represent a relevant risk based on low vapour pressure / application rate and low ability for locomotion of very young babies.

Therefore the endpoint coming from the dietary DNT study with zeta-cypermethrin is considered relevant for human risk assessment and can be considered as sufficient and realistic case scenario.

Based on the already peer reviewed studies of alpha-cypermethrin, complemented with the DNT study with zeta-cypermethrin and the 90 day dietary study with alpha-cypermethrin, the following EU endpoints are considered relevant.

Neurotoxicity / Delayed neurotoxicity

Effects observed	Toxic for CNS and peripheral motor nerves; neurobehavioral changes are reversible within 3 days following single dose
Acute rat study	NOAEL = 4 mg/kg bw (in corn oil)
4 week oral rat study	NOAEL = 10 mg/kg bw/day (DMSO) **STOT RE 2**
13 week dietary rat study	NOAEL > 36 mg/kg bw/day (dietary)
Developmental neurotoxicity	Maternal and developmental NOAEL: 9 mg/kg bw /day based on reduced bw gain (zeta-cypermethrin) No developmental neurotoxic potential

Remark: Decisions from other authorities: EPA
The generation of a guideline developmental neurotoxicity study with dietary administration of alpha-cypermethrin was clearly not recommended based on the huge database available for pyrethroids as evaluated by the EPA in the Memorandum "Pyrethroids: Evaluation of Data from Developmental Neurotoxicity Studies and consideration of Comparative Sensitivity" [see KCA 5.7/9 2010/1232203]. In this Evaluation, including zeta-cypermethrin and three other Typ II pyrethroids, it was concluded, that guideline DNT studies with dietary administration do not provide useful toxicity data beyond that available from the developmental and reproductive guideline studies, and therefore proposed that future submissions cite currently available data rather than conducting additional DNT studies with pyrethroids. It was concluded that available DNT and reproduction studies do not indicate that juveniles are more susceptible to pyrethroid toxicity compared to adults.
In a second step EPA targeted on study proposals to identify and quantify potential juvenile sensitivity. In the Memorandum "Re-evaluation of the FQPA Safety Factor for Pyrethroid Pesticides" [see KCA 5.7/10 2011/1269851] this was done via PBPK modelling and in-vivo behavioural tests. A comprehensive discussion of the age dependent pharmacodynamic and pharmacokinetic differences of alpha-cypermethrin toxicity was done.
Based on this evaluation EPA used an acute neurotoxicity study generated with cypermethrin and retained a 3X FQPA Factor for children from birth to 6 years of age, whereas all other population groups were considered adequately protected with an FQPA Safety Factor of 1X. It is stated that: "*The enzymes which metabolize and detoxify the pyrethroids are present in rats and humans at birth. As a result, both juveniles and adults are able to tolerate low doses of pyrethroids when the internal dose, or the amount of pyrethroid at the sodium channel, is low. However, the activity of these enzymes increases with age, conveying in adults a greater capacity to detoxify pyrethroids compared to juveniles.*"(EPA, 2011; [see KCA 5.7/10 2011/1269851]

CA 5.7.1 Neurotoxicity studies in rodents

Copy from the Literature data discussed within the ECCO Peer review meeting - Full Report on alpha-cypermethrin [see KCA 5.7/1 AL-901-031, page 164-172 of 222].

Neurodevelopmental toxicity of pyrethroids:

In a first draft of the monographs of cypermethrin and a cypermethrin, the Belgian authorities concluded that both cypermethrin and alphacypermethrin may induce neurodevelopmental effects based on data reported in open literature and more specifically the papers of Cantalamessa et al (1998), which reported an impairment in expression of renal dopaminergic receptors in Wistar rats following gavage at dose of 1 mg/kg bw/d from PND 10 to PND16. Additionally, we cited the papers of Husain et al (1992) which reported delays in the onset of several neurobehavioral parameters in pups following oral (gavage) administration of cypermethrin to female rats at 15 mg/kg bw from GD 5 to GD 21 and finally the paper of Malaviya et al (1993) which reported neurobiochemical changes in brain, including reductions in AchE and Na^+ - ATPase activities in 3 week old rat following gavage treatment of female rats with cypermethrin at 15 mg/kg bw during lactation days 1 to 21.
The applicant gave a critical evaluation of the different studies as well as an additional report of studies from open literature which support the lack of consistency in results obtained from neurobiochemical assays with different pyrethroids, as well as the lack of neurobehavioral correlates to apparent small changes in neurobiochemical parameters. Papers of Eriksson were evaluated.

Summary of the discussion:

Study of Cantalamessa et al: the authors reported a reduced affinity and increased density of dopamine D1-like receptors in kidneys of rats by treatment with cypermethrin (d10-d17 post natal) (1 mg/kg bw/d) suggesting that the compound impairs dopamine DI like receptors mediated function.
According to the authors, these receptors show a developmental pattern similar to that of central dopamine receptors.
Dopamine exerts important renal action, including diuresis, via binding to dopaminergic receptors in the kidney.
Comments from the applicant: in the 3 generation reproduction study with cypermethrin, no clinical signs related to changes in urinary output were noted for animals exposed *in utero* and during lactation period at concentration up to 500 ppm (25 mg/kg bw/d).
Following the termination of one litter interval, animals were 126 days of age and rats were observed in the Cyanamid study at ages comparable to the ages at which changes in dopaminergic receptors were reported in the paper by Cantalamessa et al. It is noteworthy that potential exposure to developing offspring in the 3-generation study with cypermethrin was significantly longer than the 6 day postnatal treatment period in the study by Cantalamessa et al. The absence of any functional correlates (i.e.changes in the volume of urinary output) to the reported changes in dopaminergic receptors in the kidney following postnatal exposure of pups to cypermethrin undermines the biological significance of the data reported by Cantalamessa et al.

Study of Husain et al, 1992: the authors reported that cypermethrin at 15 mg/kg bw/d had morphological developmental effects in rats such as delayed incisor eruption, ear opening, eye opening, primary coat and delayed grip strength.
Comments from the applicant: in this study, it is likely that deliveries were only recorded during a typical 8-hour workday. As such, pups from different litters born on the same day may be considered by laboratory technicians to be one day apart in age, because one litter may have been born in the morning and another litter may have been born in the later part of the evening following the 8-hour working day. Moreover, the day of occurrence of neurobehavioral parameters was likely evaluated only once per day. As such, if a large percentage of control litters was born in the evening, prior to the next 8 hour work day, one could predict that the day of occurrence for a given neurobehavioral parameter for these control litters was recorded as one day earlier than, rather than the same day as, that for litters in cyp treated group born in the morning. Thus, a one-day increase in day of occurrence of neurobehavioral parameters in cyp exposed animals should not be considered treatment related. In another study of Kavlock et al (Journal Environ. Pathol. Toxicol., 2 : 751-765, 1979), no treatment related increases in day of occurrence of neurobehavioral parameters (pups evaluated for survivability, litter size, body weight and day of occurrence of neurobehavioral parameters including startle response, eye opening, righting reflex) were observed for pups following exposure of dams to decamethrin, another type H pyrethroid, at 2.5 or 5 mg/kg bw/d from gestation day 7 to lactation day 15, an exposure period substantially longer than that utilised by Husain et al.

The study reported by Malaviya et at (1993) suggest that disturbances in dopaminergic and cholinergic pathways may lead to a functional delay in brain maturation after cypermethrin or fenvalerate. Effect on neurotransmitter receptors was examined by In utero exposure to cypermethrin at 15 mg/kg , or fenvalerate at 10 mg/kg bw from day 5 of gestation until day 21.Fenvalerate decreases significantly H^3 spiroperidol or 3H QNB binding to striatal brain membranes in pups and cypermethrin was without effect. An increase of 3H —QNB binding and spiroperidol binding was observed in pups exposed to Fv or Cyp during the lactation period (15 mg/kg cyp day 1 Post natal to 3 week).

These effects suggest, according to the authors, a delayed maturation of cerebral cortex.

Comments from the applicant: it is unlikely that cypermethrin would elicit no changes in enzyme activity in the brain of 3 week old rats following gestation exposure, while a second type II pyrethroid, fenvalerate would increase or decrease enzyme activities following the same pattern of exposure. Changes (increases or decreases) in receptor binding noted for cyp or fenv appear dependent on the period of exposure to either pyrethroid and it is unlikely that these changes in receptor binding are biologically significant. The ligand used (H-QNB) is a non-selective ligand because it does not distinguish between various subtypes of muscarinic receptors. As five different subtypes of muscarinic receptors have been identified and selective radioligands are available for several subtypes, these results should be interpreted with caution. Finally, it is unlikely that modulation of muscarinic receptor binding is relevant or causal endpoint for potential pyrethroid induced neurotoxicity. Because Ac binds to muscarinic receptors, changes in muscarinic receptor binding of acetylcholine may be a more relevant endpoint for cholinesterase inhibitor induced neurotoxicity.
Results from the 3 generation reproduction study with cypermethrin indicate no neurobehavioral effects (clinical signs of neurotoxicity) for animals which had been exposed to cyp in utero and during the lactation period at dietary cc up to 500 ppm. However, at 500 ppm (25 mg/kg bw/d), a statistically significant reduction in parental bw for all generations during the premating treatment period were observed. Animals were likely evaluated for clinical signs of toxicity from approx. 3 weeks of age, the age at which changes in enzyme activity and /or receptor binding were reported in the study of Malaviya et al. As such, the slight changes in enzyme activity and receptor binding in the brain following gestation or lactation exposure to cypermethrin do not appear to be correlated with any neurobehavioral finding.

Results from additional studies with pyrethroids:
Paper of Eriksson and Fredriksson (toxicology and applied pharmacol, 1991, 108, 78-85): the authors reported a decrease in the density of muscarinic receptors in the brain of the adult NMRI mice and an increase in motor activity in these mice following following gavage of pups with either 0.7 mg/ kg bw/d bioallethrin (type I) or 0.7 mg/kg bw/d of deltamethrin (type II) administered in a fat emulsion vehicle from PND 10 to PND 16.
In an attempt to repeat these findings, Muhammad and Ray (study in addendum) conducted a study in which bioallethrin and delatmethrin was administered to neonatal NMRI mice from PND 10 to 16. The test material was administered by gavage as a fat emulsion at 0.7 or 3.5 mg/kg bw/d for bioallethrin and 0.7-mg/kg bw/d for deltamethrin. Muscarinic receptor binding and motor activity were evaluated in 4-month mice. Results from this study showed changes in muscarinic receptor density in adult mice, which had received bioallethrin during the postnatal period. However, these changes were small, inconsistent across 3 trials (increase in one and similar to controls in the 2 other), and not clearly dose related. For animals which had received deltamethrin during postnatal period, muscarinic receptors density at 4 months of age was significantly decreased in one trial, but not in another trial, when compared to controls. Adult animals, which had been exposed to either bioallethrin or deltamethrin during postnatal period, exhibited minimal changes in motor activity in some experiments but not in other experiments. Moreover, the behavioural results for mice that had been exposed to bioallethrin were not dose-related.

The collective results from the paper by Muhammad and Ray show that neurobiochemical and neurobehavioral findings are not reproducible for the same pyrethroid in different experiments conducted at the same laboratory. Moreover, the neurobiochemical findings reported were inconsistent for the two pyrethroids tested (i.e. increased muscarinic receptor binding density in one trial for bioallethrin and decreased muscarinic receptor density in one trial for deltamethrin). Further, the increase in muscarinic receptor density observed by Muhammad and Ray in one trial following exposure of postnatal mice to bioallethrin was in opposition to that observed by Eriksson and

Fredriksson (i.e. decreased muscarinic receptor density) following exposure of postnatal mice to this identical pyrethroid.

The lack of consistent and reproducible findings from the muscarinic receptor binding assay (i.e. density determinations) reported by Eriksson end Fredriksson and Muhammad and Ray suggest that small changes in neurobiochemical parameters are likely not related to treatment with pyrethroids, as these changes were **not sufficiently robust to survive minor intra- and inter-laboratory variations** in experimental conditions. Even if exposure to pyrethroids did result in small changes in neurobiochemical parameters i.e. receptor densities, these changes would be expected to have little functional consequence given the rather large receptor reserve capacity of the brain. In fact, correlation's between changes in muscarinic receptor density and changes in motor activity reported by Eriksson and Frederiksson were **not confirmed** in the experiments conducted by Muhammad and Ray.
Results from inhalation studies conducted by Bayer AG with several pyrethroids support the lack of correlation between changes in muscarinic receptor density and neurobehavioral findings following postnatal exposure to pyrethroids. In these studies, neonatal NMRI mice were administered one of five different pyrethroids via inhalation exposure (6 hours/day) from PND 10 through 16, and animals were evaluated for spontaneous motor activity and muscarinic receptor density at both 17 days and 4 months of age. Data from these studies showed that for the majority of compounds tested, slight increase in muscarinic receptor density were noted for mice on PND 17 with no correlating changes in spontaneous motor activity for these animals. As no consistent neurobehavioral changes could be correlated with the subtle, inconsistent and non-reproducible neurobiochemical changes reported, the neurobiochemical changes, even if treatment related, are likely not biologically significant.
Importantly, results from inhalation studies by Bayer AG showed no changes in muscarinic receptor density or spontaneous motor activity in adult (4 month old) mice for the majority of pyrethroids following postnatal inhalation exposure. These data from adult (4 month old) mice further refute the findings by Eriksson and Fredriksson.
The data reported by Bayer AG are noteworthy as the inhalation route of exposure can be considered as worst-case route of exposure compared to the oral route. This is because the inhalation route of exposure bypasses the first-pass effect of the liver (i.e. detoxification) and generally results in complete (i.e. 100%) absorption of a chemical into the systemic circulation.
The detailed analyses of neurobehavioral findings and biochemical changes in the brain and kidney, as presented in the above sections, indicate that prenatal and post natal exposure to pyrethroids does not result in any significant changes in neurotransmitter release or receptor number in the brain or kidney which can be correlated with adverse effects on neurobehavioral or physiological function. Based on the lack of conclusive (i.e. reproducible and consistent) evidence from the open literature, which suggests that pyrethroids are developmental neurotoxicants, a developmental neurotoxicity study with alphacypermethrin is not scientifically justified.

Additional support which negates the requirement for a developmental study with alphacypermethrin:

An evaluation of the collective results from the developmental toxicity studies with alphacypermethrin and the developmental and the 3-generation reproduction studies with cypermethrin also negates the requirement for a developmental neurotoxicity study with alphacypermethrin. Results from the developmental toxicity study in Sprague-Dawley rats with alphacypermethrin technical support a NOAEL for maternal toxicity of 9 mg/kg bw/d, based on clinical signs of neurotoxicity, decreased food consumption, and a weight gain reduction of 22% for dams during the treatment period (gestation day 6 to 15) at 15/18 mg/kg bw/d, the highest dose tested. The NOEL for developmental toxicity in this study was also 9-mg/kg bw/d, based on decreased foetal weights at 15/18-mg/kg bw/d.

In New Zealand rabbits with alphacypermethrin a NOAEL for maternal toxicity of 15 mg/kg bw/d is based on reductions in food consumption and body weight loss of 0.03 kg during treatment period (gestation day 7 to 19) at 30 mg/kg bw /d, the highest dose tested.
Results from the developmental toxicity study in Sprague Dawley rats with cypermethrin support a NOEL for maternal toxicity of 17.5 mg/kg bw/d, based on a 12% reduction in body weight gain during the treatment period (gestation day 6 to 15) at 35 mg/kg bw/d, the next highest dose tested. Results from developmental toxicity study with rabbits with cypermethrin support NOELs for maternal and developmental toxicity of 120-mg/kg bw/d, the highest tested dose.
Lastly, results from the 3 —generation reproduction study with cypermethrin in Wistar rats support NOELs for parental and offspring toxicity of 100 ppm (5 mg/kg bw/d), based on reductions in food consumption and mean body weights during the premating treatment periods for all parental

generations and reduced mean pup weights on PND 21 for F3B males at 500 ppm, the highest tested dose.

The results from these studies demonstrate that neither alphacyp nor cyp is a selective developmental toxicant or a teratogenic agent in either the rat or rabbit. Because neither alphacyp nor cyp is selectively toxicant to the developing fetus or offspring, there is no increased sensitivity of developing offspring following prenatal or postnatal exposure to alphacyp or cyp. In the absence of any selectivity or increased sensitivity to the developing fetus or offspring, a developmental neurotoxicity study with alphacyp is not warranted.

A pyrethroid ad hoc working group sponsored by Industrieverband Agra e. V. developed a white paper which discussed the potential neurotoxicity of pyrethroids to developing animals. This paper was submitted to the German regulatory authorities (BBA/BGA) in June 1994. A retrospective of the multigenerational and developmental toxicity studies conducted with many different pyrethroid insecticides was included in this paper. Results from this analysis support the conclusions drawn from the developmental toxicity and reproductive toxicity with alphacyp and cyp, namely, that neither pyrethroid is selectively toxic to developing offspring. A summary of the results from reproductive toxicity studies with numerous pyrethroids, including deltamethrin and zetacypermethrin, is included (table 1), appendix C and a summary of the results is included in table (2) appendix C. Finally a comparison of the results from the rat reproductive toxicity and rat developmental toxicity studies conducted with these pyrethroids is included in table 3 of appendix C

The results presented in table 1 of appendix C show that the majority of NOELs for offspring's toxicity are equal or higher than the NOELs for parental toxicity. Moreover, results from the majority of these studies show no functional or morphological changes in the nervous system of offspring. Although the multigenerational studies were not specifically designed to assess potential peri- or postnatal neurotoxicity following pyrethroids, these studies can, in fact, be used to identify any potential effects on the developing nervous system. Specifically, if pyrethroids were developmental neurotoxicants, pups exposed to pyrethroids in utero and during lactation would be expected to show impairment of physical development and growth and maturation, as well as inadequate nestbuilding (for rearing of subsequent generations) and lactation. In fact ad hoc working group reported that rearing and nursing behaviour of dams from all parental generations, as well as social and suckling behaviour of neonates, were not affected by treatment and, except in cases where clearly toxic (i.e. sublethal) dose were tested, there were no disturbances in the physical development of offspring during lactation and post-weaning periods.

In table 2, the NOELs for embryotoxicity are generally equal to or higher than the NOELs for maternal toxicity for both rats and rabbits. These results presented in table 3 show that for nearly all pyrethroids, the NOEL for toxicity to offspring from reproduction study in rats is of similar order of magnitude as the NOEL for developmental toxicity from the rat developmental toxicity study. These data indicate that there is no enhanced toxicity to developing offspring following an extended exposure period (including exposure during postnatal development) as well as an evaluation of additional endpoints of potential offspring toxicity, in the multigenerational reproduction studies conducted with pyrethroids.

Taken together, the retrospective analysis presented in the white paper prepared by the pyrethroid ad hoc working group provides strong evidence that treatment of lactating and/or pregnant females with pyrethroids is not associated with developmental neurotoxicity.

Our evaluation:

Age-dependent differences in susceptibility to pyrethroid insecticides (Sheets, L. (Neurotoxicology, 21(1-2): 57-64, 2000).

The effect of pyrethroids on the startle response in 21-day-old animals (Long-Evans rats)was compared with effects in adult rats of same strain. The study was performed in the same lab. and results are summarised in table 1. The results indicate that there are no differences between pups and adults in startle response.

In this study a difference between pups and adults was seen: higher doses selected for startle responses generally evoked clinical signs in pups and not in adults. This age-dependent difference in response led to determinations of the LD50 for pups and adults. (Table 2) Differences were seen for deltamethrin and cypermethrin and for 21 day old rats, the LD50 was approximately 7 fold lower than for the adult with both Type II pyrethroids.

Deltamethrin was used as a model type II pyrethroid to investigate target tissue brain levels and cc of deltamethrin was measured in whole brain, the presumed target tissue under the circumstances that reduce the startle response by approximately 50% (2 h after 4 mg/kg bw):

The results showed that brain level adult deltamethrin conc. were << than brain level in weaning.

At the respective LD50 dose, the same brain level is observed in adults and weaning rats, which received a seven time lower dose.
The main age related differences that the applied dose associated with a lethal brain cc is much lower in pups than in adults and this difference only occur at high dose levels that exceed the neonate's capacity for detoxification. This difference seems to be specific for type II pyrethroids and is consistent with metabolism as basis for the difference since these compounds contain alpha cyano moiety that slow down their degradation to less toxic metabolites.

In conclusion, neonates are more sensitive than adults to acute lethal dose of certain pyrethroids but not to the much lower levels that are relevant for dietary risk assessment. Target sites in the brain of the neonates are not inherently more sensitive than in the adult over a wide range of exposure. These results indicate that susceptibility of neonates is associated with the neonate's relatively limited metabolic capacity for detoxification.

Table 1.

	control	Low dose	Middle dose	High dose
		% increase or decrease in response amplitude relative to control values		
Cismethrin	50±2	0 (3 mg/kg bw)	-16(6 mg/kg bw)	-24(12 mg/kg bw)
In adults		2 to 3 fold increase in startle response is observed at 6 or 12 mg/kg bw.		
Permethrin	38+4	+5 (30 mg/kg bw) no effect	+65 (60 mg/kg bw)	+22 (120 mg/kg bw)
In adults			in adult also increase 2-fold	in adult also increase 2-fold
Deltamethrin	58+6	-20* (1 mg/kg)	-39*(2 mg/kg)	-59*(4 mg/kg)
In adults		Decrease in response amplitude of 50% at 4 mg/kg		
cypermethrin	41±5	-40* (9 mg/kg)	-45*(19 mg/kg)	-55*(38 mg/kg)
In adults		Decrease in response amplitude of 50% at 38 mg/kg		

*P<0.05

Table 2. Lethality in neonatal, weaning and adult male rats

	PND11	PND21	PND72
	LD50 (mg/kg bw)		
Cismethrin	119	32	55
Permethrin	254	201	406
Deltamethrin	5	11	81
Brain level at 4 mg/kg		0.052 tig/g brain	0.023 pg/g brain
Brain level after lethal dose		0.129 Rig after lethal dose of 12 mg/kg bw	0.145 tig/g after lethal dose of 80 mg/kg bw
cypermethrin	18	73	439

Neurobiology, Gomeza et al, PNAS, 1999, **96, 10483-10488.**

Coordinated locomotor activity depends on the proper balance of muscarinic cholinergic and dopaminergic neurotransmission in the **striatum.** The M4 muscarinic receptor is expressed abundantly in the striatum. **M4 receptor** deficient M4 -/- mutant mice showed a significant increase in basal **locomotor activity** (by about 30% as compared with their wild type littermates) as measured by the total number of consecutive photobeam interruptions during a 60-min test period. M4 receptor **KO** mice were more active than their wild type. The M4 receptor subtype is colocalised with D1 and D2 dopamine receptors on striatal projection neurons. D1 receptor —mediated increases in locomotor activity are potentiated in the absence of functional M4 receptors. In mice, **M2** receptor stimulation is primarily responsible for muscarinic receptor dependant **analgesia.** Muscarinic receptor mediated **salivation, tremor, and hypothermia** responses are dependent of **M3** subtype receptor. **M1** receptors may play a role in higher cognitive functions such as **memory and learning.** Antagonists that display high affinity for MI receptors also show high affinity for M4 receptors. Members of the muscarinic acetylcholine receptor family(M I -M5)are widely expressed in the central nervous system and in body

periphery. Central muscarinic receptors are known to play key roles in memory and learning as well as in the regulation of many sensory, motor and autonomic processes. In the periphery, muscarinic receptors mediate the well known activities of acetylcholine released from parasympathetic nerves. The precise functional role of the individual muscarinic receptor species remain to be determined.
At the molecular level, the M4 subtype is expressed abundantly in the striatum (caudate-putamen) and are also present, though at lower levels, in several other brain regions including cerebral cortex and hippocampus. In the striatum, a region known to be critically involved in extrapyramidal motor control, the M4 as well as other muscarinic receptor subtypes are coexpressed with D1 and D2 dopamine receptors on striatal projection neurons. Considerable evidence suggests that complex interactions between these two neurotransmitter receptor systems are critical for the proper regulation of motor control.. in M4 receptor deficient mice an increase in basal locomotor activity and hypersensitivity to stimulatory locomotor effects of D1 receptor activation were observed, indicative of functional interaction between these two receptor systems.3 H QNB is a non selective muscarinic antagonist which labels all five muscarinic receptor subtypes. Olfactory bulb contains also high levels of QNB labeled receptors, comparable to striatum

Eriksson and Fredriksson, toxicol and applied pharmacol., 108, 78-85,1991.

Spontaneous behavior was tested in male mice at the age of 17 days and 4 months : motor activity, locomotion, rearing were measured. Receptor assay was performed in cerebral cortex, hippocamus and striatum measuring receptor density using 3H QNB as antagonist for labeling of all muscarinic receptors.
Results: neonatal exposure to bioallethrin or deltamethrin between day 10 and day 16 postnatal day produces significant behavioral deviations in the adult mouse at 4 months about 1 week after behavioral tests on 4 month mice, they were killed and MAChR were assayed in Ccortex, hippocampus and striatum. There was a significant decrease and tendency toward decrease in the amount of 3H QNB binding sites in cerebral cortex of mice receiving bioallethrin (0.7 mg)and deltamethrin (0.7 mg, at 10 days for 7 days)respectively. In the other brain regions, hippocampus and striatum, no significant changes were observed.

	Densities of muscarinic receptors in adult mouse		
	cortex	hippocampus	striatum
Control	1266 ±82	1332±134	1539±91
Bioallethrin	1132±188*	1377±108	1477±127
deltamethrin	1204±88*	1354±76	1546±122

Recently, the authors reported that neonatal exposure to bioallethrin and deltamethrin, administered in the same manner and at the same doses as in the present study, affected the MAChR in the 17 day old mouse. The bioallethrin exposure caused an increase in the density of MAChR in cerebral cortex and an increase and decrease in proportion of binding sites. Deltamethrin treatment also caused an increase in the density of MAChR, but the proportions of HA and LA binding sites were reversed.

Densities of muscarinic receptors in cerebral cortex and hippocamus of neonatal mice

		Deltamethrin mg/kg bw Type II		Bioallethrin mg/kg bw Type I	
	control	0.71	1.2 clinical signs	0.71	72 clinical signs
Cortex	805±60	869±78*	815±48	875±47*	801±66
Hippocampus	923±110	890±73	854±51*	875±77	907±65

(toxicol. Applied Pharmacol, 102, 456-463)

Malaviya et al, 1993: pregnant rats were given 15 mg/kg bw cypermethrin daily from day 5 until day 21 of gestation. Synaptosomal membranes were prepared from corpus striatum. Binding was assayed with spiroperidol or QNB. No clinical signs were noted. QNB binding was markedly *decreased* ; no significant changes in binding in spiroperidol to striatal membranes in 4 week of age pups exposed during gestation day 5-21.
Another group was exposed during lactation from day 1 to 3 week to 15 mg/kg bw cypermethrin. ^{3}H QNB binding to striatal membranes was statistically significantly *increased.* Spiroperidol binding was not modified.

In the developing rat brain, mAChRs appear between gestational day 16 and 18. Their density increases gradually after birth, reaching adult levels at approximately 30-45 days of age. MeHg which increases mAChR density (up-regulation) in adult rat brain (hippocampus and cerebellum) decreased temporaly binding in the immature rat brain. MeHg inhibits development of mAChRs when administered before they appear (Coccini et al, environ.health perspect. 108, 2000, 29-33).

Our conclusion: coordinated locomotor activity depends on the proper balance of muscarinic cholinergic and dopaminergic neurotransmission in the striatum. M4 receptor subtype is particularly abundant in the striatum and receptor deficient M4 -/- mutant mice showed a significant increase in basal locomotor activity, It is *likely* that increased locomotor responses are a result of altered striatal activity.
In the paper of Eriksson, a slight decrease (statistically significant ?) is observed in the cortex. Therefore, a correlation between locomotor activity modification and decrease in receptor density in striatum is not established.
In the paper of Malaviya, exposure to cypermethrin at 15 mg/kg bw/d during gestation, 3H QNB binding is decreased and during lactation, 3H QNB binding is increased. The results reported with fenvalerate are not in the same direction. (!)
Based on this study, the NOAEL is < or = 15 mg/kg bw/d .
In the 3 generation rat study, the NOAEL = 10 mg/kg bw/d.

Vulnerable periods during ontogenesis of CNS can be divided into 2 major courses of events :

1. early brain development : brain aquires its general adult shape and precursors of glial cells and neurons proliferate.
2. Brain growth spurt : rapid fundamental changes including maturation of axonal and dendritic outgrowth; establishment of neural connections; synaptogenesis, cell, axon and dendrite death proliferation of glial cells with accompanying myelinization = cytoarchitectural changes are accompanied by a vast number of biochemical changes. Fetal-neonatal brain -+ mature adult. Stage of development when animal acquire many new motor and sensory faculties and spontaneous motor behavior. In adult animal, induction of behavioral and cholinergic disturbances is limited to a short period namely around post natal day 10.

In human, BGS occurs maximally at the 3rd trimester of pregnancy up to 2 years of life. In mouse and rat: this period is neonatal spanning the first 3-4 week of life.
Early postnatal exposure in the rodent encompasses a time span equivalent to peri-neonatal exposure in the human. Therefore, by administering substances to mice during the 10-16 postnatal day period, there will be a situation, which is not identical to that in humans but still involves a sensitive period of postnatal life. Although the rodent seems to be an appropriate model for testing the postnatal neurotoxic effects of pyrethroids to make extrapolations to human, the use of only one single mouse strain gives a rather narrow view.
In order to affect brain receptors, the compound must in a first time reach brain ; no specifically high level of radioactivity was observed in rat brains. It should be important to know if cypermethrin accumulates in brain and more specifically in a particular brain region (cortex ?)
Adaptative changes involving mACHrs in target organ may occur after prolonged overexposure and neurotransmitter receptors are regulated by homeostatic mechanisms that compensate for changes in the amopunt of agonist or antagonist to which they are exposed. For example, although mAChRs overstimulation induces a receptor density decrease (down-regulation), prolonged treatment with muscarinic antagonists cause receptor increase and supersensitivity to muscarinic agonists. It is increasingly evident that adaptation of neurotransmitter receptors may be a primary mechanism mediating the long term actions of drugs and chemicals in the CNS. In the developing rat brain, mAChRs appear between gestation day 16 and 18. Their density increases gradually after birth, reaching adult levels at 30-45 days of age. It seems however that the postulated up regulation mechanism which in the adult compensates for the complex interactions of compounds with cholinergic system is not functional in the immature brain. (Coccini et al, 2000).

Which kind of animal study should be designed to gain more information relevant for clarifying the mechanisms of action and the endpoints for determining a dose effect relationship ?

End of Copy from the Literature data discussed within the ECCO Peer review meeting report
Alpha-cypermethrin, hens, route not given, 0, 70, 140 and 700 mg/kg bw. Reversibility after 700 mg/kg bw ([redacted] 989c) (Dossier Gharda)

Guidelines:	Partly in compliance with the test method OECD 418 (1984)
Deviations:	Test should include a positive control group and a concurrent control group of animals. Route of administration is not given. Statistics were not performed. Absence of complete raw data.
GLP:	No
Acceptance:	not stated

5 Young adult female domestic white leghorn hens received a single dose alpha-cypermethrin (99.0%) at 0, 70, 140 and 700 mg/kg bw /d in DMSO. One group exposed to 700 mg/kg bw was observed for reversibility of toxic effects.
Findings: no effect was seen on behavior (no ataxia developed), food consumption or body weight. Histopathology revealed a dose-related pattern of effects on brain, spinal cord, and sciatic nerve. Inflammatory reaction in the brain accompanied with astrocytic proliferation (140 mg/kg bw) or vacuolated neuron cells with astrocytic activation, slight changes in certain fields, and edema (700 mg/kg bw).
Conclusion: At high doses, alpha-cypermethrin is toxic for the CNS and peripheral motor nerves. The NOAEL for neurotoxicity under the conditions of this study was <70 mg/kg bw.

Alphacypermethrin, rat. oral by gavage 0, 4, 20 and 40 mg/kg bw (1993b, see CA 5/13 AL-451-004) (Dossier Cyanamid)

Guidelines:	The study was performed according to the EPA Pesticide guideline subdivision F. Addendum 10. Neurooxicity Series (1991) and is therefore not in compliance with OECD guideline 418 (1984)
Deviations:	Hens must be used and observed during 21 days. There is no positive control.
GLP:	Yes
Acceptance:	The study was recommended to be used for acute reference dose setting.

10 Charles River (Crl:CD:BR)rats /sex/dose received by gavage a single dose alpha-cypermethrin(ST91/243) at 0, 4, 0, and 40 mg/kg bw/d in corn oil. Animals were observed 14 days following treatment.
Findings: In the range-finding study for the acute neurotoxicity study in rats deaths were observed at 50 and 60 mg/kg bw. Profound signs of neurotoxicity were observed at 40, 50, and 60 mg/kg bw. In the main study each one animal died in the 20 and 40 mg/kg bw dose groups. Effects observed included severe clinical and neurobehavioural changes shortly after dosing. Most changes were completely reversed by 3 days after dosing in the surviving animals. The only histopathological finding showing a relationship to treatment was sporadic fibre degeneration in the sciatic nerve, which occurred in the control and dose groups, but with an incidence that was statistically significant at 20 and 40 mg/kg bw only. Fore and hind limb gripstrength, hind limb landing footsplay, motor activities were not affected.

Table B.5.7.1-1 : Neurotoxic effects of alpha-cypermethrin: FOB conducted 5 hours post-dosing observations showing overall statistical significant difference.

Endpoint/dose	0	4 mg/kg bw/d		20 mg/kg bw/d		40 mg/kg bw/d	
		♂	♀	♂	♀	♂	♀
Mortality				1		1	
Body weight		all animals gained weight between the day of dosing and day 14.					
Functional observational							
battery findings							
posture in home cage				*		*	
removal from home cage				*			
handling in hand				*			
fur appearance						*	
salivation				*			
arousal							**
static limb position						*	
abnormal gait type						*	
fore limb extension						*	
hind limb extension						*	
righting reflex						***	

Statistically significant * p<0.05 **; p <0.01; *** p<0.001 (Chi-squared or Wilcoxon's test)

Table B.5.7.1-2 : Occurrence of fibre degeneration in proximal and distal sciatic nerves of rats dosed with alpha-cypermethrin (number affected animals/number examined)

Fibre degeneration in sciatic nerve			4 mg/kg bw/d		20 mg/kg bw/d		40 mg/kg bw/d	
	♂	♀	♂	♀	♂	♀	♂	♀
proximal :	0/5	0/5	0/5	0/5	5/5	4/5	5/5	0/5
distal:	0/5	0/5	0/5	0/5	2/5	0/5	3/5	0/5
proximal or distal:	0/5	0/5	0/5	0/5	5/5	4/5	5/5	0/5

Conclusion: At high doses alpha-cypermethrin is toxic for the CNS and peripheral motor nerves, but does not produce signs similar to the OP-induced delayed neuropathy. A single oral dose of 4 mg/kg bw (NOAEL) did not cause neurotoxicity. The NOAEL is 4 mg/kg bw.

Remark from BASF: Below is the supplemental study which was discussed in the DAR of alpha-cypermethrin (1999) in chapter 5.8.2.1. Based on placing this study in the list of endpoints in the EFSA conclusion (2004) under the point "Neurotoxicity study" it is shifted to this chapter with the hope to support clarity.

- Rat, oral administration by gavage:
-Phase I: 10 doses (37.5 mgtkg) + 10 doses (25 mg/kg), 4 weeks
-Phase II: 10, 20,40 mg/kg, 4 weeks (1983, AL-451-002)
(Dossier Cyanamid)

Guidelines: MOA-study, no guideline
GLP: No
Acceptance: The study was accepted

Phase I: 40 Wistar rats/sex/group received alpha-cypermethrin (b.n.°7; 96.6%) in DMSO by intubation. 1 dose per day, 5 day /week for 2weeks (37.5 rng/kg bw) and in arachis oil for 2 weeks (25 mg/kg bw). Control rats received DMSO for 2 weeks (l0 doses) fofollowed by a further 10 doses of arachis oil.
Phase II: 10 Wistar female rats /roup received 20 doses of alpha-cypermethrin (b.n.° 7; 96.6%) in DMSO by intubation over a 4 week period (5 dav/week) at 10, 20 or 40 mg/kg bw.

In Phase I over 80% of the treated animals ahowed some signs of intoxication . 21% of the treated animals died. Signs of intoxication were abnormal gait, ataxia, piloerecion, lethargy, chromodacryorrhoea, salivation and hypersensitivity to sensory stimuli. The glucuronidase and beta-galactosidase activities in the sciatic posterior tibial nerve (SPTN) were increased at 5.6 and 8 weeks after commencement of dosing, elevated maximal at week 5 and had recovered by the end of the 12 week study. No significant enzyme changes were found in the trigeminal ganglia and trigeminal nerve.
In Phase II signs of intoxication were similar to those reported in Phase I at 20 and 40 mg/kg bw. No signs of intoxication were at 10 mg/kg bw and enzyme activity was also not different from control. Dose dependent enzyme changes in the SPTN and in the trigeminal ganglia and nerve were seen in the 20 and 40 mg/kg bw group.
Conclusion: Biochemical changes consistent with sparse axonal degeneration were found in the SPTN of rats which had received 20 doses of alpha-cypermethrin at 40 mg/kg bw . At 20 mg/kg bw no consistent biochemical changes were found in the SPTN of animals. The enzyme changes in the trigeminal nerve and ganglia were smaller than in the SPTN, would therefore, not be expected to be related to any patholgical change.
The NOAEL = 10 mg/kg bw;

Additional data taken from the open literature:

Taken from the DAR of Zeta-cypermethrin:

Report: CA 5.7.1/1
Anonymous, 2006a
Draft Assessment Report (DAR) Zeta-Cypermethrin - Volume 1 to volume 3 - Initial risk assessment provided by the RMS Belgium of the third stage (part A) of the review programme referred to in article 8(2) of council directive 91/414/EEC
2006/7013990
Guidelines: none
GLP: no

Development neurotoxicity study of zetacypermethrin in rats [redacted] 2005)

Page 6-80 to -84

Remark: **This already peer-reviewed and on European level accepted study is considered accurate to address the request for DNT investigations with alpha-cypermethrin based on the bridging rationale presented in CA 5.00.** An additional conversion factor is not considered necessary. Based on the proven lactational transfer of zeta-cypermethrin within this study and via the study presented under CA 5.8.2/5 the demand for direct pup dosing with any cypermethrin in general is not justified according to the OECD Guideline for the Testing of Chemicals No. 426. In paragraph 21 it is stated that direct dosing of pups should be considered in those cases where there is a lack of evidence of continued exposure to offspring. Continuous exposure is definitely shown for zeta-cypermethrin, it is supported by data generated with beta-cypermethrin (see DAR beta-cyperemehrin, multigenerationstudy) and transfer into milk was shown for alpha-cypermethrin in goat and cow (see chapter CA 6.2.3) and is indicated in the DNT study with alpha-cypermethrin (see CA 5.7.1/3). Based on this study it is concluded that lactational exposure to zeta-cypermethrin does not occur at sufficient quantities to induce adverse effects in pups up to maternal toxic doses. Based on the bridging approach this is considered valid for alpha-cypermethrin as well.

Development neurotoxicity study of zetacypermethrin in rats [redacted] 2005)

Executive Summary

Neurotoxicity of zetacypermethrin in rats was evaluated in a developmental neurotoxicity study. 25 female rats per group were exposed to zetacypermethrin from gestation day 6 through lactation day 21 at dietary concentrations of 50, 125, 300 ppm. On PND 4, surplus pups were selected for culling and litters were standardized to 20 pups/litter.
No mortality and no abnormalities at necropsy were observed in the maternal rats and the offspring. Body weight, body weight gain, and food consumption were reduced in the F0 females of the 300 ppm group at various time points. Mean body weight for F1 male and female offspring in the 300 ppm group were lowered. No effects were seen on F1 age of attainment of balanopreputial separation or vaginal patency or on mean bw at the age of attainment. Occasional statistically significant differences were obtained in some offspring behavioral parameters and in one of 32 brain morphometric measurements, these apparent alterations did not occur to a degree or in a manner suggesting a compound-related pattern of neurotoxicity. In conclusion, a NOAEL for maternal and developmental toxicity was set at 125 ppm considering the decreased body weight, body weight changes and food consumption in dams and decreased body weight and body weight changes in litters.

I. MATERIAL AND METHODS

A. MATERIALS

1 Test Material:

Description:	*zetacypermethrin*
Lot/Batch no.:	*PL03-0427*
Purity:	*81.8%*
Stability:	*Stability and homogeneity were confirmed*

2 Dosing:

Dose levels:	*Range-finding study: 0, 50, 80, 300, 350 ppm (in diet)* *Main study: 50, 125, 300 ppm (in diet)*
Dosing period:	*GD 6 – LD 21*

3 Test animals

Species:	*Rat*
Strain:	*Crl:CD(SD) IGS BR*
Sex:	*range-finding study: Female (15/group)* *range-finding study: Female (25/group)*

B. STUDY DESIGN AND METHODS

Range-finding study:
10 females rats per group received zeta-cypermethrin at dietary concentrations of 0, 50, 80, 300, 350 ppm on gestation day 6 through lactation day 21. On PND 4, surplus pups were selected for culling and litters were standardized to 8 pups/litter. All F1 pups were observed daily and motor activity was assessed for 1 pup/sex/litter on PND 21. Milk samples were collected on LD 4, 11 and 17/18 and the same dams were used for blood collection on LD 5 and 21. The culled pups from the same five dams were utilized for blood collection on PND 5. Quantification of zetacypermthrin in plasma was performed with GC/μECD. The remaining F0 females were euthanized and necropsied on LD 21. A storage stability study has shown that cypermethrin was stable in milk and liver for at least 3 months under frozen conditions. Rat milk and plasma samples were analyzed within 30 days of receipt.

25 female rats per group were exposed to zetacypermethrin from gestation day 6 through lactation day 21 at dietary concentrations of 50, 125, 300 ppm. On PND 4, surplus pups were selected for culling and litters were standardized to 20 pups/litter. Homogeneity and stability of zetacypermethrin in diet was demonstrated. The study was perfomed under GLP (no attest of competent authority) and in compliance with OECD guideline 426 (adopted 2003).

II. RESULTS AND DISCUSSION

A MATERNAL OBSERVATIONS

Range-finding study:

No clinical signs were observed at any dose. Lower mean lactation bw and bw gain was observed at 300 and 350 ppm.
Body weight: in the 350 ppm group were slightly reduced throughout gestation period, without affecting statistically the mean gestation body weight.
Food consumption was decreased at 350 ppm during gestation. Lower mean food consumption was observed during lactation at 300 and 350 ppm.
No compound-related effects were seen on mean number of pups born, former implantation sites and unaccounted sites. Mean live F1 litter size, % male at birth, postnatal survival and general physical condition of pups were unaffected.
Main study:

Mortality: all F0 treated females survived until the scheduled necropsies.
Body weight and body weight gain in F0 females were lowered at 300 ppm beginning late in gestation and continuing through lactation day 21.
Food consumption: was reduced for F0 females at 300 ppm during the first three days of treatment and throughout lactation period.
Necropsy: no abnormalities were seen in F0 females.

B LITTER DATA

Range-finding study:

Mean male and female pup body weight gains in the 350 ppm group were lower during preweaning, and post weaning.
There was a decreasing trend in motor activity patterns (total activity and/or ambulatory activity counts) in all exposed males and females on PND 21.

Main study:
No compound-related effects were seen on mean number of offspring born, live litter size or % males /litter at birth. Offspring survival throughout the postnatal period was unaffected by the compound. Mean body weight for F1 male and female offspring in the 300 ppm group were lowered. No effects were seen on F1 age of attainment of balanopreputial separation or vaginal patency or on mean bw at the age of attainment. No effects were seen at necropsy.

FOB:
No consistent dose-related trends were noted when components of the FOB were evaluated on PND 4, 11, 22, 45 and 60. Forelimb grip strength value for males at 300 ppm was lower on PND 22. They were however within the historical control data of the company (174.8±35; 146.8±30.86; 109±35, control; 300 ppm and historical control respectively). Such an observation was not seen at subsequent intervals and there were no indications of neuromuscular deficits in other aspects of FOB assessments or in other behavioral tests. A higher number of females from the 50 ppm group were observed with eyelids wide open on PND 22, and lower urination counts were noted for males from the 50 and 125 ppm groups on PND 45. These isolated findings were not considered treatment related as no effects were recorded at top dose.

Motor activity:
On PND 17, significant treatment group by time interactions were found for both ambulatory and total activity counts in animals of both sexes. In males at 300 ppm, significantly higher levels compared to control values were noted only during the fourth session interval (45-60 minutes) in both measures of activity. For females on PND 17, significantly lower levels of activity were noted during the first interval (0-15 min) in rats of the 300 ppm group and during the second interval (16-30 min) in offffsprings from 125 and 300 ppm. Statistically significantly mean overall lower ambulatory and total activity counts were noted in females from the 300 ppm group on PND21. Apparent decrease in motor activity in females on PND 21 were considered more likely due to a slight shift in the timing on the normal inverted U-shaped developmental activity pattern, than to the zetacypermethrin effect of maternal and/or direct offspring consumption during pre-weaning period.

On PND 61, there were no changes in ambulatory or total activity counts that were considered biologically meaningful. No statistically significantly effects were seen in females. In males, an overall higher interval mean value was noted in total session counts (13% > control) and a significant treatment group by time interaction occurred in ambulatory counts for males at top dose (15% > control). These slight changes were not considered to be biologically significant.
Acoustic startle response was performed on PND 20 and 60. Vmax was not altered but on PND 20, the overall Tmax was longer for females at 300 ppm (26.5±3.36 ms compared to control 24.9±3.24 and historical data of 22.3-27.3 mean of 24.2±1.44 ms). This isolated finding was not considered biologically meaningful.
Neuropathology: no gross findings were seen in brain or spinal cord on PND 21.
Brain weight/brain measurements: no compound related findings were seen.
Histopathology: a statistically significant increase in the mean height of the vertical thickness of the cortex (5.6%) in the 300 ppm females was not accompanied by any alteration in cortical height nor was there any changes in cortical thickness or height, or any other morphometric measurements for either male or females at PND 72.

C ANALYSIS OF ZETACYPERMETHRIN IN MILK

Range-finding study:

Zetacypermethrin was present in dam milk and plasma, increasing with increasing dose level. Litter plasma indicated similar dose proportionality on PND 5 and PND 21 and levels were similar to those observed in dams. Milk concentration of zetacypermethrin was proportional to administered dose. The presence of zetacypermethrin in maternal milk indicates that zetacypermethrin is transferred from maternal vasculature into milk, and thus, milk is a route of excretion. Milk concentrations increased from lactation day 4 to 11 and remained relatively constant from day 11 to day 17.

Table 5.7.1-1: Range-finding study dietary administration of zetacypermethrin from GD 6 to LD 21.

Endpoints/dose	***0***	***50 ppm***	***80 ppm***	***300 ppm***	***350 ppm***
Compound ingestion *during gestation Day 6-20 (mg/kg bw/d)*	*0*	*3.5*	*5.5*	*20.8*	*24.1*
Compound ingestion during lactation Day 1-21 (mg/kg bw/d)	*0*	*8.2*	*13.3*	*47.6*	*54.6*
No. females on study	*10*	*10*	*10*	*10*	*10*
No. gravid					*10*
Bw *day 20 gestation (g)*					*(↓4%)*
Bw lactation day 11				*(↓6%)*	*↓8%*
Bw lactation day 14				*(↓6%)*	*(↓10%)*
Bw lactation day 17				*(↓5%)*	*(↓6%)*

Endpoints/dose	***0***		***50 ppm***		***80 ppm***		***300 ppm***	***350 ppm***
Bw changes *during GD 0-20*							*(↓2%)*	*(↓3%)*
GD 6-9							*(↓23%)*	*(↓46%)*
LD 1-21	*42*		*32*		*39*		*27*	*35*
Food consumption *during gestation (g/animal/day) Day 6-9*							*(↓10%)*	*(↓13%)*
Food consumption during lactation (g/animal/day) Day 1-21							*↓11%*	*↓16%*
Implantation sites mean	*15.6*		*16.6*		*15.1*		*15.7*	*15.1*
No. born	*15.1*		*16.2*		*14.0*		*15.2*	*14.5*
Unaccounted sites	*0.4*		*0.4*		*1.1*		*0.5*	*0.6*
Live litter size PND 0	*15.1*		*16.0*		*14.0*		*15.1*	*14.4*
Litter survival % PND 7-14	*100*		*100*		*100*		*100*	*100*
Litter weight changes (g)	*m*	*f*	*m*	*f*	*m*	*f*	*m*	*f*
PND 7-11								*↓20%*
PND 11-14							*↓24%*	
PND 21-22					*↓83%*	*↓87%*	*↓93%*	*↓92%*
Litter weight (g)								
PND 22							*↓19%*	*↓18%*
PND 23							*↓20%*	*↓17%*
PND 24							*↓16%*	*↓14%*
PND 25							*↓16%*	*↓13%*
Zetacypermethrin level (ppm)								
Milk:								
Day 4			*0.37-1.09*		*1.01-3*		*2.5-8.97*	*6.19-17.7*
Day 11			*0.77-1.12*		*0.44-2.06*		*3.37-5.02*	*3.37-9.27*
Day 17			*0.29-0.84*		*0.90-3.87*		*7.44-19.5*	*3.55-14.5*
Plasma:								
Dam *LD 5*			*0.11-0.29*		*0.19-0.29*		*0.51-1.10*	*0.67-1.17*
LD 21			*0.11-0.27*		*0.06-0.56*		*0.15-1.86*	*0.52-1.58*
Pup *LD 5*			*0.09-0.16*		*0.12-0.18*		*0.26-0.45*	*0.32-0.52*
LD 21			*0.08-0.27*		*0.10-0.45*		*0.40-2.65*	*0.23-1.14*

Table 5.7.1-2: Dietary administration of zetacypermethrin from GD 6 to LD 21, main study.

Endpoints/dose	***0***		***50 ppm***		***125 ppm***		***300 ppm***	
Compound ingestion *during gestation Day 6-20 (mg/kg bw/d)*			*3.6*		*9*		*21.1*	
Compound ingestion during lactation Day 1-21 (mg/kg bw/d)			*8.7*		*21.4*		*48.7*	
No. females on study	*25*		*25*		*25*		*25*	
No. gravid	*25*		*25*		*24*		*25*	
Females with viable pups	*24*		*25*		*23*		*25*	
Bw *day 20 gestation (g)*							*(↓4%)*	
Bw lactation day 1, 4, 7, 11, 14, 17, 21							*↓4-6%*	
Bw changes *during GD 6-20*							*(↓10%)*	
Bw changes *during GD 0-20*							*↓11%*	
Food consumption *during gestation (g/animal/day) Day 6-20*							*↓4%*	
Food consumption *during lactation (g/animal/day) Day 4-21*							*↓9-13%*	
Litter data:								
Body weight:	*m*	*f*	*m*	*f*	*m*	*f*	*m*	*f*
PND 13							*(↓5%)*	*↓8%*
PND 17							*(↓6%)*	*↓10%*
PND 21							*(↓7%)*	*↓10%*
Body weight changes								
PND 11-13							*↓13%*	*↓14%*
PND 13-17							*(↓10%)*	*↓14%*
PND 4-21							*(↓7%)*	*↓12%*
FOB: *forelimb gripstrength PND 22*							*↓*	
Motor activity:								
Ambulatory motor activity counts								
PND 17 time interval							*↑46-60 min*	*↓0-15 min*
PND 21 time intervall								*↓overall*
Total motor activity counts								
PND 17 time interval					*↓16-30 min*		*↑46-60 min*	*↓0-15 min* *↓16-30 min*
PND 21 time intervall								*↓overall*

Endpoints/dose	***0***		***50 ppm***		***125 ppm***		***300 ppm***	
Acoustic startle response								
PND 20								*↑time*
Eyelids *wide open PND 22*	*15*	*13*	*17*	*19**	*14*	*15*	*12*	*17*
Urination *counts PND 45*	*0.6*		*0.1**		*0.1**		*0.2*	
mean height vertical ***thickness cerebral cortex*** *PND 21 level 2*								*↑6%*

**↑↓ Statistically significantly different from control*

III. CONCLUSION

Range-finding study:

The analytical determinations of zetacypermethrin concentrations in milk samples of dams exposed to diets containing 50, 80, 300 and 350 ppm and in plasma samples from pups confirmed that pups were adequately exposed to the compound during lactation. Based on lower F0 body weights and/or food consumption during gestation and/or lactation in the 300 and 350 ppm groups, lower mean F1 male and female pup weights in the 300 and 350 ppm groups, and a decreasing trend in motor activity at all dose levels for males and at 300 and 350 ppm for female pups, dietary concentrations of 50, 125 and 300 ppm zetacypermethrin were selected for the main study.
Although occasional statistically significant differences were obtained in some offspring behavioral parameters and in one of 32 brain morphometric measurements, these apparent alterations did not occur to a degree or in a manner suggesting a compound-related pattern of neurotoxicity. According to the applicant, the results of the behavioral testing battery and the neuropathology assessments suggested no CNS domain-, age- or dietary-related adverse pattern of developmental neurotoxicity in the offspring.

The RMS agrees with the setting of NOAEL maternal and developmental toxicity at 125 ppm and consideres that the few measures affected at top dose in conjunction with other signs of toxicity including systemic toxicity, such as decreased body weight and body weight changes are not persuasive evidence of a direct neurotoxic effect.
NOAEL maternal and developmental toxicity = 125 ppm (9mg/kg bw/d) based on the decreased body weight, body weight changes and food consumption in dams and decreased body weight and body weight changes in litters.

Additional data taken from the open literature:

Taken from the DAR of beta-cypermethrin

Report: CA 5.7.1/2
Anonymous, 2013a
Draft Assessment Report - Beta-Cypermethrin - Volume 3, Annex B.6: Toxicology and metabolism
2013/1418020
Guidelines: none
GLP: no

Report: -456003
A dose range-finding oral toxicity study of beta-cypermethrin in rat pups (listed in the DAR of beta-cypermethrin under Annex B.6.7 c

Report -456005
An oral (gavage) developmental neurotoxicity study of beta-cypermethrin in rats (listed in the DAR of beta-cypermethrin under Annex B.6.7 d)

Remark: In bilateral discussions with the Rapporteur Belgium it was agreed to perform a targeted study with alpha-cypermethrin to address the critical endpoints found for beta-cypermethrin and thereby address the concerns of the rapporteur. At this point it is referred to the range finder and the main DNT study in the DAR from Beta-cypermethrin, the executive summary as given in the DAR is presented, and the relevant endpoints found in the beta-cypermethrin study are summarized in table form to specify the critical endpoints to be addressed in the screening DNT study with alpha-cypermethrin. The study with alpha-cypermethrin is presented under CA 5.7.1/3.

Report -456003*: A dose range-finding oral toxicity study of beta-cypermethrin in rat pups (listed in the DAR of beta-cypermethrin under Annex B.6.7 c*

Executive summary

Beta-cypermethrin was administered by oral gavage to CRl:CD(SD)BR strain pre-weanling, rat pups from postnatal day (PND) 10-16. Initially (Phase I)doses of 0,3, 12, and 30 mg/kg bw/day were administered to 10 pups/sex/group, however following mortalities at the top two doses this phase of the experiment was terminated. In Phase II, new pups were assigned to treatment groups and dosed from PND 10 to 16.
Treatment related mortalities occurred between PND10-13 and were observed in pups at doses ≥4 mg/kg bw/day. Of the 4 males receiving 12 mg/kg bw/day, thee were found dead on PND11 and the remaining male was euthanized and discarded without examination on day 11.

Report -456005: ***An oral (gavage) developmental neurotoxicity study of beta-cypermethrin in rats (listed in the DAR of beta-cypermethrin under Annex B.6.7 d)***

Executive Summary

The developmental neurotoxicity of beta-cypermethrin in Crl: (SD)Br strain rats was investigated in two separate phases. In Phase I, 25 dams/dose were dosed by oral gavage from gestation day (GD) 6 through to PND 20 with 3, 12, 30 mg/kg bw beta-cypermethrin in aqueous CMC. Pups of this group were potentially exposed in utero and through nursing during lactation. In Phase II, 25 dams/dose were dosed by oral gavage from GD 6 through to PND 9 with 0.5 mg/kg bw. Pups in Phase II were potentially exposed in utero and through nursing until day 9 and were then administered 0.5 mg/kg bw/day by oral gavage from PND 10-16. No mortality and no abnormalities at necropsy were observed in the maternal rats. Dams of the 30 mg/kg bw dose group showed treatment related effects including clinical signs of toxicity and reduction in body weight gain during lacation at doses >= 12 mg/kg bw. Treatment related effects in pups of Phase I were limited to increased motor activity at 30 mg/kg bw/day on day 13 after lactational exposure. Pups of Phase II (direct gavage group) produced clinical signs of toxicity, decreases in body weight during direct treatment, clonic convulsions, altered grooming and deaths.

Determination of endpoints considered relevant to be addressed within a targeted screening DNT study with alpha-cypermethrin

The DNT study with beta-cypermethrin has induced a limited set of target parameters which are characteristic for acute neurotoxicity. A summary of effects is listed in the following table.

Table 5.7.1-3: Summary of effects found in the DNT study performed with beta-cypermethrin as taken from the respective DAR

Substance	Beta-cypermethrin (CA 5.7.1/2)	
Strain of rats	SD rats	
Exposure of dams administration route vehicle dose [mg/kg bw/day] Period of exposure	 gavaged aqueous CMC 0-3-12-30 GD6-PND20	 gavaged aqueous CMC 0-0.5 GD6-PND9
Exposure of pups administration route vehicle dose [mg/kg bw/day] Period of exposure	No direct exposure but via milk	 gavage aqueous CMC 0-0.5 PND10-16
Mortality	-	yes (see Range finder in not-pre-exposed pups at a dose ≥ 4mg/kg bw)
Detailed clinical observations n pups	No clinical signs	PND10-16: tremors, clonic convulsions, carangiform ataxia, straub tail
Litter weight	-	PND 10-17:↓ **BW 93%,** PND 10-17:↓ **BWG 91%**

Substance	Beta-cypermethrin (CA 5.7.1/2)	
FOB (PND 4, 11, 21, 35, 45, 60)	-	↑ grooming counts (PND11)
Total motor activity counts:	30 mg/kg bw: PND13: trend for ↑MA PND 21: - PND 61:-	PND13: trend for ↓MA ♂/ ↑MA ♀ PND 21: ↑ MA ♂
Sensory function - Acoustic startle response (PND 20 & 60)	-	↓ startle response (PND 20 & PND 60)
Biel maze test (PND 22 & 62)	-	↓ mean time and error ♀ (PND 62)*
Neuropathology (PND21 & 72)	-	-
Brain weight and dimension (PND21/72)	-	-
Histopathology CNS / Peripheral NS (PND 21/72)	-	-

-: parameter was not affected in the study;
*: a reduction of mean time and error in the biel maze test is not considered adverse but reflects normal variability of this parameter.

Beta-cypermethrin induced mainly acute neurotoxic effects in the direct dosing phase and on PND21 combined with some changes of body weight / body weight development. Effects seen at a later stage around PND60 were effects of questionable relevance as for example the effects seen in the Biel maze test which indicated reduced time and error in females (not considered adverse), or reduced startle response, which was also seen on PND20. Furthermore, no macroscopic or microscopic changes of the central or peripheral nervous system or changes of the brain weight and dimension were induced.

Based on the request of the rapporteur, BASF agreed to perform a targeted developmental neurotoxicity study copying the study design of beta-cypermethrin with the exception to go not over the period PND21 as the data suggest main . In the following the data on alpha-cypermethrin are presented.

Report:	CA 5.7.1/3 [redacted] ~~2014~~ 2015 a BAS 310 I (Alphacypermethrin) - Developmental neurotoxicity screening study in Wistar rats - Oral administration of the dams and pups (gavage) 2015/1001621
Guidelines:	~~OPPTS~~ EPA 870.6300, OECD 426
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Remark (Jan-2015): ~~The study is not finalized yet and data on stability, homogeneity and concentration control, pathological examinations of animals and neuro-histopathology are missing. The data were summarized by the author of the dossier to the best of knowledge from the raw data. Due to the change of dose regimen within the dose groups the values were manually split to present the data of Cohort 1 and Cohort 2-4 independently. Statistic was performed. The data are not QAU-checked and might change slightly in the QAU-checked final report.~~

Update (Nov- 2015): The update adds data on stability, homogeneity and concentration control, pathological examinations of dams and pups and examination of pups brain (weight, length and width). Furthermore, in a separate study (Doc ID 2015/1175543, included in the dossier in Chapter CA 4 /Analytical methods) data on the alphacypermethrin content in the pups, separated into brain and carcass, are presented. The update is highlighted in yellow.

Executive Summary

The aim of this study was to investigate the clinical effect of BAS 310 I (alpha-cypermethrin) on the embryonic, fetal and pre-weaning development of the nervous system. For this purpose, 25 pregnant female Wistar rats were treated with BAS 310 I by oral gavage in aqueous CMC from gestation day (GD) 6 through postnatal day (PND) 8 and their offspring from PND10 to PND 21. The study was splitted in two phases: Cohort 1 dams and pups were administered 0.5, 2, or 5 mg/kg bw/day. Because of excessive toxicity and lethality in the high-dose pups (test group 3: 5mg/kg bw/d) after direct dosing, the high dose test group was terminated. As the directly dosed mid- and low-dosed pups still exhibited adverse clinical symptoms in a dose-dependent fashion, the dose in cohort 2-4 pups was reduced to a constant dose of 0.25 mg/kg bw, irrespective of the pre-treatment of dams at 0.5 or 2 mg/kg bw/day. Furthermore, the body burden of PND4 and PND10 pups prior to direct treatment was determined.

Neurotoxicity was neither induced in dams nor in pups prior to the onset of direct pup dosing. This is in line with the analytical determination of alpha-cypermethrin in PND 4 and 10 pups, which revealed that alphacypermethrin is mainly deposed in the carcass of the pups, but does not reach the brain in relevant amounts.
Acute treatment-related mortality was induced by direct pup dosing at 2 and 5 mg/kg bw/day starting on PND10 while signs of neurotoxicity were observed down to 0.5 mg/kg bw/day. A 50% reduction of the pup dose to 0.25 mg/kg bw/day - while remaining the pre-treatment of dams until PND 8 at 0.5 and 2 mg/kg bw/day - resulted in similar signs of neurotoxicity, however reduced in incidence and severity in dependence of the maternal pretreatment.

Apart from the clinical symptoms, pup development was rather unaffected in the surviving animals in all test groups as shown by unchanged pup weights/weight gain and absence of necropsy findings. Motor activity measurements did not reveal any significant treatment-related changes. Regarding neuropathology, no treatment-related findings in brain weights, gross pathology and length and width measurements of the brain were found in any of the groups.

The analytical investigations of PND 10 pups revealed a body burden of 7 µg/kg bw in low dose pups. This internal body burden, corrected for bioavailability of 46%, together with the external dose of 250 µg/kg bw can be considered as **calculated LOAEL at 265 µg/kg bw/day**. Considering the internal dose of 101 µg/kg bw at PND4 as systemic NOAEL, and corrected for bioavailability of 46% ends up in a **calculated NOAEL of 220 µg/kg bw**.

Altogether, the data show that there occurs exposure via milk which still leads to measureable amounts of alphacypermethrin after a two-days treatment-free interval. However, direct pup dosing is an extreme method for pre-weanling pups, and of no direct relevance to the overall human or ecotoxicological risk assessment. Therefore, as discussed in the "Reasoning for the appropriateness of dietary DNT studies for alpha-cypermethrin" (see above) this study is not used for the human risk assessment but the dietary developmental neurotoxicity study with zeta-cypermethrin is considered adequate.

(DocID 2015/1001621)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I (Alphacypermethrin)
Test substance:	01/0265-5
Description:	solid / white
Lot/Batch #:	PMAM000622
Purity:	99.2%
Stability of test compound:	The test substance was stable under storage conditions over the test period (expiry date 12 Dec 2015)
2. Vehicle:	CMC (1%)
3. Test animals:	
Species:	Rat (animals time-mated by the breeder)
Strain:	Crl:WI(Han)
Sex:	Female
Age:	About 10-12 weeks (at arrival)
Weight at dosing (range of means):	females: 200.3 – 204.9
Source:	Charles River Laboratories, Research Models and Services, Germany GmbH
Acclimation period:	6 days
Diet:	Ground Kliba maintenance diet mouse/rat "GLP", ad libitum

Water:	water, ad libitum
Housing:	single housing in polycarbonate cages type III (exception: during rearing up to weaning 1 dam with her litter)
Environmental conditions:	
Temperature:	20 - 24°C
Humidity:	30 - 70%
Air changes:	15 per hour
Photo period:	12 h light / 12 h dark (06:00 - 18:00 / 18:00 - 06:00)

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: 08-Sep-2014 – 22 Sep 2015
(In life dates: 14-Sep-2014 (start of administration) to 02-Dec-2014 (necropsy)

Analytical report Doc ID 2015/1175543:
Dates of experimental work: 21-Jul-2015 - 11-Sep-2015

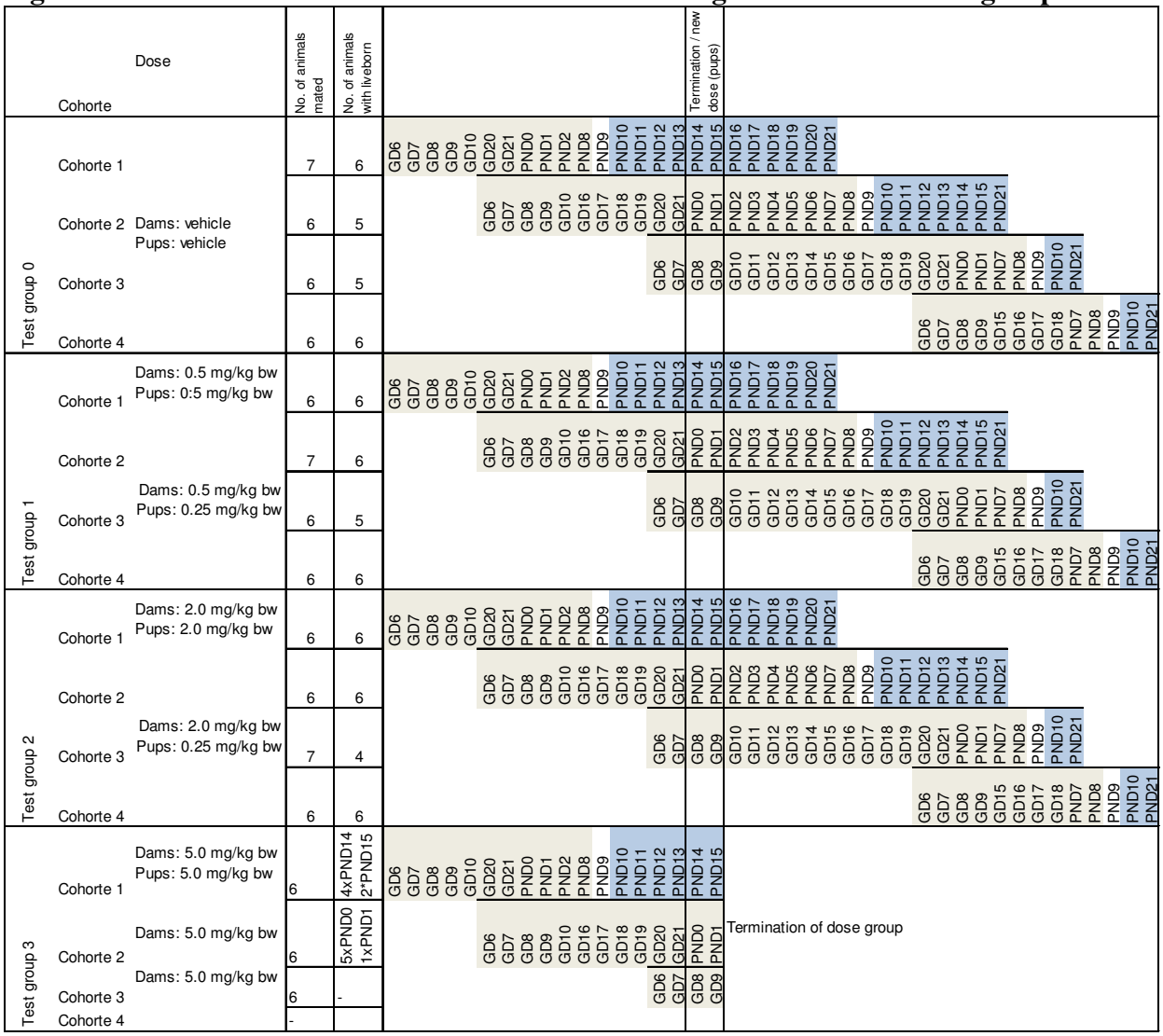
2. Animal assignment and treatment:
The animals paired by the breeder (time-mated animals) were supplied at noon on the day of evidence of mating; this day is referred to as GD 0 and the following day as GD 1. The dams were allowed to deliver and rear their pups until postnatal days (PND) 4 or 22. BAS 310 I was administered by oral gavage to groups of 25 dams at concentrations of 0.5, 2, and 5 mg/kg bw/day from GD 6 to PND 8. The administration volume was 2 mL/kg bw. Animals during parturition were not treated. Thereafter, pups of Cohort 1 were dosed by oral gavage with 0.5, 2, and 5 mg/kg bw from PND 10 onwards. Based on the severe clinical findings in the pups of dose group 3 (5 mg/kg bw/day) all group 3 animals were killed for humane reasons. Furthermore the dose in group 1 and 2 was reduced in a way, that the pups of cohorts 2, 3 and 4 were dosed equally with 0.25 mg/kg bw.

Table 5.7.1-4: Overview on dam assignment to cohorts and applied doses

Cohort 1					
	Group	**0**	**1**	**2**	**3**
	Dose dams	**0 mg/kg**	**0.5 mg/kg**	**2 mg/kg**	**5 mg/kg**
	Dose pups	**0 mg/kg**	**0.5 mg/kg**	**2 mg/kg**	**5 mg/kg**
	Cohort 1 dams	1-7	26-31	51-56	76-81
Cohort 2-4					
	Group	**0**	**1**	**2**	**-**
	Dose dams	**0 mg/kg**	**0.5 mg/kg**	**2 mg/kg**	**-**
	Dose pups	**0 mg/kg**	**0.25 mg/kg**	**0.25 mg/kg**	**-**
	Cohort 2 dams	8-13	32-38	57-62	82-87
	Cohort 3 dams	14-19	39-44	63-69	88-93
	Cohort 4 dams	20-25	45-50	70-75	-

Figure 5.7.1-1 shows the chronological order of the Cohort 1-4 treatment. The time point of termination of group 3 is marked and illustrates that PND14 and PND15 as well as PND0 and PND1 pups and dams of GD8 and 9 were affected by the test group 3 termination.

Figure 5.7.1-1: Schematic illustration of animal assignment to treatment groups



3. Test substance preparation and analysis:
For the test substance preparations in 1% Carboxymethylcellulose suspension in drinking water, the analytical results of the stability verification were taken into account. The specific amount of test substance was weighed, topped up with 1% Carboxymethylcellulose (CMC) suspension in drinking water in a calibrated beaker and intensely mixed with a homogenizer. During administration, the preparations were kept homogeneous with a magnetic stirrer.
The stability of the test substance in 1% CMC suspension in drinking water at room temperature over a period of 7 days had been verified prior to the start of the study (Project No.: 01Y0265/01Y014) for the concentration 0.025 g/100 mL and after study commencement for the reduced concentration of 0.0125 g/100 mL (Project No.: 01Y0265/01Y016). From the low- and high-dose formulations each, 3 samples were withdrawn from the top, middle and bottom of the preparation vessel for homogeneity and concentration control analyses. In addition, three samples each were withdrawn from the adapted dosing formulations for pups (test group 1 and 2).

4. Statistics:
Means and standard deviations (S.D.) of each test group were calculated for several parameters whenever possible. The following statistical analyses were carried out or will be carried out in the final:

Parameter	Statistical test
Food consumption (parental animals), body weight and body weight change (parental animals and pups; for the pup weights, the litter means were used), duration of gestation, number of pups delivered per litter,	DUNNETT test (two-sided)
Different indices (e.g. mating, fertility, gestation index), Females with liveborn, stillborn, all stillborn pups; Number of live, stillborn, dead pups, pups cannibalized, sacrificed moribund and different indices (e.g. viability index, lactation index) and number of litters with necropsy findings in pups	FISHER´s exact test
Proportion of pups with necropsy findings per litter	WILCOXON test (one-sided)
Motor activity, brain weights	KRUSKAL-WALLIS test (two-sided) and WILCOXON test (two-sided)
Brain measurements length, width, separately for cohort 1 and cohort 2-4	WILCOXON-test (one-sided-) with Bonferroni-Holm-Adjustment

C. METHODS

1. Observations:

Clinical examinations of dams

During the study, all animals were observed for any clinically abnormal signs. A check for moribund and dead animals was made twice daily from Mondays to Fridays and once daily on weekends and public holidays. A cage side examination was performed at least once daily for any signs of morbidity, pertinent behavioral changes and/or signs of overt toxicity.
The parturition and lactation behavior was generally evaluated in the morning in combination with the daily clinical inspection of the dams.

Clinical examinations of pups

Status (sex, liveborn or stillborn) and number of all pups delivered were determined as soon as possible after birth. At the same time, the pups were also examined for gross-morphological changes.
Regarding viability/mortality a check was made for any dead or moribund pups twice daily on workdays or once daily on weekends or public holidays.
All live pups were examined daily for clinical symptoms (including gross-morphological findings) during the clinical inspection of the dams.

Detailed clinical examinations of pups

A detailed clinical observation (DCO) was performed in all pups on PND 4 after standardization and thereafter on PND 10, 12, 14, 16, 18 and 20. For observation, the animals were removed from their cages and placed in a standard arena (50x37.5 cm with a lateral border of 25 cm) for at least 15 seconds per animal. The examination was carried out about 3-4 hours after test substance administration based on the time of appearance of clinical findings during the daily clinical observations. The scope of examinations and the scoring of the findings that were observed were based on the current index of findings in GROSSE-Reprotox software and includes but is not limited to the following parameters listed:
Abnormal behavior in handling, fur, skin, posture, salivation, respiration, activity/arousal level, tremors, convulsions, abnormal movements, gait abnormalities, lacrimation, palpebral closure, exophthalmos, assessment of the urine discharged during the examination, pupil size.

Motor activity measurement of pups

10 males and 10 females per group were selected. The motor activity (MA) was carried out on PND 17 and 21 (in a randomized sequence on each examination day). For this purpose, the animals were placed in polycarbonate cages type III for the time of measurement. The examinations were performed using the TSE Labmaster System supplied by TSE Systems GmbH, Bad Homburg, Germany. Eighteen beams are allocated per cage. The number of beam interrupts was counted over 12 intervals of 5 min each. The sequence at which the animals were placed in the cages was at random. The animals were given no food or water during the measurements.

2. Body weight:

Body weight of the parental females was determined on GD 0, 6, 8, 10, 13, 15, 17, 19, and 20. Females with litter were weighed on the day of parturition (PND 0) and on PND 4, 7, 9, 14, and 21.
The pups were weighed on the day after birth (PND 1) and on PND 4, 7, 10, 11, 13, 15, 17, 19, and 21.

3. Food consumption:

Food consumption of the dams was determined for GD 0-6, 6-13, and 13-20. Food consumption of the females, which gave birth to a litter, will be determined for PND 0-4, 4-7, 7-14 and 14-21.

4. Pathology and histopathology:

Necropsy

On PND 22, the dams were anesthetized with isoflurane and decapitated and assessed by gross pathology. Pups with scheduled sacrifice were anesthetized with isoflurane and sacrificed with CO_2, except for those 10 male and female pups necropsied with regard to the question of neuropathology, which were subjected to deep anesthesia with isoflurane and sacrified by perfusion fixation. SOERENSEN phosphate buffer was used as rinsing solution, and neutrally buffered 4% formaldehyde solution was used as a fixative.
Afterwards, the organs were assessed macroscopically, and in case of notable findings, were evaluated on a case-by-case basis, depending on the type of finding noted.

All stillborn pups and all pups that died before weaning were examined externally, eviscerated and their organs were assessed macroscopically.

The perfusion fixed pups were necropsied with regard to the question of neuropathology, and the visible organs were assessed by gross pathology as accurately as possible after a perfusion fixation. The cranial vault and the spinal cord were opened and the skin was removed from both hind extremities. In this state, the perfused animals were stored in neutrally buffered 4% formaldehyde solution for at least 48 h.

Pup brain weights, length and width

The terminal brain weight (including olfactory bulb) was determined after its removal but before further preparation. The terminal body weights were recorded to calculate the relative organ weights. The length and maximum width of the brain was measured in all animals (length: on a line extending from the rostral end of the frontal lobe to the caudal medulla oblongata of the cerebellum; width: pituitary region).

Pup organ/tissue fixation

The following organs/tissue specimens were carefully removed and processed histotechnically:

1. All gross lesions
2. Brain with olfactory bulb
3. Eyes with retina and optic nerve
4. Nose (nasal cavity)
5. Pituitary gland
6. Trigeminal ganglia (s. Gasserian)

The animals and the tissues or organ material remaining after trimming were stored in neutrally buffered 4% formaldehyde solution.

Neurohistopathology

The histotechnical processing of brain (olfactory bulb, prosencephalon with frontal lobe, diencephalon with parietal lobe, mesencephalon with occipital lobe and temporal lobe, pons, cerebellum and medulla oblongata) and brain-associated organs/tissues (eyes with retina and optic nerve), pituitary gland, trigeminus ganglia with part of nerve and nose were prepared in all animals per sex and group and gross lesions were prepared in all affected animals.

5. Blood/tissue sampling/

As many male and female surplus pups per group as available on PND4 and each 10 male and female pups on PND10 before treatment were sacrificed after isoflurane anesthesia with CO2, shock-frozen on dry-ice and stored at -80°C. From the thus collected frozen pups the brains were dissected in toto and brains as well as bodies were shipped frozen to PTRL Europe GmbH (Helmholtzstr. 22. Science Park I, 89081 Ulm, Germany) for the concentration investigation of the test substance.

These analyses were conducted as a separate GLP-study (PTRL Europe Study ID P3731G, BASF Doc ID 2015/1175543) but the results are reported here:

Analytical determination of alpha-cypermethrin in pup brain and body sample

The analytical determination of the concentrations of alpha-cypermethrin (Cis II) and possibly its isomers Cis I, Trans III and Trans IV in the pups brain and body samples was performed at PTRL Europe. The procedure was developed and validated at PTRL Europe based on QuEChERS extraction with subsequent clean-up and LC/MS/MS. All body samples were homogenized frozen using a knife mill. Brain samples from PND10 were homogenized frozen by hammering the respective samples in a plastic bag. Brain samples from PND 4 were homogenized frozen by pestling the tissue samples with a spatula.
The method used for analyzing the test material in the samples involved extraction and fortification with a solvent followed by LC/MS/MS analysis with an external standard.

II. RESULTS AND DISCUSSION

A. TEST SUBSTANCE ANALYSES

Based on analytical results the stability of BAS 310 I in 1% CMC in drinking water over a period of 7 days at room temperature is given at a concentration of 0.2 mg/ml and for the reduced concentration of 0.125 g/L. All determined concentrations were in the range of 90 % - 110 % of the nominal concentration.

Concentration control analysis of all dose levels were determined at the beginning of the dam administration period and at the beginning of the F1 pup dosing.
The homogeneity of the low and high dose as well as of the dose used for F1 pup dosing (Cohorte 2-4) was determined with the samples used for concentration control analysis. Three samples (one from the top, middle and bottom in each case) were taken from the beaker with a magnetic stirrer running. Additional reserve samples were retained.

Table 5.7.1-5: Analysis of test-item concentration and homogeneity

BAS 310 I nominal conc. [g/100 mL]	Date of sampling	Date of analysis	Sample [#]	Analytical concentration [g/100mL]	% of nominal concentration	Mean ± RSD
0	14-Sep-2014	21-Oct-2014	2	0		
0,025	14-Sep-2014	21-Oct-2014	3	0,022	88,3	85,9 ± 3,4
			4	0,021	82,6	
			5	0,022	86,8	
0,10	14-Sep-2014	21-Oct-2014	6	0,092	92,5	
0,25	14-Sep-2014	21-Oct-2014	7	0,226	90,3	89,4 ± 0,9
			8	0,223	89,1	
			9	0,222	88,9	
0	23-Oct-2014	10-Dec-2014	11	0		
0,0125	23-Oct-2014	10-Dec-2014	12	0,012	95,2	87,2 ± 13,9
			13	0,01	73,2	
			14	0,012	93,1	
0,025	17-Nov-2014	10-Dec-2014	17R	0,021	83,8	
0,10	17-Nov-2014	10-Dec-2014	18R	0,075	74,7	
0,0125	02-Dec-2014	10-Dec-2014	19	0,012	93,5	

No test substance was detected in the vehicle control samples (samples 2 and 11).
The mean values of the test-item concentrations in the homogeneity samples were in the range of 85.9 – 89.4% of the target nominal concentrations and thus within the acceptable range of ± 15% of the target concentration for liquid suspensions.
The concentration in the single samples (6,17R, 18R and 19) was in the range of 74,7- 93,5% of the nominal concentration and two (17R, 18R) were below the 15% trigger of acceptable range for liquid suspensions. However, they still can be considered acceptable when all results are taken into account during the course of the study.

The relative standard deviations (RSD) of the samples 3-5 and samples 7-9 of 3.4% and 0.9% indicated the homogenous distribution of BAS 310 I in 1% CMC in drinking water. The RSD of the pup dosing samples for Cohorte 2-4 (sample 12-14) with a nominal concentration of 0.125 mg/ml was slightly higher with 13,9%. However, this slight deviation from the acceptance criterion (RSD up to 10%) is considered acceptable given the very low test substance concentration.

It is concluded that the results demonstrate the correctness of the concentrations of BAS 310 I in 1% CMC in drinking water.

B. OBSERVATIONS OF DAMS

1. Mortality

No dam died treatment-related during gestation or lactation. Due to premature termination of Group 3 based on severe pup toxicity 18 dams were premature sacrificed (Cohort 3 dams on GD 8 (6 dams); Cohort 2 dams on LD 1 (5 dams) and 2 (1 dam) and Cohort 1 dams on LD 14 (4 dams) and 15 (2 dams)). Cohort 4 animals of Group 3 were not started. All other dams survived until scheduled necropsy.

2. Clinical signs of toxicity

No treatment related clinical signs of toxicity were reported in dams during the entire study period up to the highest dose of 5 mg/kg bw.
Two high-dose dams (Nos.: 78 and 79) had no more pups alive on PND 11.

Comparison with beta-cypermethrin data as presented in the DAR 2013: Treatment related clinical signs of toxicity were observed in dams at doses ≥ 12 mg/kg bw. Therefore the data of alpha-cypermethrin are comparable at 5 mg/kg bw.

C. BODY WEIGHT AND BODY WEIGHT GAIN OF DAMS

Bodyweight was not affected in any treatment group compared to control neither during gestation nor during lactation phase (see Table 5.7.1-6).

The body weight change of the high-dose parental females was statistically significantly below the concurrent control values during GD 19 - 20 (about 25%, 9.4 g compared to 12.5 g in control (3.1 g less)) and during PND 8 - 14 (-11.4 g [vs.] 9.5 g in control).
The significantly reduced BWG from GD 19-20 is not considered treatment related but incidental because the BWG in the complete treatment phase GD6-20 was unaffected and the lower BWG of group 3 in the period GD 19-20 is a compensation of the higher body weight increase in the phase GD 6-19, which is 3g higher than the respective control. So the difference of 3 g is exactly that what group 3 dams have not gained in the period GD19-20.

Body weight gain during the lactation phase showed no significant change in the treatment phase between LD 0-8. There was no dose-related consistent trend observable between the treatment groups.

The significantly reduced body weight gain during PND 8 - 14 reflects the reduced food intake during this time period which is considered to be subsequent to reduced caloric needs (due to the low number of breast-fed pups), rather than an adverse effect of its own. In addition, this episode of reduced body weight gain fell into the treatment–free period of the dams from PND 8 onwards, which also indicates that this apparent effect is not treatment-related or adverse.
The body weight change of the mid- and low dose parental females was comparable to the concurrent control values throughout the entire study period.

Table 5.7.1-6: Summary of maternal bodyweight and bodyweight gain after administration of alpha-cypermethrin to rats in a DNT study

	Females			
Group	**0**	**1**	**2**	**3**
mg/kg bw/day	**0**	**0.5**	**2**	**5**
F_0 females: mean bodyweight [g] / [% of control]				
No. of animals#	**22**	**23**	**23**	**17**
Gestation: day 0	169.0	169.0 / 100	169.3 / 100	172.7 / 102
day 6	201.4	200.3 / 99	200.9 / 100	204.9 / 102
day 19	281.2	282.8 / 101	282.8 / 101	287.7 / 102 [12]
day 20	293.7	296.0 / 101	295.6 / 101	297.1 / 101 [12]
Lactation: day 0	231.9	228.1 / 98	234.1 / 101	233.4 / 101 [12]
day 7	249.9	249.4 / 100	252.5 / 101	268.2 / 107 [6]
day 8	254.9	255.3 / 100	260.2 / 102	269.7 / 106 [6]
day 21	258.1	259.2 / 100	264.0 / 102	- / -
F_0 females: mean bodyweight gain [g] / [% of control]				
Gestation: day 0-6	32.4	31.3 / 96	31.6 / 97	32.2 / 99
day 6-20	92.3	95.7 / 104	94.8 / 103	92.3 / 100 [12]
day 6-19	79.8	82.5 / 103	81.9 /103	82.8 / 104 [12]
day 19-20	12.5	13.1 / 105	12.8 / 102	**9.4** / 75** [12]
Lactation: day 0-4	6.1	9.3 / 152	4.6 / 76	8.6 / 141 [6]
day 4-7	11.9	12.0 / 101	13.8 / 116	14.5 / 122 [6]
day 7-8	5.0	5.9 / 118	7.7 / 154	1.4 / 29 [6]
day 8-14	9.5	9.2 / 97	7.3 / 76	**-11.4**/ -120 [6]**
day 14-21	-6.3	-5.3 / 84	-3.4 / 55	- / -
day 0-21	26.2	31.1 / 118	29.9 / 114	- / -

*: $p \leq 0.05$; **: $p \leq 0.01$ (Dunnetts´ test; two-sided)

#The number of animals which were pregnant and used for value derivation is given for the different groups. Based on the subdivision into 4 cohorts and the premature termination of group 4 only a variable number of animals were available for group 3 value determination. Any change in number of animals used for value derivation is given in square brackets.

Figure 5.7.1-2: Mean body weight of F0 parental females during gestation of F1 litters

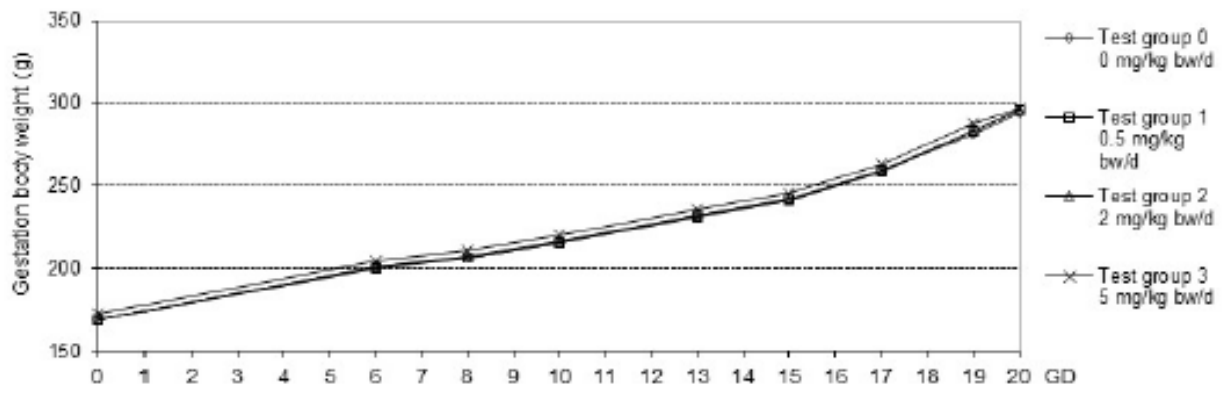
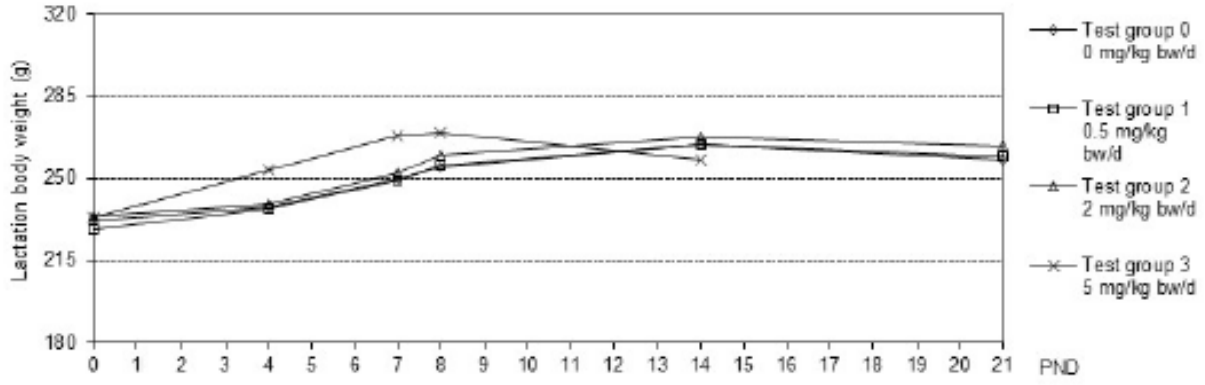


Figure 5.7.1-3: Mean body weight of F0 parental feamles during lactation of F1 litters



Comparison with beta-cypermethrin data as presented in the DAR 2013: Beta-cypermethrin showed as well no treatment related changes of body weight or body weight gain.

D. FOOD CONSUMPTION OF DAMS

No treatment-related or adverse differences of food intake were observed in any group during gestation and lactation .

The statistically significantly reduced food consumption in the high-dose parental females during LD 7-14 (-27%) clearly correlates with the corresponding number of breast-fed pups, keeping in mind that 2 out of 6 dams did not have any pups from PND11 onwards. Therefore this reduced food consumption is considered to reflect an adaption of food intake subsequent to reduced caloric needs rather than an adverse effect on its own. In addition this episode of reduced food consumption fell into the treatment–free period of the dams from PND 8 onwards, which also indicates that this apparent effect is not treatment-related or adverse.

Table 5.7.1-7: Summary of food consumption data after administration of alpha-cypermethrin to rats in a DNT study

	Females			
Group	0	1	2	3
mg/kg bw/day	0	0.5	2	5
F_0 females: mean food consumption [g/animal/day] / [% of control]				
No. of animals#	22	23	23	17
Gestation: day 0-6	17.4	17.8 / 102	17.9 / 103	18.0 / 103
day 6-13	19.7	20.0 / 101	19.9 / 101	20.6 / 104 [12]
day 13-20	22.2	22.5 / 101	22.7 / 102	23.5 / 105 [12]
day 6-20 (mean of means)	21	21.2 / 101	21.3 / 102	22.0 / 105
Lactation: day 0-4	25.0	26.7 / 107	25.7 / 103	26.0 / 104 [6]
day 4-7	39.8	41.6 / 104	42.0 / 106	44.4 / 111 [6]
day 7-14	51.7	54.1 / 105	53.6 / 104	37.7** / 73 [6]
day 14-21	60.8	63.5 / 104	63.1 / 104	- / -
day 0-21	44.3	46.5 / 105	46.1 / 104	- / -

*: $p \leq 0.05$; **: $p \leq 0.01$ (Dunnetts´ test; two-sided)

#The number of animals which were pregnant and used for value derivation is given for the different groups. Based on the subdivision into 4 cohorts and the premature termination of group 4 only a variable number of animals were available for group 3 value determination. Any change in number of animals used for value derivation is given in square brackets.

Comparison with beta-cypermethrin data as presented in the DAR 2013: Beta-cypermethrin showed as well no treatment related changes of food consumption.

E. FEMALE REPRODUCTION AND DELIVERY DATA

Female reproduction and delivery was unaffected up to the highest dose. As all females delivered by the breeder had successfully paired (plugs detected) the mating index was assumed to be 100% in all test groups. No effect on fertility index, or gestation index or duration of gestation was seen, number of live born or stillborn pups was comparable as well as the pup sex ratio.

Table 5.7.1-8: Summary of female reproduction and delivery data after administration of alpha-cypermethrin in the screening DNT study[#]

	Females			
Group	0	1	2	3
mg/kg bw/day	0	0.5	2	5
Females on study	25	25	25	18
Females mated	25	25	25	18
Female Mating Index	100	100	100	100
Females Pregnant	22	23	23	17
Female Fertility Index	88	92	92	94
Females allowed to deliver[#]	22	23	23	12
Females with Liveborn Pups[#]	22	23	23	12
Gestation Index	100	100	100	100[#]
Females with Stillborn Pups (%)	2 (9.1)	1 (4.3)	0 (0.0)	2 (17.0)
Females with all Stillborn (%)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Pups delivered				
Mean ± SD	10.2 ± 1.45	10.7 ± 1.40	10.2 ± 1.07	10.0 ± 1.54
Total	225	245	234	120
Liveborn	223	244	234	118
Live Birth Index	99	100	100	98
Stillborn (%)	2 (0.9)	1 (0.4)	0 (0.0)	2 (1.7)
Duration of gestation (days)	21.6	21.7	21.6	21.8
Sex ratio -day 0				
- males (%)	58.3	47.5	50. 9	56.8
-females (%)	41.7	52.5	49.1	43.2
Sex ratio -day 21				
- males (%)	58.7	48.5	49.0	n.a.
-females (%)	41.3	51.5	51.0	

*: $p \leq 0.05$ ** : $p <= 0.01$ (Fisher´s exact test; one-sided)

[#]The lower number in Test group 3 is based on the premature termination of this group. The 7 animals of the 4th (last) cohort were not placed into the study and the 6 animals of the 3rd cohort were terminated in early pregnancy. Thus only 12 gestating females were available for the delivery of pups. The raw data indicated that the parameter "females with liveborn pups" is significantly reduced resulting in an gestation index of 71%, but this is based on study termination and no substance related effect, therefore the data were adapted to the number of females allowed to deliver. Due to termination of the study all parameters addressing a PND observation were not available.

n.a.: not applicable due to termination of the group 3

Comparison with beta-cypermethrin data as presented in the DAR 2013: The data are comparable showing in both cases no effect on reproduction and delivery data.

F. OBSERVATIONS OF PUPS

1. Mortality

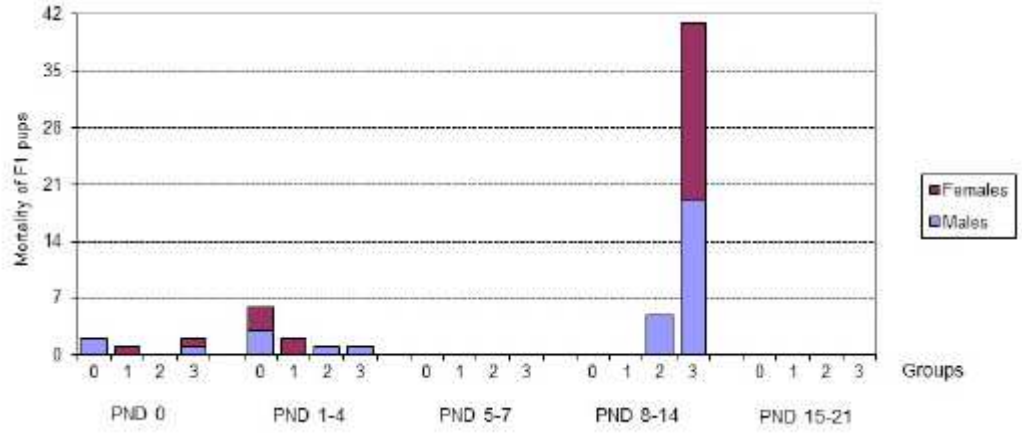
In test group 3 (5 mg/kg bw/d) 42 pups died or were cannibalized, 41 of the decedents died after pup dosing commencing on PND 10. This high mortality rate and the severe clinical observations after direct dosing of the pups led to the termination of the high-dose group. Therefore, no viability or lactation indices were calculated for test group 3.
For test group 1 and 2 the viability index and the lactation index were comparable with the concurrent control (see litter data in Table 5.7.1-9 and Figure 5.7.1-4).

Table 5.7.1-9: Summary of mortality in pups

Group	0	1	2	3
Dose dams [mg/kg bw/day]	0	0.5	2	5
Pups liveborn	223	244	234	118
Pups died	4	0	3	**25****
Pups cannibalized	3	2	3	**17****
Pups accidental death	0	0	1	0
Pups dead day 0	0	0	0	0
Pups dead day 0-4 (%)	6 (2.7)	2 (0.8)	1 (0.4)	1 (0.4)
Pups dead day 8-14	0 (0)	0 (0)	5 (2.1)	41 (35)
Pups surviving PND0-4	217	242	233	n.a.
Viability index (%)	97	99	100	-
Pups surviving PND4-21	184	202	196	n.a.
Lactation index (%)	90	91	88	-

*: p ≤ 0.05 ** : p <= 0.01 (Fisher´s exact test; one-sided)
n.a.: not applicable due to termination of the group 3

Figure 5.7.1-4: Mortality of F1 pups



The increased mortality was restricted to Cohort 1, where dams (dosed from GD6-PND8) and pups (from PND10 onwards) were gavaged with the same dose. The treatment-related mortalities in pups occurred between PND10 and PND14 and were observed in male pups at doses ≥ 2 mg/kg bw/day and in female pups at doses ≥ 5 mg/kg bw/day. Around 40% of the high dose pups died on PND 10 in both sexes. On PND11 additionally 7-8 % died and around 20 % were found cannibalized, followed by some further cannibalized pups on PND12 and 14. Based on the equal distribution of dead pups between high dose males (73%) and females (82%) a definite sex dependent susceptibility is not anticipated although already some mortality was seen in the mid dose males. However, on PND10 and 11 each 1 male pup died whereas 3 male pups suffered cannibalism on PND11. No mortality was seen in the group receiving 0.5 mg/kg bw/day.

Table 5.7.1-10: Mortality of cohort 1 pups before and during direct treatment[#]

	Male pups				**Female pups**			
Group	0	1	2	3	0	1	2	3
mg/kg bw/day (dams)	0	0.5	2	5	0	0.5	2	5
mg/kg bw/day (pups)	0	0.5	2	5	0	0.5	2	5
Pups- cohort 1: no. of pups / % of group (# no. of litter)								
Number of pups liveborn	36 (#1-6)	23 (#26-31)	24 (#51-56)	26 (#76-81)	17 (#1-6)	29 (#26-31)	28 (#51-56)	27 (#76-81)
PND1				1z / 3.9% (#80)				
PND2						1z / 3.6% (#28)		
PND3	2d / 5.6% (#2)				1d / 5.9% (#2)			
PND4	1z / 2.8% (#2)				1z / 5.9% (#2)			
PND 10			1d / 4.2% (#56)	10d / 38.5% (#76-79)				11d / 40.7% (#76-80)
PND 11			1d / 4.2% (#52) 3z/12.5% (#52, 55, 56)	2d / 7.7% (#79,81) 5z / 19.2% (#77, 78, 80, 81)				2d / 7.4% (#81) 6z / 22.2% (#76,77,79, 81)
PND 12				2z / 7.7% (#76)				1z / 4.1% (#80)
PND 14								2z / 7.4% (#80)
Pups died	3 not **treatment -related**	0	5 (21%) **treatment -related**	**1 not,** 19 (73%) **treatment-related**	2 not **treatment -related**	1 not **treatment -related**	0	22 (82%) **treatment-related**

[#] The cohort 1 contained each 6 pregnant animals of each dose group. Based on the mortality seen the treatment was changed for cohort 2-4; no statistical evaluation was performed on these data.

d- died; z-cannibalized

Based on the high mortality in cohort 1 the study design was changed: The high dose group was terminated while the dams dosed with 0, 0.5, and 2 mg/kg bw were continued. Though the pup doses were reduced to a constant dose of 0.25 mg/kg bw.

With this study design cohort 2-4 pups, treated with 0.25 mg/kg bw/day, showed no more treatment-related mortality either with dams pre-exposure to 0.5 or 2 mg/kg bw.

Table 5.7.1-11: Mortality of cohort 2-4 pups before and during direct treatment#

	Male pups			Female pups		
Group	0	1	2	0	1	2
mg/kg bw/day (dams)	0	0.5	2	0	0.5	2
mg/kg bw/day (pups)	0	0.25	0.25	0	0.25	0.25
Pups- cohort 1: no. of pups* / % of group (pups from litter)						
Number of pups liveborn	75 (#8-25)	75 (#32-50)	78 (#57-75)	61 (#8-25)	77 (#32-50)	73 (#57-75)
PND1	0	0	1d (#69)	1z (#20)	0	0
PND4	0	0	0	0	1z (#43)	0
PND 10 -21	0	0	0	0	0	0
Pups died	0	0	1	1	1	0

* no. of pups not considering pups culled on PND4 or with scheduled sacrifice on PND10;

#Cohort 2-4 were those pups from litters #8-25, 32-50, and 57-75.

In conclusion, taking all the data of Cohort 1-4 together, it can be stated that no mortality occurred in pups after bolus administration of 0.25 or 0.5 mg/kg bw/day subsequent to a pre-exposure of dams to a dose of 0.5 mg/kg bw. In addition no mortality occurred when pups were administered 0.25 mg/kg bw and pre-exposure in dams was increased to 2 mg/kg bw.

Comparison with beta-cypermethrin data as presented in the DAR 2013: Mortality in pups with beta-cypermethrin was investigated at 1, 4 and 12/8 mg/kg bw/day in a range finder study. Mortality occurred in this test at a dose of 4 mg/kg bw/day (10% of animals died) and at 8 mg/kg bw/day (5/6 died) and at 12 mg/kg bw/day on (3/4 animals died). The main study with pre-exposed pups was performed at 0.5 mg/kg bw/day and induced no mortality. This is comparable to alpha-cypermethrin.

2. Clinical signs of toxicity

Cohort 1 as well as Cohort 2-4 pups showed no clinical signs during the lactation period until direct treatment started. Therefore the lactation exposure up to PND 9 did not induce adverse effects in pups up to a maternal dose of 5 mg/kg bw/day.

In Cohort 1 all litters treated with 0.5, 2 and 5 mg/kg bw were affected after commencement of pup dosing (see Table 5.7.1-12). The number of affected pups and the severity of findings increased dose-dependently. The number of affected pups was 23/51 (45%) at 0.5 mg/kg bw and 100% at 2 and 5 mg/kg bw. Treatment-related signs were restricted to tremors, twitching, convulsions and excessive grooming in group 1 (0.5 mg/kg bw), while group 2 pups (2 mg/kg bw) showed in addition hypothermia, vocalization and lateral position. Dose groups 3 (5 mg/kg bw/day) was terminated based on severity of effects and is not further considered for clinical observation.

Interestingly, in group 1 (0.5 mg/kg bw) the highest incidence of clinical signs was not observed directly after first treatment but in the phase PND14-17. Half of the litters (3/6) were unaffected during the first 4 days of treatment and one third (2/6) were unaffected in the last third of treatment (PND17-21). Excessive grooming was more pronounced in the first third of treatment, while twitching was rather seen at the end of the treatment.

Table 5.7.1-12: Clinical signs of F1-cohort 1 pups before and during direct treatment*

	Cohort 1			
Group	0	1	2	3
Dams & Pups mg/kg bw/day	0	0.5	2	5
Number of litters in total (number of pups in total)	6 (51)	6 (51)	6 (52)	6 (53)
Number of litters affected				
- PND 0-21	0	6	6	6
- PND 0-9	0	0	0	0
- PND 10-13 /14-17* / 18-21*	0 / 0 / 0	3 / 6 / 4	6 / 6 / 6	6 / n.a. / n.a.
Tremors	0 / 0 / 0	1 / 4 / 4	6 / 6 / 6	4 / n.a. / n.a.
Twitching	0 / 0 / 0	0 / 0 / 3	0 / 0 / 6	0 / n.a. / n.a.
Convulsions Tonic Clonic	0 / 0 / 0	0 / 1/ 0	6 / 5 / 3	6 / n.a. / n.a.
Excessive Grooming	0 / 0 / 0	2 / 2 / 0	0 / 1 / 0	0 / n.a. / n.a.
Hypothermia	0 / 0 / 0	0 / 0 / 0	2 / 0 / 0	3 / n.a. / n.a.
Vocalisation	0 / 0 / 0	0 / 0 / 0	4 / 0 / 0	6 / n.a. / n.a.
Lateral position	0 / 0 / 0	0 / 0 / 0	2 / 0 / 0	1 / n.a. / n.a.
No more pups alive PND 11	0 / 0 / 0	0 / 0 / 0	0 / 0 / 0	2 / n.a. / n.a.
- PND 10-21 Incidence litter (pup)				
Tremors		6 (18)	6 (49)	4 (15)
Twitching		3 (5)	6 (36)	
Convulsions Tonic Clonic		1 (1)	6 (41)	6 (34)
Excessive Grooming		3 (6)	1 (1)	
Hypothermia			2 (3)	4 (10)
Vocalisation			4 (6)	6 (20)
Lateral position			2 (3)	1 (1)
Number of litters not affected				
- PND 10-13 /14-17* / 18-21*	6 / 6 / 6	3 / 0 / 2	0 / 0 / 0	0 / n.a. / n.a.
- PND 10-21	6 (100 %)	0 (0%)	0 (0%)	0 (0%)
Number of pups not affected				
- PND 10-21	**51 (100 %)**	**28 (55%)**	**0 (0%)**	**0 (0%)**

*Based on premature killing of Group 3 dams/pups on PND 14 and 15 the data were restricted on PND10-13; single data when available were disregarded
n.a. not applicable based on termination of dose group

In Cohort 2-4 only marginal differences were seen on litter base; with 3 litters were free of symptoms in group 1 and 2 litters were free of symptoms in group 2 (see Table 5.7.1-13). A clear dose dependency was seen on pup level, where 21.2 % of the pups were affected when dams were treated with 0.5 mg/kg bw and 44% of pups were affected when dams were pre-treated with 2 mg/kg bw alpha-cypermethrin. An early effect in the low dose group on litter base was twitching, while tremors increased in the mid third of treatment. 14 of the 17 litters (82%) were free of symptoms in the last third of treatment. Pups reared by 2 mg/kg bw-treated dams showed in addition convulsion and a much higher incidence of tremors and twitching directly in the first treatment interval (PND10-13), whereas in the last third of treatment the effects were comparable with the mid dose group (13/17 litters were unaffected).

The effects seen in the different treatment groups indicate that lactational exposure plays a major role in development of different characteristics at least up to PND17. In the later period of treatment the effects seen are comparable and might represent rather the direct exposure to the bolus administered dose of pups. Whether this later phase represents the real symptomatic induced by the bolus administration alone or is still interfered with lactation exposure, or whether metabolic capacity or adipose tissue ratio is increased, remains open.

Table 5.7.1-13: Clinical signs of F1-cohort 2-4 pups before and during direct treatment*

	Cohort 2-4		
Group	0	1	2
Dams [mg/kg bw/day]	0	0.5	2
Pups [mg/kg bw/day]	0	0.25	0.25
Number of litters in total (number of pups in total)	**16 (135)**	**17 (151)**	**17 (150)**
Number of litters affected			
- PND 0-21	0	14	15
- PND 0-9	0	0	0
- PND 10-13 / 14-17 / 18-21	0 / 0 / 0	8 / 9 / 3	15 / 13 / 4
Tremors	0 / 0 / 0	4 / 7 / 0	10 / 10 / 0
Twitching	0 / 0 / 0	7 / 7 / 3	9 / 10 / 4
Convulsions Tonic Clonic	0 / 0 / 0	0 / 0 / 0	1 / 0 / 0
- PND 10-21 Incidence litter (pup)			
Tremors		10 (16)	14 (41)
Twitching		12 (21)	13 (37)
Convulsions Tonic Clonic			1 (1)
Number of litters not affected			
- PND 10-13 /14-17 / 18-21	16 / 16 / 16	9 / 8 / 14	2 / 4 / 13
- PND 10-21	16 (100 %)	3 (21.4%)	2 (11.8%)
Number of pups not affected			
- PND 10-21	**135 (100 %)**	**119 (78.8%)**	**84 (56%)**

3. Clinical signs in pups in chronological order

A more detailed look into the characteristics of the effects induced by bolus administration of pups to 0.5 mg/kg bw/day and 0.25 mg/kg bw/day is performed in the following.

Bolus administration of 0.5 mg/kg bw/day to pups pre-treated by dams treated with 0.5 mg/kg bw induced effects in 45% of cohort 1 pups (see Table 5.7.1-12). Effects were first observed on PND11 in one pup showing severe tremors, followed by 19 pups showing slight tremors between PND12 and 20 (37.3%). Tremors were the predominant symptom and were observable over the complete interval of treatment with a slightly higher occurrence from PND15-20. Under this dose regimen pups showed excessive grooming (11.8%) in the early phase of direct dosing, while twitching was seen rather at the end (seen in 9.8% of pups). The data indicate that symptoms were distributed over the whole treatment group and only a small number of animals was repeatedly seen affected during the observation period, often with varying symptom. The percentage of affected pups per observation day stayed below 14%.

Table 5.7.1-14: Overview clinical signs in F1-Cohort 1/ group 1 pups dosed with 0.5 mg/kg bw (dam dosed with 0.5 mg/kg bw): individual clinical observations in chronological order

	Affected pup in a litter at day of observation [# litter-pup]											
Observed effect (Pup incidence)	PND 10	PND 11	PND 12	PND 13	PND 14	PND 15	PND 16	PND 17	PND 18	PND 19	PND 20	PND 21
Total number of litters and pups available for clinical observation at PND10: 51 pups in 6 litters available												
Tremors slight **(37.25%)**			26-8, 26-9		31-9	26-5, 30-6, 30-7, 30-8	29-3 30-7	27-3, 27-7	30-4, 31-3, 31-10	26-6, 31-7, 31-11	26-5, 28-1, 31-3, 31-9	
Tremors severe **(2%)**		26-3										
Twitching **(9.8%)**										31-9	26-6, 26-9, 30-3	30-4
Convulsions Tonic Clonic **(2%)**					28-1							
Excessive Grooming **(11.8%)**			29-7	30-1, 30-2, 30-4, 30-8	28-4, 29-7							
Affected pups (%) **litters (%)**	**0** **0**	**2** **13.6**	**6** **16.6**	**8** **16.6**	**8** **50**	**8** **33**	**4** **33**	**4** **16.6**	**6** **33**	**8** **33**	**14** **67**	**2** **16.6**
Affected pups in total (%) **Affected litters in total (%)**	**23/51 = 45.1%** **6/6 = 100%**											

Clinical observations after reduction of the pup dose to 0.25 mg/kg bw/day while keeping pre-treatment of dams at 0.5 mg/kg bw induced effects in 32 of 151 pups (21.2%) (see Table 5.7.1-11). Symptoms were clearly reduced and less severe than in Group 1 pups of Cohort1, as they were restricted to slight tremors and twitching, which is a less severe form of tremors (see Table 5.7.1-13). Both symptoms were directly observed on PND10 and remained mainly until PND16 which showed the highest incidence with 6.6% of pups showing clinical signs. The average percentage of affected pups was around 3% allocated to 2-6 litters each day. From PND17 onwards tremors disappeared and twitching was the only reported clinical sign with no more than 2% of the pups being affected.
Compared to the pups of Cohort 1 which got the double dose by bolus application the spectrum of clinical signs here and the severity was clearly reduced under this dose regime and pup incidence was reduced from 45% to 21.2%. The dominant phase of clinical sign was PND 10 – 16.

Table 5.7.1-15: Overview clinical signs in F1-Cohort 2-4/ group 1 pups dosed with 0.25 mg/kg bw (dam dosed with 0.5 mg/kg bw): individual clinical observations in chronological order

	Affected pup in a litter at day of observation [# litter-pup]											
Total number of litters and pups available for clinical observation at PND10: 151 pups in 17 litters available												
Observed effect (Pup incidence)	PND 10	PND 11	PND 12	PND 13	PND 14	PND 15	PND 16	PND 17	PND 18	PND 19	PND 20	PND 21
Tremors slight (**10.6%**)	47-7	46-3	41-4 48-2 48-5		32-4 40-8 43-2	41-5	33-3 33-5 37-3 37-6 41-8 44-4 44-5					
Twitching (**13.9%**)	44-4 44-5 48-9 49-3 49-9	41-5 45-5 46-8		42-8 48-5	40-9 41-3	33-9 47-7	43-3 44-10 50-8		43-4 44-11 47-2		44-9	
Affected pups (%) **litters (%)**	**4** **24**	**2.7** **18**	**2.0** **12**	**1.3** **12**	**3.3** **24**	**1.9** **18**	**6.6** **35**	**0** **0**	**2.0** **18**	**0** **0**	**0.7** **6**	**0** **0**
Affected pups in total (%) **Affected litters in total (%)**	**32/151 = 21.2% (12 females, 20 males)** **14/17 = 82.4%**											

The clinical sings in pups from dams pre-treated with 2 mg/kg bw were more frequent and more severe (see Cohort 2-4, group 2, Table 5.7.1-14) compared to those seen in pups from mothers predosed with 0.5 mg/kg bw/day. This increased incidence was mainly seen in the first part of direct pup dosing (PND10-17) and is most probably due to the higher amount transferred with the milk. The percentage of pups showing tremors increased from 10.6 % to 27.9% (slight and moderate tremor incidence added). The percentage of pups showing twitching increased from 13.9% to 24.7% (see Table 5.7.1-14). Additionally clonic tonic convulsions were reported in one animal. Symptoms were directly observed on PND10 and were predominant in the first interval until PND17 with around 4 to 10% of pups affected in slightly less than half of the litters. After PND17 tremors ceased completely and only single pups showed twitching. The percentage of affected pups after PND17 stayed below 1.3% and was thereby comparable with the pups of the same Cohort but with the lower pretreatment of dams (0.5 mg/kg bw, see Table 5.7.1-15). This indicates that rather the later part reflects the direct dosing of pups.

Table 5.7.1-16: Overview clinical signs in F1-Cohort 2-4/ group 2 pups dosed with 0.25 mg/kg bw (dam dosed with 2 mg/kg bw): individual clinical observations in chronological order

	Affected pup in a litter at day of observation [# litter-pup]											
Total number of litters and pups available for clinical observation at PND10: 150 pups in 17 litters available												
Observed effect	PND 10	PND 11	PND 12	PND 13	PND 14	PND 15	PND 16	PND 17	PND 18	PND 19	PND 20	PND 21
Tremors slight (25.3%)	59-9 64-7 73-1 74-7 75-2	59-8 64-7 75-10	70-1 72-2 74-4 75-2	57-9 68-1 68-2 69-2	61-4 62-1 62-3 66-7 69-1 69-2 75-3	59-5 59-8 62-8 64-2 66-3 69-2 69-10	59-3 59-7 60-2 60-3 64-1 69-9 74-5	57-2 57-6 57-8 59-2 59-4 66-4				
Tremors moderate (2.6%)	57-1 57-3 57-6 57-7											
Twitching (24.7%)	60-6 60-7 64-4 73-2 75-4	57-8 69-10 75-8	60-10 66-7 67-6 67-8	60-4 62-4 64-8 67-2 69-10 69-11	59-7 64-3 66-9 67-6 69-7 75-2	64-5 64-7 64-9 67-2 67-4 69-9 70-1 72-3	57-1 57-6 57-7 64-10 67-4 69-4 74-7		69-4 69-7	57-3	66-7 72-4	
Convulsions Tonic Clonic (0.6%)	57-4											
Affected pups (%) **litters (%)**	**10** **41**	**4** **29**	**5.3** **41**	**6.7** **41**	**8.7** **47**	**10** **47**	**9.3** **41**	**4** **18**	**1.3** **6**	**0.7** **6**	**1.3** **6**	**0** **0**
Affected pups in total (%)	**66/ 150 = 44%**											
Affected litters in total (%)	**15/17 = 88.2 %**											

The different characteristics within the first interval (PND10-17) of Cohort 2-4 group 1 and 2 pups (both dosed at 0.25 mg/kg bw/day) are clearly attributed to double exposure via lactation exposure. The abrupt reduction of symptoms around PND 17 in pups dosed at 0.25 mg/kg bw/day in both, the 0.5 mg/kg bw/day pre-treated and the 2 mg/kg bw/day pre-treated dams to less than 2% affected foetuses might indicate the end or a lower level of lactation exposure to alpha-cypermethrin. It remains to be determined if not pre-treated pups or pups reared from 0.25 mg/kg bw/day pre-treated dams would show symptoms.

4. Detailed clinical observation (DCO) of pups

A detailed clinical observation of pups was performed on PND 4, 10, 12, 14, 16, 18, and 20 in a standard arena.
Cohort 1 mid and high dose pups showed toxicity resulting in mortality, so that these groups were only summarized in the following table but were not further discussed.
Low dose bolus administration of 0.5 mg/kg bw/day to pups delivered from dams pre-treated with 0.5 mg/kg bw (Cohort 1, Group 1) induced slight tremors in 8 male and 8 female pups (see

Table 5.7.1-18) from all litters. Tremors were seen from PND12 onwards in males, and from PND 14 onwards in females. No effect was seen in both sexes at PND20. No significant sex difference was obvious. The percentage of affected pups was slightly lower (31.4%) than that in the general clinical observations (45.1%). Control animals did not show any abnormal findings besides one animal that died before DCO on PND 4.

Cohorte 1 Group data:

Table 5.7.1-17: Detailed clinical observation in F1-Cohort 1/ group 1,2,3 pups dosed with 0.5-2-5 mg/kg bw/day (dam dosed similar) compared to control

		males: ~~total number of pups 23~~							females: ~~total number of pups: 28~~						
		Post natal day (PND)													
	Group	4	10	12	14	16	18	20	4	10	12	14	16	18	20
# of animals examined	0	119	118	108	108	108	108	108	87	86	76	76	76	76	76
	1	26	26	23	23	23	23	23	31	31	28	28	28	28	28
	2	27	27	23	19	19	19	19	31	31	28	28	28	28	28
	3	28	28	20	6	0	0	0	30	30	7	0	0	0	0
Normal															
Nothing abnormal detected	0	118	108	108	108	108	108	108	86	76	76	76	76	76	76
	1	26	23	21	17	23	21	23	31	28	28	23	26	27	28
	2	27	16	4	9	18	13	13	31	21	11	11	20	20	22
	3	28	11	1	0	-	-	-	30	13	2	0	-	-	-
Dead															
Died before DCO	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0
	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	**4**	0	0	0	0	0	0	0	0	0	0	0
	3	0	0	**14**	0	-	-	-	0	0	**17**	**2**	-	-	-
Found Dead	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	**1**	0	0	0	0	0	0	0	0	0	0	0	0
	3	0	**6**	0	0	-	-	-	0	**3**	0	0	-	-	-
Premature sacrifice	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	3	0	0	0	3	-	-	-	0	0	0	3	-	-	-
Body position															
Lateral	3	0	1	0	0	-	-	-	0	5	0	0	-	-	-
Dorsal	3	0	1	0	0	-	-	-							

Activity															
Tremor	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	1	0	0	2	6	0	2	0	0	0	0	5	2	1	0
	2	0	8	14	10	1	5	2	0	6	17	17	8	8	0
	3	0	6	5	3	-	-	-	0	5	5	2	-	-	-
Convulsion	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	1	0	0	0	0	0	1	0	0	0	0	0
	3	0	5	0	0	-	-	-	0	9	0	0	-	-	-
Twitching	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	1	4	0	0	0	0	0	0	6
	3	0	0	0	0	-	-	-	0	0	0	0	-	-	-
General state															
Hypo-thermia	3	0	5	0	0	-	-	-	0	8	0	1	-	-	-

Table 5.7.1-18: Detailed clinical observation in F1-Cohort 1/ group 1 pups dosed with 0.5 mg/kg bw/day (dam dosed with 0.5 mg/kg bw/day)

	Affected pup in a litter at day of observation [# litter-pup]													
	Total number of litters: 6 litters available													
	males: ~~total number of pups 23~~							females: ~~total number of pups: 28~~						
total number of pups	26	26	23	23	23	23	23	31	31	28	28	28	28	28
Observed effect	PN D4	PN D10	PN D12	PN D14	PN D16	PN D18	PN D20	PN D4	PN D10	PN D12	PN D14	PN D16	PN D18	PN D20
Tremors slight			27-3 28-1	27-3 28-1 28-5 29-2 29-4 30-4		28-4 29-3					26-4 26-7 26-9 28-7 31-11	31-8	30-8 26-6	
Affected pups (%) **litters (%)**	**0** **0**	**0** **0**	**8.7** **33**	**26** **66**	**0** **0**	**8.7** **33**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**17.9** **50**	**3.6** **17**	**7.1** **33**	**0** **0**
Affected pups in total (%) **Affected litters in total (%)**	**16/51= 31.4%** **6/6 = 100%**													

Cohorte 2-4 Group data:

Bolus administration of 0.25 mg/kg bw/day to pups delivered from dams pre-treated with 0.5 mg/kg bw (Cohort 2-4, Group 1; see Table 5.7.1-19) revealed in 1 male pup twitching of both hindlimbs. No other effect was seen. Females did not show any signs of neurotoxicity. As both sexes showed no significant differences no sex difference was obvious and this dose is considered near to the no observed effect level with a percentage of affected pups in DCO of 0.6%. Whether this dose would be a NOAEL in case of dams would be pre-treated with 0.25 mg/kg bw/day remains open.

Table 5.7.1-19: Detailed clinical observation in F1-Cohort 2-4/ group 1 pups dosed with 0.25 mg/kg bw (dam dosed with 0.5 mg/kg bw)

	Affected pup in a litter at day of observation [# litter-pup]													
	Total number of litters: 17 litters available													
	males: ~~total number of pups: 75~~							females: ~~total number of pups: 76~~						
total number of pups	82	82	75	75	75	75	75	84	83	76	76	76	76	76
Observed effect	PN D4	PN D10	PN D12	PN D14	PN D16	PN D18	PN D20	PN D4	PN D10	PN D12	PN D14	PN D16	PN D18	PN D20
Twitching hindlimbs both		44-5												
Affected pups (%)	**0**	**1.3**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
litters (%)	**0**	**5.9**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
Affected pups in total (%)	**1/151= 0.6%**													
Affected litters in total (%)	**1/17 = 5.8%**													

Bolus administration of 0.25 mg/kg bw/day to pups delivered from dams pre-treated with 2 mg/kg bw (Cohort 2-4, group 2; see Table 5.7.1-20) revealed slight tremors in 4 male pups between PND 10 and PND14. No effect was seen from PND 16 onward. The percentage of affected pups in the DCO was considerably lower (2.7%) than the percentage of affected pups in the general clinical observations (21.2%).

Table 5.7.1-20: Detailed clinical observation in F1-Cohort 2-4/ group 2 pups dosed with 0.25 mg/kg bw (dam dosed with 2 mg/kg bw)

	Affected pup in a litter at day of observation [# litter-pup]													
	Total number of litters: 17 litters available													
	males: ~~total number of pups 77~~							females: ~~total number of pups: 73~~						
total number of pups	84	84	77	77	77	77	77	80	80	73	73	73	73	73
Observed effect	PN D4	PN D10	PN D12	PN D14	PN D16	PN D18	PN D20	PN D4	PND 10	PN D12	PN D14	PN D16	PN D18	PN D20
Tremor slight		57-3 59-2	57-3 60-2	62-1										
Affected pups (%) **litters (%)**	**0** **0**	**2.6** **11.8**	**2.6** **11.8**	**1.3** **5.9**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**0** **0**	**0** **0**
Affected pups in total (%)	**4/150= 2.7%**													
Affected litters in total (%)	**4/17 = 23.5%**													

Comparison with beta-cypermethrin data as presented in the DAR 2013: The clinical signs in pups treated with beta-cypermethrin as described in the DAR 2013 were clonic convulsions, carangiform ataxia and straub tail at 0.5 mg/kg bw. With alpha-cypermethrin dosed to dams and pups equally at 0.5 mg/kg bw slight and moderate tremors, twitching, clonic convulsions, and excessive grooming were reported. However, no permanent seizures like straub tail were seen with alpha-cypermethrin.
Both alpha- and beta-cypermethrin are considered comparable in the development of neurotoxic symptoms.

G. BODY WEIGHT AND BODY WEIGHT GAIN OF PUPS

Body weights of male or female pups were not significantly affected in any treatment group of cohort 1 when compared to control (see Table 5.7.1-21 and Figure 5.7.1-5). The relative body weights of the treated pups were within 95% – 103 % at PNDs 1, 4, 7, 10, 11, 13, 15, 17, and 21, with the exception of the high dose male and female pups, which changed to 88% compared to control animals in males and 108% in females after the commencement of treatment.
Body weight gains of these high dose pups showed statistically significant reductions of mean body weight gains during PND 11-13 and corresponding increases during PND 11-15 which are related to the intoxication and death of a big proportion of high-dose pups after commencement of direct pup dosing.

The increased body weight gain in the mid-dose group during PND 15-17 is considered as incidental findings, as the respective body weight and the weight gain over the complete weaning period was unaffected. Further evidence is based on the litter weight, which was in addition unaffected at PND15 and 17 with 100 % and 102% of control (see Table 5.7.1-22).

Table 5.7.1-21: Mean body weight of cohort 1 pups administered alpha-cypermethrin

	Males				Females			
Group	0	1	2	3	0	1	2	3
Dose [mg/kg bw/day]	0	0.5	2	5	0	0.5	2	5
F_1 - cohort 1: bodyweight [g] / [% of control]								
PND 1	6.6	6.6 / 100	6.5 / 98	6.6 / 100	6.3	6.5 / 103	6.1 / 97	6.3 / 100
PND 4	10.2	10.3 / 101	9.7 / 95	10.2 / 100	9.8	10.4 / 106	9.4 / 96	9.9 / 101
PND 7	15.4	15.8 / 103	14.9 / 97	15.7 / 102	14.9	15.8 / 106	14.5 / 97	15.3 / 103
PND 10	21.4	22.1 / 103	21.3 / 100	21.9 / 102	20.8	22.2 / 107	20.7 / 100	21.4 / 103
PND 11	23.8	24.7 / 104	23.4 / 98	23.6 / 99	23.0	24.4 / 106	22.7 / 99	21.7/ 94
PND 13	28.6	29.7 / 104	28.3 / 99	30.6 / 107	27.7	29.4 / 106	27.6 / 100	24.4 / 88
PND 15	33.2	34.1 / 103	33.1/ 100	35.8 / 108	32.0	33.6 / 105	32.3 / 101	29.1 / 91
PND 17	37.3	38.3 / 103	38.1 / 102	n.a.	35.8	37.7 / 105	37.0 / 103	n.a.
PND 21	49.2	49.7 / 101	48.6 / 99	n.a.	46.8	48.5 / 104	47.0 / 100	n.a.
F_1 - cohort 1: bodyweight gain [g] / [% of control]								
PND 1 - 4	3.6	3.7	3.2	3.6 / 97	3.6	3.8	3.3	3.6 / 95
PND 10-11	2.4	2.6	2.2	1.9 / 79	2.2	2.2	2.0	**0.5**/23**
PND 11-13	4.8	4.9	4.9	**6.2*/ 129**	4.6	5.0	4.9	**2.6*/57**
PND 13-15	4.5	4.4	4.8	**8.5**/ 189**	4.3	4.2	4.7	**6.5**/151**
PND 15-17	4.1	4.2	**5.0*/ 122**	n.a.	3.8	4.1	**4.7**/124**	n.a.
PND 4 - 21	38.9	39.4 / 101	38.9 / 100	n.a.	37	38.2 / 103	37.6 / 102	n.a.

Statistics: Dunnett-test (two-sided) * p≤0.05; p**≤0.01; n.a. not applicable based on termination of dose group

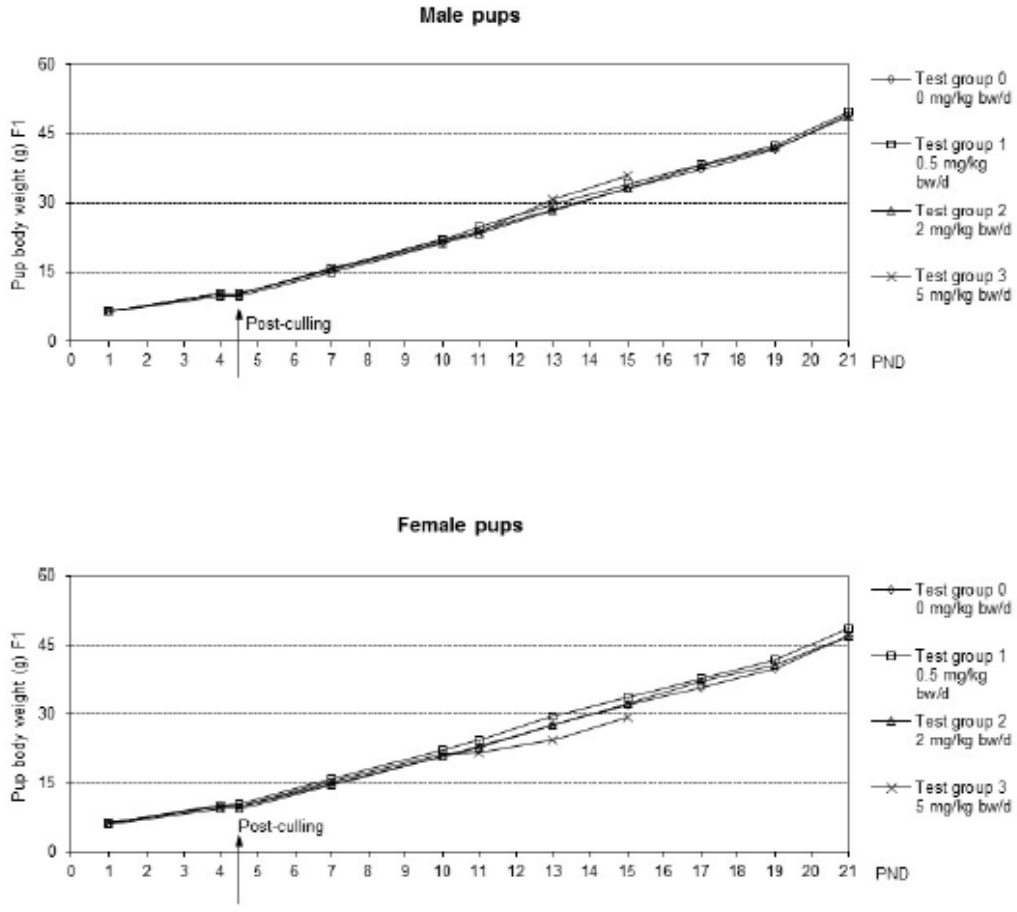
Table 5.7.1-22: Cohort 1 mean litter weight after administration of alpha-cypermethrin

	Litter			
Group	0	1	2	3[#]
mg/kg bw/day (dams)	0	0.5	2	5
mg/kg bw/day (pups)	0	0.5	2	5
F_1 - cohort 1: litterweight [g] / [% of control]				
PND 1	6.5	6.5 / 100	6.3 / 96	6.5 / 97
PND 4	10.1	10.3 / 102	9.6 / 95	10.0 / 99
PND 7	15.2	15.8 / 104	14.7 / 97	15.5 / 102
PND 10	21.2	22.1 / 104	21.0 / 99	21.7 / 102
PND 11	23.5	24.5 / 104	23.0 / 98	22.0 / 94
PND 13	28.3	29.5 / 104	27.9 / 98	24.6 / 87 [4]
PND 15	32.7	33.8 / 103	32.6 / 100	29.7 / 91 [2]
PND 17	36.7	38.0 / 104	37.4 / 102	n.a.
PND 21	48.3	49.1 / 102	47.6 / 99	n.a.
F_1 - cohort 1: litterweight gain [g] / [% of control]				
PND 1 - 4	3.6	3.8 / 106	3.3 / 92	3.7 / 103
PND 4 - 21	38.2	38.8 / 102	38.0 / 99	n.a.

[#]The lower numbers of litters indicated in square brackets in Test group 3 are based on the mortalities.

n.a.: not applicable due to termination of the group 3

Figure 5.7.1-5: Mean body weight of F1 pups (Cohort 1)



Bodyweights of male or female pups as well as litter weights were not affected in any treatment group of cohort 2-4 when compared to control (see Table 5.7.1-23, Table 5.7.1-27 and Figure 5.7.1-6). The relative body-/litter weights of the treated pups of groups 1 and 2 were within 99% - 105% and 99% - 103% of the body-/litter weights of the control animals at PNDs 1, 4, 7, 10, 11, 13, 15, 17, and 21.

Furthermore, body weight gains as well as litter weight gain were also not affected by treatment. Body weight- and Litter weight gains ranged between 98% - 100% compared to control animals. There was no dose-related consistent trend observable between the treatment groups regarding body weight or body weight gain.

Data from treatment group 3 were not available for cohort 2-4 as the dose group was terminated at PND 1.

Table 5.7.1-23: Mean pup body weight of cohort 2-4 pups administered alpha-cypermethrin

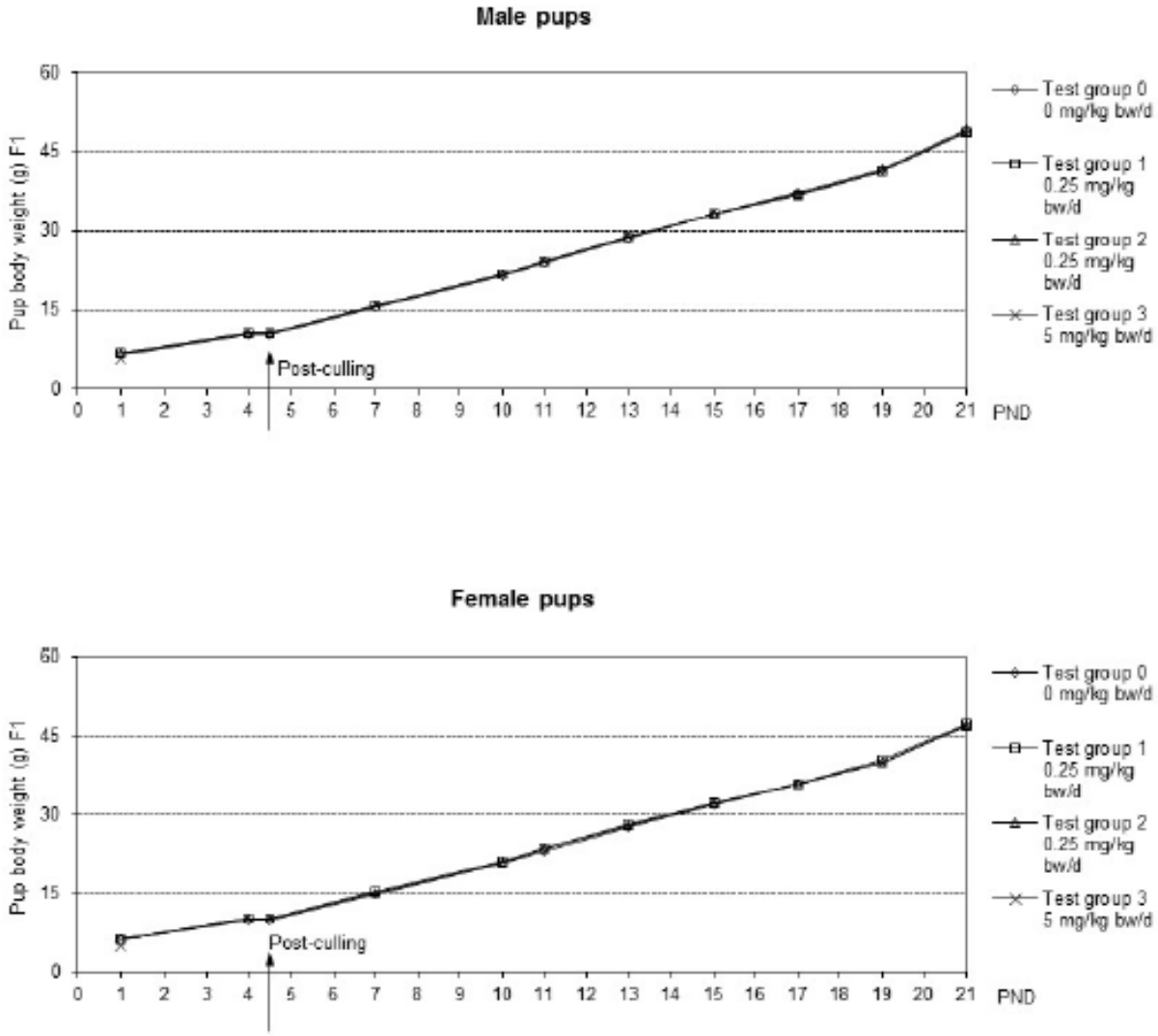
	Males			Females		
Group	0	1	2	0	1	2
mg/kg bw/day (dams)	0	0.5	2	0	0.5	2
mg/kg bw/day (pups)	0	0.25	0.25	0	0.25	0.25
F_1 - cohort 2-4: bodyweight [g] / [% of control]						
PND 1	6.6	6.9 / 105	6.9 / 105	6.3	6.5 / 105	6.5 / 105
PND 4	10.2	10.6 / 104	10.6 / 104	9.8	10.2 / 103	10.1 / 103
PND 7	15.4	15.8 / 103	15.8 / 103	14.9	15.3 / 103	15.2 / 102
PND 10	21.4	21.8 / 102	21.7 / 101	20.8	21.2 / 102	20.9 / 100
PND 11	23.8	24.2 / 102	24.1 / 101	23.0	23.6 / 103	23.4 / 102
PND 13	28.6	28.8 / 101	28.7 / 100	27.7	28.1 / 101	27.9 / 101
PND 15	33.2	33.1 / 100	33.1 / 100	32.0	32.2 / 100	32.2 / 100
PND 17	37.3	36.8 / 99	37.1 / 99	35.8	35.9 / 100	35.9 / 100
PND 21	49.2	48.8 / 99	49.0 / 100	46.8	47.2/ 101	47.0 / 100
F_1 - cohort 2-4: bodyweight gain [g] / [% of control]						
PND 1 - 4	3.6	3.7 / 100	3.7 / 100	3.6	3.7 / 100	3.6 / 100
PND 4 - 21	38.9	38.2 / 98	38.4 / 99	37.0	37. 0 / 101	36.8 / 100

Statistics: Dunnett-test (two-sided) * p≤0.05; p**≤0.01;

Table 5.7.1-24: Cohort 2-4 mean litter weight after administration of alpha-cypermethrin

	Litter			
Group	0	1	2	
mg/kg bw/day (dams)	0	0.5	2	
mg/kg bw/day (pups)	0	0.25	0.25	
F_1 - cohort 2-4: litterweight [g] / [% of control]				
PND 1	6.5	6.7 / 103	6,7 / 103	
PND 4	10.1	10.4 / 102	10.4 / 102	
PND 7	15.2	15.6 / 103	15.5 / 102	
PND 10	21.2	21.5 / 101	21.3 / 100	
PND 11	23.5	23.9 / 102	23.8 /101	
PND 13	28.3	28.5 / 101	28.3 /100	
PND 15	32.7	32.7 / 100	32.7 / 100	
PND 17	36.7	36.4 / 99	36.5 / 99	
PND 21	48.3	48.0 / 99	48.0 / 99	
F_1 - cohort 2-4: litterweight gain [g] / [% of control]				
PND 1 - 4	3.6	3.7 / 100	3.7 / 100	
PND 4 - 21	38.5	37.7 / 98	37.6 / 98	

Figure 5.7.1-6: Mean body weight of F1 pups (Cohort 2-4)



Comparison with beta-cypermethrin data presented in the DAR 2013: Body weight reduction was seen in pups treated with 0.5 mg/kg bw/day beta-cypermethrin during direct administration of the test substance (PND10-16) but restored after cessation of dosing to values within historical control. With alpha-cypermethrin at 0.5 mg/kg bw no effect on body weight or bodyweight gain was seen during the treatment period (PND 10-21).

H. Clinical assessment of neurotoxicity

MOTOR ACTIVITY (MA)

Motor activity measurements of Cohort 1 (low and mid dose) and Cohort 2-4 pups on PND17 and PND 21 revealed two significantly reduced individual values (see Table 5.7.1-25). One significant lower MA was seen reduced at the onset of measurement on PND21 in Cohort 2-4 female pups from group 1 (dosed with 0.25 mg/kg bw/day; dams pre-dosed with 0.5 mg/kg bw). The other lower MA was seen at interval 7 on PND21 in Cohort 2-4 female pups from group 2 (dosed with 0.25 mg/kg bw/day; dams pre-dosed with 2 mg/kg bw). Both changes are isolated findings. These two alterations did not occur to a degree or in a manner suggesting a compound-related pattern of neurotoxicity.
No significant change as indication of treatment-related effect was seen in the mean total motor activity counts (see Table 5.7.1-26). The respective graphs are listed below.

Table 5.7.1-25: Mean motor activity on PND 21 in female pups of Cohort 2-4 (N = 7 pups)

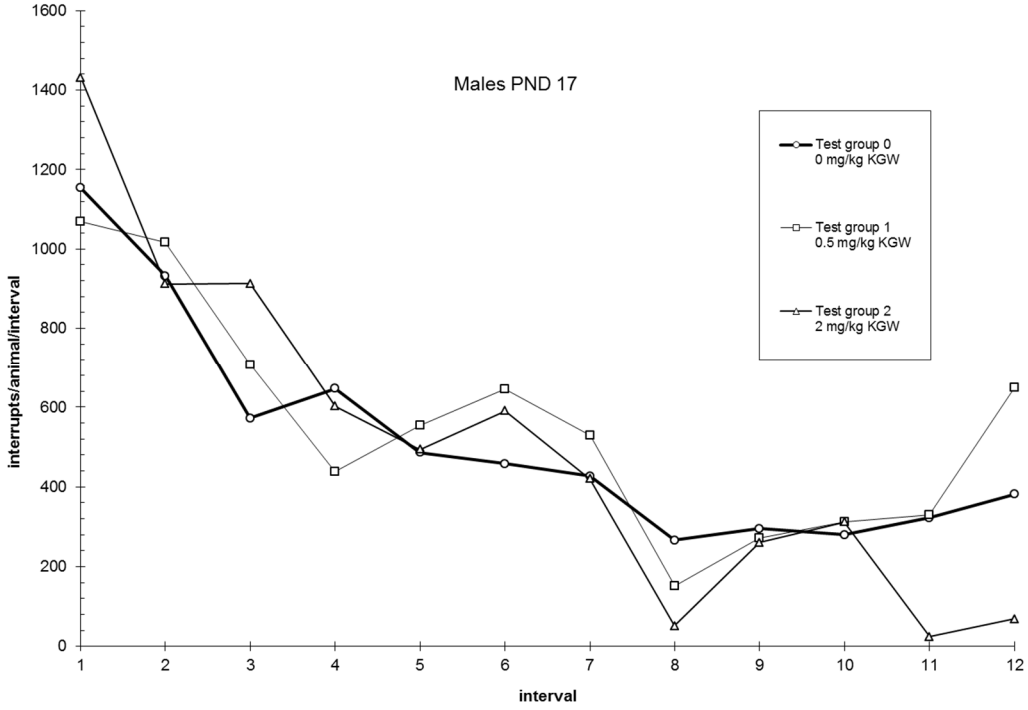
Dam/ Pup Dose	Interval												
	1	2	3	4	5	6	7	8	9	10	11	12	1-12
0/0	1076.4	464.0	324.9	339.6	186.9	134.0	161.2	283.1	104.7	83.1	133.1	108.0	3399.0
0.5/ 0.25	**810.1****	465.0	277.6	165.7	128.6	119.1	87.0	76.0	30.9	50.1	48.1	153.3	2402.7
2.0/ 0.25	1009	519.4	410.3	98.4	56.0	76.9	**30.9***	26.7	28.3	20.6	41.1	269.9	2587.4

Kruskal-Wallis + Wilcoxon-tests (two sided): * p≤0.05; p**≤0.01 (Statistical unit= animal);

Table 5.7.1-26: Mean total motor activity counts [1/hour] of F_1-males and -females of cohort 1 and cohorts 2-4.

	Males				Females			
Group (cohort 1)	**0**	**1**	**2**	**3**	**0**	**1**	**2**	**3**
mg/kg bw/day (dams)	0	0.5	2	5	0	0.5	2	5
mg/kg bw/day (pups)	0	0.5	2	5	0	0.5	2	5
F_1 - cohort 1: Motor activity counts [total] / [% of control]								
PND 17	6219	6674 / 107	6078 / 98	- / -	6712	5118 / 76	6923 / 103	- / -
PND 21	3230	4228 / 131	4094 / 127	- / -	3399	3590 / 106	2630 / 77	- / -
Group (cohort 2-4)	**0**	**1**	**2**	**3**	**0**	**1**	**2**	**3**
mg/kg bw/day (dams)	0	0.5	2	5	0	0.5	2	5
mg/kg bw/day (pups)	0	0.25	0.25	-	0	0.25	0.25	-
F_1 - cohort 2-4: Motor activity counts [total] / [% of control]								
PND 17	6219	5401 / 87	5826 / 94	- / -	6712	6245 / 93	6453 / 96	- / -
PND 21	3230	3668 / 114	3311 / 102	- / -	3399	2403 / 71	2587 / 76	- / -

Table 5.7.1-27: Time course showing mean total motor activity counts of F_1-males and -females of cohort 1 on PND 17.



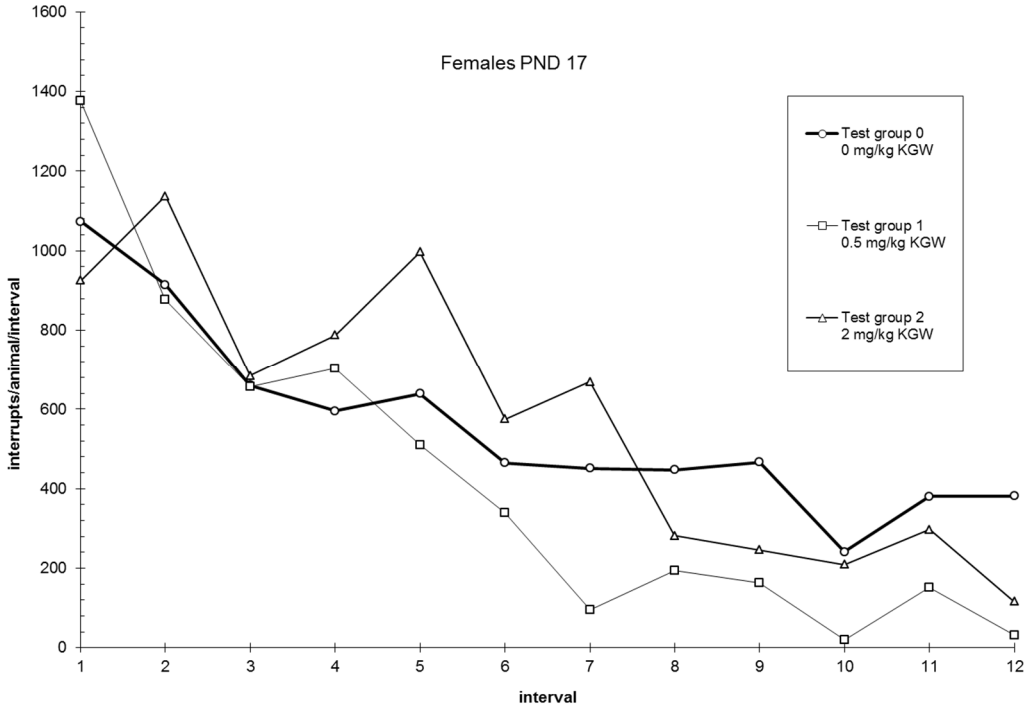
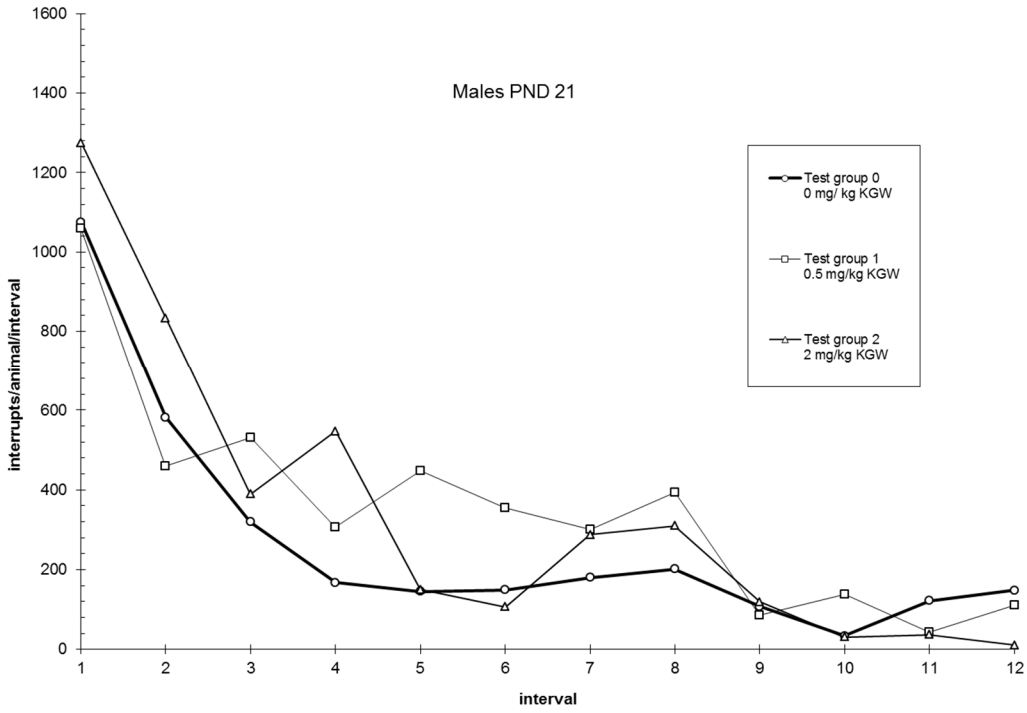


Table 5.7.1-28: Time course showing mean total motor activity counts of F_1-males and -females of cohort 1 on PND 21.



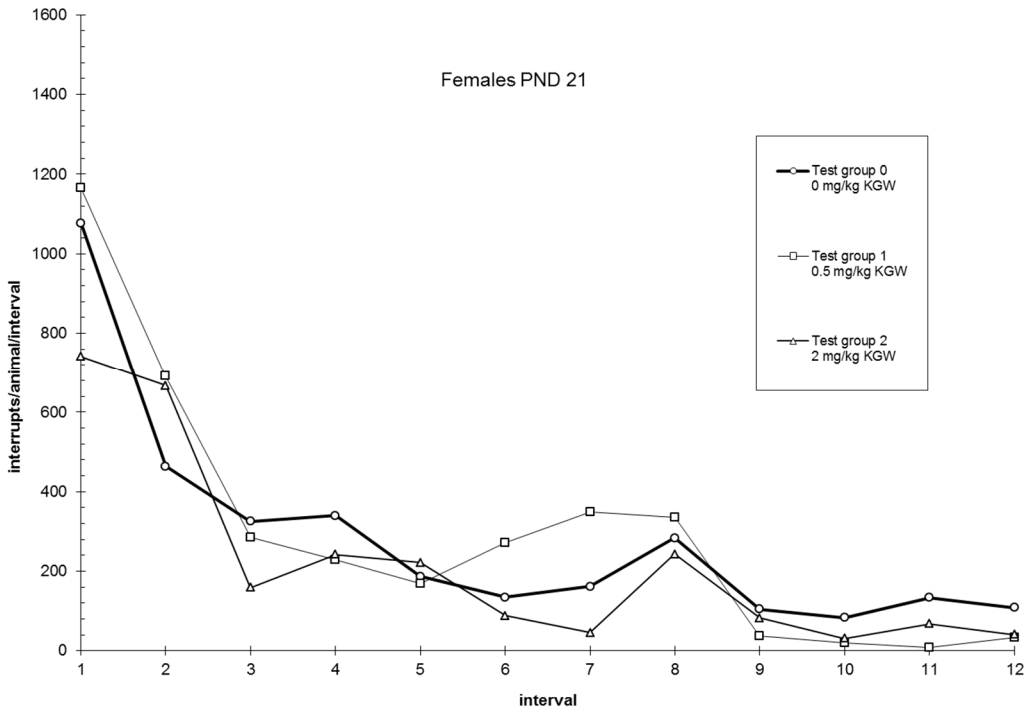
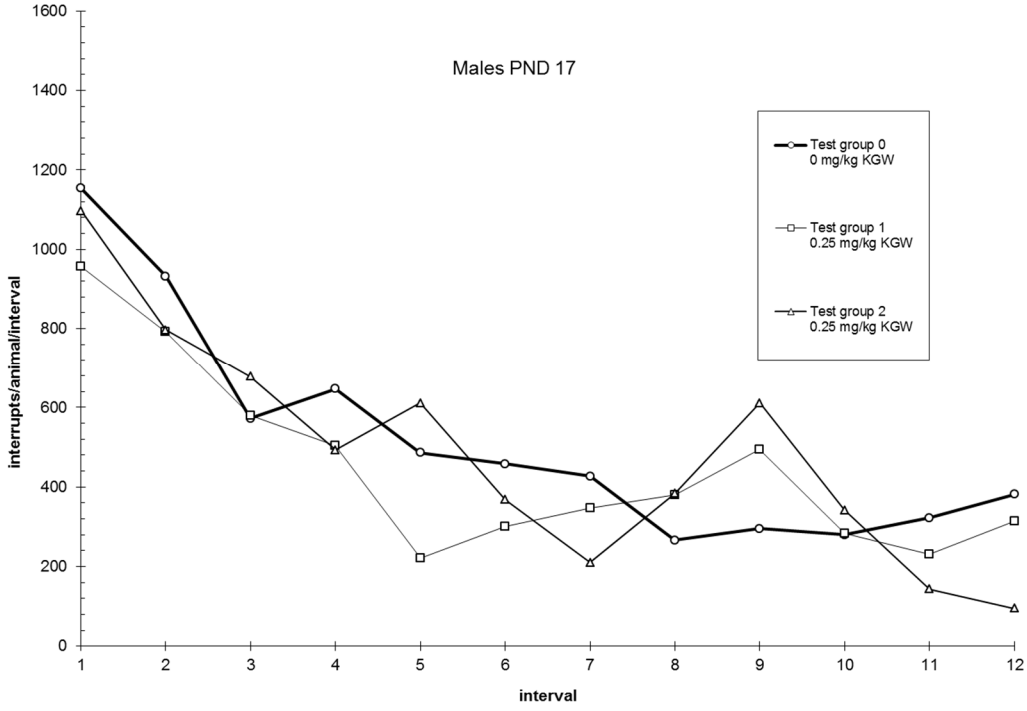


Table 5.7.1-29: Time course showing mean total motor activity counts of F_1-males and -females of cohort 2-4 on PND 17.



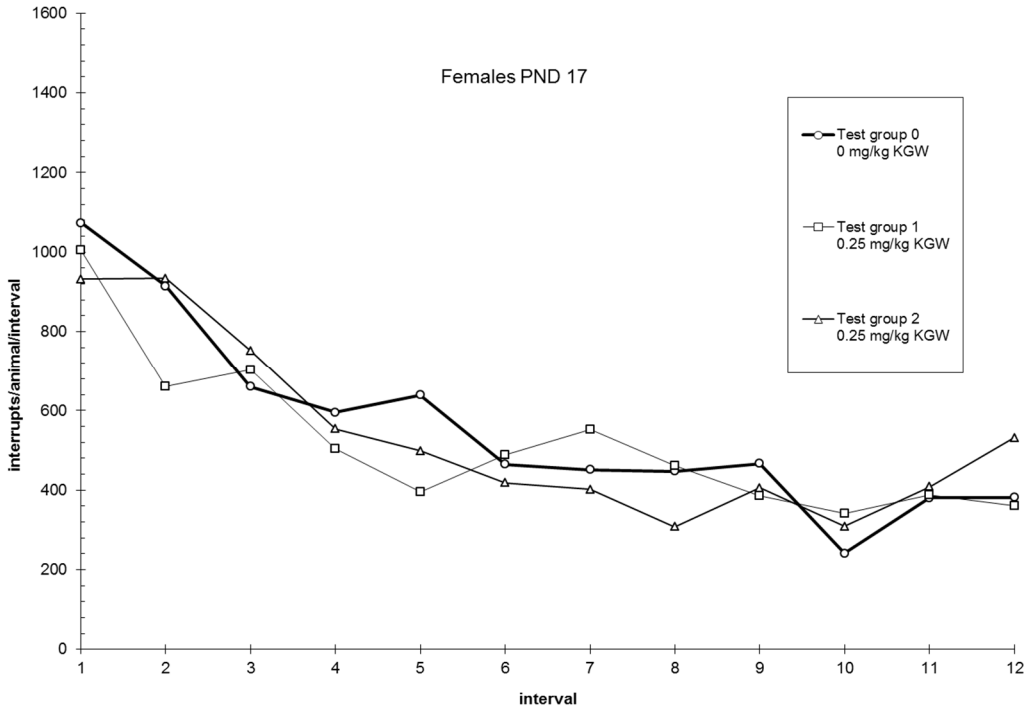
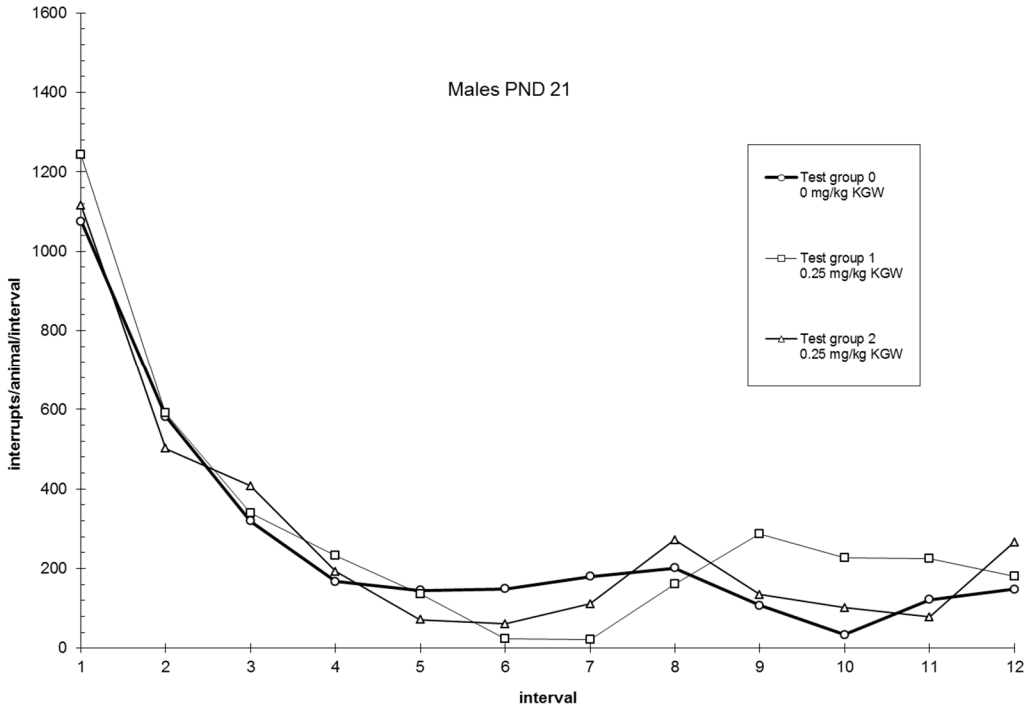
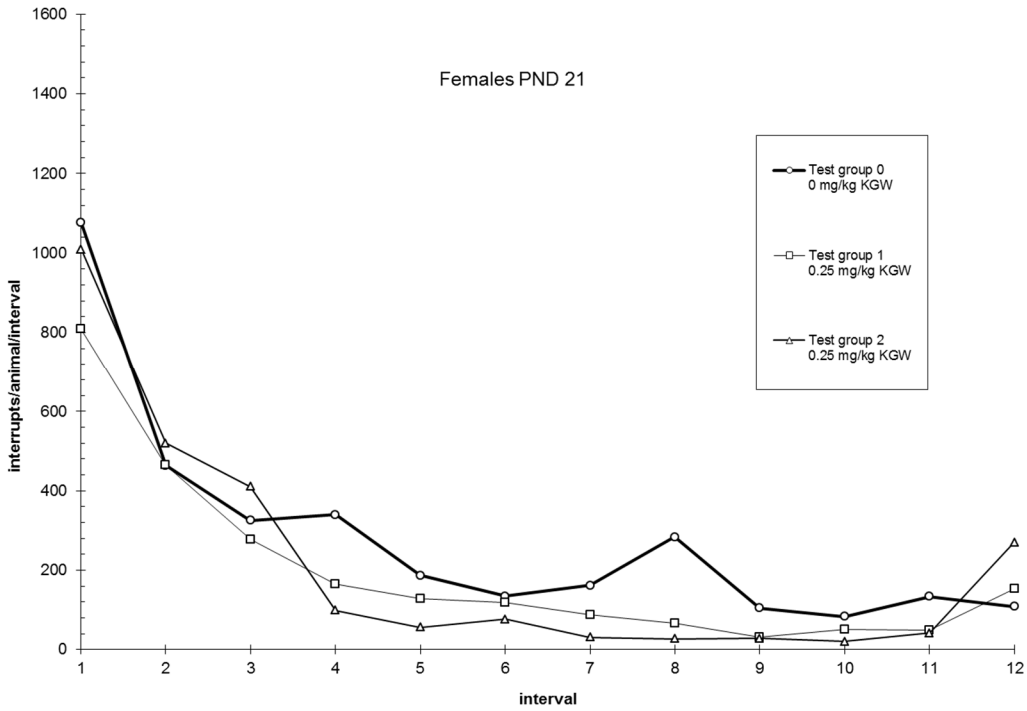


Table 5.7.1-30: Time course showing mean total motor activity counts of F_1-males and -females of cohort 2-4 on PND 21.





I. NECROPSY

1. Necropsy

Necropsy of dams on lactation day 21

No treatment related findings were observed.

Necropsy of Cohort 1 Pups on PND 21

In the high-dose F1 pups increased affected pups per litter rates were observed for gas bubble (31.2% vs. 0% in control), blood coagulum in renal pelvis (15.2% vs. 0% in control) and hemorrhagic testis (1.9% vs. 0% in control). This resulted in a higher rate of total pup necropsy observations (39.7% vs. 7.3% in control).

A few F1 control pups showed spontaneous findings at gross necropsy, such as incisors sloped, hemorrhagic thymus, empty stomach, dilated renal pelvis and fluid-filled thoracic cavity.

Table 5.7.1-31: Summary of Pup Necropsy Observations in cohort 1 pups

	Cohort 1			
Group	0	1	2	3
Dams & Pups (mg/kg bw/day)	0	0.5	2	5
~~**(mg/kg bw/day)**~~	~~0~~	~~0.5~~	~~2.0~~	~~5~~
Litters (pups) evaluated	22 (169)	6 (48)	6 (44)	6 (39)
Individual necropsy findings				
- Total pup necropsy observations				
- Fetal incidence [No. (%)]	12 (7.1%)	0	0	15 (38 %)
- Litter incidence [No. (%)] F	8 (36%)	0	0	5 (83%)
- Affected fetuses/litter (Mean ± SD) [%] W	7.3 ± 12.25	0	0	**39.7 ±27.75 ****
- Gas Bubble in stomach				
- Fetal incidence [No. (%)]	0	0	0	12 (31%)
- Litter incidence [No. (%)] F	0	0	0	**4 (67%)****
- Affected fetuses/litter (Mean ± SD) [%] W	0	0	0	**31.2 ± 30.87 %****
- Blood coagulum in renal pelvis				
- Fetal incidence [No. (%)]	0	0	0	5 (13%)
- Litter incidence [No. (%)] F	0	0	0	**4 (67%)****
- Affected fetuses/litter (Mean ± SD) [%] W	0	0	0	**15.2 ±15.1 % ****
- Hemorrhagic Testis				
- Fetal incidence [No. (%)]	0	0	0	1 (2.6%)
- Litter incidence [No. (%)] F	0	0	0	1 (17%)
- Affected fetuses/litter (Mean ± SD) [%] W	0	0	0	**1.9 ± 4.54***

Statistics: F= fisher's exact test; W= Wilcoxon-test, *: p<=0.05; ** = p<=0.01;

Necropsy of Cohort 2-4 Pups on PND 21
No treatment related findings were observed.

A few F1 pups showed spontaneous findings at gross necropsy. Incisors sloped, hemorrhagic thymus, empty stomach and fluidfilled thoracic cavity were seen in control animals only while dilated renal pelvis showed no relation to dosing. Diaphragmatic hernia was an isolated finding in one group 2 pub. Although the incidence is slightly above the historic control (pup incidence 0.7% vs. 0.5% HCD, Litter incidence 5.9% vs 4.2% HCD) the affected Pups/litter incidence is within the Historic control data (0.7% vs. 2.1% HCD). Furthermore this finding was not seen in Cohort 1 animals despite higher dose levels. Therefore all these findings were not considered to be associated with the test substance.

2. Gross lesions

No gross lesions were observed in Cohort 1 and Cohort 2-4 pups.

3. Organ weight

Neither cohort 1 nor cohort 2-4 pups showed significant differences of brain weight or brain to body weight when compared to the control group (see Table 5.7.1-38).

Table 5.7.1-32: Absolut and relative Brain Weights of Cohort 1 and Cohort 2-4 pups

Sex		Males					Females				
Organ weight	Dose Dam/Pup [mg/kg bw]	Absolute weight	Δ%	Relative weight [% of bw]	Δ% #	Note	Absolute weight	Δ%	Relative weight [% of bw]	Δ% #	Note
Cohort 1; 3 pups											
Terminal weight [g]	0	51,667					45,9				
	0,5/0,5	49,833	(-4)				49,8	(+8)			
	2,0/2,0	46,467	(-10)				49,933	(+9)			
Brain [g]	0	1,68		3,252			1,637		3,639		
	0,5/0,5	1,717	(+2)	3,445	(+6)		1,633	(±0)	3,284	(-10)	
	2,0/2,0	1,743	(+4)	3,804	(+17)		1,637	(±0)	3,295	(-9)	
Cohort 2-4; 7 pups											
Terminal weight [g]	0	51,971					49,414				
	0,5/0,25	53,771	(+3)				45,514	(-8)			
	2,0/0,25	52,457	(+1)				49,543	(±0)			
Brain [g]	0	1,62		3,13			1,556		3,163		
	0,5/0,25	1,649	(+2)	3,077	(-2)		1,571	(+1)	3,487	(+10)	
	2,0/0,25	1,647	(+2)	3,156	(+1)		1,631	(+5)	3,299	(+4)	

Statistics: Kruskal-Wallis and Wilcoxon test (two-sided): *: p<=0.05; ** = p<=0.01;

4. Length and width of brain

No statistical differences were found between control and treated pups regarding brain measurements of width and length, neither for Cohort 1 nor Cohort 2-4 pups (see Table 5.7.1-33).

Table 5.7.1-33: Brain width and length of Cohort 1 and Cohort 2-4 pups

Brain measurement		Males				Females			
	Dose Dam/Pup [mg/kg bw]	Length	Δ%	Width	Δ%	Length	Δ%	Width	Δ%
Cohort 1;	0	1,848		1,499		1,835		1,482	
	0,5/0,5	1,843	±0%	1,503	±0%	1,840	±0%	1,497	+1%
	2,0/2,0	1,860	+1%	1,500	±0%	1,850	+1%	1,443	-3%
Cohort 2-4	0	1,848		1,499		1,835		1,482	
	0,5/0,25	1,817	-1%	1,489	+2%	1,817	-1	1,457	**-2%**
	2,0/0,25	1,859	±0%	1,503	+1%	1,844	+1	1,479	±0%

Statistics: Wilcoxon test with Bonferoni-Holm-Adjustment (one-sided): *: p<=0.025; ** = p<=0.005;

5. Histopathology / Neurohistopathology

Neurohistopathology was not investigated as this was not affected in the DNT studies with Beta- and Zeta-cypermethrin.

J. Analytical determination of BAS 310 I in PND 4 and PND 10 pups before treatment

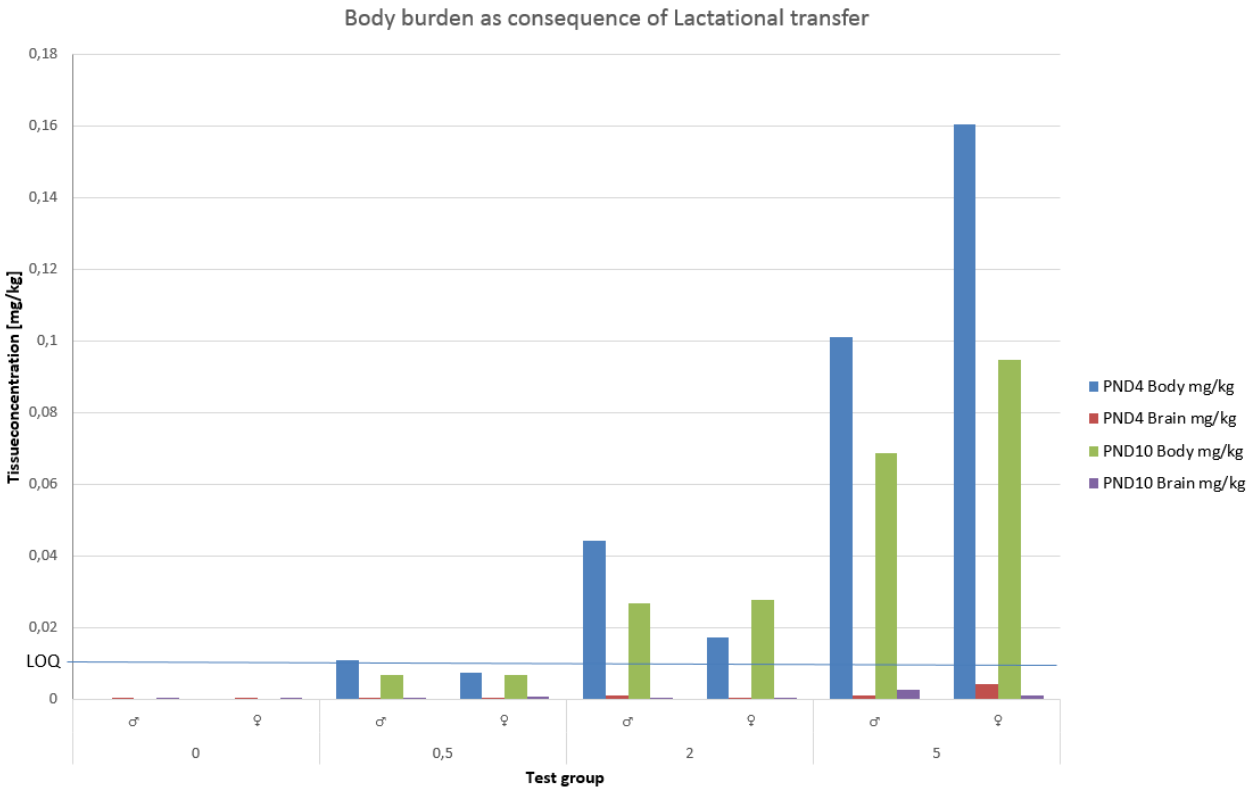
The residues of alpha-cypermethrin in the pups brain and residual body at PND 4 and PND10 are summarized in Table 5.7.1-39 given as mg alphacypermethrin per kg tissue. The measurement was done before the direct pup dosing started, therefore the Cohort 1 and 2-4 animals could be taken together as they were up to this timepoint treated equally. However, due to the termination of the high dose group after excessive toxicity seen in cohort 1 pups, there were less animals available for high dose investigations. Therefore, the high dose group at PND 10 consists only of 3 instead of 10 animals /sex, while the PND4 groups were determined by the number of superplus animals.

Table 5.7.1-34: Concentration of BAS 310 I in brain and body of pups

Maternal Dose [mg/kg bw/day]	Sex of pups	PND4			PND10		
		Number of pups	Body [mg/kg]	Brain [mg/kg]	Number of pups	Body [mg/kg]	Brain [mg/kg]
0 Group 0	♂	3	nd	0.000	10	nd	0.000
	♀	4	nd	0.000	10	nd	0.000
0.5 Group 1	♂	6	0.011	0.000	10	0.007	0.000
	♀	7	0.007	0.000	10	0.007	0.001
2 Group 2	♂	6	0.044	0.001	10	0.027	0.000
	♀	4	0.017	0.000	10	0.028	0.000
5 Group 3	♂	2	0.101	0.001	3	0.069	0.003
	♀	1	0.160	0.004	3	0.095	0.001

nd: < LOD; LOD= 0.0002 mg/kg for rodent brain and 0.001 mg/kg for rodent body ; LOQ=0.01 mg/kg;

Figure 5.7.1-7: Concentration of BAS 310 I in brain and body of pups PND 4 and PND 10



Alpha-cypermethrin was found at PND 4 and PND10 mainly in the **carcass specimen**.
The mean body burden at **PND4** was with one exception (low dose female pups) above the limit of quantification (10 µg/kg) and increased with the maternal dose up to values of 101/160 µg/kg tissue in the high dose pups without inducing any effects in the pups. The value of 160 µg/kg tissue is however based on one single female carcass and was with regard to the more conservative male value not used for NOAEL calculations. On **PND 10**, 2 days after the last maternal dosing, the carcass still exhibited measureable amounts of alpha-cypermethrin. The body burden with 69 / 95 µg/kg tissue in high dose males and female pups, respectively, was slightly lower compared to the level found at PND 4 (about 60-70%). The low dose pups on PND10 exhibited 7 µg/kg tissue, which is a value slightly below the LOQ used for LOAEL calculations below.

Only very low amounts of alpha-cypermethrin were detectable in **brain specimens** at PND 4 and PND 10, all below the limit of quantification, with the highest value found in the single high dose female pup at PND 4 with 4 µg/kg tissue.

No detectable isomerization of alpha-cypermethrin (Cis II) could be observed in all specimens.

A bioavailability of 46% is considered in order to transfer these body burden values into an external dose. The PND 4 level detected in high dose male pups (101 µg/kg tissue) is considered the systemic no observed effect levels, while the PND 10 low dose level (7 µg/kg tissue) was sufficient to lead to acute neurotoxicity in 21.2% of the pups after direct treatment with 250 µg/kg free alphacypermethrin in CMC. This ends up in a NOAEL of 220 µg/kg bw (101µg/kg / 46%) and a LOAEL of 265 µg/kg bw (7 µg/kg / 46% +250 µg/kg).

The robustness of this LOAEL calculation is questioned by the one high dose female at PND 4 which was free of any clinical signs despite a body burden of 160 µg alphacypermethrin/kg tissue, representing an calculated external dose of 350 µg/kg bw (160 µg/kg / 0,46). However, it demonstrates the conservatism of this calculation considering the Dose Response. Only 21.2% of the pups showed slight clinical signs, thereof 10.6% slight tremors after administration of 250 µg/kg bw to the low dose pups (external dose: 265 µg/kg bw). 44-45 % of the pups showed effects after administration of 250 µg/kg bw to the mid dose animals or 500 µg/kg bw to the low dose animals (external dose: 308-515 µg/kg bw). The effects increased with regard to the incidence of tremors from 29 % to 41% and in regard to severity from up to moderate to up to severe tremors. 100 % of the animals showed clinical sings in the mid dose after administration of 2000 µg/kg bw (external dose: 2059 µg/kg bw). Therefore, the high dose female pup with an systemic dose of 160 µg/kg bw (external dose of 350 µg/kg bw) can be considered as one animal of the 50% less sensitive animals in this dose range (perhaps due to a higher amount of fat or due to a higher metabolic capacity).

In summary, the detection of alpha-cypermethrin in pups at PND 4 and 10 prior to any direct contact to alpha-cypermethrin is the proof of lactational transfer. The availability of alpha-cypermethrin in pups 48 hours after the last maternal dosing indicates a rather slow elimination at least from some tissue compartments and a still ongoing lactational transfer. The extremely low amounts in brain correlates with the lack of adverse effects in pups during lactational exposure. Converting the systemic dose based on the bioavailability of 46% ends up in a NOAEL of 220 µg/kg bw (101µg/kg / 46%) while the LOAEL is 265 µg/kg bw (7 µg/kg / 46% +250 µg/kg).

III. CONCLUSIONS

The administration of BAS 310 I (alpha-cypermethrin) to pregnant female Wistar rats by oral gavage from gestation day (GD) 6 to postnatal day (PND) 8 at doses of 0.5, 2, or 5 mg/kg bw induced no clinical or neurotoxic signs in the dams or the offspring. The analytical processing of the pups on PND 4 and PND 10 confirmed that the body burden of the pups correlates with the maternal dose and that a two-days treatment-free interval between the last exposure of dams and the first exposure of offspring is obviously not enough to eliminate the test substance completely.

However, lactational exposure up to PND 10 did not induce adverse effects in pups up to a maternal dose of 5 mg/kg bw, which resulted in a pup body burden of up to 101/160 µg/kg bw on PND4 in male and female pups, respectively. Considering PND4 as at least similar sensitive as later time points based on the increasing metabolic capacity, 101 µg/kg bw can be considered as systemic NOAEL.

Substance administration via direct pup dosing from PND10 to PND21 induced acute treatment-related mortality at 2 and 5 mg/kg bw/day, while signs of neurotoxicity were observed down to 0.5 mg/kg bw/day (tremors, tonic-clonic convulsions, excessive grooming and twitching, together with hypothermia, lateral position and vocalization at the mid and high dose).
A 50% reduction of the pup dose to 0.25 mg/kg bw/day while remaining the pre-treatment of dams until PND 8 at 0.5 and 2 mg/kg bw/day resulted in similar signs of neurotoxicity, however reduced in incidence and severity.

Apart from the clinical symptoms, pup development was rather unaffected in the surviving animals in all test groups as shown by unchanged pup weights/weight gain and absence of necropsy findings. Motor activity measurements did not reveal any significant treatment-related changes. Regarding neuropathology, no treatment-related findings in brain weights, gross pathology and length and width measurements of the brain were found in any of the groups.

The analytical investigations of PND 10 pups after the two-days treatment-free interval still revealed a body burden of 7 µg/kg bw in low dose pups. This internal body burden, corrected for bioavailability of 46%, together with the external dose of 250 µg/kg bw can be considered as **calculated LOAEL at 265 µg/kg bw/day**. Considering the internal dose of 101 µg/kg bw at PND4 as systemic NOAEL, and corrected for bioavailability of 46% ends up in a **calculated NOAEL of 220 µg/kg bw**.

Altogether, the data show that there occurs exposure via milk which still leads to measureable amounts of alphacypermethrin after a two-days treatment-free interval. However, direct pup dosing is an extreme method for pre-weanling pups, and of no direct relevance to the overall human or ecotoxicological risk assessment. Therefore, as discussed in the "Reasoning for the appropriateness of dietary DNT studies for alpha-cypermethrin" (see above) this study is not used for the human risk assessment but the dietary developmental neurotoxicity study with zeta-cypermethrin is considered adequate.

Report: CA 5.7.1/4
[redacted] 1994a
WL85871 (Fastac): A 90 day dietary toxicity study in the rat - Volume 1 of 3
AL-425-007
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Executive Summary

The aim of this study was to determine the effect of WL85871 (BAS 310 I) on neurotoxicity to rats after administration via the diet for 90 days. The test substance was administered to groups of 15 male and female Crl:CD(SD)BR rats at dietary concentrations of 50, 250, and 500 ppm. Clinical signs were recorded daily and functional observational battery (FOB) and measurements of motor activity, forelimb and hindlimb gripstrength and hindlimb landing footsplay were made before the start of dosing and in weeks 4, 8 and 13. At termination, 10 animals underwent a conventional necropsy and the remaining animals were subjected to whole body glutaraldehyde perfusion fixation in order to investigate central and peripheral nervous system tissues, eyes and muscle.
The administration of WL85871 at a dietary level of 500 ppm had an adverse effect on bodyweight gain and food intake and was associated with minor reductions in red blood cell parameters (RBC, HGB, HCT) and plasma total protein. Although relative liver weight was increased in males treated at this dose level, there was no microscopic evidence of hepatotoxicity and this response was therefore considered to be an adaptive one. Relative kidney weight was also slightly increased, but again, in the absence of morphological changes, this response and the not strictly dose related increases in blood urea nitrogen seen at 500 ppm and lower dose levels, were considered to be of no toxicological significance. At 250 ppm possible effects of treatment were limited to a transient reduction in food intake and slight reductions in red blood cell parameters and total plasma protein. No treatment related alterations in neurobehavior (FOB), motor activity or hind limb landing foot splay were observed. Lower forelimb grip strength in females fed 500 ppm at week 4 was considered attributed to a lower body weight.
In conclusion, the no observed effect level for neurotoxicity was concluded to be 500 ppm whereas that for general toxicity was 50 ppm WL85871.

(DocID 1994/7001607)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin (FASTAC, WL85871)

Description:	solid (powder) / white
Lot/Batch #:	02156
Purity:	95.6%
Stability of test compound:	The test substance was stable in the diet at room temperature for a period of 4 weeks, as determined in an earlier experiment with FASTAC (Report SBTR.93.002).

2. Vehicle and/or positive control: None

3. Test animals:

Species:	Rats
Strain:	Crl:CD(SD)BR
Sex:	Male and female
Age:	35 days (start of administration)
Weight at dosing (mean):	males: 208 g females: 173 g
Source:	Charles River Laboratories, Manston, UK
Acclimation period:	7 days
Diet:	pelleted diet (Rat and Mouse Maintenance Diet No. 1, Expanded, Special Diets Services)
Water:	water, ad libitum
Housing:	in groups (5 animals per cage) during acclimatization and single housing during the rest of the study period in grid bottomed polypropylene cages (pattern RB3R North Kent Plastics, Dartford)
Environmental conditions:	
Temperature:	19 - 23°C
Humidity:	45 - 70%
Air changes:	NA
Photo period:	12 h light / 12 h dark (06:00 - 18:00 / 18:00 - 06:00)

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: 06-May-1993 - 23-Mar-1994
(In life dates: 06/07-May-1993 (start of administration)
to 05/13-Aug-1993 (necropsy)

2. Animal assignment and treatment:
WL85871 was administered to groups of 15 male and 15 female mice at dietary concentrations of 0, 50, 250 and 500 ppm for at least 90 days.

3. Test substance preparation and analysis:
The test substance is applied via the oral route in the diet. Three batches of the substance preparations were prepared before the beginning of the study and twice during the study to assure stability of test substance preparations. For each dietary concentration separately, a weighed amount of test article was dissolved in acetone and poured over 50 kg of diet. The diet was mixed for 2 minutes in a high speed mixer. Control diet had an equivalent volume of acetone added (5 mL/kg) and was also mixed for 2 minutes.
The stability of the test substance in the diet at room temperature for a period of weeks was determined in an earlier experiment. Homogeneity was re-confirmed by taking sextuplicate samples in a semi-random manner from each of the first batch of treated diets prepared.
Concentrations of treated diets were assessed on a second occasion in this study by analysis of duplicate samples of the third batch of diets prepared. For control diet, a single sample of each of the first and third batches prepared was analysed. HPLC (all concentrations) and GC (50 and 250 ppm) analysis were performed.

4. Statistics:
For neurobehavioural responses the analyses were designed to identify effects in treated groups, relative to controls, taking into account changes over time. Analyses were first applied to test for differences in profiles over time for the various treatment groups. Secondly, differences between groups at each of the time points were tested so as to ascertain when in the experiment differences first became statistically significant.
Means and standard deviations (S.D.) of each test group were calculated for several parameters.

C. METHODS

1. Observations:
The animals were examined for clinical changes, for abnormal pattern of behavior and for gross deviations in food and water consumption and mortality twice daily on working days and once daily on weekends and public holidays.

2. Body weight:
Body weight was determined before the start of the administration period and and thereafter in weekly intervals.

3. Food consumption and compound intake:
Individual food consumption was determined in weekly intervals.

4. Clinical assessment of neurotoxicity:
Observations and activity measurements were made before the start of dosing and again in weeks 4, 8 and 13. These observations were applied to 10 male and 10 female animals and consisted of a Functional Observation battery (FOB), forelimb and hindlimb grip strength, a hindlimb landing footsplay test, and a motor activity assessment.

5. Hematology and clinical chemistry:
Blood samples were taken by orbital-sinus puncture from 10 males and 10 females of each group in week 12 after an overnight fasting.
The following hematological and clinical chemistry parameters were determined for all animals:

Hematology:		
Red blood cells	*White blood cells*	*Clotting Potential*
✓ Erythrocyte count (RBC)	✓ Total leukocyte count (WBC)	NA
✓ Hemoglobin (Hb)	✓ Differential blood count	
✓ Hematocrit (Hct)	✓ Platelet count (PLT)	
✓ Mean corp. volume (MCV)	✓ Plateletcrit	
✓ Mean corp. hemoglobin (MCH)	✓ Mean platelet volume	
✓ Mean corp. Hb. conc. (MCHC)	✓ Platelet distribution width	
✓ Reticulocytes (RET)		
✓ Red cell distribution width		

Clinical chemistry:		
Electrolytes	*Metabolites and Proteins*	*Enzymes:*
✓ Calcium	✓ Albumin	✓ Alanine aminotransferase (ALT)
✓ Chloride	✓ Albumin/globulin ratio	✓ Aspartate aminotransferase (AST)
✓ Phosphorus (inorganic)	✓ Bile acids	✓ Alkaline phosphatase (ALP)
✓ Potassium	✓ Bilirubin (total)	✓ γ-glutamyl transpeptidase (γ-GT)
✓ Sodium	✓ Cholesterol	
	✓ Creatinine	
	✓ Glucose	
	✓ Protein (total)	
	✓ Triglycerides	
	✓ Urea nitrogen	

6. Urinalysis:
For urinalysis the individual animals were transferred to metabolism cages during week 11 and urine was collected overnight from 10 males and females. No food or water was supplied during urine collection.

The following quantitative or semi-quantitative parameters were determined for all animals:

Urinalysis					
	Quantitative parameters:		*Semi quantitative parameters*		
✓	Urine volume	✓	Bilirubin	✓	Protein
✓	Osmolality	✓	Hemoglobin	✓	pH-value
		✓	Color and turbidity	✓	Urobilinogen
		✓	Glucose	✓	Sediment (microscopical exam.)
		✓	Ketones		

7. Ophthalmoscopy:
Both eyes of all animals were examined before the start of treatment and those of all control and high dose animals were examined again in week 12. Examinations were made using an indirect ophthalmoscope after instillation of a mydriatic agent.

8. Sacrifice and pathology:
Five males and five females of each group were assigned to while body perfusion fixation and the remainder were assigned to conventional necropsy and immersion fixation.

Perfusion fixation:
The animals were fasted before necropsy. They were each anaesthetized with an intraperitoneal injection of sodium pentobarbitone and the heart was exposed to allow incision in both the left ventricle and right atrium. Heparinised saline was first infused into the aorta via the left ventricle, followed by infusion of 2.5% buffered glutaraldehyde. There was no macroscopic examination of internal organs or organ weighing in these animals. Carcasses of perfused animals were stored refrigerated for at least one hour before the tissues listed overleaf were dissected into fresh fixative.

The following organs were examined after perfusion fixation:
- Brain (6 transverse slices taken between the frontal lobes and the medulla oblongata)
- Eyes with optic nerve (left and right)
- Sciatic nerve (right and left*, proximal and distal tibial, longitudinal and transverse)
- Skeletal muscle (left, femoral, gastrocnemius with sural)
- Spinal cord (cervical, thoracic and lumbar, longitudinal and transverse)
- Spinal ganglia (cervical and lumbar)
- Sural nerve (right and left, longitudinal and transverse)

*taken for resin embedding

Immersion fixation:
The animals were fasted before necropsy. They were a lethal intraperitoneal injection of sodium pentobarbitone, weighed and then exsanguinated.
Post mortem examination included physical examination of the external body surface, all orifices, carcass, cranial cavity, thoracic, abdominal and pelvic cavities with their associated organs and tissues and the neck with its associated organs and tissues.

The following organs were sampled, weighed and examined histopathologically after immersion fixation:

Pathology:

The following organs were collected (column C), weighed (W) and examined histopathologically (H, ✓: all groups, #: control and top dose).

C	W	H		C	W	H		C	W	H	
✓	✓	#	adrenals	✓	✓	#	kidneys			#	skeletal muscle
✓		#	aorta								skin
✓		#	bone[§]					✓		#	spinal cord
✓	✓	#	brain	✓	✓	#	liver	✓	✓	#	spleen
✓		#	caecum	✓		#	lung				sternum w. marrow
✓		#	cervix	✓		#	lymph nodes[#]	✓		#	stomach (fore- & glandular)
✓		#	colon								
✓		#	duodenum	✓		#	mammary gland	✓	✓	#	testes
✓		#	epididymides				nose/nasal cavity	✓	✓	#	thymus
								✓		#	tongue
✓			esophagus	✓	✓	#	ovaries	✓		#	thyroid/parathyroid
✓		#	eyes (with optic nerve)	✓		#	pancreas	✓		#	trachea
			gall bladder				pharynx	✓		#	urinary bladder
✓		✓	gross lesions	✓		#	pituitary gland	✓		#	uterus
✓		#	Harderian glands	✓		#	prostate				vagina
✓	✓	#	heart	✓		#	rectum				
				✓		#	salivary gland (sub maxillary)		✓		body (anesthetized animals)
✓		#	ileum	✓		✓	Sciatic nerve (proximal and distal – tibial)				
✓		#	jejunum	✓		#	seminal vesicles				

[§] from knee joint, femur, sternum, 3 x lumbar vertebrae; [#]mesenteric and submandibular

II. RESULTS AND DISCUSSION

A. TEST SUBSTANCE ANALYSES

Diets were found to be homogenous and of correct concentrations (within 7% of nominal).

B. OBSERVATIONS

1. Clinical signs of toxicity

No treatment related clinical signs of toxicity were reported. Those signs that were observed, generally minor skin lesions such as occasional scabs or fur thinning, were seen in control animals also and represented typical background clinical signs.

2. Mortality

No animal died during the study.

C. BODY WEIGHT AND BODY WEIGHT GAIN

Bodyweight gain was not adversely affected in either sex treated at 50 ppm or, in males treated at 250 ppm. Females dosed at 250 ppm showed marginally lower weight gain than controls from the onset of treatment. Over the last few weeks of the study the difference in bodyweight between these groups became statistically significant, with females treated at 250 ppm having a terminal bodyweight 7% lower than the control value.

A more marked reduction in weight gain was noted in both sexes at 500 ppm. In the first week of treatment bodyweights were 13% and 10% lower in males and females, respectively, when compared to the control. Weights were consistently lower than the control values throughout the study with reductions in mean final bodyweight of 8% and 9% for males and females, respectively.

Table 5.7.1-35: Mean body weight of rats administered WL85871 for 90 days

	Males				Females			
Dose level [ppm]	0	50	250	500	0	50	250	500
Body weight [g]								
- Week 0	207.6	213.6	210.3	208.9	173.1	177.7	174.0	172.0
- Week 1	265.8	270.0	254.8	231.7** (87.2%)	203.4	207.8	195.4	182,2** (89.6%)
- Week 7	456.8	466.6	454.6	412.2** (90.2%)	293.8	300.2	278.9	262.1** (89.2%)
- Week 9	488.0	496.8	488.3	446.1** (91.4%)	311.3	317.1	291.5* (93.6%)	277.9** (89.3%)
- Week 13	519.3	530.3	523.4	477.0** (91.9%)	323.0	332.8	301.3	293.2** (90.8%)

* = p≤0.05; ** = p≤0.01

D. FOOD CONSUMPTION AND COMPOUND INTAKE

No differences of food intake were observed in the 50 ppm group. At 250 ppm a moderate reduction of food intake of 16% and 11% during the first week was observed, but was similar to control in the following weeks. This pattern was considered consistent with the introduction of an unpalatable diet. At 500 ppm a moderate reduction of food intake of during the first week was observed, lasting until the last 2 (males) or 4 (females) weeks of the study, when differences were marginal and no longer statistically significant.

The calculated average test article intake at 50, 250 and 500 ppm over the study was 3.7, 17.9 and 36.1 mg/kg bw/day for males, and 4.2, 21 and 42 mg/kg bw/day for females.

E. CLINICAL ASSESSMENT OF NEUROTOXICITY

The FOB yielded no statistically significant alterations in animals given 50 ppm. Isolated observations (number of defaecations, faecal consistency, startle/"click" response, number of pools of urine passed) were seen, but were not dose related, not consistent for both sexes, and were only significant at the $p<0.05$ level, and given the large number of observations made, may have arisen purely by chance. It is concluded, therefore, that there were no toxicologically significant neurobehavioural differences manifested in the FOB.

Furthermore, there were no alterations in forelimb or hindlimb gripstrength for either sex at 50 or 250 ppm or for males dosed at 500 ppm. Females of the high dose group had significantly lower forelimb grip strength at week 4, but was normal at week 8 and 13. Overall hindlimb grip strength was also lower ($p<0.05$) than that of the controls but, when individual occasions were considered, no significant differences were apparent. Since females dosed at 500 ppm were lighter animals than controls, this initial difference in grip strength may simply have reflected the reduced bodyweight gain.

There were no significant intergroup differences in hindlimb landing footsplay.

No treatment related alterations in motor activity were observed. Although the total number of movements made by females treated with 50 ppm was significantly lower than the control values at week 4, the lack of dose response relationship as well as the between occasion variation in control values suggested that this single significant finding was fortuitous.

Table 5.7.1-36: Mean total number of movements for females

	Week number			
Concentration [ppm]	-1	4	8	13
0	140	361	377	266
50	191	242*	341	255
250	220	443	492	329
500	197	323	299	343

F. BLOOD ANALYSIS

1. Hematology

Males treated at 250 or 500 ppm had marginally (≤5%) lower mean red blood cell counts, haemoglobin concentrations and haematocrits than control animals. These minor differences, although statistically significant, were not associated with abnormal cell types or with alterations in other haematological parameters.

The minimal (3%) reduction in haemoglobin concentration alone in males dosed at 50 ppm was considered to be of no toxicological significance.

There were no significant haematological variations in female groups.

Table 5.7.1-37: Group mean haematology data in male animals at week 13

Concentration [ppm]	RBC [10E12/L]	HGB [g/dL]	HCT [%]
0	9.03	16.4	50.8
50	8.69	15.9*	49.4
250	8.63*	15.8*	48.9*
500	8.56*	15.7**	48.8*

* = p≤0.05; ** = p≤0.01

2. Clinical chemistry findings

With the exception of a raised mean blood urea nitrogen level in females, there were no treatment related alterations in blood chemistry at the 50 ppm dietary level.

In males dosed at 250 or 500 ppm and in all treated female groups, blood urea nitrogen levels were moderately raised, but not in a dose related manner. There was no accompanying increase in plasma creatinine or significant changes in concentration of plasma electrolytes to suggest a nephrotoxic effect. Furthermore, there were no histopathological changes related to treatment. The increases in BUN were therefore considered not related to treatment.

The plasma total protein levels of females (250 and 500 ppm) were slightly (≤6%) but significantly lower than the control values. For high dose females this reduction may ave reflected the lowered food intake of this group.

The apparent decreases in plasma bilirubin (females) and alkaline phosphatase (males) noted at 500 ppm were considered to be of no diagnostic significance since a rise in these markers is normally sought as evidence of hepatic dysfunction.

Table 5.7.1-38: Group mean clinical chemistry data at week 13

Concentration [ppm]	Urea N. [mmol/L]	Protein [g/L]	Calcium [mmol/L]	Alkaline phosphatase [I.U./L]	Bilirubin [µmol/L]
Males					
0	5.2	65.1	2.70	248	2.0
50	5.4	64.8	2.72	259	2.0
250	6.0*	65.2	2.72	212	2.0
500	5.8*	64.4	2.72	212	2.0
Females					
0	5.4	71.1#	2.80	175	1.9
50	7.0**	68.2	2.77	147	2.0
250	7.0**	67.5*	2.79	175	1.8
500	6.9**	67.0*	2.72*	139	1.5*

* = p≤0.05

** = p≤0.01; # = F19 excluded for statistical reasons

G. URINALYSIS

There were no toxicologically significant changes in urine cellularity or composition. Slight variations in urinary osmolality, pH and glucose in females of the 50 ppm dose group were considered spontaneous as no dose response was observed.
An apparent fall in urinary triple phosphates in males dosed 500 ppm was of doubtful toxicological significance.

H. Ophthalmoscopy

There were no ocular changes or abnormalities that could be attributed to treatment.

I. NECROPSY

1. Organ weight

When compared to the control group (set to 100%) the mean absolute weights were not significantly changed (see Table 5.7.1-39). Expressed as a percentage of bodyweight, a number of organs of animals treated at 500 ppm showed significant weight variations. The increase in relative testes weight noted in males was considered to be a direct effect of growth retardation. Not attributable to reduced bodyweight were the slight increases in relative kidney weight and the moderate increase in relative liver weight in males.

Table 5.7.1-39: Group mean relative organ weight data (% of bodyweight) for those organs showing statistically significant variations.

Concentration [ppm]	Brain	Kidneys	Liver	Adrenals	Testes
Males					
0	0.44	0.61	2.56	0.011	0.71
50	0.43	0.60	2.69	0.010	0.73
250	0.44	0.61	2.72	0.012	0.72
500	0.48*	0.67*	2.91**	0.012*	0.83*
Females					
0	0.67	0.64	2.79	0.024	-
50	0.64	0.62	2.72	0.023	-
250	0.72	0.65	2.88	0.027	-
500	0.72	0.69*	2.97	0.027	-

* = p≤0.05; ** = p≤0.01

2. Gross lesions

No macroscopic lesions were recorded.

3. Histopathology

There were no microscopic changes to account for the increased liver and relative kidney weights at 500 ppm and no evidence of neuropathological changes or other microscopic changes associated with the test substance.

III. CONCLUSIONS

The administration of WL85871 induced no treatment-related changes in FOB, motor activity or hindlimb landing foosplay. Changes in forelimb gripstrength were considered related to lower body weight of the 500 ppm females. Macroscopic and microscopic investigations revealed no neuropathological lesions at any dose.
At a dietary level of 500 ppm bodyweight gain and food intake were reduced and minor reductions in red blood cell parameters (RBC, HGB, HCT) in both sexes and total plasma protein was slightly lowered. Although relative liver and kidney weight was increased in males treated at this dose level, there was no microscopic evidence of morphological changes, both these responses were considered adaptive and not of toxicological relevance. Moderately but not strictly dose related increases in blood urea nitrogen seen in all females and in males at 250 and 500 ppm were considered to be not treatment related based on missing accompanying increases in parameters that suggest a nephrotoxic effect.
At 250 ppm effects of treatment were limited to a transient reduction in food intake and slight reductions in red blood cell parameters and total plasma protein.

The no observed effect level for neurotoxicity was concluded to be 500 ppm, which is equivalent to 36.1 and 42 mg/kg bw/day for males and females, respectively, whereas that for general toxicity was concluded to be 50 ppm WL85871 (3.7 / 4.2 mg/kg bw for males / females, respectively).

CA 5.7.2 Delayed polyneuropathy studies

As alpha-cypermethrin does not belong to a chemical class suspected to induce delayed neuropathies, no study is considered to be necessary and thus no further study was conducted.

EU Dossier update (Oktober 2015) contains data which were not present at the dossier submission in February 2015, these are: For the studies discussed in chapter CA 5.8.1/37 & 38 concerning the metabolite M310I011 (PBA) separate analytical reports (Doc ID 2015/1029517, Doc ID 2015/1003984 & 2015/1032402) are now available and the data are highlighted.

CA 5.8 Other Toxicological Studies

CA 5.8.1 Toxicity studies of metabolites

Studies evaluated in the monograph of the rapporteur member state Belgium of September 1999:

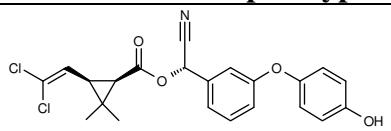
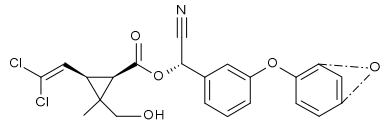
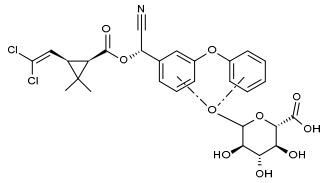
No toxicological studies were presented for metabolites during the former European assessment of alpha-cypermethrin. Thus, the following conclusion was drawn in the Annex I listing of alpha-cypermethrin:

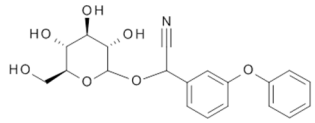
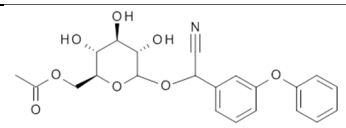
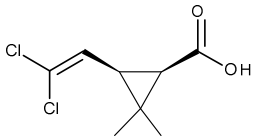
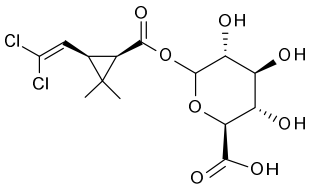
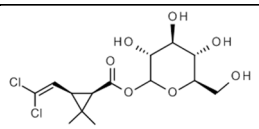
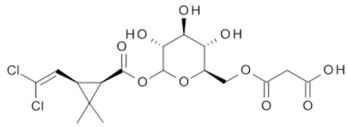
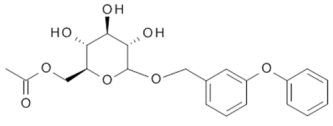
Other toxicological studies	No data – not required

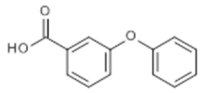
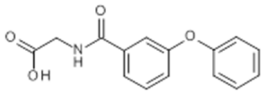
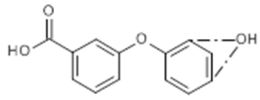
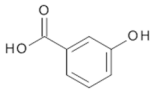
Meanwhile further insights into behavior of alpha-cypermethrin in plants, animals and in the environment has been obtained and thus several metabolites were identified which require further consideration (see section KCA 6.9).

With regard to potential toxicological relevance the following metabolites were considered and are addressed in this dossier section.

Table 5.8.1-1: Alpha-cypermethrin metabolites considered for potential toxicological relevance assessment

Metabolite	Structure	Reason for relevance assessment
Group A) Hydroxylated derivatives of alpha-cypermethrin and their conjugates		
M310I017 Reg. No. 6002320 WL 48394 (cis) CL 194198		Livestock metabolite (goat) Plant metabolite (wheat, cabbage)
M310I015		Livestock metabolite (goat)
M310I021		Livestock metabolite (goat)

Metabolite	Structure	Reason for relevance assessment
Group B) Conjugated hydroxy-(3-Phenoxy-phenyl)acetonitrils		
M310I005		Plant metabolite (lettuce)
M310I007		Plant metabolite (lettuce)
Group C) DCVA, hydroxylated DCVA and their conjugates		
M310I001 Reg. No. 4080830 CAS number: 59042-49-8 DCVA DCVC acid Cis-DVA CL 912554 CL 194198 (cis)		Livestock metabolite (goat, hen) Plant metabolite (wheat)
M310I003		Livestock metabolite (goat, hen)
M310I004		Livestock metabolite (goat)
M310I008		Plant metabolite (lettuce)
M310I009		Plant metabolite (lettuce)
Group D) Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates		
M310I024 CAS-No. 13826-35-2 Reg.No. 207323 PBAlc WL 40673 CL 206128		Plant metabolite (wheat, cabbage)
M310I006		Plant metabolite (lettuce)
M310I018 Reg.No. 4080665 CAS-No. 39515-51-0 EC No. 254-487-1 PBAld		Plant metabolite (wheat)

Metabolite	Structure	Reason for relevance assessment
WL 42049 CL 206969 3-PBAD 3-PBAD III m-PBAD CPBT		
M310I011 Reg. No. 130213 CAS No.: 3739-38-6 3-phenoxybenzoic acid PBA m-PB acid 3-PB acid WL 44607 CL 206128		Livestock metabolite (goat, hen) Plant metabolite (wheat)
M310I010 Reg. No. 4108084 N-(3-phenoxybenzoyl)glycine 3PBA glycine WL 46194 CL 117585		Livestock metabolite (goat) Plant metabolite (wheat)
M310I026 Reg.No. 4110960 3-phenoxy-benzoyl glutamic acid CL 949371		Plant metabolite (wheat)
M310I013 WL 46114 CL 213336		Plant metabolite (cabbage)
M310I025 Reg.Nr.4110493		Plant metabolite (wheat)
Group E) 3-Hydroxybenzoic acid		
M310I019 CAS-No. 99-06-9 EC-No. 202-726-5 FL-no. 08.132		Livestock metabolite (hen)

Thus, the conclusion for relevant endpoints for the current re-registration was drawn as follows:

Other toxicological studies (SANCO/11802 data point 5.8)

Toxicity studies of metabolites as referred to in the introduction	**Group A) Hydroxylated derivatives of alpha-cypermethrin and their conjugates:** **M310I017 (**livestock and plant metabolite) – by weight of evidence not genotoxic in vitro and in vivo – due to close structural relationship covered by the toxicological database of alpha-cypermethrin. Conclusion: **not toxicologically relevant** based on grouping approach **M310I015 (**livestock metabolite) – by weight of evidence not genotoxic in vitro and in vivo and exposure – below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. – due to close structural relationship covered by the toxicological database of alpha-cypermethrin. Conclusion: **not toxicologically relevant** based on grouping approach **M310I021 (**livestock metabolite) – by weight of evidence not genotoxic in vitro and in vivo based on data reported for the aglycone – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. – due to close structural relationship covered by the toxicological database of alpha-cypermethrin. Conclusion: **not toxicologically relevant** based on grouping approach **Group B) Conjugated hydroxy-(3-Phenoxy-phenyl)acetonitrils:** **M310I005** (plant metabolite) – not genotoxic in vitro based on data reported for the aglycone – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. Conclusion: **not toxicologically relevant** based on grouping approach

	M310I007 (plant metabolite) - not genotoxic in vitro based on data reported for the aglycone - exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. Conclusion: **not toxicologically relevant** based on grouping approach **Group C) DCVA, hydroxylated DCVA and their conjugates** **M310I001** (livestock & plant metabolite) - rat metabolite of alpha-cypermethrin. - not genotoxic in vitro - exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. - no relevant endocrine activity Conclusion: covered by toxicological testing of parent and **not toxicological relevant** based on grouping approach. **M310I004** (livestock metabolite) - major rat metabolite of alpha-cypermethrin. - not genotoxic in vitro based on data reported for the aglycone DCVA and for alpha-cypermethrin. - exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. Conclusion: covered by toxicological testing of parent and **not toxicological relevant** based on grouping approach. **M310I008** (plant metabolite) - expected cleavage to aglycone in mammalian gastrointestinal tract. - not genotoxic in vitro based on data reported for the aglycone DCVA and for alpha-cypermethrin. - exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. Conclusion: covered by toxicological testing of parent and **not toxicological relevant** based on grouping approach.

	M310I009 (plant metabolite) - expected cleavage to aglycone in mammalian gastrointestinal tract. - not genotoxic in vitro based on data reported for the aglycone DCVA and for alpha-cypermethrin. - exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. Conclusion: covered by toxicological testing of parent and **not toxicological relevant** based on grouping approach. **Group D) Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates** **M310I024** (plant metabolite) – not genotoxic in vitro (reported in DAR of lambda-cyhalothrin – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. – no relevant endocrine activity Conclusion: **not toxicological relevant** **M310I006** (plant metabolite) – not genotoxic in vitro based on data for the aglycone M310I024 – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compound. – no relevant endocrine activity based on data for the aglycone M310I024 Conclusion: **not toxicological relevant** **M310I018** (plant metabolite) – not genotoxic in vitro and in vivo – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. – Moderate acute toxicity – Not irritating to skin and eye – May be a skin sensitizer – 90-day rat study: NOEL 50 mg/kg bw/day, increased liver and kidney weights attributed to metabolic activation considered not adverse – no relevant endocrine activity Conclusion: **not toxicological relevant**

	M310I011 (livestock and plant metabolite) – by weight of evidence not genotoxic – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. – moderate acute toxicity – not acutely neurotoxic – no relevant endocrine activity Conclusion: **not toxicological relevant** **M310I010** (livestock and plant metabolite) – by weight of evidence not genotoxic based on data for the unconjugated derivatives – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. – moderatel acute toxicity based on data for the unconjugated derivatives – not acutely neurotoxic based on data for the unconjugated derivatives – no relevant endocrine activity based on data for the unconjugated derivatives Conclusion: **not toxicological relevant** based on grouping approach. **M310I026** (plant metabolite) – by weight of evidence not genotoxic based on data for the unconjugated derivatives – exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. – moderatel acute toxicity based on data for the unconjugated derivatives – not acutely neurotoxic based on data for the unconjugated derivatives – no relevant endocrine activity based on data for the unconjugated derivatives Conclusion: **not toxicological relevant** based on grouping approach.

M310I013 & M310I025 (plant metabolite)
- by weight of evidence not genotoxic based on data for non-hydroxylated M310I011
- exposure below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds.
- moderatel acute toxicity based on data for non-hydroxylated M310I011
- not acutely neurotoxic based on data non-hydroxylated M310I011
- no relevant endocrine activity based on data non-hydroxylated M310I011
- conjugates of hydroxylated M310I013 are rat metabolites

Conclusion: **not toxicological relevant** based on grouping approach

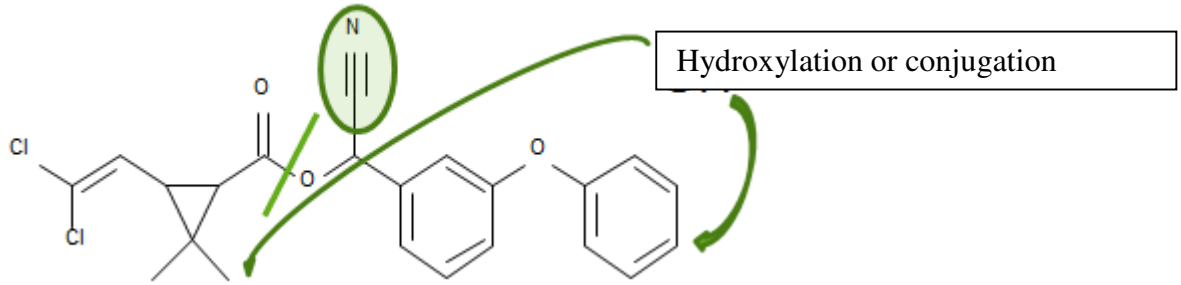
Group E) 3-Hydroxybenzoic acid

M310I019 (livestock)
- Evaluated by EFSA as food flavor being a Cramer Class 1 compound with low toxicity: TTC 1800 µg/person/day
- Toxicological evaluation by EFSA based on grouping approach:
  - By weight of evidence no safety concern with respect to genotoxicity
  - Acute toxicity, repeated dose toxicity data and developmental / reproductive toxicity data of group candidate and supporting substances were consistent with the group evaluation
- Low acute toxicity
- No relevant estrogenic activity

Conclusion: **not toxicological relevant** based on EFSA grouping approach for food flavours.

Strategy to assess metabolite relevance – grouping approach

Although there are differences in the metabolic pathways in plants, animals and the environment the formed degradation products are the same or closely related. Metabolism of alpha-cypermethrin in plants and animals is mostly characterized by cleavage of the cyano-group, ester cleavage and subsequent hydroxylation and/or conjugation of the degradates.



In order to avoid unnecessary animal testing and use of resources a grouping approach was setup.

The grouping proposal takes into account:

A) Chemical similarity,
B) Coverage by mammalian toxicity studies conducted with alpha-cypermethrin or other cypermethrins
C) Information on chemical reactivity (structural alerts)

In a further step key structures were selected for toxicological relevance assessment within every metabolite group.

D) Selection of key structures for hazard assessment

A Chemical Similarity

With regard to evaluation of chemical similarity the general proposals given by e.g. by the EFSA Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment [EFSA Journal 2012;10(07):2799] were followed. The following general molecular modifications were considered to probably not cause higher toxicity of the metabolites:

- Simple demethylation of the ring or side chain
- Simple hydroxylation of the ring system without any cleavage of the ring
- Hydroxylation of another ring position
- Conjugation of metabolites with amino acids

Comparison was made to parent as well as to the grouped metabolites in order to select key metabolites for testing. In addition consideration of increased hydrophilicity and thus considered faster excretion of the grouped metabolites as compared to the tested key metabolites and/or parent was taken into account. Conjugates were considered as their aglycons/parent metabolites if cleavability could be assumed, (e.g. for O-glucosides)

Generally there is some overlap between functional groups covered by metabolites of different metabolite groups. Therefore on a case by case basis only one of the metabolites with this functional group was considered for testing and considered suitable to allow evaluation of the metabolite with the same functional group included in another group, where such consideration was taken into account this is indicated in the respective grouping evaluation.

B Coverage of metabolites of concern by mammalian toxicity studies

An overlap exists in the metabolism of alpha-cypermethrin or other cyppermethrins in animals, plants and soil. Thus general conclusions drawn from the animal metabolism of the cypermethrins as well as overlaps of metabolite structures were taken into consideration when grouping the metabolites with regard to human exposure from the food residues. It was considered whether either the metabolites under consideration or similar structures were formed in the metabolisms studies conducted in mammals.
Moreover, it is taken into consideration that metabolites after uptake in to the body could be transformed by known metabolic pathways into structures that have been identified in the mammalian metabolism studies conducted.

C Presence of Structural alerts - QSAR evaluation of metabolites

For all unconjugated metabolites identified with potential relevance, the presence for potential structural alerts was evaluated with different SAR/QSAR models. Models used, were OASIS TIMES, and VEGA. However, the QSAR predictions obtained are limited in reliability as most of the structures evaluated were not in the prediction domain. Thus, given the structural relationship of the metabolites evaluated inter alia and in relation to the parent molecule alpha-cypermethrin, the predicted alerts were compared to those for the parent and those metabolites were toxicological data were available in order to overcome the limitations of the predictions made.

OASIS TIMES

OASIS TIMES is a hybrid statistical and knowledge-based model for toxicity prediction. The Tissue Metabolism Simulator (TIMES), developed by LMC (Bourgas University, Bulgaria; http://oasis-lmc.org/) integrates on the same platform a metabolic simulator and QSAR models for predicting toxicity of selected metabolites. The metabolic simulator generates plausible metabolic maps from a comprehensive library of Dettenbiotransformations and abiotic reactions. It allows prioritization of chemicals according to toxicity of their metabolites. Of OASIS TIMES the prediction models for Ames test and in vitro chromosome aberration were considered and therefore predictivity is limited to these test systems only. The reports for the evaluations made are available under [KCA 5.8.1/1 2014/1289314] for Ames mutagenicity and [KCA 5.8.1/2 2014/1289315] for prediction of chromosomal aberration in vitro. Q(SAR) Model Reporting Formats (QMRF) for both endpoints are provided in [KCA 5.8.1/3 2013/1414242 and [KCA 5.8.1/4 2013/1414460.

The reactivity model describing interactions of chemicals with DNA is based on an alerting group approach. Only those toxicophores extracted from the training set having clear interpretation for the molecular mechanism causing the ultimate effect included in the model. The mechanistic interrelation between alerts and related parametric ranges generalizing the effect of the rest of the molecules on the alert is also considered. The structural component of the model is based on the structural similarity between chemicals in the training set which were correctly predicted by the model. The structural neighborhood of atom-centered fragments is used to determine this similarity. The training set consists of 1514 chemicals for Ames and 808 chemicals for chromosomal aberration.

The derived model is combined with metabolic simulator TIMES used for predicting metabolic activation of chemicals with the S9 mix. The metabolic simulator is trained to reproduce documented maps for mammalian liver metabolism for 261 chemicals. Parent chemicals and each of the generated metabolites are submitted to a battery of models to screen for a general effect and mutagenicity mechanisms. Thus, chemicals are predicted to be mutagenic as parents only, parents and metabolites, and metabolites only. Mutagenicity could be due to the parent chemical only or as a result of its metabolic activation (i.e., the parent is inactive but it is transformed to a mutagenic metabolite), or both parent structure and metabolites could be mutagenic.

This OASIS QSAR system is also included in the OECD Toolbox (but not in combination with TIMES), in order to make use of (Q)SAR approaches also in the assessment of chemicals under REACH (http://www.oecd.org/chemicalsafety/risk-assessment/). The BASF-internal version has the advantage that it is capable to consider metabolic transformation.

VEGA

Using the VEGA platform, access to a series of QSAR (quantitative structure-activity relationship) models for regulatory purposes was obtained. Of the models offered by VEGA [http://www.vega-qsar.eu/] only the two independent statistical prediction models for mutagenicity (Ames) were selected. The data obtained for alpha-cypermethrin and its metabolite can be found in [see KCA 5.8.1/5 2014/1289317], [see KCA 5.8.1/6 2014/1319956] and [see KCA 5.8.1/7 2014/1320536]. This report includes all three models used (CAESAR, SarPy, Toxtree). CAESAR makes predictions based on the comparison of the structure of interest to the CAESAR database of mutagenicity data of substances in the structure database. A score is provided for the match of the structures, and the mutagenicity data of the closest related substances compared to the structure of interest. Consequently, if a structure is not adequately presented in the database, the prediction is only of very limited validity.

The second algorithm SarPy searches for isolated structural alerts of substructures in the molecule. Again this is based on the mutagenicity data provided in the structure database. Only predictivity for mutagenicity based on the Ames test was generated.

The third algorithm Toxtree is based on Benigni-Bossa Mutagenicity rules of structural alerts for mutagenicity. It works as a decision tree for estimating carcinogenicity, based on a list of structural alerts (SAs). The SAs for mutagenicity are molecular functional groups or substructures known to be linked to the mutagenic activity of chemicals. As one or more SAs embedded in a molecular structure are recognised, the system flags the potential mutagen of the chemical. The model goes through a first step in which a set of 12 SAs related to mutagenicty is checked The SAs are the following (SA numbers refer to the original Benigni/Bossa study):
- SA 1: Acyl halides
- SA 6: Propiolactones or propiosultones
- SA 11: Simple aldehyde
- SA 12: Quinones
- SA 13: Hydrazine
- SA 14: Aliphatic azo and azoxy
- SA 16: alkyl carbamate and thiocarbamate
- SA 18: Polycyclic Aromatic Hydrocarbons
- SA 21: alkyl and aryl N-nitroso groups
- SA 22: Azide and triazene groups
- SA 25: Aromatic nitroso group
- SA 28bis: Aromatic mono- and dialkylamine
- SA 29: Aromatic diazo

CAVEAT on reliability of QSAR modules implied

With regard to the QSAR evaluations as implied in OECD TIMES and in VEGA it should be noted that for nearly all analysis the algorithm reported an out of structural domain error. Each of this QSAR models is build on a set of chemicals that forms its chemical domain, space or applicability domain. That means that the prediction is best if a structure of interest is represented in the original baseline dataset. Substances outside of the dataset are evaluated in comparison to the chemical space, and only in case that the chemical space adequately covers all structural elements or the queried structure, the prediction is considered to be adequately covered by experimental data. Predictions outside of the applicability domain have far lower predictability. In addition all mentioned QSAR models check for structural alerts, like those identified by the Benigni-Bossa rules that have been implicated in mutagenic actions.

As a consequence, the predictivity is solely based on the proposed DNA-interaction via the structural alert, not (VEGA) or not appropriately (OASIS TIMES) taking into account possible functional group interaction and stereochemical hindrance. It is well established that structure elements have to be evaluated within the context of a structure.

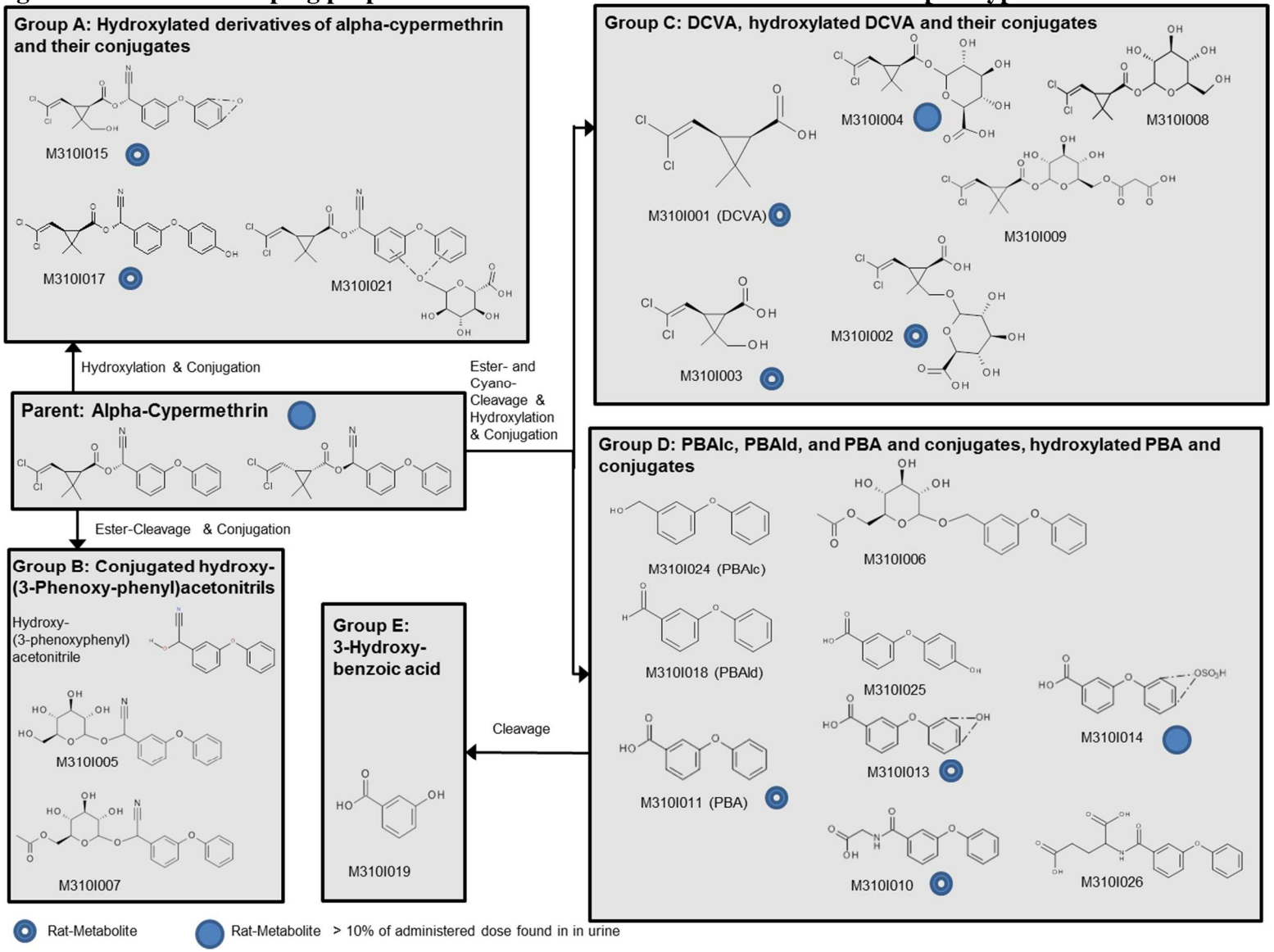
In general, the predictivity of various QSAR models for genotoxicity equilvalent to the Ames test has been considered to be reasonable accurate. Predictivity rates expressed as accuracy and specificity are usually >80%. This is in particular true, if information from more than one QSAR model is combined.

QSAR models for chromosome aberration however are far less well established. One of the underlying limitations is that typical in vitro assays for chromosome aberration, like chromosome aberration in V79 cells or the in vitro micronucleus assay have false discovery rates of approximately 30%. This means that three out of ten molecules are falsely categorized. In addition a high number of in vitro positive substances are negative in adequate in vivo assays. The later is often a function of the underlying mode of action and the kinetic behavior of a substance. Both are not adequately covered by QSAR predictions.

Predictions for chromosome damage were performed with OASIS Times. The prediction model used in the OECD toolbox is similar to that of OASIS Times, with the major difference being the different underlying database of reference compounds and the combination with the metabolism generator in OASIS Times.

The VEGA system - with the CAESAR SarPy and ToxTree modules used - only predicts bacterial mutagenicity.

Figure 5.8.1-1: Grouping proposal for relevance assessment of metabolites of alpha-cypermethrin



The groups were named as follows:

A) Hydroxylated derivatives of alpha-cypermethrin and their conjugates
B) Conjugated hydroxy-(3-Phenoxy-phenyl)acetonitrils
C) DCVA, hydroxylated DCVA and their conjugates
D) Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates
E) 3-Hydroxybenzoic acid

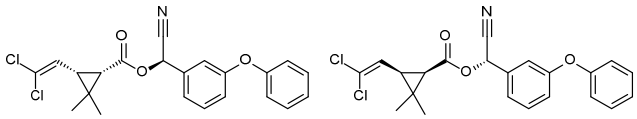
D Selection of key structures hazard assessment and toxicological testing strategy

As proposed by the EFSA scientific opinion on evaluation of the toxicological relevance of pesticide metabolites for dietary risk assessment the predicted exposure levels are taken into consideration. The proposed threshold levels for chronic exposure are 0.0025 µg/kg bw/day for potentially genotoxic compounds, 0.3 for presumably neurotoxic, and 1.5 µg/kg bw/day for non-genotoxic Cramer Class III compounds. These threshold levels are based on the threshold of toxicological concern (TTC) concept.
Furthermore an acute exposure assessment was considered necessary since an ARfD has been allocated for alpha-cypermethrin. The corresponding TTC value for acute exposure would be 5 µg/kg bw/day.

The TTC-concept is an approach to assess whether chemical structures, for which no or only limited information on the toxicological profile is available, are of concern. The TTC [Cramer et al. 1978a, [see KCA 5.8.1/8 1978/1001324]; Kroes et al. 2004a, [see KCA 5.8.1/9 2004/1036074]; Munro et al 1996a, [see KCA 5.8.1/10 1996/1005180] has meanwhile been considered in the EU e.g. for the evaluation of chemicals under the REACH regulation [*ECHA (2012) Guidance on information requirements and chemical safety assessment Chapter R.8: Characterisation of dose [concentration]-response for human health*] and has been employed or considered for the evaluation of food flavourings [*EFSA, 2010e. Guidance on the data required for the risk assessment of flavourings to be used in or on foods. European Food Safety Authority. The EFSA Journal 8(6): 1623. Available at: http://www.efsa.europa.eu/en/efsajournal/doc/1623.pdf*] or pesticidal degradation products by EFSA [*EFSA, 2012b. Scientific Opinion: Exploring options for providing preliminary advice about possible human health risks based on the concept of Threshold of Toxicological Concern (TTC). European Food Safety Authority (EFSA) Scientific Committee, Parma, Italy; EFSA Journal, 10(7), 2750 and EFSA, 2012a. Scientific opinion on evaluation of the toxicological relevance of pesticide metabolites for dietary risk assessment. EFSA Panel on Plant Protection Products and their Residues (PPR), European Food Safety Authority (EFSA), Parma, Italy; EFSA Journal, 10(07), 2799*].

The available toxicological database for metabolites of each group will be discussed in the following paragraphs. This starts with an assessment of the QSAR predictions of the parent alpha-cypermethrin and is followed by the evaluation of metabolites structured by groups as indicated above.

Alpha-cypermethrin (other denominators: BAS 310 I, Reg. No 4078193, WL 85871, CL 900049



Alpha-cypermethrin is a racemate of (S)-cyano-3-phenoxybenzyl (1R,3R) 3 (2,2 dichlorovinyl)-2,2 dimethylcyclopropanecarboxylate and (R)-cyano-3 phenoxybenzyl (1S,3S) 3 (2,2 dichlorovinyl)-2,2 dimethylcyclopropanecarboxylate. With regard to QSAR evaluations the stereoisomeric properties could not be taken into account as the available tools do not allow evaluation of stereoselective effects.

Weight of evidence approach for genotoxicity of alpha-cypermethrin QSAR prediction in relation to toxicological testing

For alpha-cypermethrin an extensive database on in vitro and in vivo genotoxicity studies is available. There is no evidence for mutagenicity or clastogenicity in any of the conducted studies in bacterial, fungal or mammalian cells in vitro or in mammals in vivo. Given the lacking evidence in vitro and in vivo the identified alerts for chromosomal for alpha-cypermethrin in OASIS Times is not considered of relevance.

Hydroxylated derivatives of alpha-cypermethrin and their conjugates

Definition of group A: Hydroxylated derivatives of alpha-cypermethrin and their conjugates

For the group of hydroxylated derivatives of alpha cypermethrin, the following molecules were taken into consideration:

M310I017
M310I015
M310I021

All three structures are closely related to alpha-cypermethrin. M310I017 is deviating by hydroxylation of the terminal aromatic benzyl-ring and for M310I015 by an additional hydroxylation of the dimethylcyclopropane moiety. M310I021 is a glucone conjugate of a ring-hydroylated derivative.

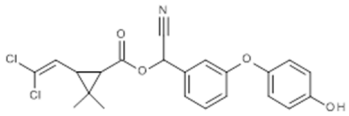
Note: for M310I017 the para-location of the hydroxylation group was confirmed. Although an exact position of the ring hydroxylation of M310I015 was not possible due to insufficient metabolite amounts recovered, it is reasonable to assume that thre ring-hydroxylation is located in the para-position also for the following reasons:

1. It is plausible that M310I015 is a metabolite successor of M310I017
2. Given the molecular structure a para-position of the terminal ring is more probable
3. Metabolic investigations on M310I011 the Phenoxy benzoic acid moity of alpha-cypermethrin in several mammalian species identified the para-hydroxylation of the phenoxy-ring i.e. 4-hydroxylation as the major metabolic pathway thereof [see CA 5.8.1/39 CY-905-031]. A ortho-position hydroxylation instead was not reported.

With regard to the glycone M310I021 again the most plausible position of the glycone conjugate is the para-position of the terminal ring for the same criteria as stated above.

Evaluation of group A members: Hydroxylated derivatives of alpha-cypermethrin and their conjugates

M310I017 (other denominators: Reg.No. 6002320; 4'hydroxy-alpha-cypermethrin, CL 194198, WL 48394 (cis), WL 48393 (trans))



M310I017 is a metabolite found in rats (small amounts in feces) and in livestock animals (goat).

QSAR Predicitions on M310I017

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 2 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]

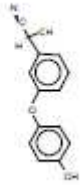
There were **no** Ames mutagenicity alerts for M310I017 or in-silico generated metabolites although several metabolites and Phenyl-OH itself had a structural alert info "Haloalkenes with Electron-Withdrawing Groups". In all cases the structures were out of domain.

For in-vitro chromosome aberration the prediction for M310I017 was **positive** (out of domain) referring to the structural alert info "Cyanohydrines". One out of 15 in-silico metabolites was predicted positive (in domain) with the alert info 'Cyanohydrines' (undetermined model reliability). The remaining in-silico metabolites were negative (out of domain).

Figure 5.8.1-2: Metabolite of M310I017 predicted positive for in-vitro chromosome aberration

2.3

Metabolite



Predicted CA with S9	in vitro CA positive
Predicted Mechanism	Interactions with topoisomerases / proteins
Alert info	Cyanohydrines
ModelReliability	Undetermined (0 <n< 10)
Total Domain	In domain

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 2 of report [see KCA 5.8.1/5 2014/1289317]]

M310I017 is out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules had positive experimental data. The other four had the prediction non-mutagen, which was experimentally confirmed. The concordance of the total underlying database was moderate (0.667) and thus is not very robust. Both the two positive (similarity 0.857 to 0.934) and the four negative molecules (similarity 0.785 to 0.976) has a reasonably similar structure to M310I017. Therefore, the chemical space does adequately cover the structure of M310I017.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 2 of report [see KCA 5.8.1/5 2014/1289317]]

M310I017 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules had positive experimental data. The other four had the prediction non-mutagen, which was experimentally confirmed. The concordance of the total underlying database was moderate (0.667) and thus is not very robust. Both the two positive (similarity 0.857 to 0.934) and the four negative molecules (similarity 0.785 to 0.976) has a reasonably similar structure to M310I017. Therefore, the chemical space does adequately cover the structure of M310I017.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 2 of report [see KCA 5.8.1/5 2014/1289317]]

M310I017 is in the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. None of the six most similar molecules had positive experimental data. The concordance of the total underlying database was high (1.0) and thus is very robust. All six negative molecules (similarity 0.757 to 0.94) have a reasonably similar structure to Phenyl-OH. Therefore, the chemical space does adequately cover the structure of Phenyl-OH.

Conclusion on QSAR evaluations of M310I017
The QSAR evaluation of M310I017 is of moderate reliability and by weight of evidence there was no conclusive alert for genotoxicity.

The similar structure to alpha-cypermethrin and the identical structural alert from QSAR applications result in the same assessment as for alpha-cypermethrin. The in vivo and in vitro data of alpha-cypermethrin indicates no chromosome damaging potential. Furthermore, in vitro data exist for the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile, (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 184-187) that demonstrate no chromosome damaging potential. Therefore, the positive prediction for CA of M310I017 (OASIS Times), based on study results obtained for the parent alpha-cypermethrin and the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile is not reflecting the toxicological database and is rejected.

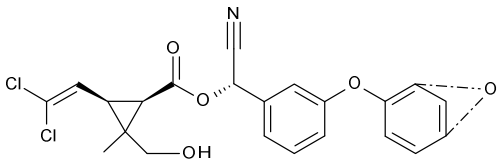
Toxicological evaluation of M310I017

No conclusive alerts for gentotoxicity were identified by the QSAR evaluation conducted. Furthermore, hydroxylation of the ring system without any cleavage of the ring is often identified as probably not causing higher toxicity of metabolites (EFSA, 2012; 10(07):2799). Thus, given the close structural relationship to the parent molecule alpha-cypermethrin and the lacking concordance with study results obtained for the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile, M310I017 is by weight of evidence not considered to be genotoxic and not considered to be more toxic than the parent molecule.

With regard to chronic consumer exposure the TTC concept for a non-genotoxic, but presumably neurotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. However, for acute exposure the TTC-threshold of 5 µg/kg bw/day would be exceeded (189% of TTC value) [see Table in section MCA 6.9]. As stated above M10I017 due to its close structural relationship to alpha-cypermethrin is considered to be covered by the toxicological database for the parent. Consequently the ARfD of 0.04 mg/kg bw/day is considered to apply also for M310I017 which will clearly cover the estimated acute dietary exposure level.

M310I017 is considered to be not genotoxic based on the available information and due to the close structural relationship to alpha-cypermethrin covered by the toxicological evaluation of the parent molecule. Therefore, M310I017 is considered to be **not toxicologically relevant**.

M310I015



M310I015 is a metabolite found in trace amounts in rats and in livestock animals (goat)

a. QSAR Predicitions on M310I015

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 3 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for M310I015 or in-silico generated metabolites although several metabolites and Dihydroxy alpha-cypermethrin itself had a structural alert info "Haloalkenes with Electron-Withdrawing Groups". In all cases the structures were out of domain. For in-vitro chromosome aberration the prediction for Dihydroxy alpha-cypermethrin was **positive** (out of domain) referring to the structural alert info "Cyanohydrines". One out of 19 in-silico metabolites was predicted positive (in domain) with the alert info 'Cyanohydrines' (undetermined model reliability). The remaining in-silico metabolites were negative (out of domain).

Figure 5.8.1-3: Metabolite of M310I015 predicted positive for in-vitro chromosome aberration

3.3

Metabolite

Predicted CA with S9	in vitro CA positive
Predicted Mechanism	Interactions with topoisomerases / proteins
Alert info	Cyanohydrines
ModelReliability	Undetermined (0 <n< 10)
Total Domain	In domain

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 3 of report [see KCA 5.8.1/5 2014/1289317]]
M310I015 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules had positive experimental data. The other four had the prediction non-mutagen, which was experimentally confirmed. The concordance of the total underlying database was moderate (0.668) and thus is not very robust. Both the two positive (similarity 0.837 to 0.911) and the four negative molecules (similarity 0.769 to 0.953) has a reasonably similar structure to Dihydroxy alpha-cypermethrin. Therefore, the chemical space does adequately cover the structure of Dihydroxy alpha-cypermethrin.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 3 of report [see KCA 5.8.1/5 2014/1289317]]
M310I015 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules had positive experimental data. The other four had the prediction non-mutagen, which was experimentally confirmed. The concordance of the total underlying database was moderate (0.668) and thus is not very robust. Both the two positive (similarity 0.837 to 0.911) and the four negative molecules (similarity 0.769 to 0.953) has a reasonably similar structure to Dihydroxy alpha-cypermethrin. Therefore, the chemical space does adequately cover the structure of Dihydroxy alpha-cypermethrin.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 3 of report [see KCA 5.8.1/5 2014/1289317]]
M310I015 is in the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. None of the six most similar molecules had positive experimental data. The concordance of the total underlying database was high (1.0) and thus is very robust. All six negative molecules (similarity 0.752 to 0.919) have a reasonably similar structure to Dihydroxy alpha-cypermethrin. Therefore, the chemical space does adequately cover the structure of Dihydroxy alpha-cypermethrin.

Conclusion on QSAR evaluations of M310I015
The QSAR evaluation of M310I017 is of moderate reliability and by weight of evidence there was no conclusive alert for genotoxicity.

The similar structure to alpha-cypermethrin and the identical structural alert from QSAR applications result in the same assessment as for alpha-cypermethrin. The in vivo and in vitro data of alpha-cypermethrin indicates no chromosome damaging potential. Furthermore, in vitro data exist for the metabolite (Hydroxy-(3-Phenoxy-phenyl)acetonitrile, see [see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 184-187] that demonstrate no chromosome damaging potential. Therefore, the positive prediction for chromosomal aberration of M310I015 (OASIS Times), based on study results obtained for the parent alpha-cypermethrin and the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 184-187) is not reflecting the toxicological database and is rejected.

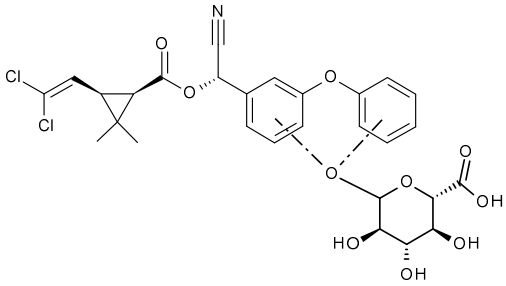
Toxicological evaluation of M310I015

No conclusive alerts for gentotoxicity were identified by the QSAR evaluation conducted. Furthermore, hydroxylation of the ring system without any cleavage of the ring is often identified as probably not causing higher toxicity of metabolites (EFSA, 2012; 10(07):2799). Thus, given the close structural relationship to the parent molecule alpha-cypermethrin and the lacking concordance with study results obtained for the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 184-187), M310I015 is by weight of evidence not considered to be genotoxic and not considered to be more toxic than the parent molecule.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated chronic exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I015 is considered to be not genotoxic based on the available information and the estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. M10I015 due to its close structural relationship to alpha-cypermethrin is considered to be covered by the toxicological database for the parent. Therefore, M310I015 is considered to be **not toxicologically relevant**.

M310I021



M310I021 is a metabolite found in in livestock animals (goat)

a. Toxicological evaluation of M310I021

After dietary uptake M310I021 is expected to be cleaved to the aglycone M310I017. No conclusive alerts for gentotoxicity were identified by the QSAR evaluation conducted for the agylcone M310I017. Furthermore, hydroxylation of the ring system without any cleavage of the ring is often identified as probably not causing higher toxicity of metabolites (EFSA, 2012; 10(07):2799). Thus, given the close structural relationship to the parent molecule alpha-cypermethrin and the lacking concordance with study results obtained for the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 184-187), M310I015 is by weight of evidence not considered to be genotoxic and not considered to be more toxic than the parent molecule.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I021 is considered to be not genotoxic based on the available information and the estimated exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. M10I021 due to its close structural relationship to alpha-cypermethrin is considered to be covered by the toxicological database for the parent. Therefore, M310I021 is considered to be **not toxicologically relevant**.

Toxicological relevance Group A: Hydroxylated derivatives of alpha-cypermethrin and their conjugates

M310I017, M310I015 and M310I021 are structurally closely related to the parent compound alpha-cypermethrin. The additional hydrolations are not considered to increase the toxicity profile of these metabolites when compared to alpha-cypermethrin as oxygen-bridged sugar conjugation which is considerd to be immediately cleaved when uptaken via the diet. The conducted evaluation on QSAR regarding genotoxicity alerts did not identify any alert considered of relevance. The only alert determined namely the structural alert for cyanohydines was also identified for parent alpha-cypermethrin and for the metabolite Hydroxy-(3-Phenoxy-phenyl)acetonitrile (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 184-187) for which both the available toxicological database did not provide any evidence to support this alert. Furthermore, hydroxylation of the ring system without any cleavage of the ring is often identified as probably not causing higher toxicity of metabolites (EFSA, 2012; 10(07):2799). Consequently the metabolites of that group are considered to be covered by the toxicological database of the parent molecule alpha-cypermethrin.

In conclusion the metabolites of the group A are by weight of evidence not considered to be genotoxic or neurotoxic and and are not considered to be toxicological relevant.

Conjugated Hydroxy-(3-Phenoxy-phenyl)acetonitrils

I. Definition of group B: Conjugated Hydroxy-(3-Phenoxy-phenyl)acetonitrils

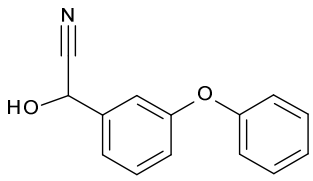
This group comprises Hydroxy-(3-Phenoxy-phenyl)acetonitrile and its glucoside-conjugates:

Hydroxy-(3-phenoxyphenyl)acetonitrile
M310I005
M310I007

It is expected that these oxygen-bridged sugar conjugates are immediately cleaved to the aglycone when orally uptaken into the body. Consequently the toxicological evaluation is based on the aglycone itself.

II. Evaluation of group B members: Conjugated Hydroxy-(3-Phenoxy-phenyl)acetonitrils

Hydroxy-(3-Phenoxy-phenyl)acetonitrile (other denominators: Aglycon of M310I005 or M310I007; CAS-Nos for stereoisomers: 71962-66-8, 61826-76-4 and 39515-47-4)



Hydroxy-(3-Phenoxy-phenyl)acetonitrile is the aglycon of the above mentioned metabolites M310I005 or M310I007 and a presumed intermediate in the plant metabolism. It was not determined in the rat metabolism studies.

a. QSAR Predicitions on Hydroxy-(3-Phenoxy-phenyl)acetonitrile

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 7 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for Hydroxy-(3-Phenoxy-phenyl)acetonitrile or in-silico generated metabolites and no structural alerts were reported. In all cases the structures were out of domain.
For in-vitro chromosome aberration the prediction for Hydroxy-(3-Phenoxy-phenyl)acetonitrile was **positive** (out of domain, model reliability undetermined (0<n<10)) referring to the structural alert info “Cyanohydrines”. None of 5 in-silico metabolites were predicted positive (out of domain).

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 7 of report [see KCA 5.8.1/5 2014/1289317]]
Hydroxy-(3-Phenoxy-phenyl)acetonitrile was out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Four of the six most similar molecules had positive experimental data. Each one of the remaining metabolites had the prediction "mutagen" and "non-mutagen". Experimental values were negative for both of them. The concordance of the total underlying database was low (0.0) and thus is not very robust. Both the positive (similarity 0.833 to 0.856) and the negative molecules (similarity 0.836 to 0.843) have a reasonably similar structure to Hydroxy-(3-Phenoxy-phenyl)acetonitrile. Therefore, the chemical space does adequately cover the structure of Hydroxy-(3-Phenoxy-phenyl)acetonitrile.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 7 of report [see KCA 5.8.1/5 2014/1289317]]
Hydroxy-(3-Phenoxy-phenyl)acetonitrile is out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The same metabolites as reported in the CAESAR model were also used in the SarPy model yielding the same results.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 7 of report [see KCA 5.8.1/5 2014/1289317]]
Hydroxy-(3-Phenoxy-phenyl)acetonitrile could be out of the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. One of the six most similar molecules had positive experimental data. The concordance of the total underlying database was low (0.496) and thus is not very robust. All six molecules (similarity 0.801 to 0.831) have a reasonably similar structure to Hydroxy-(3-Phenoxy-phenyl)acetonitrile. Therefore, the chemical space does adequately cover the structure of Hydroxy-(3-Phenoxy-phenyl)acetonitrile.

Conclusion on QSAR evaluations of Hydroxy-(3-Phenoxy-phenyl)acetonitrile
For Hydroxy-(3-Phenoxy-phenyl)acetonitrile results from an Ames test, Mouse Lymphoma Assay, and an in vitro chromosome aberration assay were reported in the DAR for lambda-cyhalothrin (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, pages 133-134 and pages 179-187). Based on the negative results for chromosome aberration in in vitro studies the positive prediction using OASIS TIMES QSAR model is rejected.

Toxicological studies on Hydroxy-(3-Phenoxy-phenyl)acetonitrile

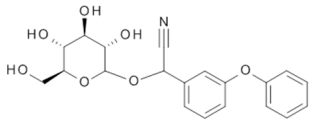
Toxicity of Hydroxy-(3-Phenoxy-phenyl)acetonitrile was assessed in the course of the renewal of lambda-cyhalothrin (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, pages 133-134 and pages 179-187). The acute oral toxicity in rats (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, pages 133-134) was >300 and < 2000 mg/kg bw. An in vitro genotoxicity package consisting of an Ames test, a mouse lymphoma assay and an in vitro chromosome aberration study gave no evidence for genotoxicity neither without nor with metabolic activation (see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, pages 179-187).

Toxicological evaluation of Hydroxy-(3-Phenoxy-phenyl)acetonitrile

The QSAR evaluation of Hydroxy-(3-Phenoxy-phenyl)acetonitrile is of moderate reliability and by weight of evidence there was no conclusive alert for genotoxicity. The lacking genotoxicity is supported by a datapackage on in vitro genotoxicity. Furthermore the acute toxicity study conducted showed only a moderate toxicity in comparison to alpha-cypermethrin.

In conclusion, Hydroxy-(3-Phenoxy-phenyl)acetonitrile is considered to be **not toxicologically relevant**.

M310I005



M310I005 is a plant metabolite found in lettuce.

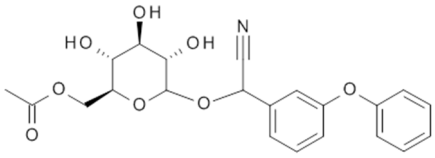
a. Toxicological evaluation of M310I005

Based on the toxicological evaluation of the aglycone of M310I005 - Hydroxy-(3-Phenoxy-phenyl)acetonitrile above – M310I005 is considered not to be genotoxic and is considered to be acutely less toxic than alpha-cypermethrin.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I005 is considered to be not genotoxic based on the available information and the estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. In conclusion M310I005 is considered to be **not toxicologically relevant**.

M310I007



M310I007 is also found as a plant metabolite in lettuce.

a. Toxicological evaluation of M310I007

Based on the toxicological evaluation of the aglycone Hydroxy-(3-Phenoxy-phenyl)acetonitrile above M310I007 is considered not to be genotoxic and and is considered to be acutely less toxic than alpha-cypermethrin.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I007 is considered to be not genotoxic based on the available information and the estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. In conclusion M310I005 is considered to be **not toxicologically relevant**.

Toxicological relevance Group B: Conjugated Hydroxy-(3-Phenoxy-phenyl)aceto-nitrils

It is expected that these oxygen-bridged sugar conjugates M310I005 and M310I007 are immediately cleaved to the aglycone Hydroxy-(3-Phenoxy-phenyl)aceto-nitrils when orally uptaken by mammalians. By weight of evidence there was no conclusive alert for genotoxicity for Hydroxy-(3-Phenoxy-phenyl)aceto-nitril. The lacking evidence is supported by toxicological data published within the evaluation of the Draft Renewal Assessement Report of Lambda-cyhalothrin of RMS Sweden of February 2013, pages 179-187) showing that there is no genotoxic potential in vitro.

In conclusion the metabolites of the group B are by weight of evidence not considered to be genotoxic or neurotoxic. They are below the relevant thresholds of toxicological concern for acute and chronic dietary exposure and thus considered to be not toxicologically relevant.

DCVA, hydroxylated DCVA and their conjugates

I. Definition of group C: DCVA, hydroxylated DCVA and their conjugates

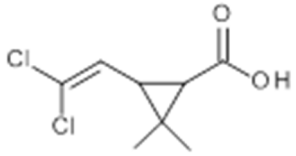
For the group of DCVA, hydroxylated DCVA and their conjugates, the following molecules were taken into consideration:

M310I001 (DCVA)
M310I004
M310I008
M310I009
M310I003 (hydroxylated DCVA)
M310I002

M310I004, M310I008 and M310I009 are o-bound glycosides of DCVA. These glycosides when uptaken into the human gastrointestinal tract via the diet are expected to be cleaved to the corresponding agylcone DCVA. Thus the glucones are considered to be covered by the toxicological evaluation of DCVA. M310I003 (hydroxylated DCVA) is structurally closely related to DCVA. With regard to the glycoside of hydroxylated DCVA (M310I002) same immediate cleavage is assumed as for the o-glycosides of DCVA. Moreover, M310I004 (the DCVA glucoronide), M310I003 (hydroxylated DCVA) and its glucuronid ester M310I002 are known rat metabolites, of which M310I004 is the major metabolite excreted via urine.

II. Evaluation of group C members: DCVA, hydroxylated DCVA and their conjugates

M310I001 (other denominators: DCVA, Reg.No. 4080830, CAS No.: 59042-49-8, 180011, CL 912554; Cis-Isomer: Cis-DVA, CL 194198 (cis), cis-3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecaroxylic acid; DCCA)



M310I001 is a metabolite found in rats, livestock animals (goat and hen) in wheat, in soil, water and sediment. This cis enantiomer respectively its racemate the trans-isomer (Trans-DCVA; CL 196336 (trans)) is also known as metabolite for e.g. Cypermethrin or Permethrin.

a. QSAR Predicitions on M310I001

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 5 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for M310I001 or in-silico generated metabolites although several metabolites and M310I001 itself had a structural alert info "Haloalkenes with Electron-Withdrawing Groups". In all cases the structures were out of domain. The same holds true for the in vitro chromosome aberration.

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 5 of report [see KCA 5.8.1/5 2014/1289317]]
M310I001 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The prediction is based on 6 molecules, of which all were predicted and actual 'non-mutagen'. The similarity to M310I001 of the molecules in the data set is moderate as indicated by similarity factors of 0.724 to 0.838.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 5 of report [see KCA 5.8.1/5 2014/1289317]]
M310I001 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The prediction is based on 6 molecules, of which 3 were predicted 'mutagen' but experimental values confirmed that all 6 were 'non-mutagen'. The similarity to M310I001 of the molecules in the data set is moderate as indicated by similarity factors of 0.724 to 0.838.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 5 of report [see KCA 5.8.1/5 2014/1289317]]
M310I001 is out of the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules were predicted 'mutagen' and three of them were positive referring to experimental data. The concordance of the total underlying database was moderate (0.518) and thus is not very robust. All six negative molecules (similarity 0.684 to 0.732) have a moderate similar structure to M310I001.

Conclusion on QSAR evaluations
No conclusive structural alert was identified for M310I001.

Toxicity studies on M310I001

Report:	CA 5.8.1/11 Hutson D.H., 1982a Pyrethroid intermediates (cis-DVA and CPBT): Short-term tests for genotoxic activity AL-470-006
Guidelines:	none
GLP:	yes
Remark:	only data for cis-DVA were considered from the study report

Executive Summary

S. typhimurium (strains TA98, TA 100, TA 1535, TA 1537 and TA 1538) and E. coli strain WP2 and WP2 uvrA were exposed to M310I001; batch: ST81-140, purity: 99%) using methanol as a solvent in the presence and absence of metabolic activation in two independent experiments. Triplicate plates were used per dose and per test condition. Vehicle and positive controls were included in each experiment. The test substance was used at concentrations of 31.25, 62.5, 125, 250, 500, 1000, 2000, and 4000 µg/plate in the two main experiments. Bacteriotoxic effects were observed in a range finding test at concentrations higher than 4000 mg/plate. Precipitation of the test substance did occur at 1000 µg/plate and at higher concentrations. A biologically relevant increase in the number of revertant colonies was not noticed in any of the strains tested in presence or absence of metabolic activation in any of the experiments. The positive controls induced the appropriate response in the corresponding strains in the presence of metabolic activation, thus demonstrating the sensitivity of the test system and the functionality of the S9 fraction. For Salmonella strains TA 1537, TA 1538, TA 98, and TA 100 no adequate control substances were used in the absence of metabolic activation. According to the results of the study M310I001 was not mutagenic in the Salmonella typhimurium / Escherichia coli reverse mutation assay under the experimental conditions of the study.

(Doc ID 1982/7000919)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material	BPDU 7, CIS-DVA (Metabolite of BAS 310 I, Alpha-Cypermethrin)
Description:	Solid, beige, crystalline
Lot/Batch #:	ST81/140
Purity:	99%
Stability:	Infra-red spectroscopic fingerprint analysis revealed stability throughout the study period.
Solvent used:	Methanol

2. Control Materials:

Vehicle control: The vehicle control with and without S-9 mix only contained the vehicle used for the test substance at the same concentration and volume for all tester strains.

Positive control compounds tested without addition of metabolic activation system:

Strain	Mutagen	Solvent	Concentration
TA 100	Benzo(a)pyrene (BP)	Methanol	20 µg/plate
TA 1535	Sodium azide (NaN_3)	Methanol	5 µg/plate
TA 1537	Neutral red (NR)	Methanol	20 µg/plate
TA 1538	Benzo(a)pyrene (BP)	Methanol	20 µg/plate
TA 98	Benzo(a)pyrene (BP)	Methanol	20 µg/plate
WP2 uvrA	NQO	Methanol	20 µg/plate
WP2	NQO	Methanol	20 µg/plate

Positive control compounds tested with addition of metabolic activation system:

Strain	Mutagen	Solvent	Concentration
TA 100	Benzo(a)pyrene (BP)	Methanol	20 µg/plate
TA 1535	Sodium azide (NaN_3)	Methanol	5 µg/plate
TA 1537	Neutral red (NR)	Methanol	20 µg/plate
TA 1538	Benzo(a)pyrene (BP)	Methanol	20 µg/plate
TA 98	Benzo(a)pyrene (BP)	Methanol	20 µg/plate
WP2 uvrA	NQO	Methanol	20 µg/plate
WP2	NQO	Methanol	20 µg/plate

3. Activation: Rat liver microsomal activation system (S9 fraction) was used for activation. No further details were given.

4. Test organisms: S. typhimurium strains: TA98, TA100, TA1535, TA1537, TA1538
E. coli: WP2, WP2 uvrA

5. Test concentrations:
Pre-incubation assay: In the first and second test triplicate plates were prepared for each concentration (neg. control; 31.25, 62.5, 125, 250, 500, 1000, 2000, and 4000 µg/plate) and condition (i.e. with and without S9) for all tester strains.

B. TEST PERFORMANCE:

1. Dates of experimental work: January, 1982

2. Dose range finding test:
In order to assess bacteriotoxic effects, a dose range finding test was conducted with Salmonella typhimurium TA 100 up to 6000 µg/plate. Up to 4000 µg/CIS-DVA per plate were shown to be non-toxic to the bacteria. Visible precipitation of the test substance was observed at 1000 µg per plate and above.

3. Plate-incorporation assay:
20 µL volumes of solution of the test substance were added to top agar mix to give final concentrations of 31.25, 62.5, 125, 250, 500, 1000, 2000, and 4000 µg/plate, as appropriate. Assays were carried out both in the presence and in the absence of rat liver S9 fraction. The cultures were incubated at 37°C for 48-72 h before the revertant colonies were counted. Means and standard deviations were calculated.

4. Statistics:
No special statistical tests were performed.

5. Evaluation criteria:
The test chemical is considered positive in this assay if the following criteria are met:

- A reproducible increase of 2.5-fold the control values or greater are considered to indicate a mutagenic response.

II. RESULTS AND DISCUSSION

A. TOXICITY
A bacteriotoxic effect was observed at concentrations higher than 4000 µg/plate in the dose range finding test. In the main experiments only concentrations were used up to the non-toxic dose of 4000 µg/plate.

B. MUTATION ASSAYS
In the 2 independent experiments with and without metabolic activation no biologically relevant increase in number of revertants was observed in any strain tested [see Table 5.8.1-2]. The positive controls yielded revertant numbers in a range expected for the respective strains and thus demonstrated the sensitivity of the test system.

Precipitation was observed at 1000 µg/plate and higher concentrations.

III. CONCLUSION

According to the results of the present study, the test substance CIS-DVA, Metabolite of BAS 310 I, alpha-Cypermethrin is not mutagenic in the Salmonella typhimurium / Escherichia coli reverse mutation assay under the experimental conditions applied.

Table 5.8.1-2: Bacterial gene mutation assay with M310I001 - Mean number of revertants

Experiment 1: Plate incorporation assay														
Strain	TA 98		TA 100		TA 1535		TA 1537		TA 1538		E. coli (WP2)		E. coli (WP2 uvrA)	
Metabol. activation	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
Neg. control (Methanol)	18.0	8.3	54.0	33.0	8.0	4.7	14.7	10.7	6.3	4.7	10.3	12.7	10.3	9.3
M310I001														
31.25 µg/plate	15.3	8.7	47.7	37.3	6.3	4.0	14.7	8.7	6.3	5.7	7.3	7.3	7.7	5.7
62.5 µg/plate	17.7	9.3	55.3	35.0	7.7	7.0	17.3	9.3	5.0	4.3	8.3	9.7	8.3	10.7
125 µg/plate	14.0	11.3	51.3	42.3	5.7	4.7	7.7	10.3	5.3	2.3	8.7	8.7	9.0	10.0
250 µg/plate	16.3	10.7	45.0	36.7	6.3	4.7	14.0	10.7	5.7	6.5	6.3	11.3	8.3	6.0
500 µg/plate	11.7	8.3	39.0	35.7	4.3	4.5	11.3	10.0	4.0	3.0	8.3	8.3	7.3	6.0
1000 µg/plate	12.3	9.7	40.0	38.7	7.0	4.0	13.0	15.5	5.0	5.3	11.7	9.0	8.7	6.7
2000 µg/plate	11.7	10.0	42.3	33.0	5.3	3.3	13.0	7.3	3.3	3.5	9.7	7.7	9.7	7.3
4000 µg/plate	10.7	9.7	40.0	28.0	7.0	2.7	13.3	10.0	1.3	3.0	5.7	7.0	10.7	5.7
Pos. control[§]	165	9.7	391	42.0	1011	1284	148	17	46	4.0	384	706	417	2.5
Experiment 2: Plate incorporation assay														
Strain	TA 98		TA 100		TA 1535		TA 1537		TA 1538		E. coli (WP2)		E. coli (WP2 uvrA)	
Metabol. activation	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
Neg. control (Methanol)	20.7	15.7	69.7	45.3	11.3	11.3	8.0	6.7	19.0	8.5	29.0	32.7	11.3	6.0
M310I001														
31.25 µg/plate	17.3	11.7	62.7	48.3	5.0	7.3	11.3	8.0	19.0	8.7	34.0	27.0	18.7	11.7
62.5 µg/plate	19.7	12.7	62.7	60.0	8.0	6.7	12.3	9.3	24.3	8.3	30.7	33.3	12.7	10.3
125 µg/plate	15.0	19.7	57.0	59.0	11.7	5.0	13.3	6.7	17.0	9.0	32.0	28.3	11.0	10.7
250 µg/plate	17.0	15.7	70.7	51.3	11.0	5.3	12.0	6.7	15.0	8.3	26.3	24.7	8.7	8.0
500 µg/plate	14.7	15.7	65.3	52.7	6.7	7.0	9.0	8.3	12.3	9.0	31.0	22.7	17.7	7.7
1000 µg/plate	21.3	10.3	70.3	45.3	8.3	5.7	10.7	5.3	8.7	9.7	27.3	25.0	16.3	7.5
2000 µg/plate	17.7	11.3	62.0	49.0	11.0	5.3	12.3	6.0	13.0	6.7	27.7	20.7	15.5	8.3
4000 µg/plate	13.7	10.7	62.0	43.3	10.3	6.0	7.0	1.0	11.3	5.3	30.3	17.7	17.3	8.0
Pos. control[§]	358	19	540	50	900	1132	142	11.0	134.7	12	288	618	870	31.3

[§] = Compound and concentrations see Material and Methods (I.A.2.) above

Report: CA 5.8.1/12
[redacted] 1982a
Toxicology of pyrethroids 871: Acute oral, percutaneous toxicity, skin, eye irritancy, skin sensitizing potential of cis-2-(2,2-dichlorovinyl)-3,3-Dimethylcyclopropane carboxylic acid (cis-DVA) and -alpha-cyano-3-phenoxybenzyl-tosylate
AL-470-001
Guidelines: none
GLP: no
Remark: only data for M310I001 were extracted

Executive Summary

M310I001 (purity 99%) was tested for its acute oral and dermal toxicity, skin and eye irritation potential as well as skin sensitizing potential. The acute toxicity studies were conducted in rats, the irritation studies in rabbits, and the GPMT was performed with guinea pigs. The tests were conducted in a manner comparable to standard OECD guidelines and no major deviations were observed that would prevent an appropriate hazard assessment. In the acute oral toxicity test two out of 12 rats died at the limit dose of 5000 mg/kg bw, and thus a LD_{50} > 5000 mg/kg bw was derived. In the acute dermal toxicity test none of 12 rats died at the top dose of 1000 mg/kg bw (applied as a 50% solution in DMSO), and thus a LD_{50} > 1000 mg/kg bw was derived. M310I001 was not irritating to the skin or eyes, and no skin sensitizing potental was observed for M310I001. In conclusion, no hazard was identified in the acute systemic or local studies performed and no classification is warranted in either category.

Acute oral toxicity

Summary
A single dose of 5000 mg M310I001 (in DMSO) per kg bodyweight was applied to 6 male and 6 female Wistar rats via gavage. Animals were observed for clinical signs various times at the day of administration and at least once daily thereafter for a total of 14 days. Two female animals died at day 2 of the study period. Accordingly, the oral LD_{50} was found to be greater than 5000 mg/kg bw for males and females, respectively:

Rat, oral: LD_{50} > 5000 mg/kg bw

Clinical signs noted were comatose condition, muscle incoordination, lethargy, salivation, and piloerection. The mean body weights of the administration groups increased throughout the study period, with a slow retardation in the first week after administration. No necropsy was performed. Under the conditions of this study the median lethal dose of M310I001 after oral administration was found to be greater than 5000 mg/kg bw in males and females, respectively.

(DocID AL-470-001)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: CIS-DVA (Metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	Solid, beige, crystalline
Lot/Batch #:	BPDU 7, ST81/140
Purity:	99%
Stability	Infra-red spectroscopic fingerprint analysis revealed stability throughout the study period. Test substance formulation in DMSO were checked for stability by means of HPLC. Stability was confirmed for at least 70 h.

2. Vehicle: DMSO

3. Test animals:

Species:	Rats
Strain:	Wistar albino rats
Sex:	male/female
Age:	10-11 weeks (at study begin)
Weight at dosing (mean):	250-380 g (males), 150-250 g (females)
Source:	Shell Toxicology Laboratory (Tunstall) Breeding Unit
Acclimation period:	7 days
Diet:	Rat food (PRD, Labsure Animal Foods, Dorset), ad libitum
Water:	Tap water, ad libitum
Housing:	Four animals in clear polycarbonate cages with stainless-steel wire-mesh floors and tops
Environmental conditions:	
Temperature:	19-25 °C
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 21-Oct-1981 - 05-Feb-1982 (Experimental period)

2. Animal assignment and treatment:
In a range-finding assay 50-5000 mg M310I001 per kg bodyweight were applied to one rat of each sex. In the main study 6 rats of each sex were dosed with 5000 mg/kg bw, which were fasted overnight (18 h). Solutions of the test substance were prepared daily. M310I001 was supplied as a 500 mg/mL solution in DMSO and was further diluted to the required volume with DMSO.
Clinical signs and symptoms were recorded several times at the day of administration and afterwards at least once daily for the individual animals up to 14 days post-administration. Individual body weights were determined shortly before administration, weekly thereafter and at the end of the study. No necropsy was performed.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality occurred in the range-finding assay when one animal of each sex was treated in the range from 50 to 5000 mg/kg bw. In the main study no mortality occurred within the males, whereby 2/6 females were found dead two days after administration of test substance.

B. CLINICAL OBSERVATIONS

In the males coma was observed within 2.5 h of dosing in 5/6 males and lasting 2 days. The male animal not suffering from coma showed muscular incoordination within 2.5 h of dosing also recovering at day 2. Further clinical signs included increased salivation, piloerection, and lethargy. All males were free from symptoms 4 days after administration.
Similar results were observed with the female animals. 4/6 females showed comatose condition, 1/6 females showed muscle incoordination and 1/6 females showed lethargy within 2.5 h after administration of test substance. Two of the comatose animals died at day 2. The remaining female animals recovered within 2 days.

C. BODY WEIGHT

The mean body weights of the administration groups increased at least in week 2 of the study period.

III. CONCLUSION

Under the conditions of this study, the oral LD_{50} in rats was determined to be greater than 5000 mg/kg bw for males and females.

Acute dermal toxicity

Summary
In an acute dermal toxicity study groups of 6 male and 6 female Wistar rats were exposed to a single dose of 1000 mg/kg bw of M310I001 (batch ST81/140, 50% solution in DMSO) to the clipped skin under occlusive conditions for 24 hours. The animals were observed for 14 days after administration. Based on the absence of mortality in this study the acute dermal LD_{50} was determined to be greater than 1000 mg/kg bw:

Rat dermal: LD_{50} > 1000 mg/kg bw

The mean body weights of the animals increased as throughout the study period. Clinical signs of toxicity included nasal bleeding and/or chromodacryrrhea (5/6 males; 6/6 females) and dermal irritation (1/6 males; 0/6 females). The majority of animals had recovered by day 2. No necropsy was performed. The available data on acute dermal toxicity of the test substance do not meet the criteria for classification according to Regulation EC Directive on dangerous preparations 1999/45/EC and Regulation (EC) No 1272/2008. Classification for acute dermal toxicity is therefore not warranted.

(DocID AL-470-001)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	CIS-DVA (Metabolite of BAS 310 I, Alpha-Cypermethrin)
Description:	Solid, beige, crystalline
Lot/Batch #:	BPDU 7, ST81/140
Purity:	99%
Stability	Infra-red spectroscopic fingerprint analysis revealed stability throughout the study period. Test substance formulation in DMSO were checked for stability by means of HPLC. Stability was confirmed for at least 70 h.
2. Vehicle:	The test substance was administered as a 50% solution in DMSO.

3. Test animals:

Species:	Rats
Strain:	Wistar albino rats
Sex:	male/female
Age:	10-11 weeks (at study begin)
Weight at dosing (mean):	250-380 g (males), 150-250 g (females)
Source:	Shell Toxicology Laboratory (Tunstall) Breeding Unit
Acclimation period:	7 days
Diet:	Rat food (PRD, Labsure Animal Foods, Dorset), ad libitum
Water:	Tap water, ad libitum
Housing:	3 animals in clear polycarbonate cages with stainless-steel wire-mesh floors and tops during the observation period. Single housing during 24-h treatment period.
Environmental conditions:	
Temperature:	19-25 °C
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 6 am to 6 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 21-Oct-1981 - 05-Feb-1982 (Experimental period)

2. Animal assignment and treatment:
In a range-finding study one animal of each sex was treated with 250-1000 mg test substance per kg bodyweight. In the main study five male and five female rats were treated with 1000 mg test substance per kg bodyweight for 24 h under occlusive conditions. The top dose of 1000 mg/kg bw was equivalent to the maximum dose volume that could be applied. The day before dosing the animals were weighed and approximately 60% of the dorsal hair was shorn with fine electric clippers. Directly before treatment the skin was visually inspected. The calculated dose was applied by syringe using a 50% test solution in DMSO. During treatment food was withheld but water was available ad libitum. At the end of the treatment the tape and aluminum foil were removed and the skin washed with warm dilute detergent solution and then dried. The animals were observed for signs of toxicity for 14 days after dosing. Body weights were recorded at day 0, day 7 and day 14.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality was observed in the range-finding and main study, respectively.

B. CLINICAL OBSERVATIONS

Clinical signs of toxicity included nasal bleeding and/or chromodacryrrhea (5/6 males; 6/6 females) and dermal irritation (1/6 males; 0/6 females). Nasal bleeding and chromodacryrrhea most often occurred within 2 hours of dosing. The majority of animals had recovered by day 2 and all animals had gained weight by the end of the 14 day observation period.

C. BODY WEIGHT

All animals had gained weight relative to their initial bodyweights by the end of the 14 day observation period.

III. CONCLUSION

Under the conditions of this study, the oral LD_{50} in rats was determined to be greater than 1000 mg/kg bw for males and females, respectively.

Acute dermal irritation

Summary

In an acute dermal irritation study, the skin irritation/corrosion potential of M310I001 (batch ST81/140, purity: 99%) was tested. The intact and abraded clipped skin of 3 male and 3 female New Zealand White rabbits was exposed to 0.5 g of the unchanged test substance for 24 hours covered with an occlusive dressing. Reactions on intact and abraded skin were quite similar, but only results from intact skin were taken into account for further assessment.

The cutaneous reactions were assessed 30 min, 48 and 72 hours after removal of the patch and then in weekly intervals until day 7 after treatment.

Slight erythema, observed in all animals immediately after removal of the patch were fully reversible in 5/6 and 6/6 animals after 48 and 72 h, respectively. Slight edema reactions at 24 h were fully reversible within 48 h in all animals. Mean scores over 24, 48 and 72 hours for each animal were within 0.16 and 0.5 for erythema and between 0.0 and 0.16 for edema. The overall 24 to 72 hour skin irritation scores were 0.39 for erythema and 0.05 for edema.

(DocID AL-470-001)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: CIS-DVA (Metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	Solid, beige, crystalline
Lot/Batch #:	BPDU 7, ST81/140
Purity:	99%
Stability	Infra-red spectroscopic fingerprint analysis revealed stability throughout the study period.

2. Vehicle: The test substance was administered unchanged.

3. Test animals:

Species:	Rabbit
Strain:	New Zealand white
Sex:	3 males / 3 females
Age:	5-11 months
Weight at dosing:	males: 3.53 – 4.57 kg, females: 3.58 – 4.42 kg
Source:	Shell Toxicology Laboratory (Tunstall), Sittingbourne, UK
Acclimation period:	14 days
Diet:	SG1 with vitamin C supplement (Grain Harvesters Ltd., Kent, UK), ad libitum
Water:	Tap water (filtered), ad libitum
Housing:	Single housing in stainless steel wire mesh cages with grating, floor area: 2881 cm²
Environmental conditions:	
Temperature:	16 - 19 °C
Humidity:	No data
Air changes:	No data
Photo period:	No data

B. STUDY DESIGN AND METHODS

1. Dates of work: 21-Oct-1981 - 05-Feb-1982 (Experimental period)

2. In-vitro pre-test: No *in vitro* pre-test was conducted.

3. Animal assignment and treatment:
The potential of CIS-DVA to cause acute dermal irritation or corrosion was assessed by a single topical application of 0.5 g of the unchanged test substance for 24 hours to the intact untreated and abraded skin of three male and female New Zealand White rabbits using a patch of 2 cm x 2 cm. The test substance was covered with a lint patch and was occluded by an impervious polythene sheet held in place by means of an elastic adhesive bandage. After 24 hours the wrappings and patches were removed. Before treatment dorsal hair between the shoulders and hindquarters was closely shorn with fine electric clippers, and two test sites, approximately 10 cm apart and located lateral to the midline, were selected.
The cutaneous reactions were assessed 30 min after removal of the patch and at 48 and 72 hours and 7 days after application.

II. RESULTS AND DISCUSSION

Slight erythema (grade 0.5-1) was observed in all animals immediately after removal of the patch. At 48 h erythema (grade 0.5) were only visible in one animal and were fully reversible in all animals at the 72 h reading time point. Slight edema (grade 0.5) was present in 2/6 animals at 24 h. Edema reactions were fully reversible within 48 hours in all 6 animals. No further skin reactions were reported. Mean scores over 24, 48 and 72 hours were between 0.16 and 0.5 for erythema and between 0 and 0.16 for edema, respectively. The overall 24 to 72 hour skin irritation scores were 0.39 for erythema and 0.05 for edema. Individual and mean irritation scores after 24 hour dermal application of CIS-DVA are presented in Table 5.8.1-3.

Table 5.8.1-3: Individual and mean skin irritation scores after 24 hour dermal application of M310I001 on non-abraded site

Readings	Animal	Erythema	Edema	Additional findings
24 h	01F	1	0.5	-
	02F	1	0	-
	03F	1	0	-
	04M	1	0.5	-
	05M	1	0	-
	06M	0.5	0	-
48 h	01	0	0	-
	02	0	0	-
	03	0	0	-
	04	0	0	-
	05	0.5	0	-
	06	0	0	-
72 h	01	0	0	-
	02	0	0	-
	03	0	0	-
	04	0	0	-
	05	0	0	-
	06	0	0	-
7 d	01	0	0	-
	02	0	0	-
	03	0	0	-
	04	0	0	-
	05	0	0	-
	06	0	0	-
Mean 24 - 72 h	01	0.33	0.16	
	02	0.33	0.0	
	03	0.33	0.0	
	04	0.33	0.16	
	05	0.50	0.0	
	06	0.16	0.0	
Mean		0.39	0.05	

III. CONCLUSION

Based on the findings of this study, M310I001 showed a minimal skin irritation potential to rabbits under the test conditions chosen.

Eye irritation

Summary
In an eye irritation study, the eye irritation/corrosion potential of M310I001 (batch ST81/140, purity: 99%) was determined by instillation of 100 mg of the unchanged test substance into the conjunctival sac of one eye of six New Zealand White rabbits. No washing was performed. The ocular reactions were assessed approximately 1, 24, 48 and 72 hours and 7 days after the administration of the test substance.
No effects on iris were observed in any animal at any time point. Very slight effects on cornea (grade 0.5) were observed at 24 h in 5/6 animals and were fully reversible within 7 days in 4/5 animals. In one animal a cornea score of 0.5 was still visible after 7 days. Moderate conjunctival redness (grade 2), observed in all animals 1 and 24 hours after application, decreased to slight at 48 and 72 h in most of the animals. Redness was fully reversible in 5/6 animals within 7 days. Only in 1/6 animals slight redness (grade 1) was still visible after 7 days. Slight conjunctival chemosis and discharge was noted at the 1 and 24 h reading. These effects were fully reversible after 72 h at the latest. The individual mean scores calculated over 24, 48 and 72 hours were within 0.0 and 0.5 for corneal opacity and 0 for iris lesions. Values for redness of the conjunctiva were within 1.33 (4/6 animals) and 2.0 (1/6 animals). Mean values for chemosis over 24, 48, and 72 h were within 0.16 and 0.83, respectively.

(DocIDAL-470- 01)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BPDU 7, CIS-DVA (Metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	Solid, beige, crystalline
Lot/Batch #:	BPDU 7, ST81/140
Purity:	99%
Stability	Infra-red spectroscopic fingerprint analysis revealed stability throughout the study period.

2. Vehicle: The test substance was administered undiluted.

3. Test animals:

Species:	Rabbit
Strain:	New Zealand white
Sex:	3 males / 3 females
Age:	5-11 months
Weight at dosing:	males: 3.53 – 4.57 kg, females: 3.58 – 4.42 kg
Source:	Shell Toxicology Laboratory (Tunstall), Sittingbourne, UK
Acclimation period:	14 days
Diet:	SG1 with vitamin C supplement (Grain Harvesters Ltd., Kent, UK), ad libitum
Water:	Tap water (filtered), ad libitum
Housing:	Single housing in stainless steel wire mesh cages with grating, floor area: 2881 cm²
Environmental conditions:	
Temperature:	16 - 19 °C
Humidity:	No data
Air changes:	No data
Photo period:	No data

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: 21-Oct-1981 - 05-Feb-1982

2. In-vitro pre-test: No in vitro pre-test was conducted

3. Animal assignment and treatment:
The potential of CIS-DVA to cause acute eye irritation/corrosion was assessed by instillation of 100 mg of the undiluted test substance into the conjunctival sac of one eye. The lids were held together for a few seconds to prevent loss of material. No washing was performed. The ocular reactions were assessed approximately 1, 24, 48 and 72 hours and 7 days after dosing. In the event of any corneal damage visualization was aided by the instillation of one drop of fluorescein solution (2%). Body weights were determined shortly prior to application.

II. RESULTS AND DISCUSSION

The instillation of undiluted M310I001 into the conjunctival sac of one eye of each of six rabbits resulted in slight initial pain in the majority (4/6) of the animals and moderate initial pain in the other 2 animals. No effects on iris were observed in any animal at any time point. Very slight effects on cornea (grade 0.5) were observed at 24 h in 5/6 animals and were fully reversible within 7 days in 4/5 animals. In one animal a cornea score of 0.5 was still visible after 7 days. Conjunctival redness (grade 2) was observed in all animals after 1 and 24 h. These effects were slowly decreasing over time and were fully reversible in 5/6 animals after 7 days. Similarly, chemosis (grade 0.5 to 2) was observed after 1 and 24 h, was also slowly decreasing over time and was fully reversible within 72 h. Slight discharge (grade 0.5 to 1) was observed at 1 and 24 h only. No further effects on the eyes of rabbits were reported. The individual mean scores calculated over 24, 48 and 72 hours were within 0.0 and 0.5 for corneal opacity and 0 for iris lesions. Values for redness of the conjunctiva were within 1.33 (4/6 animals) and 2.0 (1/6 animals). Mean values for chemosis over 24, 48, and 72 h were within 0.16 and 0.83, respectively.
For details regarding the individual and mean scores as well as additional findings see Table 5.8.1-4.

Table 5.8.1-4: Individual and mean eye irritation scores after ocular application of M310I001

Readings	Animal	Cornea		Iris	Conjunctiva			Additional findings
		Opacity	Area involved		Redness	Chemosis	Discharge	
1 h	01F	0	0	0	2	1	0.5	-
	02F	0	0	0	2	1	0.5	-
	03F	0	0	0	2	2	1	-
	04M	0	0	0	2	1	0.5	-
	05M	0	0	0	2	0.5	0.5	-
	06M	0	0	0	2	2	1	-
24 h	01	0.5	4	0	2	1	0	-
	02	0	0	0	2	1	0	-
	03	0.5	4	0	2	1.5	0	-
	04	0.5	4	0	2	1.5	0.5	-
	05	0.5	2	0	2	0.5	0	-
	06	0.5	4	0	2	2	0.5	-
48 h	01	0	0	0	1	0	0	-
	02	0	0	0	1	0	0	-
	03	0	0	0	1	0	0	-
	04	0	0	0	1	0	0	-
	05	0.5	4	0	2	0	0	-
	06	0.5	2	0	2	0.5	0	-
72 h	01	0	0	0	1	0	0	-
	02	0	0	0	1	0	0	-
	03	0	0	0	1	0	0	-
	04	0	0	0	1	0	0	-
	05	0.5	2	0	1.5	0	0	-
	06	0.5	2	0	2	0	0	-
7 d	01	0	0	0	0	0	0	-
	02	0	0	0	0	0	0	-
	03	0	0	0	0	0	0	-
	04	0	0	0	0	0	0	-
	05	0.5	4	0	0	0	0	-
	06	0	0	0	1	0	0	-
Mean 24 - 72 h	01	0.16	-	0	1.33	0.33	-	
	02	0	-	0	1.33	0.33	-	
	03	0.16	-	0	1.33	0.50	-	
	04	0.16	-	0	1.33	0.50	-	
	05	0.5	-	0	1.83	0.16	-	
	06	0.5	-	0	2.00	0.83	-	
Mean		0.25	-	0	1.53	0.44	-	

III. CONCLUSION

Based on the findings of this study, M310I001 is mildly irritating to the eye of rabbits under the test conditions chosen. The available data on acute eye irritation of the test substance do not meet the criteria for classification according to Regulation EC Directive on dangerous preparations 1999/45/EC and Regulation (EC) No 1272/2008. Classification for acute eye irritation is therefore not warranted.

Skin sensitization

Summary
For the determination of potential sensitizing properties of M310I001 (batch: ST81/140, purity: 99%) a maximization test in guinea pigs ("P" strain) was conducted. Based on the results of a pre-test, the intradermal induction was performed with a 0.05% test item preparation in light liquid paraffin into the neck region of the animals. The epicutaneous induction (7 days after intradermal induction) and the challenge exposure (14 days after epicutaneous induction) were performed with a 50% test item preparation in petroleum jelly. The study was performed in 5 control and 10 test group animals per sex. Control group animals were treated with the same injection scheme as the test group animals but replacing the test item by the vehicle. Regarding epicutaneous induction, the control group animals were treated with the vehicle. 14 days after the last induction, the challenge was carried out. 0.3 mL of the 50% test substance preparation was applied for 24 hours to the intact skin of the flank under occlusive conditions. 24 and 48 hours after removal of the patch, skin readings were performed. No positive control was included in the study and no information regarding a periodic reliability check of the laboratory was given.
The intradermal induction with 0.05% test substance preparation caused positive reactions in all test group animals of the pre-test. No reactions were observed after epidermal induction with 50% test substance preparation in the animals during the pretest. No data were given for the animals of the main test. The challenge revealed no reactions in any animal of the control or test group, indicating that M310I001 has no skin sensitizing properties in the guinea pig maximization test under the conditions applied.

(DocID AL-470-001)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: CIS-DVA (Metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	Solid, beige, crystalline
Lot/Batch #:	BPDU 7, ST81/140
Purity:	99%
Stability:	Infra-red spectroscopic fingerprint analysis revealed stability throughout the study period. Solutions were stable for at least 7.5 h (expert statement)

2. Vehicle / Positive control:

Vehicles:
Light liquid paraffin, petroleum jelly
Positive control: not available

3. Test animals:

Species:	Guinea Pig
Strain:	"P"
Sex:	male/female
Age:	9 - 12 weeks
Weight at dosing:	437-792 g
Source:	Shell Toxicology Laboratory (Tunstall), Sittingbourne, UK
Acclimation period:	at least 14 days
Diet:	SG1 with vitamin C supplement, Grain Harvesters Ltd., Kent, UK, ad libitum
Water:	Tap water (filtered), ad libitum
Housing:	Groups of 2-3 animals were housed in stainless steel wire mesh cages with a floor area of approximately 1674 cm^2
Environmental conditions:	No details given

B. STUDY DESIGN AND METHODS

2. Dates of experimental work: 21-Oct-1981 - 05-Feb-1982

2. Animal assignment and treatment:
The skin sensitizing potential of M310I001 was assessed using the Maximization Test based on the method of Magnusson and Kligman. For this, male and female guinea pigs were randomly allocated to groups. Five animals per sex were used as control group animals and 10 animals per sex in the test group. Based on the results of a pre-test, animals were intradermally induced with 0.05% test substance preparations. Epidermal induction and challenge were conducted with 50% test substance preparations. The animals were closely shorn in the shoulder region using electric clippers followed by an electric razor; two rows of three injections were made, one on each side of the midline.

3. Clinical observation:
The animals were observed for a few days during the intradermal range finding test on general toxicity. No information is available from the main test.

4. Body weights:
Individual body weights were determined on day 0.

5. Pre-test:
100 µL of several dilutions of the test substance (0.05, 0.1, 0.5, and 1.0% in light liquid paraffin) were injected intradermally to 2 male and 2 female animals. The animals were examined over the next few days to determine the maximum concentration that could be used without causing untoward toxicity. Further groups of 2 male and 2 female animals were used to determine the irritation potential of the test substance applied at 25 and 50% in light paraffin and 60% in petroleum jelly. Therefore 0.3 mL of the test substance dilution was applied to the shaved flank of the animals and was covered with a "Poroplast" elastic adhesive bandage for 24 h. After exposure the skin reactions were assessed.

6. Main study – intradermal induction:
Based on the results of the pretest, test group animals received intradermal injections of 0.05% test substance preparations. A 0.05% test substance preparation (TS in light liquid paraffin) was intradermally injected to ten animals per sex. 6 intradermal injections were applied at the neck region of each animal: front row: 2 injections each of 0.1 mL Freund´s complete adjuvant without test item; middle row: 2 injections each of 0.1 mL of a test item preparation in vehicle at the selected concentration; back row: 2 injections each of 0.1 mL Freund's complete adjuvant / vehicle (1:1) with test item at the selected concentration. Skin reactions were assessed 24 hours after the beginning of the application. Control group animals received the same injections but with the test substance preparation being replaced by the vehicle.

7. Main study – epicutaneous induction:
One week after intradermal induction, 0.3 mL of the 50% test
light liquid paraffin caused a positive response in each 2 male and female animals. After topical item preparation (in petroleum jelly) was applied to each test group animal under the same conditions as described in the epidermal pretest but for 48 h. The control animals were treated similarly with vehicle only.

8. Main study - challenge:
The challenge was carried out 14 days after the epicutaneous induction. 0.1 mL of the 50% test item preparation was applied to the test and control group animal. The animals were exposed under occlusive conditions as described above for 24 hours and skin readings were performed 24 and 48 h after removal of the patch.

9. Evaluation of results
The number of animals with skin findings at 24 and/or 48 hours after the removal of the patch was taken into account for the determination of the sensitization rate. The evaluation "sensitizing" results if at least 30% of the test animals exhibit skin reactions.

10. Positive controls
No information on positive controls was given.

II. RESULTS AND DISCUSSION

A. PRE-TEST

Injections of 0.05, 0.1, 0.5, and 1.0% test substance preparations in induction no response was observed after treatment with 25 and 50% test substance preparation in light liquid petroleum or with a 60% preparation in petroleum jelly, respectively.

B. OBSERVATIONS

No abnormalities were observed during general observation.

C. BODY WEIGHTS

Body weights at start of treatment were within 437-792 g in the animals of the test group and within 503-693 g in the animals of the control group. No final body weights were given.

D. SKIN REACTIONS AFTER INTRADERMAL INDUCTION

According to the results from the pretest, intradermal induction with 0.05% test substance preparation in light liquid paraffin leads to a positive response. No details from the main study regarding intradermal induction were available.

E. SKIN REACTIONS AFTER EPICUTANEOUS INDUCTION

According to the results from the pretest, topical application of 50% test substance preparation in petroleum jelly leads to no dermal response. No details from the main study regarding topical induction were available.

F. SKIN REACTIONS AFTER CHALLENGE

The challenge with a 50% test substance preparation in petroleum jelly did not cause any skin reactions in animals of the control group and test group 24 and 48 hours after removal of the patch (see Table 5.8.1-5). Since no borderline results were observed, a 2nd challenge was not performed.

Table 5.8.1-5: cis-DVA - Skin reactions after challenge

	Challenge			
Skin findings	Control group		Test group	
	24 h	48 h	24 h	48 h
Grade 0	10/10#	10/10	19/20*	19/20*

x/y = number of findings / number of animals tested;
* = one animal was killed for humane reasons prior to topical induction stage

G. POSITIVE CONTROL

No information regarding positive control data was given.

III. CONCLUSION

Based on the results of this study it is concluded that M310I001 does not have sensitizing properties in the guinea pig maximization test under the test conditions chosen. 0% of the animals were considered positive after challenge application.

Endocrine modulation

Information with regard to the endocrine potential of M310I001 was found in the open literature. A summary table of the available literature is provided in Table 5.8.1-6 below. The literature is however described in more detail in the chapter CA 5.8.3.

In vitro studies:

M310I001 showed no estrogenic activity in yeast or mammalian ER-dependent transactivation assays, and no effect on androgen or thyroid receptor, neither as agonist nor as antagonist.

However M310I001 displayed anti-estrogenic effects when tested in competition with 17β-estradiol (1E-9M). While the anti-estrogenic effect starts in yeast cells in the µ-molar range, an antagonistic activity in mammalian cells is seen at concentrations of 7.23 nM. A significant reduction about 50% of the estradiol activation is seen in the low µM range.
M310I001 is one of the main metabolites of alpha-cypermethrin and when taking its phase II conjugation products into consideration as being formed out of M310I001 as well, this metabolite should have been in the system at a rate above 10% of the administered dose of alpha-cypermethrin. Therefore its toxicological properties are considered covered in the rat studies with alpha-cypermethrin. A relevant anti-estrogenic activity of this main metabolite would therefore be expected to be noticed in sensitive reproductive and developmental processes in studies with alpha-cypermethrin, or with any representative within the group of cypermethrins. Mainly one would expect effects like a delay of sexual maturation and vaginal opening. However, none of the regulatory studies indicate anti-estrogenic activity, and also literature data on cypermethrin were, if at al, discussing rather estrogenic activity of pyrethroids.

Considering all available data on endocrine potential M310I001 is considered to display some anti-estrogenic activity in vitro, however this is not mirrored in the regulatory in vivo studies in rats. Therefore this property is not considered relevant for further assessment.

Table 5.8.1-6: Endocrine activity studies with M310I001

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference Doc ID
In vitro assays				
ER-activation	hER transactivation in Yeast (lacZ)	ER-agonist: Not active ER-antagonist: Active LOIC: 6±4E-05 M IC_{50}: 6.5±3.5E-04 M	Agonist: E2: EC50: 2.1±0.4E-10 M Antagonist:4HO-TAM: IC_{50}: 2.8±0.7E-6 M	Tyler et al., 2000 2000/1024078
ER-activation	hER transactivation in CV-1 cells (Luciferase)	ER-agonist: Not active ER-antagonist: Active RIC_{20}: 7.23E-09 M IC_{50}: approx. 1E-06M	Agonist: 17β-E2: EC_{50}: 3.7E-9 M (REC: 100% = 1E-9M 17β-E2)	Du et al., 2010 2010/1232195
AR-activation	hAR transactivation in Yeast (lacZ)	AR-agonist: Not active AR-antagonist: Not active	Agonist: DHT: EC_{50}: 9.7±0.3E-10 M Antagonist: Flutamide: IC_{50}: 6.8±0.7E-6 M	Tyler et al., 2000 2000/1024078
AR-activation	hAR transactivation in MDA-kb2 cells (Luciferase)	AR-agonist: Not active AR-antagonist: not active	Agonist: DHT: EC_{50}: 3.99E-10 M (REC: 100% = >1E-8M DHT) Antagonistic: n.a.	Du et al., 2010 2010/1232195
TRβ-activation	TRβ transactivation in CV-1 cells (Luciferase)	TR-agonist: Not active TR-antagonist: Not active	Agonist: T3: EC50: 2.94E-9 M (REC: 100% = >1E-7M T3) Antagonistic: n.a.	Du et al., 2010 2010/1232195

PE: Proliferative effect; E2: 17 beta-Estradiol; EE: ethinyl estradiol; 4HO-TAM: 4-Hydroxy-tamoxifen; RPE: relative proliferative effect calculated as the ratio (PE-1) of the test chemical over (PE-1) of E2 (x 100); RIE: The relative inductive efficiency (RIE) is the ratio between the maximal up-regulation of pS2 expression level by the test compound to that of E2 (x100) or the relative inhibitory effciency of down-regulation of ERalpha expression level by the test compound compared to that of E2 (x 100); EC50/IC50: Effect concentration /Inhibitor concentration at which 50% increase /decrease is found compared to control; LOIC: Lowest observed inhibitory concentration

Toxicological evaluation of M310I001

The QSAR evaluation of M310I001 is of low reliability and by weight of evidence there was no conclusive alert for genotoxicity.

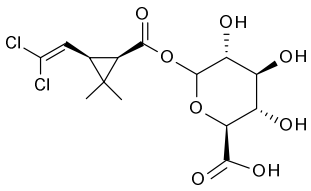
M310I001 is considered to be not genotoxic based on the in vitro studies conducted. Referring to the negative Ames test (see CA 5.8.1/1) the negative prediction for genotoxicity does reflect the experimental evidence. The acute toxicity studies conducted did not provide any evidence for significant acute systemic toxicity and/or significant effects on local tolerance.

M310I001 is considered to display some anti-estrogenic activity in vitro; however this is not mirrored in the regulatory in vivo studies in rats. Therefore this property is not considered relevant for further assessment.

With regard to chronic consumer exposure the TTC concept for a non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day. Moreover, the o-glycoside of M310I001 i.e. M310I004 is the major rat metabolite, and M310I001 the presumed precursor is expected to be immediately glucorinized when formed or uptaken in mammals and as such excreted. In conclusion the potential toxic properties of M310I001 are considered to be coverd by the testing of the parent molecule.

M310I001 is considered to be not genotoxic based on the available information. The in vitro evidence for anti-estrogenic activity is not considerd relevant due to the lacking evidence in apical in vivo studies. M310I001 is considered to be covered by the toxicological database for the parent. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. Therefore, M310I001 is considered to be **not toxicologically relevant**.

M310I004



M310I004 is a major metabolite found in rats and livestock animals (goat).

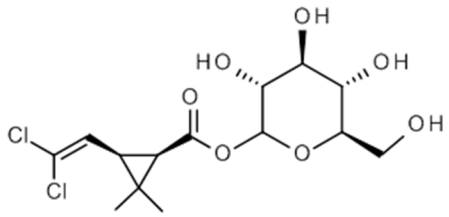
a. Toxicological evaluation of M310I004

Hydrolytic cleavage of the ester bound and elimination of the cis and trans cyclopropanecarboxylic acid moieties in the free and conjugated form is a major route of metabolism of cypermethrin in rats and in man [see section CA 5.1.1 of this dossier]. The cis and trans cyclopropanecarboxylic acid moiety were almost exclusively conjugated and rapidly eliminated as the ester glucuronide M310I004 (30% of the dose in male, 47% of the dose in females; trans representing 51.4% and cis 32.8% of the administered dose) with some free acid (twice as much trans as cis) being found in the urine (4%) together with traces of the trans-hydroxymethyl cyclopropyl acids (2%). Thus metabolite M310I004 as a major mammalian metabolite is covered by the toxicological testing of alpha-cypermethrin in mammals. M310I004 when uptaken into the gastrointestinal tract via the diet it is expected to be cleaved to the aglycone i.e. M310I001.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100% utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I004 is considered to be not genotoxic based on the available information on M310I001. M310I004 is considered to be covered by the toxicological database for the parent. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. Therefore, M310I004 is considered to be **not toxicologically relevant**.

M310I008



M310I008 is a plant metabolite (lettuce).

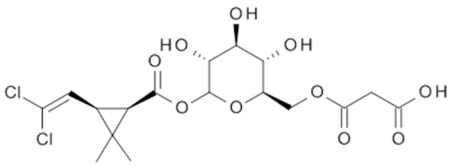
a. Toxicological evaluation of M310I008

Hydrolytic cleavage of the ester bound and elimination of the cis and trans cyclopropanecarboxylic acid moieties in the free and conjugated form is also a major route of metabolism of cypermethrin in the plant [see section CA 6.2 of this dossier]. In lettuce the presumed cis and trans cyclopropanecarboxylic acid moiety was glucose conjugated forming M310I008. When M310I008 is uptaken into the mamalian gastrointestinal tract via the diet it is expected to be cleaved to the aglycone i.e. M310I001 and thus to be covered by the toxicological evaluation of M310I001.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I008 is considered to be not genotoxic based on the available information on M310I001. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. Consequently M310I008 is considered to be **not toxicologically relevant.**

M310I009



M310I009 is a major metabolite found in rats and livestock animals (goat).

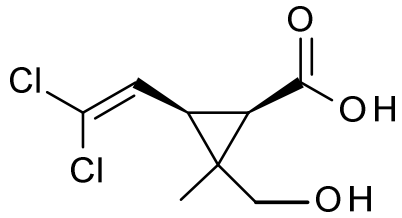
a. Toxicological evaluation of M310I009

Hydrolytic cleavage of the ester bound and elimination of the cis and trans cyclopropanecarboxylic acid moieties in the free and conjugated form is also a major route of metabolism of cypermethrin in the plant [see section CA 6.2 of this dossier]. In lettuce the presumed cis and trans cyclopropanecarboxylic acid moiety is glucose conjugated forming M310I008. This glycosylated compound is by malonylation on the glucose moiety metabolized into M310I009. When M310I009 is uptaken into the mamalian gastrointestinal tract via the diet it is expected to be cleaved to the aglycone i.e. M310I001 and thus to be covered by the toxicological evaluation of M310I001.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I009 is considered to be not genotoxic based on the available information on M310I001. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. Consequently M310I009 is considered to be **not toxicologically relevant**.

M310I003 (other denominators: HO-DCVA)



M310I003 is a mammalian metabolite found rat and livestock animals (goat, hen).

a. QSAR Predicitions on M310I003

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 6 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]9315]]
There were **no** Ames mutagenicity alerts for M310I003 or in-silico generated metabolites. Structural alerts were reported for the parent substance, namely "Haloalkenes with Electron-Withdrawing Groups". The same structural alerts were observed with several metabolites. For metabolite 6.1 the structural alert "Four-and Five-Membered Lactones" was reported.
The same holds true for in-vitro chromosome aberration. In all cases the structures were out of domain.

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 6 of report [see KCA 5.8.1/5 2014/1289317]]
M310I003 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. One of the six most similar molecules had positive experimental data. The other five molecules were predicted 'non-mutagen', which was experimentally confirmed. The concordance of the total underlying database was moderate (0.675) and thus is not very robust. Both the positive (similarity 0.755) and the negative molecules (similarity 0.742 to 0.802) have a reasonably similar structure to M310I003. Therefore, the chemical space does adequately cover the structure of M310I003.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 6 of report [see KCA 5.8.1/5 2014/1289317]]
M310I003 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The same metabolites as reported in the CAESAR model were also used in the SarPy model. Of the five molecules that showed negative experimental data two were predicted positive.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 6 of report [see KCA 5.8.1/5 2014/1289317]]
M310I003 is in the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. All of the six most similar molecules were predicted 'non-mutagen', which was experimentally confirmed. The concordance of the total underlying database was high (1.0) and thus is very robust. All six molecules (similarity 0.703 to 0.728) have a reasonably similar structure to M310I003. Therefore, the chemical space does adequately cover the structure of M310I003.

Conclusion on QSAR evaluations
No conclusive structural alert was identified for M310I003.

Toxicological evaluation of M310I003

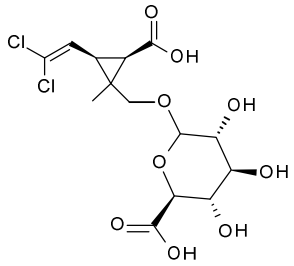
The QSAR evaluation of M310I003 is of moderate reliability and by weight of evidence there was no conclusive alert for genotoxicity.

Referring to the negative Ames test (see CA 5.8.1/1) of the structural similar substance M310I001 and the negative prediction for genotoxicity in various QSAR models, M310I003 is not considered to be genotoxic.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100% utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I003 is considered to be not genotoxic based on the available information on M310I001. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the relevant TTC-threshold.
In conclusion, M310I003 is considered to be **not toxicologically relevant**.

M310I002



M310I002 is a rat metabolite.

a. Toxicological evaluation of M310I002

Being a rat metabolite the toxicity of M310I002 is intrinsically covered by the toxicological testing of alpha-cypermethrin.

In conclusion M310I002 is considered to be **not toxicologically relevant**.

Toxicological evaluation of the Group C metabolites (DCVA, hydroxylated DCVA and its conjugates)

M310I004, M310I008 and M310I009 are o-bound glycosides of M310I001. These glycosides when uptaken into the human gastrointestinal tract via the diet are expected to be cleaved to the corresponding agylcone M310I001. The hydroxylated derivative of M310I001 i.e. M310I003 is due to its close structural relation to M310I001 not considered to be more toxic than M310I001. By weight of evidence M310I003 is not considered to be genotoxic. Thus the M310I001 derivatives, the glucones and M310I003 are considered to be covered by the toxicological evaluation of M310I001. M310I001 is not considered to be of toxicological relevance. Moreover M310I004 the glucuronid-ester of M310I001 is a major mammalian metabolite which is e.g. built in the rat at more than 30% of the applied dose. M310I003 and its glucuronid-ester M310I002 were also determined in the rat (1-2% of the applied dose). As the glucuronide ester M310I002 is formed from M310I003 whose precursor is the M310I001 itself the determination of the glucuronid ester in the rat further supports the coverage of the hydroxylated derivative M310I003 by the grouping approach.
Overall the group members are intrinsically covered by the mammalian metabolism and toxicological testing of the parent molecule alpha-cypermethrin. The central structure M310I001 and its derivatives did not provide and evidence for neurotoxicity and genotoxicity.

In conclusion the metabolites of the group C are by weight of evidence not considered to be genotoxic or neurotoxic. They are below the relevant thresholds of toxicological concern for acute and chronic dietary exposure and thus considered to be not toxicologically relevant.

Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates

I. Definition of group D: Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates

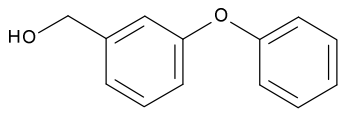
For the group of Group D) Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates, the following molecules were taken into consideration:

M310I024 (PBAlc)
M310I006 (o-glucoronide of PBAlc)
M310I018 (PBAld)
M310I011 (PBAcid)
M310I010 (glycine-conjugate of PBAcid)
M310I026 (glutamate-conjugate of PBAcid)
M310I013 and M310I025 (hydroxylated PBAcid)
M310I012 (glycine-conjugate of hydroxylated PBAcid)
M310I014 (sulfate-conjugate of hydroxylated PBAcid)

The redox-chain of phenoxy benzoic acid (PBAcid; M310I011), i.e. the aldehyde (M310I018) and the alcohol (M310I024) were grouped together. An o-glucoronide of M310I024 was included in this group. Furthermore the hydroxylated derivative of the phenoxy benzoic acid (M310I013/M310I025) was included here. For both acid metabolites M310I011 and M310I013/M310I025 glycine conjugates were determined in rat, livestock animals and/or plants i.e. M310I010 and M310I012. Furthermore a glutamate-conjugate of M310I011 was determined as a wheat metabolite (M310I026) and a sulfate conjugate of M310I013/M310I025 was determined as a rat metabolite (M310I014).

II. Evaluation of group D members: Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates

M310I024 (other denominators: CAS-No. 13826-35-2, Reg. No. 207323, PBAlc, 3-Phenoxybenzyl alcohol, WL 40673, CL 206128)



M310I024 is a plant metabolite found in wheat

a. QSAR Predicitions on M310I024

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 10 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for M310I024 or in-silico generated metabolites and no structural alerts were reported. In all cases the structures were out of domain.
For in-vitro chromosome aberration the prediction for M310I024 was **negative** (out of domain). None of the 6 in-silico metabolites was predicted positive (out of domain) and no structural alerts were reported.

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 10 of report [see KCA 5.8.1/5 2014/1289317]]
M310I024 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Three of the six most similar molecules had positive experimental data. The other three had the prediction non-mutagen, which was experimentally confirmed. The concordance of the total underlying database was moderate (0.67) and thus is not very robust. Both the positive (similarity 0.854 to 0.882) and the negative molecules (similarity 0.85 to 0.863) has a reasonably similar structure to M310I024. Therefore, the chemical space does adequately cover the structure of M310I024.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 10 of report [see KCA 5.8.1/5 2014/1289317]]
M310I024 is out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Three of the six most similar molecules had positive experimental data. The other three had the prediction non-mutagen, which was experimentally confirmed. The concordance of the total underlying database was low (0.33) and thus is not very robust. Both the positive (similarity 0.854 to 0.882) and the negative molecules (similarity 0.85 to 0.863) have a reasonably similar structure to M310I024. Therefore, the chemical space does adequately cover the structure of M310I024.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 10 of report [see KCA 5.8.1/5 2014/1289317]]
M310I024 could be out of the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. One of the six most similar molecules had positive experimental data. The concordance of the total underlying database was moderate (0.511) and thus is not very robust. All six molecules (similarity 0.827 to 0.87) have a reasonably similar structure to M310I024. Therefore, the chemical space does adequately cover the structure of M310I024.

Conclusion on QSAR evaluations
No conclusive structural alert was identified for M310I024.

Toxicological data of M310I024

For M310I024 results of toxicological studies are reported in the the DAR for lambda-cyhalothrin [see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013].

ADME

Supplemental information from open literature about toxicokinetics of pyrethroid metabolits in males and female rats was evaluated in the DAR or lambda-cyhalothrin [see Ueyama et al., 2010; discussed in Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 125-129] indicating that females eliminate M310I024 more rapid than male rats.

Acute toxicity

In non-accepted non-guideline studies with limited number of animals indication is given that the acute oral toxicity is greater than 500 mg/kg bw/day and the acute dermal toxicity > 4 ml/kg, that the compound is not irritant to the skin but irritating to the eye [see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 137-138, 140-141, 145-146, 150-151]. These studies were not considered acceptable and indicate that M301I024 is less toxic than alpha-cypermethrin.

Genotoxicity

Acceptable studies for genotoxicity in vitro - an Ames test, a Mouse Lymphoma Assay, and an in vitro chromosome aberration assay - were reported in the DAR for lambda-cyhalothrin [see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 188-187]. **No indications of mutagenic or clastogenic properties** were observed in these tests.

Endocrine modulation

Information with regard to the endocrine potential of M310I024 was found in the open literature. A summary table of the available literature is provided in Table 5.8.1-7 below. The Literature is however described in more detail in the chapter CA 5.8.3.

Endocrine modulation In vitro studies:
M310I024 has been reported to show distinct estrogenic effects on mRNA expression level for estrogen receptor and pS2 at approx. 1000-fold higher concentrations than beta-E2 (Jin et al., 2010; [see CA 5.8.3/17 2010/1232199]). Additionally it showed week estrogenic activity in two ER-dependent transactivation assays in the yeast (Tyler et al., 2000; [see CA 5.8.3/2 2000/1024078] and McCarthy et al., 2006; [see CA 5.8.3/16 2006/1051135]) but not in MCF-7 cells up to 10 µM (Laffin et al., 2010; [see CA 5.8.3/16 2010/1232196] and Tange et al., 2014; [see CA 5.8.3/2 2000/1024078]). The potency of M310I024 in yeast was about 100.000-fold lower than that of the positive control 17β-E2. No ER-antagonistic properties were found in yeast cells. Inconsistent data were reported from E-Screen assay in MCF-7 cells, one time inducing proliferation at 0.1 µM (Jin et al., 2010, [see CA 5.8.3/17 2010/1232199]), the other time showing no activity up to 10 µM (Laffin et al., 2010, [see CA 5.8.3/16 2010/1232196]). An induction of proliferation of MCF-7 cells is not directly attributable to an estrogenic effect; therefore the relevance of this information is questionable.

M310I024 was not active as androgen receptor agonist in yeast (Tyler et al., 2000; [see CA 5.8.3/2 2000/1024078]) and in mammalian cells (Tange et al., 2014; [see CA 5.8.3/9 2014/1242695]). An anti-androgenic potential was seen in yeast cells in the µ-molar range, however in mammalian cells the transactivation of human androgen receptor was not reduced to 50% even at 100 µM. Therefore this finding is rather considered unrelevant for further consideration.

Endocrine modulation In vivo studies:
Laffin et al., 2010 [see CA 5.8.3/16 2010/1232196] evaluated the estrogenic activity in vivo using the uterotrophic assay in Sprague-Dawley rats. Ovariectomized animals were orally gavaged at 1, 5, and 10 mg/kg bw of M310I024 dissolved in corn oil.once daily for 3 days. No effect on uterine wet weigth or body weight was found. In addition no effect on organ weights, onset of puberty and sexual maturation was found in a pubertal assay in Sprague-Dawley rats at 0, 1, 5 or 10 mg/kg bw/d administered from weaning (PND 22) until detection of the onset of puberty by vaginal opening.

Considering all available data on endocrine potential M310I024 can be considered as non active with regard to anti-estrogenic and androgenic potentail. Some anti-androgenic properties were found in yeast, but in mammalian cells only at higher concentrations so that this effect is not considered relevant for the in vivo situation. The estrogenic effect which was repeatedly found in vitro was not substantiated in a pubertal female assay or in an uterotrophic assay.

Table 5.8.1-7: Endocrine activity studies with M310I024

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference Doc ID
In vitro assays				
ER-dependent regulation of mRNA expression	Estrogen Receptor (ERα) mRNA levels in MCF-7	Estrogenic active RIE: 88.7% at 1µM	E2: ↓(at 1nM) RIE set to 100%	Jin et al., 2010 2010/1232199
ER-dependent regulation of mRNA expression	pS2 mRNA levels in MCF-7	Estrogenic active RIE: 81.8% at 1µM	E2: 4-fold ↑ (1 nM) RIE set to 100%	
ER-activation	hER transactivation in Yeast (lacZ)	ER-agonist: Active EC_{50}: 2±0E-05 M ER-antagonist: Not active	Agonist: E2: EC_{50}: 2.1±0.4E-10 M Antagonist:4HO-TAM: IC_{50}: 2.8±0.7E-6 M	Tyler et al., 2000 2000/1024078
ER-activation	hER-α transactivation in Yeast (lacZ)	ER-agonist: Active EC_{50}: 6.67±3.1E-6M	Agonist: E2: EC_{50}: 0.35±0.16E-9M	McCarthy et al., 2006 2006/1051135
ER-activation	hER transactivation in MCF-7 cells (Luciferase)	ER-agonist: Not active (up to 10 µM)	Agonist: E2 (10 nM):2.3-fold induction	Laffin et al., 2010 2010/1232196
ER-activation	hER transactivation in MCF-7 cells (Luciferase)	ER-agonist: not active EC20 at 20 µM	Agonist: E2 EC20: 2.4 E-8M	Tange et al., 2000 2000/1024078
ER-dependent proliferation	E-Screen (MCF-7)	Proliferative effect: not active (at 1nM-10 µM)	E2: 3-fold ↑ at 1 nM	Laffin et al., 2010 2010/1232196
ER-dependent proliferation	E-Screen (MCF-7)	Proliferative effect: active at 0.1 µM PE: 1.7 RPE: 62.5%	E2: 1 nM PE: 2.12 RPE: 100%	Jin et al., 2010 2010/1232199
AR-activation	hAR transactivation in Yeast (lacZ)	AR-agonist: Not active AR-antagonist: active LOIC: 3.5±0.5E-06 M IC_{50}: 3.7±0.7E-05 M	Agonist: DHT: EC_{50}: 9.7±0.3E-10 M Antagonist: Flutamide: IC_{50}: 6.8±0.7E-6 M	Tyler et al., 2000 2000/1024078
AR-activation	hAR transactivation in CHO cells (Luciferase)	AR-agonist: not tested AR-antagonist: active IC_{20}: 1.15E-6 M IC_{50}: n.d. (up to 100 µM)	Antagonist: Flutamide: IC_{20}: 0.16E-6 M IC_{50}: 0.62E-6M	Tange et al., 2014 2014/1242695

In vivo assays				
Uterus weight	Uterotrophic assay in SD rats	Ostrogenic acivity: Not active	Agonist: EE 0.6µg/kg bw Uterine wet weight: 5x ↑	Laffin et al., 2010 2010/1232196
Age at VO, Age at first diestrus	Pubertal female assay in SD rats	Ostrogenic acivity: Not active	No control	

PE: Proliferative effect; E2: 17 beta-Estradiol; EE: ethinyl estradiol; 4HO-TAM: 4-Hydroxy-tamoxifen; RPE: relative proliferative effect calculated as the ratio (PE-1) of the test chemical over (PE-1) of E2 (x 100); RIE: The relative inductive efficiency (RIE) is the ratio between the maximal up-regulation of pS2 expression level by the test compound to that of E2 (x100) or the relative inhibitory effciency of down-regulation of ERalpha expression level by the test compound compared to that of E2 (x 100); EC50/IC50: Effect concentration /Inhibitor concentration at which 50% increase /decrease is found compared to control; LOIC: Lowest observed inhibitory concentration

Toxicological evaluation of M310I024

There is no evidence for M310I024 for substantial acute toxicity.

The QSAR evaluation of M310I024 is of moderate reliability and by weight of evidence there was no conclusive alert for genotoxicity.

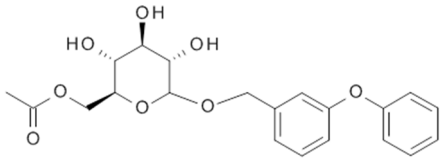
For M310I024 results from an Ames test, Mouse Lymphoma Assay, and an in vitro chromosome aberration assay were reported in the DAR for lambda-cyhalothrin [see Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 188-187]. No indications of mutagenic or clastogenic properties were observed in these tests. Therefore, based on the negative in vitro data regarding mutagenicity and clastogenicity in combination with the negative prediction for M310I024 in the various QSAR models, M310I024 is considered to be not genotoxic.

Furthermore M310I024 is considered to have no relevant endocrine activity.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I024 is not considered to be genotoxic and not considered to bear relevant endocrine activity. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. In conclusion, M310I024 is considered to be **not toxicologically relevant**.

M310I006



M310I006 is a plant metabolite (lettuce). Structurally is an o-glucuronide of M310I018.

a. Toxicological evaluation of M310I006

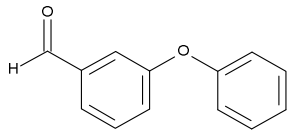
When M310I006 is uptaken into the mammalian gastrointestinal tract via the diet the o-glucuronide is expected to be cleaved to the aglycone i.e. M310I024 and thus to be covered by the toxicological evaluation of M310I024.

Based on the reported negative in vitro data regarding mutagenicity and clastogenicity for the aglycone M310I024 in combination with the negative prediction for M310I024 in the various QSAR models, M310I006 is considered to be not genotoxic.

Furthermore M310I006 based on the data available for the aglycone is considered to have no relevant endocrine activity.
With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I006 is considered to be not genotoxic based on the available information on M310I024. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the TTC-threshold. Consequently M310I006 is considered to be **not toxicologically relevant**.

M310I018 (other denominators: CAS-No. 39515-51-0, Reg. No. 4080665, PBAld, EC No. 254-487-1, WL 42049, CL 206969, 3-phenoxybenzaldehyde)



M310I018 is a metabolite found in plants (wheat) and is build by photolysis/degradation in water. M310I018 is registered at ECHA as an intermediate.

a. QSAR Predicitions on M310I018

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 13 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for M310I018 or in-silico generated metabolites and no structural alerts were received. In all cases the structures were out of domain. The same holds true for the in vitro chromosome aberration.

VEGA: Mutagenicity model (CAESAR, version 2.1.12) [see molecule 6 of report [see KCA 5.8.1/6 2014/1319956]]
3-Phenoxybenzaldehyde could be out of the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts however the result show some critical aspects. The prediction is based on 6 molecules, of which three were predicted and actual 'mutagen'. The similarity to 3-Phenoxybenzaldehyde of the molecules in the data set is quite good as indicated by similarity factors of 0.848 to 0.871. The concordance of the total underlying database was bad (0.333) and thus is not very robust.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 6 of report [see KCA 5.8.1/6 2014/1319956]]
3-Phenoxybenzaldehyde could be out of the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The prediction is based on 6 molecules, of which three showed positive experimental values. The similarity of 3-Phenoxybenzaldehyde to the molecules in the data set is quite good as indicated by similarity factors of 0.848 to 0.871. The concordance of the total underlying database was bad (0.333) and thus is not very robust.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 6 of report [see KCA 5.8.1/6 2014/1319956]]
3-Phenoxybenzaldehyde is out of the model applicability domain. The prediction is **'mutagen'** with the specific structural alerts SA11 (Simple aldehyde) but with a low reliability as the most related structure of the model dataset having the same fragment was actual non-mutagen. The overall prediction is based on 6 molecules, of which two were predicted and actual 'mutagen' while the other four were non-mutagen. The similarity to 3-Phenoxybenzaldehyde of the molecules in the data set is quite good as indicated by similarity factors of 0.823 to 0.882. The concordance of the total underlying database was moderate (0.483) and thus is not very robust.

Conclusion on QSAR evaluations
A structural alert for mutagenicity was identified with the Toxtree module while there was no alert in the other models applied. However, the genotoxicity studies discussed below on M310I018 do not provide any evidence for genotoxity.

Toxicity studies on M310I018

The datapackage published on the ECHA homepage for registration as an intermediate (Joint Sumbission) comprises of acute and local tolerance studies only [http://apps.echa.europa.eu/registered/data/dossiers/DISS-9eb9f3aa-13ef-2d26-e044-00144f67d031/AGGR-afb51bdd-4549-4bc9-9043-798d39a91cab_DISS-9eb9f3aa-13ef-2d26-e044-00144f67d031.html#L-089e8c2d-346d-4d75-b2cf-5d474bd41acf]. Based on these studies a classification Acute Tox 4 H302: Harmful if swallowed and Acute Tox 2 H330: Fatal if inhaled is proposed. The acute dermal toxicity study and the skin and eye irritation studies did not guide to a hazard classification.

An extensive datapackage on M310I018 is available to the applicant from the TSCATS database. A summary table of the available studies is provided in Table 5.8.1-8 below. The acute studies and local tolerance studies although not discussed in detail support the classification proposal on the ECHA homepage. However there is additional evidence that M310I018 might be skin sensitizer. Studies of the TSCATS database considered relevant for dietary metabolite assessment are described in more detail in the following section.

Table 5.8.1-8: Summary of TSCATS toxicological studies on M310I018

Study Title	Substance	Species/Sex	Dose range (vehicle)	Result	Guideline/GLP	Reference (BASF DocID)
Ninety Day Subchronic Oral Toxicity Study of m-PBAD in albino rats	M310I018, Lot No.: 82-92, Purity: 97%	Sprague-Dawley rats (M/F)	50, 150, 300 mg/kg bw/day (corn oil)	NOEL: 50 mg/kg bw	Equivalent to OECD 408, not GLP, QAU	1980/1001707
Histopathology results from 90-day oral rat study with meta-phenoxybenzaldehyde (final report)	M310I018, Lot No.: 82-92, Purity: 97%	Sprague-Dawley rats (M/F)	50, 150, 300 mg/kg bw/day (corn oil)	NOEL: 50 mg/kg bw		1980/1001708
Disposition of 14C-labeled meta-phenoxybenzaldehyde following oral dosing in rats (final report)	^{14}C-labelled M310I018 radiochemical purity: 99.9%, specific activity 11.3 mCi/mmol	Sprague-Dawley rats (M/F)	150 mg/kg bw/day (corn oil)	Readily absorbed and distributed to various organs and tissues. Higher urinary excretion in males vs. higher fecal excretion in females.	-	KCA 5.8.1/13 1981/1001421
Activity of 3-PBAD in a test for differential inhibition of repair deficient and repair competent strains of Eschericia coli: DNA repair	M310I018	E. coli: W3110/polA+ E. coli: P3478/polA-	2, 10, 20 µL	Negative	No guideline available, not GLP	KCA 5.8.1/14 1978/1001604
In vitro mutation assay in the presence/absence of metabolic activation.	M310I018, T1407 A/B	Balb/3T3 cells	1/20000, 1/40000, 1/80000 dilution	negative with/without metabolic activation	No guideline available, not GLP	KCA 5.8.1/15 1979/1001664, KCA 5.8.1/16 1979/1001665

Study Title	Substance	Species/Sex	Dose range (vehicle)	Result	Guideline/GLP	Reference (BASF DocID)
In vitro cell transformation assay in the presence/absence of metabolic activation.	M310I018, T1408 A/B	Balb/3T3 cells	1/20000, 1/40000, 1/80000 dilution	no morphological transformation was observed	No guideline available, not GLP	KCA 5.8.1/17 1979/1001666, KCA 5.8.1/18 1979/1001667
Activity of T1409 in the in vivo cytogenetics assay in rodents	M310I018, T1409, purity: 97%	Sprague-Dawley rats (M)	2.2, 22, 220 µL/kg bw (corn oil)	Little or no clastogenic activity was observed. No dose-response relationship was observed.	Similar to OECD 475, not GLP, QAU	CA 5.8.1/32 1979/1001668
Dominant lethal assay	M310I018, T1410	Sprague-Dawley rats (M/F)	2.16, 21.6, 216 mg/kg bw/day (corn oil)	Little or no mutagenic activity was observed. No influence on fertility up to 216 mg/kg bw	Similar to OECD 478, not GLP	CA 5.8.1/33 1979/1001669
Ames Salmonella/Microsomal plate test (EPA/OECD) PH301-ET-002-05 with meta-phenoxybenzaldehyde with attachments and cover letter dated 112191	M310I018	S. typhimurium strains: TA 98, TA 100, TA 1535, TA 1537, TA 1538	3, 10, 30, 100, 300 µg/plate (DMSO)	Negative (±S9)	Similar to OECD 471, GLP (see page 14 of report) & QAU	CA 5.8.1/31 1985/1002161

Study Title	Substance	Species/Sex	Dose range (vehicle)	Result	Guideline/GLP	Reference (BASF DocID)
Toxicity of WL 43775 intermediates: Acute toxicity, skin and eye irritancy and skin sensitization of 3-Phenoxybenzaldehyde	M310I018	Acute oral/dermal: CD rat Skin/eye irritation: NZW rabbits Sensitization: guinea pig (P-strain)	Acute oral/dermal: 500/300 mg/kg bw (0.5% CMC) Skin/eye irritation: 0.5 mL/0.2 mL (undiluted) Sensitization: intradermal induction: 0.1% (corn oil) Topical induction: 25% (corn oil) Topical challenge: 15% (corn oil)	Acute oral/dermal: LD_{50} > 500/>300 mg/kg bw Skin/eye irritation: Not irritating Sensitization: not sensitizing to skin	Acute oral/dermal: Similar to OECD 401/402 Skin/eye irritation: Similar to OECD 404/405 Sensitization: Similar to OECD 406 (GPMT) No GLP, no QAU	KCA 5.8.1/19 FE-470-006 (1976)
Acute oral toxicity study in rats with meta-phenoxybenzaldehyde with attachments and cover letter dated 112591	M310I018, Lot No. 1	Sprague-Dawley rats	5000 mg/kg bw (water)	LD_{50} > 5000 mg/kg bw	Equivalent to OECD 401, GLP & QAU	KCA 5.8.1/20 1985/1002156

Study Title	Substance	Species/Sex	Dose range (vehicle)	Result	Guideline/GLP	Reference (BASF DocID)
Acute oral toxicity study in rats (14 days): m-PBAD, Lot 450 (Finale Report) with attachments and cover letter dated 112191 LD_{50} test	M310I018, Lot No. 450	Sprague-Dawley rats	100, 1250, 1600, 2500 mg/kg bw (water)	LD_{50} = 1499 mg/kg bw	Equivalent to OECD 401, GLP & QAU	KCA 5.8.1/21 1985/1002153
Acute oral toxicity study in rats (14 days): m-PBAD-III, Lot 1 (Finale Report) with attachments and cover letter dated 112191 LD_{50} Limit test & LD_{50} test	M310I018, Lot No. 1	Sprague-Dawley rats	LD_{50} limit test: 5000 mg/kg bw (water) LD_{50} test: 800, 1000, 1250, 1600, 2500 mg/kg bw (water)	LD_{50} limit test: LD_{50} < 5000 mg/kg bw (mortality: 10/10) Necropsy findings (in dead animals): red lungs, lesions in the stomach, fluid filled intestine and discoloured adrenals LD_{50} test: LD_{50} = 1222 mg/kg bw	Equivalent to OECD 401, GLP & QAU	KCA 5.8.1/22 1985/1002152
Acute dermal toxicity test in rabbits with meta-phenoxybenzaldehyde with attachments and cover letter dated 112591	M310I018, Lot No. 1	New Zealand White Rabbits	5000 mg/kg bw (undiluted)	LD_{50} > 5000 mg/kg bw	Equivalent to OECD 402, GLP & QAU	KCA 5.8.1/23 1985/1002154

Study Title	Substance	Species/Sex	Dose range (vehicle)	Result	Guideline/GLP	Reference (BASF DocID)
Acute dermal toxicity test in rabbits with meta-phenoxybenzaldehyde with attachments and cover letter dated 112591	M310I018, Lot No. 450	New Zealand White Rabbits	5000 mg/kg bw (undiluted)	LD_{50} > 5000 mg/kg bw	Equivalent to OECD 404, GLP & QAU	KCA 5.8.1/24 1985/1002155
Acute eye irritation test with meta-phenoxybenzaldehyde with attachments and cover letter dated 112591	M310I018, Lot No. 450	New Zealand White Rabbits	0.1 mL (undiluted)	Not irritating to the eye	Equivalent to OECD 405, GLP & QAU	KCA 5.8.1/25 1985/1002157
Primary dermal irritation study in rabbits: m-PBAD-III Lot No. 1 (Final Report) with attachments and cover letter dated 112591	M310I018, Lot No. 1	New Zealand White Rabbits	0.5 mL (undiluted)	Not skin irritating	Equivalent to OECD 404, GLP & QAU	KCA 5.8.1/26 1985/1002158
Primary dermal irritation study in rabbits: m-PBAD Lot No. 450 (Final Report) with attachments and cover letter dated 112591	M310I018, Lot No. 450	New Zealand White Rabbits	0.5 mL (undiluted)	Not skin irritating	Equivalent to OECD 404, GLP & QAU	KCA 5.8.1/27 1985/1002159

Study Title	Substance	Species/Sex	Dose range (vehicle)	Result	Guideline/GLP	Reference (BASF DocID)
Four hour acute aerosol inhalation toxicity study in rats of m-PBAD-III (Final Report) with attachments and cover letter dated 112191	M310I018, Lot No. 1	Crl:CD(SD)BR	0.44, 0.91, 1.51, 1.68, 2.79 mg/L (aerosol)	LD_{50} (combined): 0.64 mg/L LD_{50} (male): 1.15 mg/L LD_{50} (female): 0.27 mg/L	Equivalent to OECD 403, GLP & QAU	KCA 5.8.1/28 1985/1002160
Four hour acute aerosol inhalation toxicity study in rats of m-PBAD (Final Report) with attachments and cover letter dated 112191	M310I018, Lot No. 450	Albino rats	0.88, 1.21, 2.74, 5.25 mg/L (aerosol)	LD_{50} (combined): 1.03 mg/L LD_{50} (male): 1.27 mg/L LD_{50} (female): 0.65 mg/L	Equivalent to OECD 403, GLP & QAU	KCA 5.8.1/29 1984/1002182
Guinea pig contact dermal irritation/sensitization: MPAD (Final Report) with cover letter dated 112191	M310I018	Guinea pig	Induction: intradermal injection (1x0.05 mL, 9x0.1 mL) Challenge: intradermal injection (1x0.05 mL)	Irritating to skin, Skin sensitizer (8/10 animals with positive reaction)	Similar to Draize test (listed in OECD 406, adopted 1981), not GLP	KCA 5.8.1/30 1979/1001663

Report: CA 5.8.1/31
Barfknecht T.R., 1991a
m-PBAD-III Lot 1 - Ames Salmonella/Microsome plate test (EPA/OECD) PH 301-ET-002-85 with meta-phenoxybenzaldehyde with attachements and cover letter
1985/1002161
Guidelines: none
GLP: no

Executive Summary

S. typhimurium (strains TA98, TA 100, TA 1535, TA 1537 and TA 1538) were exposed to M310I018; Lot No. 1) using DMSO as a solvent in the presence and absence of metabolic activation in one plate incorporation test. Triplicate plates were used per dose and per test condition. Vehicle and positive controls were included in each experiment. The test substance was used at concentrations of 3, 10, 30, 100, and 300 µg/plate in the presence and absence of S9-mix. Bacteriotoxic effects (pindot colonies) were observed in a range finding test at concentrations of 500 µg/plate or higher in TA 100 and TA 1535. In TA 100 a reduced number of revertant colonies was observed at the 160 µg/plate level. A biologically relevant increase in the number of revertant colonies was not noticed in any of the strains tested in presence or absence of metabolic activation in any of the experiments. The positive controls induced the appropriate response in the corresponding strains in the absence or presence of metabolic activation. According to the results of the study the test substance the test substance was not mutagenic in the Salmonella typhimurium mutation assay under the experimental conditions of the study.

(Doc ID 1985/1002161)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material 3-PBAD III (Meta-phenoxybenzaldehyde (Metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	liquid / yellow
Lot/Batch #:	Lot 1
Purity:	N.A.
Stability of test compound:	The test substance was stable over the study period

2. Control Materials:

Vehicle control: The vehicle control with and without S-9 mix only contained the vehicle used for the test substance at the same concentration and volume for all tester strains.

Positive control compounds tested without addition of metabolic activation system:

Strain	Mutagen	Solvent	Concentration
TA 100	Sodium azide (NaN_3)	Water	10 µg/plate
TA 1535	Sodium azide (NaN_3)	Water	5 µg/plate
TA 1537	9-Aminoacridine	DMSO	150 µg/plate
TA 1538	2-Nitrofluorene	DMSO	5 µg/plate
TA 98	2-Nitrofluorene	DMSO	5 µg/plate

Positive control compounds tested with addition of metabolic activation system:

Strain	Mutagen	Solvent	Concentration
TA 100	2-Aminoanthracene	DMSO	5 µg/plate
TA 1535	2-Aminoanthracene	DMSO	5 µg/plate
TA 1537	2-Aminoanthracene	DMSO	5 µg/plate
TA 1538	2-Aminoanthracene	DMSO	5 µg/plate
TA 98	2-Aminoanthracene	DMSO	5 µg/plate

3. Activation:

S9 was produced from the livers of Sprague-Dawley rats treated with Aroclor 1254. The S9-mix was prepared freshly prior to each experiment. For this purpose, a sufficient amount of S9-fraction is thawed at room temperature and 1 volume of S9-fraction is mixed with 9 volumes of S9-supplement (cofactors). This preparation, the so-called S9-mix, was kept on ice until used.

The rat liver S9-mix was prepared immediately before use and had the following composition:

Component	Concentration
Sodium phosphate buffer (pH 7.4)	100 mM
Glucose 6-phosphate	5 mM
NADP	4 mM
KCl	33 mM
$MgCl_2$	8 mM
S9	10 %

No information regarding the efficiency of the S9-batch, in general, was provided (i.e. incubation with B[a]P).

4. Test organisms:

S. typhimurium strains: TA98, TA100, TA1535, TA1537, TA1538

5. Test concentrations:

Preliminary experiment: Duplicate plates were prepared for each concentration (solvent control; 50, 166, 500, 1666, and 5000 µg/plate) in the absence of S9-mix in Salmonella strains TA 1535 and TA 100.

Plate incorporation assay: Triplicate plates in one experiment were prepared for each concentration (neg. control; 3, 10, 30, 100, and 300 µg/plate) and condition (i.e. with and without S9) for all tester strains.

B. TEST PERFORMANCE:

1. Dates of study period: 15 January, 1985 – 4 March 1985

2. Preliminary toxicity test:

In order to assess bacteriotoxic effects, a dose range finding test was conducted with Salmonella typhimurium TA 100 and TA 1535 up to 5000 µg/plate test substance concentration. Top agar, used as an overlay, was reconstituted into a molten state and supplemented with 0.5 mM histidine – 0.5 mM biotin at a volume of 0.1 mL/mL of agar, and maintained at 45°C until used. Two mL of top agar solution, 0.1 mL of tester strain and 0.1 mL of the appropriate concentration of the test compound were added to a sterile tube, vortexed and poured onto minimal glucose plates.

3. Plate-incorporation assay:

100 µL volumes of solution of the test substance were added to top agar mix to give final concentrations of 3, 10, 30, 100, and 300 µg/plate. The procedure was identical to that described for the preliminary toxicity test. Assays were carried out both in the presence and in the absence of rat liver S9 fraction. The cultures were incubated at 37°C for 48 h before the revertant colonies were counted. Means and standard deviations were calculated.

4. Statistics:

No special statistical tests were performed.

5. Evaluation criteria:

The test chemical is considered positive in this assay if the following criteria are met:

- A reproducible and significant dose-related increase of revertant colonies
- An increase of revertant colonies equal to or greater than three times the solvent control value, if the solvent control values were within the 95% confidence limit of the historical control data.

A negative result is defined as the absence of a reproducible increase in the number of histidine-independent colonies.

II. RESULTS AND DISCUSSION

A. PRELIMINARY TOXICITY TEST

Cytotoxicity (pindot colonies) was observed at 500, 1666, and 5000 µg/plate for TA 100 and TA 1535 in the preliminary toxicity test. Strain TA 100 also exhibited a reduced number of revertant colonies at the 160 µg/plate level. Based on these findings the top dose selected for the main experiment was 300 µg/plate.

B. MAIN EXPERIMENT

In the main experiment with and without metabolic activation no biologically relevant increase in number of revertants was observed in any strain tested [see Table 5.8.1-9]. The positive controls yielded revertant numbers in a range expected for the respective strains and thus demonstrated the sensitivity of the test system.

III. CONCLUSION

According to the results of the present study, the test substance M310I018 is not mutagenic in the Salmonella typhimurium assay under the experimental conditions applied.

Table 5.8.1-9: Bacterial gene mutation assay with M310I018 – Mean number of revertants

Experiment 1: Plate incorporation assay										
Strain	TA 98		TA 100		TA 1535		TA 1537		TA 1538	
Metabol. activation	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
Neg. control (DMSO)	43	37	211	238	14	22	15	14	23	14
M310I018										
3 µg/plate	45 ± 6	40 ± 11	194 ± 20	179 ± 10	16 ± 6	22 ± 6	16 ± 4	14 ± 3	27 ± 2	15 ± 2
10 µg/plate	47 ± 4	31 ± 6	182 ± 12	216 ± 12	16 ± 3	20 ± 4	16 ± 1	14 ± 4	30 ± 4	15 ± 6
30 µg/plate	51	27 ± 10	162 ± 23	183 ± 21	12 ± 4	22 ± 5	16 ± 3	7 ± 3	28 ± 2	11 ± 3
100 µg/plate	43 ± 6	30 ± 4	140 ± 24	181 ± 36	13 ± 4	20 ± 6	20 ± 8	15 ± 3	30 ± 3	10 ± 2
300 µg/plate	32 ± 11	12 ± 5	142 ± 35	11 ± 7	15 ± 6	9 ± 6	13 ± 6	3 ± 3	16 ± 2	1 ± 1
Pos. control[§]	1523	319	1818	868	260	904	432	891	1260	563

[§] = Compound and concentrations see Material and Methods (I.A.2.) above

Report: CA 5.8.1/32
Schechtman L.H., 1991a
m-PBAD - Activity of T1409 in the in vivo cytogenetics assay in rodents (final report with cover letter)
1979/1001668
Guidelines: none
GLP: no

Executive Summary

M310I018 (purity: 97%) was tested in vivo for the ability to induce chromosome and numerical aberrations in bone marrow cells of male Sprague-Dawley rats. Five animals per dose group were treated with 0.22, 2.2, and 22 µL/kg bw once daily by gavage for 5 consecutive days, with an administration volume of 4 mL/kg bw (corn oil). Two hours after administration of the final treatment, the animals were injected intraperitoneally with 4 mg/kg bw colchicine. Four hours after treatment with colchicine, the animals were sacrificed by carbon dioxide asphyxiation and both femurs and tibias were removed. Bone marrow cells were isolated, fixed with Carnoy´s fixative, and stained with Giemsa. Vehicle (corn oil) and positive control (TEM, 0.5 mg/kg bw) were included to demonstrate the sensitivity of the test system.

One animal of dose group 3 demonstrated a gradual weight loss and died after administration of the fifth dosage. On necropsy, there were no gross lesions observed. Otherwise, there were no adverse effects from the treatment and other animals died as a result of the dosing regimen. There was no change in ploidy in the treatment groups relative to the negative control. The damage was so severe in the positive control that the chromosome number could not be counted. The mitotic index of all treatment groups was unchanged from that of the negative control. TEM was a moderate mitotic inhibitor. The percentage of cells with aberrations for all treatment groups was not significantly ($p>0.05$) increased relative to the negative control. The group receiving TEM demonstrated severe damage, with approximately 74% of all cells analyzed containinig one or more aberrations. The aberrations per cell for all treatment groups were not significantly increased ($p>0.05$) relative to the negative control using the t-test. The group receiving TEM demonstrated severe damage, with approximately 5.86 aberrations per cell.

Based on the results of this study, it can be stated that under the experimental conditions reported, M310I018 did not induce chromosome aberrations in bone marrow cells from Sprague-Dawley rats.

(BASF DocID 1979/1001668)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: T1409 (3-Phenoxybenzaldehyde)

Description:	Liquid, pale yellow
Density:	1.49
Lot/Batch #:	not reported
Purity:	97%
Stability of test compound:	not reported
Solvent used:	Corn oil

2. Control Materials:

Negative control:	A negative control was not employed in this study.
Solvent control:	Corn oil
Positive control:	Triethylenemelamine (TEM)

3. Test animals:

Species:	Rat
Strain:	Sprague-Dawley
Sex:	male
Age:	4-6 weeks
Weight at dosing:	125 - 200 g
Source:	Charles River Breeding Farms, Wilmington, USA
Acclimation period:	Not reported (10-14 days of quarantine)
Diet:	Purina Rat Chow, ad libitum
Water:	Tap water, ad libitum
Housing:	Group housing (5 animals/cage)
Environmental conditions:	
Temperature:	not reported
Humidity:	not reported
Air changes:	not reported
Photo period:	not reported

4. Test concentrations:

Main experiment:	0.44, 4.4, and 44 µL/kg bw (at a dosing volume of 4 mL/kg bw of T1409 in corn oil)

B. STUDY DESIGNS AND METHODS:

1. Dates of experimental work: 23-July-1979 to 06-Aug-1979

2. Main experiment:

Treatment

Five animals per group were dosed with 0.44, 4.4, and 44 µL/kg bw of the test substance once daily by oral gavage for 5 consecutive days. The calculated total dose was 2.2, 22, and 22 µL/kg bw. The solvent control animals received corn oil at 4 mL/kg bw by gavage for 5 consecutive days. Positive control animals received Triethylenemelamine (TEM) intraperitoneally as a single dose of 0.5 mg/kg bw one day prior to sacrifice. Two hours after administration of the final treatment, the animals were injected intraperitoneally with 4 mg/kg bw colchicine.

Preparation of the animals

Four hours after treatment with colchicine, the animals were sacrificed by carbon dioxide asphyxiation and both femurs and tibias were removed. The epiphysis was broken and the marrow aspirated into HBSS. The cells were centrifuged at 1000 rpm for 8-10 minutes, the supernatant decanted, and the cell pellet resuspended in 8 mL of 0.075 M KCl. The tubes were incubated in a 37°C water bath for 20-30 min, centrifuged at 1000 rpm for 8-10 min and the supernatant decanted. The cells were resuspended in 5 mL Carnoy´s fixative and incubated at 4°C for 2 h. Afterwards cells were centrifuged at 1000 rpm for 8-10 min, the supernatant discarded, and the cells resuspended in 5 mL Carnoy´s fixative. After an overnight (16-20 h) incubation at 4°C, the cells were centrifuged at 1000 rpm for 8-10 min and the supernatant decanted. The cell pellet was resuspended to opalescence in Carnoy´s fixative. Two to 5 slides were prepared from each animal. Slides were stained with Giemsa and permanently mounted.

Analysis of metaphase cells

A minimum of 50 metaphase spreads from each animal was examined and scored for chromatid and chromosomal gaps and breaks, structural arrangements, ploidy, fragmentation and pulverization. The mitotic index was recorded for each rat as the number of cells in mitosis/100 cells observed. Each cell was classified into a single category (chromatid or chromosome gaps, breaks, structural rearrangements, fragments and pulverizations) according to the most severe damage that occurred. The percentage of damaged cells in the total population of cells examined was calculated for each treatment group. Chromatid and chromosome gaps are not included in the total percentage of cells with one or more aberrations. The severity of damage within the cells is reported as the number of aberrations per cell per treatment dose.

3. Statistics:

Chi-square analysis using a 2x2 contingency table was used to ascertain significant relationships between the number of cells with aberrations in the treatment group relative to the negative control. The t-test was used to compare pairwise the number of aberrations per cell of the treatment group with that of the negative control. Each comparison was considered to be between two independent, random samples of unequal variance and a significant increase in the treatment mean relative to the negative control (one-sided) was being sought.

4. Acceptance and evaluation criteria:

Acceptance criteria

The study was considered valid as the following criteria are met:

- The mitotic index for all animals of the negative control group must be at least 8%.
- The number of cells demonstrating chromosome and chromatid breaks and gaps must be less than or equal to 2% of the total cells analysed for each negative control animal.
- The mitotic index for the positive control is expected to be between 1-4%. The average number of cells demonstrating chromosome or chromatid aberrations must be at least 55% of the total cells analyzed in the positive control group.

Evaluation criteria

The test chemical is classified mutagenic if it induces significant damage (number of aberrations per cell/percentage of damaged cells). No further details were reported.

II. RESULTS AND DISCUSSION

A. MAIN EXPERIMENT:

One animal of dose group 3 demonstrated a gradual weight loss and died after administration of the fifth dosage. On necropsy, there were no gross lesions observed. Otherwise, there were no adverse effects from the treatment and o other animals died as a result of the dosing regimen.
There was no change in ploidy in the treatment groups relative to the negative control. The damage was so severe in the positive control that the chromosome number could not be counted. The mitotic index of all treatment groups was unchanged from that of the negative control. TEM was a moderate mitotic inhibitor.
The percentage of cells with aberrations for all treatment groups was not significantly ($p>0.05$) increased relative to the negative control. The group receiving TEM demonstrated severe damage, with approximately 74% of all cells analyzed containinig one or more aberrations.
The aberrations per cell for all treatment groups were not significantly increased ($p>0.05$) relative to the negative control using the t-test. The group receiving TEM demonstrated severe damage, with approximately 5.86 aberrations per cell.

Table 5.8.1-10: M310I018: Percentage of cells with aberrations

Experimental group	% of total cells analyzed				
	Gap	Break	Rearrangement	>10	with aberrations (without gaps)
Negative control	0.4	0.8	9.2	0	10.0
M310I018					
220 µL/kg bw	0	0	13.2	0	13.2
22 µL/kg bw	0	0	15.0	0	15.0
2.2 µL/kg bw	0.4	0	11.6	0	11.6
Positive Control TEM	0.4	2.0	18.0	53.6	73.6

Table 5.8.1-11: M310I018: Severity of damage as aberration per cell (250 cells scored)

Experimental group	Gaps per cell	Breaks per cell	Rearrangements per cell	Aberrations >10	Aberrations per cell (without gaps)
Negative control	0.004	0.008	0.108	0	0.116
M310I018					
220 µL/kg bw	0	0	0.172	0	0.172
22 µL/kg bw	0	0	0.185	0	0.185
2.2 µL/kg bw	0.004	0	0.136	0	0.136
Positive Control TEM	0.004	0.020	0.484	134	5.864

Table 5.8.1-12: Cytogenetic analysis of bone marrow cells (50 cells counted per animal) from dosed rats

Experimental group	No. of rat	Mitotic index	Chromatid		Chromosome		Rearrangements					multiple aberration (>10 per cell, excluding gaps)
			Gaps	Breaks	Gaps	Breaks	EX	DIC	RG	FRAG	PULV	0
Negative control	**1**	13	0	0	0	0	6	0	3	0	0	0
	2	14	1	0	0	1	2	0	1	1	0	0
	3	14	0	0	0	0	5	0	2	0	0	0
	4	12	0	0	0	0	3	0	1	0	0	0
	5	15	0	0	0	1	1	0	2	0	0	0
M310I018	**6**	14	0	0	0	0	6	0	2	0	0	0
220 µL/kg bw	**7**	11	0	0	0	0	8	0	1	0	0	0
	8	13	0	0	0	0	7	0	1	0	0	0
	9	13	0	0	0	0	7	3	3	0	0	0
	10	14	0	0	0	0	3	3	2	0	0	0
M310I018	**13**	13	0	0	0	0	6	0	3	0	0	0
22 µL/kg bw	**14**	14	0	0	0	0	6	0	2	0	0	0
	15	-	-	-	-	-	-	-	-	-	-	-
	16	14	0	0	0	0	8	0	1	0	0	0
	17	12	0	0	0	0	7	0	0	0	0	0
M310I018	**18**	11	0	0	0	0	6	0	2	0	0	0
2.2 µL/kg bw	**19**	12	0	0	0	0	2	0	2	0	0	0
	20	13	0	0	0	0	6	0	2	0	0	0
	21	15	0	0	0	0	7	0	1	0	0	0
	22	13	0	0	0	0	4	0	2	0	0	0
Positive Control	**51**	7	0	0	0	0	13	1	6	0	0	27
TEM	**52**	8	0	0	0	0	25	0	3	0	0	33
	53	8	0	3	3	1	3	2	2	0	0	24
	54	6	0	0	0	1	3	0	9	0	0	26
	55	6	0	0	0	0	40	0	15	0	0	24

Discussion
The negative (corn oil) controls were within the normal range of values for this species and strain as observed in this laboratory. The positive control (TEM) exhibited its usual severe genetic damage. The chromosome number varied due to severe damage, 74% of the cells were characterized by chromosomal aberrations, and an average of 5.86 aberrations per cell was recorded. In this study, the test compound M310I018 appeared to exhibit little or no clastogenic activity. No dose response was observed.

III. CONCLUSION

Under the conditions employed in the assay described in this report, the data suggest that the test compound exhibits little or no mutagenic activity in the in vivo cytogenetics assay.

Report: CA 5.8.1/33
Putman D.L., 1991a
m-PBAD - Activity of T1410 in the dominant lethal assay in rodents for mutagenicity (final report with cover letter)
1979/1001669
Guidelines: none
GLP: no

Executive Summary

A dominant lethal assay was performed with male and female SD rats. Three groups of male rats (10 per group) were given 5 intragastic treatments with the test substance at levels of 216 mg/kg bw (LD5), 21.6 mg/kg bw (LD0.5), and 2.16 mg/kg bw (LD0.05). Corn oil served as negative control and triethylene melamine was used as positive control. After the treatment period each male was mated with 2 females for 5 days, rested for 2 days, and was again mated with 2 females. This procedure was repeated until each male had been mated for 7 weeks with 2 new females. Fourteen days from the mid-week of mating, the females were sacrificed, and the females were examined for corpora lutea, fetal deaths and total implantations, respectively. The negative controls were within the acceptable range of values for the species and strain when compared to the laboratory historical control data. The positive control exhibited its usual severe genetic damage between weeks 2 and 7 as shown by greater preimplantation losses, increased number of dead implants per female, and an increase in females with one or more dead implants. Treatment with M310I018 exhibited minimal activity in the number of total implantations, preimplantation losses, and number of live implants per pregnant female. However, the effect on total or live implants per pregnant female occurred at a single dose level at week 7 only. The variation in corpora lutea at week 5 does not reflect any biological activity of the doses tested but rather is a variation in the female animals. The increase seen in pre-implantation losses at week 6 is probably a result of the unusually high corpora lutea counts for this time period rather than actual biological activity of the doses tested. No statistically significant dose response was observed. No effects were observed regarding fertility index, number of dead implants per pregnant female or per total implants, respectively. Under the conditions employed in the assay the data suggest that the M310I018 exhibits little or no mutagenic activity in the dominant lethal test.

Report: CA 5.8.1/34
[redacted] 1991a
m-PBAD - Ninety-day subchronic oral toxicity study of meta-phenoxybenzaldehyde in albino rats (final report with cover letter)
1980/1001707
Guidelines: none
GLP: no

Report: CA 5.8.1/35
[redacted] 1991a
m-PBAD - Histopathologic evaluation of tissuues and organs from albino rats in a 90-day subchronic oral dosing study with meta-phenoxybenzaldehyde (final report)
1980/1001708
Guidelines: none
GLP: no

Executive Summary

The oral administration of M310I018 to male and female Sprague-Dawley rats over a period of 90 days did not reveal test substance related adverse signs of systemic toxicity at dose levels of 50, 150, or 300 mg/kg bw/d. Increased liver and kidney weights were observed, but were considered to be related to increased metabolic activation rather than to substance specific effects.
The mean weekly body weights by sex for all groups were virtually identical throughout the study period. However, beginning at week 3 group 3 and 4 weekly food consumption means were significantly greater when compared to the control. Clinical pathology results indicated elevation of AST, ALT, and ALP in groups 3 and 4. Furthermore, there was a dose related increase in liver weight and liver/body weight and liver/brain weight ratios for group 4. Similarly, the kidneys were affected in groups 3 and 4. These findings indicate an increase in metabolic activity of these organs with increased production of serum enzymes. Histopathology did not reveal an increase in non-neoplastic lesions in albino rats under the conditions of the study.
The no observed effect level (NOEL) under the conditions of the present study was 50 mg/kg bw/day in male and female Sprague-Dawley rats.

(DocID 1980/1001707, 1980/1001708)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Meta-phenoxybenzaldehyde (Metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	liquid / yellow
Lot/Batch #:	82-92 and 189
Purity:	97%
Stability of test compound:	The test substance was stable over the study period

2. Vehicle and/or positive control: Mazola® corn oil

3. Test animals:

Species:	Rat
Strain:	Sprague-Dawley
Sex:	Male and female
Age:	no data
Weight at dosing:	160-260 g
Source:	Gibco Animal Resources, Madison, Wisconsin, USA
Acclimation period:	3 weeks
Diet:	Purina Certified Rodent Chow 5002®, ad libitum
Water:	Drinking water, ad libitum
Housing:	Single housing in stainless-steel wire mesh cages

Environmental conditions:

Temperature:	no data
Humidity:	no data
Air changes:	no data
Photo period:	no data

B. STUDY DESIGN

1. Dates of experimental work: 03-Jan-1980 - 04-Apr-1980

2. Animal assignment and treatment:
M-PBAD was administered to groups of 20 male and 20 female Sprague-Dawley rats at concentrations of 0, 50, 150, and 300 mg/kg bw/day via gavage for 90 days. The animals were randomized and assigned to the treatment groups. At the end of the administration period the animals were sacrificed after a fasting period of at least 16 hours. Ten rats per sex were used in a range-finding study to establish baseline clinical pathology values.

3. Test substance preparation and analysis:
Freshly mixed solutions were used for administration and were analyzed before administration to test animals. Assays of stock solutions were within 10% of the specified concentration. The dose volumes were 5, 5, 1.5, and 3 mL/kg for the rats of group 1 (control), 2 (50 mg/kg bw), 3 (150 mg/kg bw), and 4 (300 mg/kg bw), respectively.

4. Statistics:
Means and standard deviations of each test group were calculated for several parameters. Further statistical analyses were performed according to following table:

Statistics applied

Parameter	Statistical test
Body weight, organ weights, feed consumption, hematology, clinical chemistry	Analysis of variance, ANOVA, and Tuke´s HSD procedure

C. METHODS

1. Observations:
The animals were examined for general appearance, elimination, behavior and mortality once daily.

2. Body weight:
The body weight of the animals was determined before the start of the administration period (in order to randomize the animals), at the start of the treatment (day 0) and at weekly intervals thereafter.

3. Food consumption:
Food consumption was determined weekly and calculated as mean food consumption for each group.

4. Hematology and clinical chemistry:
Predose blood samples were taken from the 10 male and female animals not assigned to treatment groups. At day 45 and 90 blood samples were collected from 10 males and 10 females randomly selected from each group. Blood was collected from the abdominal aorta at predose and at day 90. At day 45 blood was collected via puncture of the retro-orbital sinus.
The following hematological and clinical chemistry parameters were determined:

Hematology:					
	Red blood cells		White blood cells		Clotting Potential
✓	Erythrocyte count (RBC)	✓	Total leukocyte count (WBC)	✓	Platelet count
✓	Hemoglobin (Hb)	✓	Differential blood count		
✓	Hematocrit (Hct)				
Clinical chemistry:					
	Electrolytes		Metabolites and Proteins		Enzymes:
✓	Calcium	✓	Albumin	✓	Alanine aminotransferase (ALT)
✓	Potassium	✓	Bilirubin (total and direct)	✓	Aspartate aminotransferase (AST)
		✓	Cholesterol	✓	Alkaline phosphatase (ALP)
		✓	Globulin (by calculation)	✓	Lactic dehydrogenase
		✓	Glucose (fasted)		
		✓	Protein (total)		
		✓	Urea		

5. Urinalysis:

Predose urine samples were taken from the 10 male and female animals not assigned to treatment groups. At day 45 and 90 urine samples were collected from 10 males and 10 females randomly selected from each group.

Urinalysis					
	Quantitative parameters:		Semiquantitative parameters		
✓	Specific gravity	✓	Bilirubin	✓	Protein
		✓	Glucose	✓	pH-value
		✓	Ketones	✓	Urobilinogen
				✓	Sediment (microscopical exam.)

6. Sacrifice and pathology:
Clinical pathology investigations were performed 1-3 days before dosing (predose), day 45 of dosing, and day 90 of dosing. After 90 days of dosing all surviving rats were fasted for at least 8 h, anesthetized, killed by exsanguination, and necropsied. The gross necropsy included examination of the carcass, the external surface, all orifices, the cranial cavity, the thoracic, abdominal, and pelvic cavities with their associated organs and tissues, and the neck with its associated organs and tissues. Organ weights for the liver, kidneys, adrenals, gonads, heart, spleen, and brain were recorded for all study animals. Organ/body and organ/brain weight ratios were calculated. Tissues from rats in group 1 and 4 were taken for histopathologic examination and from group 2 and 3 rats for fixation. A histopathologic examination of 44 tissues per animal was conducted in the control and high dose group. Among them were tissues of the reproductive system as epididymides, seminal vesicles, prostate, testes, fallopian tubes, cervix, vagina, mammary gland, ovaries, and uterus. In addition, tissues of the nervous system were examined including pons, optic nerve, peripheral nerve, and spinal cord. Furthermore, liver, kidney, and heart were evaluated from all animals of 50 and 150 mg/kg bw dose groups. The organs were sampled, weighed, and examined histopathologically as indicated in the table below.

Pathology:
The following organs were collected (column C), weighed (W) and examined histopathologically (H, ✓: all groups, #: control and top dose).

C	W	H		C	W	H		C	W	H	
✓	✓	#	adrenals	✓	✓	✓	liver	✓		#	thyroid glands
✓		#	aorta	✓		#	lung	✓		#	trachea
✓		#	bone marrow§	✓		#	lymph nodes#	✓		#	urinary bladder
✓	✓	#	brain	✓		#	mammary gland (♀)	✓		#	uterus with cervix
✓		#	caecum	✓		#	muscle, skeletal		✓		body (anesthetized animals)
✓		#	colon	✓		#	nerve, peripheral (sciatic n.)				
✓		#	duodenum	✓		#	pancreas	✓		#	epididymides
✓		#	esophagus	✓		#	parathyroid glands	✓		#	seminal vesicles
✓		#	eyes (with optic nerve)	✓		#	pituitary	✓		#	testes
✓		#	gonad	✓	✓	#	prostate	✓		#	fallopian tubes
✓		#	gross lesions	✓		#	salivary glands (submaxillary)	✓		#	cervix
✓		#	Harderian gland	✓		✓	skin	✓		#	vagina
✓	✓	✓	heart	✓		✓	spinal cord (3 levels)	✓		#	ovaries
✓		#	ileum	✓	✓	✓	spleen	✓		#	optic nerve
✓		#	jejunum (w. Payer's plaque)	✓		✓	sternum w. marrow				
✓	✓	✓	kidneys	✓		✓	stomach (cardia, fundus and pylorus)				

§ from femur

The organs or tissues were fixed in 10% formalin solution (eyes and testes in Bonin´s solution). The hematoxylin-eosin (HE) stained slides were examined and assessed by light microscopy.

II. RESULTS AND DISCUSSION

A. OBSERVATIONS

1. Clinical signs of toxicity

22 animals (14 females, 8 males) exhibited small white corneal opacities in one or both of their eyes. These lesions were present at the initiation of the study, were virtually unchanged throughout the study, and were not treatment related. Other observations, such as ocular discharge, sneezing, unthrifty appearance, alopecia, broken skin, etc. appeared for a short duration in all groups early in the study, but tended to be more common in the high treatment groups.

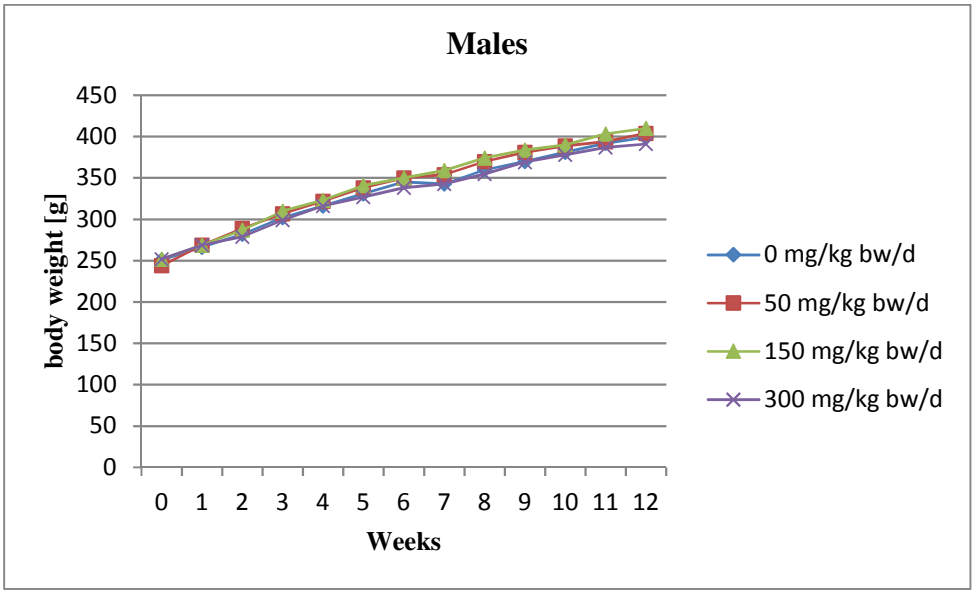
2. Mortality

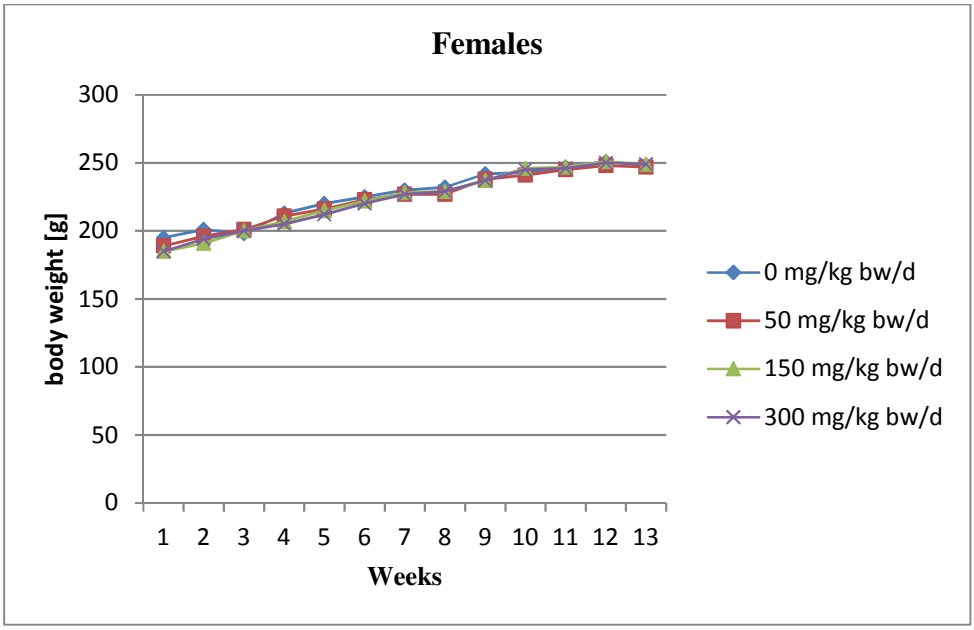
Nineteen animals died before study termination. These deaths were considered to be not related to substance administration, rather attributed to injuries from gavaging or accidental injuries.

B. BODY WEIGHT AND BODY WEIGHT GAIN

The mean body weight of the control group was not significantly different when compared to the treatment groups. In several instances, a statistically significant difference in means for weekly weight gain was seen between control and treatment groups [see Figure 5.8.1-4].

Figure 5.8.1-4: Body weight development of rats administered M310I018 for 90 days





C. FOOD CONSUMPTION

Both mean and individual feed consumption data for group 3 and 4 (150 and 300 mg/kg bw) of both sexes showed a trend of increasing food consumption over time. Statistically significant differences were seen in comparing group means for food consumption for group 3 and 4 with that of the control (see Table 5.8.1-13).

Table 5.8.1-13: Cumulative food consumption of rats administered M310I018 for 90 days

	Males				Females			
Dose level [mg/kg bw/day]	0	50	150	300	0	50	150	300
Cumulative food consumption [g/animal]								
Day 0 to 90[#]	1373±206	1439±121	**1715±128**	**1733±148**	1114±115	1141±96	**1300±130**	**1410±179**
Δ% (compared to control)[#]		+4.8	**+24.9***	**+26.2***		+2.4	**+16.7***	**+26.6***

[#] Values were calculated based on mean individual daily food. Values may not calculate exactly due to rounding of mean values

* Significantly different from control ($p<0.05$)

D. BLOOD ANALYSIS

1. Hematological findings

Group mean data for hematology parameters showed no apparent abnormalities or trends. Incidental variations are listed in Table 5.8.1-14.

Table 5.8.1-14: Selected hematology findings in rats administered M310I018 for 90 days (group means)

	Study day	RBC [10^6]	HGB [mmol/L]	HCT [g/dL]
	Males			
Control	Day 90	9.25 ± 0.17	17.42 ± 0.63	50.70 ± 1.89
50 mg/kg bw/day	Day 90	**8.90 ± 0.53***	16.79 ± 0.65	49.80 ± 2.15
150 mg/kg bw/day	Day 90	9.22 ± 0.24	16.97 ± 0.60	50.20 ± 2.15
300 mg/kg bw/day	Day 90	9.26 ± 0.15	17.08 ± 0.61	**49.33 ± 1.80***
	Females			
Control	Day 90	8.51 ± 0.28	16.58 ± 0.47	48.70 ± 1.57
50 mg/kg bw/day	Day 90	**8.09 ± 0.25***	**15.92 ± 0.47***	**46.10 ± 3.85***
150 mg/kg bw/day	Day 90	8.22 ± 0.39	16.16 ± 0.71	47.00 ± 2.00
300 mg/kg bw/day	Day 90	8.75 ± 0.35	16.85 ± 0.34	48.00 ± 0.82

* Significantly different from control ($p<0.05$)

2. Clinical chemistry findings

At day 45 mean globulin levels were significantly decreased in group 3 and 4 males when compared with the control. At the same time mean albumin/globulin ratios were significantly elevated in group 4 males (see Table 5.8.1-15). At day 45 and day 90 the mean for the level of alkaline phosphatase was significantly increased in group 4 males. At day 90 group 3 and 4 females showed a statistically significant difference in mean values for alkaline phosphatase. Mean AST values for males increased with dosage level at day 90, which was statistically significant for group 3 and 4 males. Likewise, ALT levels were increased with dosage level, being significant for group 4 males (see Table 5.8.1-16).

Table 5.8.1-15: Selected clinical chemistry findings in rats administered M310I018 for 45 days (group means)

	Males				Females			
Dose [mg/kg bw/d]	0	50	150	300	0	50	150	300
Albumin [g/dL]	4.24 ± 0.20	4.19 ± 0.27	4.27 ± 0.26	4.13 ± 0.17	4.17 ± 0.17	4.37 ± 0.24	4.37 ± 0.17	4.47 ± 0.15
Globulin [g/dL]	2.36 ± 0.18	2.15 ± 0.16	**2.13 ± 0.17***	**1.87 ± 0.24***	2.14 ± 0.27	2.12 ± 0.32	2.11 ± 0.14	2.01 ± 0.23
A/G ratio	1.81 ± 0.20	1.95 ± 0.13	2.02 ± 0.20	**2.25 ± 0.34***	1.96 ± 0.20	2.10 ± 0.3	2.08 ± 0.18	2.25 ± 0.28

* Significantly different from control ($p<0.05$)

Table 5.8.1-16: Selected clinical chemistry findings in rats administered M310I018 for 90 days (group means)

	Males				Females			
Dose [mg/kg bw/d]	0	50	150	300	0	50	150	300
ALT [IU/L]	44.4 ± 12.9	52.8 ± 15.1	**64.5 ± 10.2***	**66.3 ± 11.1***	63.5 ± 17.6	75.5 ± 42.4	70.9 ± 24.1	52.9 ± 14.2
AST [IU/L]	17.4 ± 3.6	18.6 ± 2.9	21.2 ± 5.0	**23.2 ± 4.9***	16.9 ± 3.6	17.3 ± 2.8	20.1 ± 2.8	20.9 ± 4.6
ALP [IU/L]	69.5 ± 15.3	87.7 ± 32.7	88.9 ± 24.4	**135.5 ± 52.6***	51.3 ± 13.6	70.3 ± 21.2	81.7 ± 27.9	**86.9 ± 25.8***

* Significantly different from control ($p<0.05$)

3. Urinalysis
Urinalysis revealed no trends or abnormalities.

E. NECROPSY

1. Organ weight
Absolute liver weights, liver/body and liver/brain weight ratios increased with dosage level for both male and female rats. Mean values for absolute liver weight, liver/body and liver/brain weight ratios were significantly different for group 4 males and females compared to the control. Absolute kidney weights, kidney/body and kidney/brain weight ratios also increased with dosage level for both male and female rats. Group 3 and 4 females showed significantly elevated means for absolute kidney weights. Statistically significant differences in mean values for kidney/body weight ratios occurred between group 4 and control males. Group 3 and 4 males and females had significantly elevated mean values for kidney/brain weight ratios compared to control males and females. No further trends were observed.

Table 5.8.1-17: Mean absolute organ weights in rats administered M310I018 for 90 days (group means)

	Terminal bw [g]	Liver [g]	Kidney [g]
	Males		
Control	366 ± 48	12.51 ± 2.73	2.58 ± 0.50
50 mg/kg bw/day	386 ± 46	12.67 ± 1.74	2.70 ± 0.23
150 mg/kg bw/day	385 ± 59	13.58 ± 2.76	2.79 ± 0.30
300 mg/kg bw/day	340 ± 58	**14.92 ± 2.48***	2.79 ± 0.26
	Females		
Control	213 ± 38	6.70 ± 1.22	1.49 ± 0.30
50 mg/kg bw/day	221 ± 38	7.38 ± 1.21	1.62 ± 0.14
150 mg/kg bw/day	225 ± 39	**8.61 ± 1.31***	**1.72 ± 0.18***
300 mg/kg bw/day	208 ± 41	**9.16 ± 1.80***	**1.71 ± 0.16***

* Significantly different from control ($p<0.05$)

Table 5.8.1-18: Mean relative organ weights (organ/body weight) in rats administered M310I018 for 90 days (group means)

	Liver [g]	Kidney [g]
	Males	
Control	3.47 ± 0.75	0.72 ± 0.12
50 mg/kg bw/day	3.32 ± 0.55	0.71 ± 0.10
150 mg/kg bw/day	3.55 ± 0.69	0.73 ± 0.09
300 mg/kg bw/day	**4.52 ± 1.13***	**0.84 ± 0.15***
	Females	
Control	3.20 ± 0.61	0.72 ± 0.20
50 mg/kg bw/day	3.47 ± 0.96	0.76 ± 0.19
150 mg/kg bw/day	4.02 ± 1.18	0.80 ± 0.18
300 mg/kg bw/day	**4.50 ± 1.03***	0.85 ± 0.18

* Significantly different from control ($p<0.05$)

Table 5.8.1-19: Mean relative organ weights (organ/brain weight) in rats administered m-PBAD for 90 days (group means)

	Liver [g]	Kidney [g]
	Males	
Control	629 ± 70	128 ± 11
50 mg/kg bw/day	657 ± 106	139 ± 16
150 mg/kg bw/day	726 ± 149	149 ± 16
300 mg/kg bw/day	**806 ± 150***	**150 ± 17***
	Females	
Control	385 ± 68	85 ± 17
50 mg/kg bw/day	426 ± 66	93 ± 6
150 mg/kg bw/day	487 ± 72	97 ± 10
300 mg/kg bw/day	**538 ± 114***	**99 ± 11***

* Significantly different from control ($p<0.05$)

2. Gross and histopathology

Gross lesions:
All macroscopic findings occurred either individually and were considered to be incidental or spontaneous in origin and without any relation to treatment.

Histopathology
The non-neoplastic histopathologic observations were generally those of spontaneous lesions occurring in albino rats, and included inflammatory lesions of the esophagus and periesophageal tissues, pericarditis, and inflammation of the myocardium. The large intestines contained evidence of parasits infection (nematodiasis). The liver contained evidence of parasitic migration (inflammation, focal, granulomatous). The eyes from several rats in each of the four treatment groups were noted during the study period to have corneal opacities, at histopathologic examinations these were determined to be vacuolar keratitis. The kidneys in all treatment groups contained small numbers of proteinaceous casts located in the proximal and distal convoluted tubules. The casts were observed with approximately equal frequency in all treatment groups. The urinary bladders of the male animals contained proteinaceous plugs composed of aggregates of sperm and prostate gland secretions.

Treatment with M310I018 did not induce a dose-related increase in neoplastic or non-neoplastic lesions in the examined tissues. No differences were observed between control groups and high dose treated animals regarding lesions in the reproductive or nervous system, respectively.

III. CONCLUSIONS

The oral administration of M310I018 to male and female Sprague-Dawley rats over a period of 90 days did not reveal test substance related adverse signs of systemic toxicity at dose levels of 50, 150, or 300 mg/kg bw/d. Increased liver and kidney weights were observed at mid and high dose level, but were considered to be related to increased metabolic activation rather than to adverse effects.
Therefore, under the conditions of the present study the no observed effect level (NOEL) was 50 mg/kg bw/day in male and female animals.
Supplemental information from open literature about toxicokinetics of pyrethroid metabolits in males and female rats was evaluated in the DAR or lambda-cyhalothrin [see Ueyama et al., 2010; discussed in Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 125-129] indicating that females eliminate M310I018 more rapid than male rats.

Information with regard to the endocrine potential of M310I018 was gathered from the open literature. A summary table of the available literature is provided in Table 5.8.1-20 below. The literature is however described in more detail in the chapter CA 5.8.3.

Only in vitro studies are available for M310I018 with regard to endocrine related mode of action. According to Jin et al., 2010 [see CA 5.8.3/17 2010/1232199] rather no, at most weak estrogenic effects on mRNA expression level for ER and pS2 were seen in MCF-7 cells. Furthermore no proliferation of MCF-7 cells was induced. PBAld showed estrogenic activity in the low micromolar range in a transactivation assay in yeast cells (McCarthy et al., 2006; [see CA 5.8.3/16 2006/1051135]), but no activity in mammalian MCF-7 cells (Tange et al., 2014; [see CA 5.8.3/9 2014/1242695]). In addition, anti-androgen activity in CHO cells was found to be rather weak, with an IC_{20} in a concentration range of 0.17 µM but an IC_{50} at 27.40 µM which is not considered anymore relevant for human exposure (Tange et al., 2014; [see CA 5.8.3/9 2014/1242695]).

Based on the lack of pronounced estrogenic or anti-androgenic activity in vitro in mammalian cells no further in vivo studies were considered necessary.

Table 5.8.1-20: Literature data for endocrine activity of M310I018

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference DocID
In vitro assays				
ER-dependent regulation of mRNA expression	Estrogen Receptor (ER) mRNA levels in MCF-7	Estrogenic. not active RIE: 7.3% (at 1µM)	E2: RIE set to 100% at 1 nM	Jin et al., 2010 2010/1232199
ER-dependent regulation of mRNA expression	pS2 mRNA levels in MCF-7	Estrogenic weak active RIE: 12.5% (at 1µM)	E2: 4-fold ↑ (1 nM)	Jin et al., 2010 2010/1232199
ER-activation	hER-α transactivation in Yeast (lacZ)	ER-agonist: Active EC_{50}: 4.8±3.4E-6M	Agonist: E2: EC_{50}: 0.4±0.2E-9M	McCarthy et al., 2006 2006/1051135
ER-activation	hER transactivation in MCF-7 cells (Luciferase)	ER-agonist: Not active up to 100 µM ER-Antagonist: Not tested	Agonist: E2: EC_{20}: 2.4E-8M	Tange et al., 2014 2014/1242695
ER-dependent proliferation	E-Screen (MCF-7)	Proliferative effect: not acive	E2: 1 nM PE: 2.12 RPE: 100%	Jin et al., 2010 2010/1232199
AR-activation	hAR transactivation in CHO cells (Luciferase)	AR-agonist: not tested AR-antagonist: active IC_{20}: 0.17E-6 M IC_{50}: 27.4E-6M	Androgen activity: DHT: 1E-10M Antagonist: Flutamide: IC_{20}: 0.16E-6 M IC_{50}: 0.62E-6M	Tange et al., 2014 2014/1242695

E2: 17 beta-Estradiol; EE: ethinyl estradiol; 4HO-TAM: 4-Hydroxy-tamoxifen; DHT: Dihydrotestostereon; PE: Proliferative effect; RPE: relative proliferative effect calculated as the ratio (PE-1) of the test chemical over (PE-1) of E2 (x 100); RIE: The relative inductive efficiency (RIE) is the ratio between the maximal up-regulation of pS2 expression level by the test compound to that of E2 (x100) or the relative inhibitory effciency of down-regulation of ERalpha expression level by the test compound compared to that of E2 (x 100); EC50/IC20/IC50: Effect concentratio /Inhibitor concentration at which 50% increase /20%/50%decrease is found compared to control;

Toxicological evaluation of M310I018

Acute toxicity studies indicate that M310I018 is toxic when inhaled, harmful when swallowed and is a skin sensitizer. M310I018 is practically not toxic by the dermal route and is not irritating to skin and eye but may be a skin sensitizer.

The QSAR evaluation of M310I018 is of low to moderate reliability. The one identified structural alersts was not supported by the other models applied.

For M310I018 results several in vitro and in vivo genotoxicity studies were available. Based on the whole datapackage and in particular considering the most relevant studies discussed in detail in section b above there is no evidence for genotoxicity of M310I018.

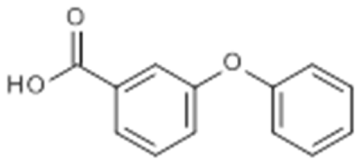
M310I018 was investigated for its estrogenic and anti-androgenic potential and is considered to show no relevant endocrine potential in vitro.

Results from a 90-day study in rats showed increased liver and kindey weights considered to be related to increased metabolic activation rather than to an adverse effect. The NOEL of the study was 50 mg/kg bw/day

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day

M310I018 is considered to be not genotoxic. The available toxicological database does not provide any concern for M310I018. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the relevant TTC-threshold. Therefore, M310I018 is considered to be **not toxicologically relevant**.

M310I011 (other denominators: PBA, PBAcid, m-PB acid, 3-PB acid, Reg.No. 130213, CAS-No. 3739-38-6, WL 44607, CL206128)



M310I011 is a metabolite found in rats, livestock (goat, hen) and in crops (wheat).

a. QSAR Predicitions on M310I011

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 11 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for M310I011 or in-silico generated metabolites and no structural alerts were received. In all cases the structures were out of domain. None of 8 in-silico metabolites was predicted positive, including M310I010 the glycine conjugate of M310I011 and a potential structure for M310I013 the hydroxylated derivative of M310I011.
For in-vitro chromosome aberration there were **no** genotoxicity alerts for M310I010 or in-silico generated metabolites and no structural alerts were received. None of 8 in-silico metabolites was predicted positive, including M310I010 the glycine conjugate of M310I011 and a potential structure for M310I013 being equal to M310I025 the para-hydroxylated derivative of M310I011.

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 11 of report, [see KCA 5.8.1/5 2014/1289317]]
M310I011 was in the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The prediction is based on 6 molecules, of which three were predicted 'mutagen', which was true for two of them regarding the experimental value. Three molecules were predicted and actual 'non-mutagen'. The similarity of the molecules in the data set to M310I011 is quite good as indicated by similarity factors of 0.850 to 0.892. Also the concordance of the total underlying database was high (1.0) and thus is very robust.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 11 of report, [see KCA 5.8.1/5 2014/1289317]]
M310I011 was in the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The similarities and the concordance scores correspond to the values of the CAESAR model (see above).

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 11 of report, [see KCA 5.8.1/5 2014/1289317]]
M310I011 was in the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The prediction is based on 6 molecules, of which one was predicted and actual 'mutagen'. The similarity to PBAcid (Reg.No.) of the molecules in the data set is moderate as indicated by similarity factors of 0.798 to 0.846. The concordance of the total underlying database was high (1.0) and thus is very robust.

Conclusion on QSAR evaluations
No conclusive structural alert was identified for M310I011.

Toxicity studies on M310I011

Report:	CA 5.8.1/36 Matsumoto K., 2000a CL206, 128: Reverse mutation test AL-435-010
Guidelines:	EPA 870.5100, JMAFF 59 NohSan No 4200
GLP:	yes (certified by Ministry of Agriculture, Forestry and Fisheries of Japan, Japan)

Executive Summary

S. typhimurium (strains TA98, TA 100, TA 1535 and TA 1537) and E. coli strain WP2 uvrA were exposed to M310I011; batch: AC11303-61, purity: 99%) using dimethylsulfoxide (DMSO) as a solvent in the presence and absence of metabolic activation in two independent experiments. Triplicate plates were used per dose and per test condition. Vehicle and positive controls were included in each experiment. In the first pre-incubation test the test substance was used at concentrations of 21, 62, 185, 556, 1667, and 5000 µg/plate. In the second pre-incubation test the test substance was used at concentrations of 156, 313, 625, 1250, 2500, and 5000 µg/plate. In both experiments a weak bacteriotoxic effect was observed depending on the strain and test conditions at concentrations from about 1250 µg/plate onward. Precipitation of the test substance did not occur up to the highest tested concentration.
A biologically relevant increase in the number of revertant colonies was not noticed in any of the strains tested in presence or absence of metabolic activation in any of the experiments. The positive controls induced the appropriate response in the corresponding strains, thus demonstrating the sensitivity of the test system.
According to the results of the study M310I011 was not mutagenic in the Salmonella typhimurium / Escherichia coli reverse mutation assay under the experimental conditions of the study.
(Doc ID AL-435-010)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material	CL206,128, 3-Phenoxybenzoic acid (Metabolite of BAS 310 I, Alpha-Cypermethrin)
Description:	Solid, white
Lot/Batch #:	AC11303-61
Purity:	99%
2. Vehicle:	Dimethylsulfoxide (DMSO)

3. Control Materials:

Vehicle control: The vehicle control with and without S-9 mix only contained the vehicle used for the test substance at the same concentration and volume for all tester strains.

Solvent/final concentration: 100 µL/plate

Positive control compounds tested without addition of metabolic activation system:

Strain	Mutagen	Solvent	Concentration
TA 100	2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide (AF-2)	DMSO	0.01 µg/plate
TA 1535	Sodium azide (NaN_3)	Water	0.5 µg/plate
TA 1537	9-Aminoacridine (9-AA)	Water	80 µg/plate
TA 98	AF-2	DMSO	0.1 µg/plate
WP2 uvrA	AF-2	DMSO	0.01 µg/plate

Positive control compounds tested with addition of metabolic activation system:

Strain	Mutagen	Solvent	Concentration
TA 100	2-aminoanthracene	DMSO	1.0 µg/plate
TA 1535	2-aminoanthracene	DMSO	2.0 µg/plate
TA 1537	2-aminoanthracene	DMSO	2.0 µg/plate
TA 98	2-aminoanthracene	DMSO	0.5 µg/plate
WP2 uvrA	2-aminoanthracene	DMSO	10.0 µg/plate

4. Activation:

S9 was produced from the livers of induced male Sprague-Dawley rats (pre-treatment with phenobarbital and β-naphthoflavone). The S9-mix was prepared freshly prior to each experiment. For this purpose, a sufficient amount of S9-fraction is thawed at room temperature and 1 volume of S9-fraction is mixed with 9 volumes of S9-supplement (cofactors). This preparation, the so-called S9-mix, was kept on ice until used.

The S9-mix was prepared immediately before use and had the following composition:

Component	Concentration
Phosphate buffer (pH 7.4)	100 mM
Glucose 6-phosphate	5 mM
NADPH	4 mM
NADH	4 mM
KCl	33 mM
$MgCl_2$	8 mM
S9	10 %

To demonstrate the efficacy of the rat liver S9 mix in this assay, the S9 batch was characterized with benzo(a)pyrene.

5. Test organisms: S. typhimurium strains: TA98, TA100, TA1535, TA1537
E. coli: WP2 uvrA

Salmonella typhimurium: The Salmonella strains are checked for the following characteristics at regular intervals: deep rough character (rfa); UV sensitivity (uvrB); ampicillin resistance (R factor plasmid). E. coli WP2 uvrA is checked for UV sensitivity.

Histidine and tryptophan auxotrophy is automatically proven in each experiment via the spontaneous rate.

6. Test concentrations:

Pre-incubation assay: In the first test triplicate plates were prepared for each concentration (neg. control; 21, 62, 185, 556, 1667, and 5000 µg/plate and positive controls) and condition (i.e. with and without S9) for all tester strains.
In the second experiment triplicate plates were prepared for each concentration (neg. control; 156, 313, 625, 1250, 2500, and 5000 µg/plate and positive controls) and condition (i.e. with and without S9) for all tester strains.

B. TEST PERFORMANCE:

1. Dates of experimental work: 04-Oct-1999 to 04-Nov-1999

2. Dose range finding test:
In order to assess bacteriotoxic effects, a dose range finding test was conducted on all strains according to the pre-incubation methods with and without metabolic activation system at 20, 78, 313, 1250, and 5000 µg/plate. A singe plate was applied for each dose level and duplicates were applied for solvent and positive control groups.

3. Pre-incubation assay:
100 µL of test solution or vehicle, 0.1 mL bacterial suspension and 0.5 mL S9 mix or phosphate buffer were incubated at 37°C for about 20 minutes. Subsequently, 2 mL of soft agar was added and, after mixing, the samples are poured onto the agar plates.
After incubation in the dark for 48 hours at 37°C each plate was checked for precipitates and status of the background lawn. Revertant colonies were counted. Means and standard deviations were calculated.

4. Statistics:
No special statistical tests were performed.

5. Evaluation criteria:
The test chemical is considered positive in this assay if the following criteria are met:

A dose-related and reproducible increase in the number of revertant colonies, i.e. about doubling of the spontaneous mutation rate in at least one tester strain either without S9 mix or after adding a metabolizing system.

A test substance is generally considered non-mutagenic in this test if:

The number of revertants for all tester strains was within the solvent control range under all experimental conditions in two experiments carried out independently of each other.

II. RESULTS AND DISCUSSION

A. TOXICITY

A bacteriotoxic effect was observed in all strains at 5000 µg/plate in the dose range finding test. In the main experiments toxicity at 5000 µg/plate was observed in the S. typhimurium strains of TA100 and TA1535 and in E. coli WP2 uvrA and at dose levels of 1667 µg/plate or more in TA98 and TA1537 in the absence of S9 mix. In the presence of S9 mix bacteriotoxicity was observed at 2500 µg/plate or more in Salmonella strains TA100, TA1535, and TA98 and in E. coli WP2 uvrA and at 1250 µg/plate or more in TA1537.

B. MUTATION ASSAYS

In the 2 independent preincubation experiments with and without metabolic activation no biologically relevant increase in number of revertants was observed in any strain tested [see Table 5.8.1-21]. The positive controls yielded revertant numbers in a range expected for the respective strains and thus demonstrated the sensitivity of the test system.

Precipitation was not observed up to the maximum concentration.

Table 5.8.1-21: Bacterial gene mutation assay with M310I011 - Mean number of revertants

Experiment 1: Plate incorporation assay										
Strain	TA 98		TA 100		TA 1535		TA 1537		E. coli	
Metabol. activation	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
Neg. control (DMSO)	30	29	95	133	7	8	10	9	19	21
M310I011										
21 µg/plate	28	26	94	126	6	6	8	9	18	16
62 µg/plate	23	30	88	118	5	8	8	14	19	15
185 µg/plate	23	24	98	113	6	9	7	13	20	16
556 µg/plate	24	24	85	109	4	8	9	12	18	17
1667 µg/plate	13	26‡	62	77	4	9	3‡	9‡	23	14
5000 µg/plate	‡	‡	‡	‡	‡	‡	‡	‡	‡	13‡
Pos. control[§]	374	670	778	567	462	573	88	492	440	105
Experiment 2: Plate incorporation assay										
Strain	TA 98		TA 100		TA 1535		TA 1537		E. coli	
Metabol. activation	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
Neg. control (DMSO)	22	20	124	126	7	7	12	8	23	19
M310I011										
156 µg/plate	21	20	118	114	11	9	12	10	21	20
313 µg/plate	21	17	120	115	3	6	8	8	23	18
625 µg/plate	22	21	106	104	6	7	9	5	20	20
1250 µg/plate	22	23	90	119	5	8	7‡	6	26	17
2500 µg/plate	15‡	22‡	61‡	88	6‡	6	‡	4‡	16‡	22
5000 µg/plate	‡	‡	‡	‡	‡	‡	‡	‡	‡	16‡
Pos. control[§]	368	747	720	595	186	560	138	758	244	118

[§] = Compound and concentrations see Material and Methods (I.A.2.) above;
‡ = toxicity was observed

III. CONCLUSION

According to the results of the present study, M310I011 is not mutagenic in the Salmonella typhimurium / Escherichia coli reverse mutation assay under the experimental conditions applied.

Report: CA 5.8.1/37
[redacted] 2014a
3-Phenoxybenzoic acid (metabolite of BAS 310 I, Alpha-Cypermethrin): Micronucleus test in Chinese Hamster V79 cells in vitro
2014/1168736

Guidelines: OECD 487 (2010), Commission Regulation EU No. 640/2012 of 06 July 2012 - B.49: In vitro Mammalian Cell Micronucleus Test

GLP: yes
(certified by Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden)

Remark Oct-2015: The pending stability analysis (Doc ID 2015/1029517) is included and highlighted**.**

Executive Summary

M310I011 (Batch: AC12251-34; Purity: 100%) was tested for chromosomal damage (clastogenicity) in Chinese Hamster V79 cells using the *in vitro* micronucleus test method. For this purpose, V79 cells were seeded in Quadriperm dishes and were incubated with the test substance at concentrations of 4.2 to 2142 µg/mL (≙10 mM) in the presence or absence of S9 mix. The cells were treated for 4 h in the presence or absence of S9 mix and for 24 h in the absence of S9 mix. After the incubation period cells were fixed and stained with Giemsa. At least 1000 cells per culture were scored for cytogenetic damage on coded slides and a proliferation index was determined for evaluation of cytotoxicity. No precipitation of the test item in the culture medium was observed, except for the highest applied concentration (2142 µg/mL) in Experiment IIB in the absence of S9 mix. No relevant influence on osmolarity or pH value was observed. In Experiment I in the absence and presence of S9 mix and in Experiment IIA in the presence of S9 mix, no cytotoxicity was observed up to the highest applied concentration. In Experiment IIA and IIB in the absence of S9 mix moderate cytotoxicity of approx. 40% of control was observed at the highest evaluated concentration. In the incubation with S9 mix and the long-term incubation without S9 mix significant increases of the micronucleus rate was observed. No increase of the micronucleus rate was observed in the short-term exposure in the absence of S9 mix. The positive control chemicals mitomycin C, griseofulvin and cyclophosphamide led to the expected increase in the micronucleus rate, thus demonstrating the sensitivity of the test system.
In conclusion, it can be stated that under the experimental conditions reported, the test item induced micronuclei as determined by the *in vitro* micronucleus test in Chinese hamster *V79* cells. Therefore, M310I011 is considered to be mutagenic in this *in vitro* micronucleus test, when tested up to the highest evaluable or the highest required concentration.

(DocID 2014/1168736)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: 3-phenoxybenzoic acid (Metabolite of BAS 310 I)
Description: solid (powder) / white
Lot/Batch #: AC12251-34
Purity/content: 100%
Stability of test compound: The test item was stable under storage conditions over the study period (expiry date 01 December 2020). The test item was stable in the vehicle for at least 4 hours (see separate report Doc ID 2015/1029517).

2. Vehicle used: DMSO (0.5%)

3. Control Materials:
Negative: No negative control was employed in this study.
Vehicle control: DMSO
Positive control (with S9): Cyclophosphamide (CPA), 15 µg/mL
Positive control (without S9): Mitomycin C, 0.1 µg/mL; Griseofulvin, 8 µg/mL

4. Activation: S9 was produced from the livers of rats (pre-treatment with phenobarbital and β-naphthoflavone). The S9-mix was prepared freshly prior to each experiment.

The S9-mix was prepared immediately before use and had the following composition:

Component	Concentration
Phosphate buffer (pH 7.4)	100 mM
Glucose 6-phosphate	5 mM
NADPH	4 mM
NADH	4 mM
KCl	33 mM
$MgCl_2$	8 mM
S9 (final protein concentration)	0.75 mg/mL

To demonstrate the efficacy of the rat liver S9 mix in this assay, the S9 batch was characterized with benzo(a)pyrene and 2-aminoanthracene.

5. Test system:

Cell line:	V79 (Chinese hamster cells) Each batch of cells is checked for mycoplasma contamination and karyotype stability.
Culture media:	MEM containing Hank´s salts, L-glutamine and Hepes (25 mM). Additionally, the medium was supplemented with 10% fetal calf serum (FCS) and 1% Pen/Strep (100 U/mL and 100 µg/mL). During the exposure period in the presence of S9 mix MEM medium without FCS was used.
Culture conditions:	Cells were grown with 1.5% CO_2 at 37°C and ≥ 90% relative humidity. Subculturing was performed twice weekly (doubling time 12-14 h). For the experiment, the cells were seeded into Quadriperm dishes containing microscopic slides. Into each chamber 1.0 x 10^5 – 1.5 x 10^5 cells were seeded.

6. Test compound concentrations:

Experiment I (4 h, ±S9 mix):	4.2, 8.4, 16.7, 33.5, 66.9, 133.9, 267.8, **535.5, 1071, 2142** µg/mL
Experiment IIa (24 h, -S9 mix):	4.2, 8.4, 16.7, 33.5, 66.9, **133.9, 267.8, 535.5**, 1071, 2142 µg/mL
Experiment IIb (24 h, -S9 mix):	33.5, 66.9, **133.9, 267.8, 401.7, 535.5, 669.4, 803.3**, 1071, 2142 µg/mL
Experiment IIa (4 h, +S9 mix):	**535.5, 803.3, 1071, 1606.5, 2142** µg/mL

Evaluated experimental points are shown in bold characters

B. TEST PERFORMANCE

1. Dates of experimental work: 15-Jul-2014 to 23-Oct-2014

2. Preliminary experiment:

A preliminary cell growth inhibition (determination of proliferation index) was performed to determine the concentrations to be used in the main experiment. The experimental conditions in this pre-experimental phase were identical to those required and described for the mutagenicity assay.

The pre-test was performed with 10 concentrations of the test item separated by no more than a factor of √10 and a solvent and positive control. All cell cultures were set up in duplicate. Exposure time was 4 hrs (with and without S9 mix). The preparation interval was 24 hrs after start of the exposure. Since the cultures fulfilled the requirements for cytogenetic evaluation, this preliminary test was designated experiment I.

3. Micronucleus test:

Pulse exposure:

The culture medium of exponentially growing cell cultures was replaced with serum-free medium containing the test item. For the treatment with metabolic activation 50 µL S9 mix per mL culture medium was added. After 4 hours the cultures were washed twice with "Saline G" (pH 7.2) containing 8000 mg/L NaCl, 400 mg/L KCl, 1100 mg/L glucose•H_2O, 192 mg/L Na_2HPO_4•2 H_2O and 150 mg/L KH_2PO_4. The cells were then cultured in complete medium containing 10 % (v/v) FBS for the remaining culture time of 20 hours.

Continuous exposure (-S9):

The culture medium of exponentially growing cell cultures was replaced with complete medium containing 10 % (v/v) FBS including the test item. The medium was not changed until preparation of the cells.

Preparation of micronuclei:

The cells were treated on the slides in the chambers with deionised water for 1 – 1.5 min at 37 °C. Afterwards the cells were fixed twice with a mixture of methanol and glacial acetic acid (3+1 parts, respectively) containing 1.25 % formaldehyde. The slides were stained with Giemsa, mounted after drying and covered with a cover slip. All slides were labelled with a computer-generated random code to prevent scorer bias.

Evaluation of cytotoxicity and cytogenetic damage:

The area of the micronucleus should not extend the third part of the area of the main nucleus. Per culture at least 1000 cells from clones with 2 - 8 cells were scored for cytogenetic damage on coded slides. The frequency of micronucleated cells was reported as % micronucleated cells.
Cytotoxicity was assessed via counting the number of clones consisting of 1 cell (c1), 2 cells (c2), 3 - 4 cells (c4), and 5 - 8 cells (c8) among the cells that were scored for the presence of micronuclei. These clusters represent the cells that have divided 1, 2, or 3 times within the experiment. From these data, a proliferation index (PI) is calculated (see formula below). Only those cultures were evaluated which showed a PI > 1.3, in order to guarantee a sufficient cell proliferation during treatment and recovery.

$$PI = \frac{(c1 \times 1) + (c2 \times 2) + (c4 \times 3) + (c8 \times 4)}{(c1 + c2 + c4 + c8)}$$

PI: Proliferation index
cx: Number of clones with x cells (with x: 1, 2, 4, or 8)

4. Statistics:
Many experiments with Chinese Hamster *V79* cells have established a range of micronucleus frequencies acceptable for control cultures. The current historical data range together with the statistical significance, confirmed by the Chi square test ($\alpha < 0.05$), should be considered for classification of the test item.

5. Evaluation criteria:
The micronucleus assay is considered acceptable if it meets the following criteria:

a) The rate of micronuclei in the solvent controls falls within the historical laboratory control data range.
b) The rate of micronuclei in the positive controls is statistically significant increased.
c) The quality of the slides must allow the evaluation of a sufficient number of analyzable cells.

A test item can be classified as non-mutagenic if:
- the number of micronucleated cells in all evaluated dose groups is in the range of the historical laboratory control data and
- no statistically significant or concentration-related increase of the number of micronucleated cells is observed in comparison to the respective solvent control.

A test item can be classified as mutagenic if:
- The number of micronucleated cells exceeds both the value of the concurrent negative control and the range of the historical negative control data.
- A significant, dose-related and reproducible increase in the number of cells containing micronuclei is observed

If the above mentioned criteria for the test item are not clearly met, the test item is classified as equivocal or a confirmatory experiment may be performed. However, results may remain questionable regardless of the number of times the experiment is repeated.

II. RESULTS AND DISCUSSION

No precipitation of the test item in the culture medium was observed, except for the highest applied concentration (2142.0 µg/mL) in Experiment IIB in the absence of S9 mix (see Table 5.8.1-22 and Table 5.8.1-23). No relevant influence on osmolarity or pH value was observed.
In Experiment I in the absence and presence of S9 mix and in Experiment IIA in the presence of S9 mix, no cytotoxicity was observed up to the highest applied concentration. In Experiment IIA and IIB in the absence of S9 mix moderate cytotoxicity of approx. 60 % of control was observed at the highest evaluated concentration.

In Experiment I (4 h exposure) in the absence of S9 mix no relevant increase in micronucleated cells was observed. In Experiment IIA (24 h exposure) in the absence of S9 mix one statistically significant increase in micronucleated cells (2.20 % micronucleated cells), clearly above the range of the laboratory historical control data (0.15 – 1.50 % micronucleated cells), was observed after treatment with 535.5 µg/mL. In Experiment IIB (24 h) in the absence of S9 mix statistically significant increases in micronucleated cells (4.05, 2.75 and 2.50 %) were observed after treatment with 401.7, 535.5 and 669.4 µg/mL but showed no dose response relationship.. The concentrations ranging from 267.8 to 803.3 µg/mL showed increases above the range of the laboratory historical control data (0.15 – 1.50 % micronucleated cells). Although without a strong evidence in relation to dose-response relationship they confirm the isolated positive finding of Experiment IIA.

In Experiment I in the presence of S9 mix one statistically significant increase in micronucleated cells (4.08 % micronucleated cells), clearly above the range of the laboratory historical control data (0.05 – 1.70 % micronucleated cells), was observed after treatment with 1071.0 µg/mL. However, no such effect was seen in the next higher concentration of 2014 µg/ml. In Experiment IIA in the presence of S9 mix all evaluated concentrations showed statistical significant increases due to the low response of the solvent control (0.15 %), but these values (0.60 – 1.40 % micronucleated cells) were clearly within the range of the laboratory historical solvent control data (0.05 – 1.70 % micronucleated cells) and no dose-dependency was observed. Thus the positive finding of Experiment I with metabolic activation could not be confirmed.

Either Griseofulvin (8.0 µg/mL), MMC (0.1 µg/mL) or CPA (15.0 µg/mL) were used as positive controls and showed distinct increases in cells with micronuclei.

Table 5.8.1-22: M310I011: Micronucleus test in V79 cells – Results of experiments without metabolic activation

Test group	Exp. No.	Concentration (µg/mL)	S9 mix	Proliferation index	Micronucleated cells (%)
4 h exposure – 24 h harvest					
DMSO	I	-	-	2.89	0.90
Mitomycin C	I	0.1	-	2.69	**5.05***
PBA	I	535.5	-	2.78	0.70
PBA	I	1071.0	-	2.69	1.43[1]
PBA	I	2142.0	-	2.83	1.15
24 h exposure – 24 h harvest					
DMSO	IIA	-	-	2.98	0.60
Griseofulvin	IIA	8	-	2.50	**9.85***
PBA	IIA	133.9	-	2.64	0.50
PBA	IIA	267.8	-	2.41	0.95
PBA	IIA	535.5	-	1.81	**2.20***
DMSO	IIB	-	-	2.56	1.60
Griseofulvin	IIB	8	-	2.66	**12.15***
PBA	IIB	133.9	-	2.81	1.10
PBA	IIB	267.8	-	2.27	2.00
PBA	IIB	401.7	-	1.85	**4.05***
PBA	IIB	535.5	-	1.75	**2.75***
PBA	IIB	669.4	-	1.52	**2.50***
PBA	IIB	803.3	-	1.45	1.55

[1] Deviating from the standard procedure (sample of 2000 cells) the number of micronucleated cells was determined in a sample of 4000 cells

* **Number of micronucleated cells statistically significantly higher than corresponding control values (Chi square test, p< 0.05)**

Table 5.8.1-23: M310I011: Micronucleus test in V79 cells – Results of experiment with metabolic activation

Test group	Exp. No.	Concentration (µg/mL)	S9 mix	Proliferation index	Micronucleated cells (%)
4 h exposure – 24 h harvest					
DMSO	I	-	+	2.45	1.45
CPA	I	15	+	1.86	**13.65***
PBA	I	535.5	+	2.42	1.55
PBA	I	1071.0	+	2.11	**4.08*[1]**
PBA	I	2142.0	+	2.38	1.40
24 h exposure – 24 h harvest					
DMSO	IIA	-	+	2.39	0.15
CPA	IIA	15	+	1.71	**12.05***
PBA	IIA	535.5	+	1.27	**0.75*‡**
PBA	IIA	803.3	+	2.27	**1.40*‡**
PBA	IIA	1071.0	+	2.25	**0.60*‡**
PBA	IIA	1606.5	+	2.46	**0.95*‡**
PBA	IIA	2142.0	+	2.52	**0.75*‡**

[1] Deviating from the standard procedure (sample of 2000 cells) the number of micronucleated cells was determined in a sample of 4000 cells

* **Number of micronucleated cells statistically significantly higher than corresponding control values (Chi square test, p< 0.05)**

‡ clearly within the range of the laboratory historical solvent control data (0.05 – 1.70 % micronucleated cells); statistical significance due to low concurrent solvent control

III. CONCLUSION

In conclusion, it can be stated that under the experimental conditions reported, the test item induced micronuclei as determined by the *in vitro* micronucleus test in Chinese hamster *V79* cells without metabolic activation.
Therefore, **M310I011** is considered to be **mutagenic** in this *in vitro* micronucleus test, when tested up to the highest evaluable or the highest required concentration.

Report: CA 5.8.1/38
[redacted]
3-phenoxybenzoic acid (metabolite of BAS 310 I, Alpha-cypermethrin) Micronucleus Assay in Bone marrow Cells of the Mouse
2015/1001101

Guidelines: OECD 474 (2014), Commission Regulation (EC) No 440/2008; B. 12
US EPA OPPTS 870.5395 (1998)

GLP: yes

~~**Remark:**~~ ~~**The final report will be supplied as soon as pending analytical data are available (End of Feb. 2015)**~~

Remark Oct-2015: The pending concentration control data (Doc ID 2015/1003984) and the Plasma Analysis (Doc ID 2015/1032402) are included and highlighted**.**

Executive Summary

M310I011 (Batch: AC12251-34; Purity: 100%) was tested for its ability to induce micronuclei in bone marrow cells of NMRI mice. For this purpose, the test substance, suspended in PEG 400, was administered once orally to groups of 7 male mice at dose levels of 500, 1000 and 2000 mg/kg body weight in a volume of 10 mL/kg body weight. The vehicle served as negative and cyclophosphamide as positive control. The animals were sacrificed 24 or 48 (additional high dose and vehicle group) hours after the administration and the bone marrow of the two femora was prepared. After staining of the preparations, 6000 polychromatic erythrocytes were evaluated per animal and investigated for micronuclei. To describe a cytotoxic effect the ratio between polychromatic and normochromatic erythrocytes was determined in the same sample and expressed in polychromatic erythrocytes per 500 erythrocytes.

The highest dose (2000 mg/kg b.w.; maximum guideline-recommended dose) was estimated by three pre-experiments to be suitable. No substantial differences between sexes in toxicity were observed. Bioavailability of the test substance in the mouse was proven in plasma samples of the treatment groups, which were dosed with 2000 mg/kg bw 2- and 4 hours after administration.

In the main study, conducted only in males, one male had to be euthanized after treatment with this dose due to severity of clinical symptoms. The test item induced systemic toxicity confirmed the systemic distribution of the compound. Thus, bioavailability of the test item under the tested conditions is given. After treatment with the test item the number of PCEs was not substantially decreased as compared to the mean value of PCEs of the vehicle control thus indicating that M310I011 did not exert any cytotoxic effects in the bone marrow. A statistically significant increase in micronuclei frequency was detected in both the low dose and the 24 h high dose group, respectively, when compared to the corresponding vehicle control group. However this was considered to be not biologically relevant, since all values obtained were very well within the range of historical vehicle control data. Moreover, no dose response relationship was observed. The positive control cyclophosphamide administered orally was used as positive control which showed a substantial increase of induced micronucleus frequency.

In conclusion, it can be stated that under the experimental conditions reported, M310I011 did not induce micronuclei as determined by the micronucleus test with bone marrow cells of the mouse.

(BASF DocID 2015/1001101)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: 3-phenoxybenzoic acid
(metabolite of BAS 310 I, Alpha-Cypermethrin)

Description:	Solid, white
Lot/Batch #:	AC12251-34
Purity:	100% (tolerance ± 1.0%)
Stability of test compound:	The stability of the test substance under storage conditions throughout the study period was guaranteed until 01 Dec 2020 as indicated by the sponsor. Stability in solvent was determined indirectly by concentration control analyses (see separate report Doc ID 2015/1003984).
Vehicle used:	PEG 400

2. Control Materials:

Negative:	No negative control was employed in this study.
Vehicle control:	Corn oil
Positive control:	Cyclophosphamide (CCP) 40 mg/kg bw (10 mL/kg bw), dissolved in sterile water

3. Test animals:

Species:	Mice
Strain:	NMRI
Sex:	Males for the main study; males and females for the range finding study
Age:	6 - 10 weeks
Mean body weight at dosing:	36.2 ± 2 g
Source:	Charles River Laboratories, Research Models and Services Germany GmbH, Sulzfeld, Germany
Number of animals per dose:	
Range finding study:	1000 mg/kg bw: 2 males/4 females
	2000 mg/kg bw: 2 males/2 females
Micronucleus assay:	7 males/dose in test group
	5 males/dose in control groups
Acclimation period:	at least 5 days
Diet:	Pelleted standard diet (certified), ad libitum
Water:	Tap water, ad libitum
Housing:	During the study the mice were housed individually in Makrolon cages (Type II) with wire mesh top

4. Environmental conditions:

Temperature:	20 - 24 °C
Humidity:	45 - 65%
Air changes:	frequency not indicated (fully air-conditioned rooms)
Photo period:	12-hour light-dark cycle (06:00 a.m. - 06:00 p.m., 06:00 p.m. - 06:00 a.m.)

5. Test compound doses:

Range finding test:	1000 and 2000 mg/kg bw
Micronucleus assay:	500, 1000 and 2000 mg/kg (at 24 h sampling) 2000 mg/kg bw (at 48 h sampling) The test substance was administered once by oral gavage using an application volume of 10 mL/kg.

B. TEST PERFORMANCE

1. Dates of experimental work: 19-Nov-2014 to 17-Dec-2014

2. Preliminary range finding test:

In a pretest for the determination of the acute oral toxicity, male and female animals were treated once by oral gavage with a test substance dose of 1000 or 2000 mg/kg bw.

3. Micronucleus test:

Treatment

Groups of male mice were treated once with the vehicle or 500, 1000 or 2000 mg test substance/kg bw by oral gavage for the 24-h sampling period. Additional test groups treated with the vehicle control and the high dose were used for the second sampling period (48-h). The application volume was 10 mL/kg bw and the volume to be applied was calculated based on actual weight on the day of administration. The positive control substance CCP was administered once by oral gavage (CCP). The animals of all dose groups, except the positive control, were examined for acute toxic symptoms at intervals of around 0-1 h, 2-4 h, 5-6 h, 24 h, and/or 48 h after administration of the test item. Sampling of the bone marrow was done 24 and 48 hours after treatment, respectively.

Preparation of the animals

The animals were sacrificed using CO_2 followed by bleeding. The femora were removed, the epiphyses were cut off and the marrow was flushed out with foetal calf serum using a syringe. The cell suspension was centrifuged at 1500 rpm (390 x g) for 10 minutes and the supernatant was discarded. A small drop of the re-suspended cell pellet was spread on a slide. The smear was air-dried and then stained with May-Grünwald/Giemsa. Cover slips were mounted with EUKITT. At least one slide was made from each bone marrow sample.

Analysis of cells

Evaluation of the slides was performed using NIKON microscopes with 100x oil immersion objectives. Per animal 6000 polychromatic erythrocytes (PCE) were analysed for micronuclei. To describe a cytotoxic effect the ratio between polychromatic and normochromatic erythrocytes was determined in the same sample and expressed in polychromatic erythrocytes per 500 erythrocytes. The analysis was performed with coded slides.

4. Statistics:

Statistical methods (nonparametric Mann-Whitney test) were used as an aid in evaluating the results, except for the high dose group at the 48-h sampling time point, for which statistical significance was evaluated by means of the one-way Analysis of Variance (ANOVA)

5. Acceptance and evaluation criteria:

Acceptance criteria

The study is considered valid as the following criteria are met:
- at least 5 animals per group could be evaluated.
- PCE to erythrocyte ratio was not less than 20 % of the negative control.
- the positive control shows a statistically significant and biological relevant increase of micronucleated PCEs compared to the vehicle control.

Evaluation criteria

A test item is classified as mutagenic if it induces either a dose-related increase or a clear increase in the number of micronucleated polychromatic erythrocytes in a single dose group above the laboratory's historical solvent control data range. Statistical methods (nonparametric Mann-Whitney test) were used as an aid in evaluating the results, if necessary. However, the primary point of consideration is the biological relevance of the results.
A test item that fails to produce a biological relevant increase in the number of micronucleated polychromatic erythrocytes is considered non-mutagenic in this system.

II. RESULTS AND DISCUSSION

A. ANALYTICAL DETERMINATIONS

The stability of the test substance in the vehicle is confirmed indirectly by dose formulation analytics (see separate report Doc ID 2015/1003984, BASF study code 04Y0418/01Y017). The homogeneity of the test substance in the vehicle was guaranteed by constant stirring and moderate heating (< 40°C) of the formulation directly before each dosing occasion and by analytical determination of 3 individual samples of each concentration. The mean concentration of these three samples were 42.1, 85.2 and 169.4 mg/g at nominal concentrations of 50; 100 and 200 mg/g, respectively. Thereby, the values are in the range of ± 15 % of the nominal concentration range and thereby acceptable for liquid test substance preparations.

Bioavailability of the test substance in the mouse was shown in plasma samples of the treatment groups which were dosed with 2000 mg/kg bw 2 and 4 hours after administration (see separate report Doc ID 2015/1032402, BASF study code 06Y0418/01Y018).

B. MICRONUCLEUS ASSAY (PRELIMINARY TEST)

In the pretest for the determination of the acute oral toxicity in males and females, the animals were treated with the test substance at doses of 1000 or 2000 mg/kg bw. Ruffled fur was observed in the majority of animals of the pretest. Partially closed eyes and abdominal posture were incidentally seen in a single animal each at one time point only. One female animal died at 1 h after dosing with 1000 mg/kg bw. No substantial differences between sexes in toxicity were observed. By the use of two additional females, the mortality observed in a female animal using 1000 mg/kg b.w. was confirmed to be not sex-specific, but most probably due to individual sensitivity, so that only male animals were used in the main experiment.

C. MICRONUCLEUS ASSAY (MAIN EXPERIMENT)

Clinical examinations

The animals treated with the low dose of the test item (500 mg/kg bw) or with the vehicle control (PEG 400), respectively, did not express any clinical symptoms. In the 1000 mg/kg bw dose group slightly reduced spontaneous activity was observed in one animal at 1 h post-treatment (see Table 5.8.1-24). In the 2000 mg/kg bw dose group clinical signs observed included slightly reduced spontaneous activity, abdominal posture, eyelid closure, apathy, and convulsions. These signs were only seen 1 hour post-dosing in a few animals (1-2). One animal of the high dose group was sacrificed approximately 40 minutes after application due to tonic seizures, apathy and cyanosis.

Table 5.8.1-24: Clinical examinations in the main experiment

	hours post-treatment (males)				
Clinical symptoms	**1**	**2-4**	**6**	**24**	**48**
High dose: 2000 mg/kg b.w. (14 males at 1 to 24 h; 7 males at 48 h)					
Abdominal posture	2	0	0	0	0
Eyelid closure	1	0	0	0	0
Slightly reduced spontaneous activity	2	0	0	0	0
Apathy	1	0	0	0	0
Convulsions	1	0	0	0	0
Death*	1	0	0	0	0
Medium dose: 1000 mg/kg b.w. (7 males)					
Slightly reduced spontaneous activity	1	0	0	0	0

*animal 23 was sacrificed approximately 40 minutes after application due to tonic seizures, apathy and cyanosis

Micronucleus test results

The highest dose (2000 mg/kg b.w.; maximum guideline-recommended dose) was estimated by three pre-experiments to be suitable. In the main study one male had to be euthanized after treatment with this dose due to severity of clinical symptoms. The test item induced systemic toxicity confirmed the systemic distribution of the compound. Thus, bioavailability of the test item under the tested conditions is given. Bioavailability has been demonstrated by [redacted] 1978 in the study of the metabolism of 3-PBA and its glucoside conjugate in rats, already evaluated in the first annex I inclusion process.

After treatment with the test item the number of PCEs was not substantially decreased as compared to the mean value of PCEs of the vehicle control thus indicating that 3-phenoxybenzoic acid did not exert any cytotoxic effects in the bone marrow (see Table 5.8.1-25).

A statistically significant increase in micronuclei frequency was detected in both the low dose and the 24 h high dose group, respectively, when compared to the corresponding vehicle control group. The mean values were 0.190% ± 0.10% and 0.120% ± 0.050% for the 500 and 2000 mg/kg bw dose group, respectively. However this was considered to be not biologically relevant, since all values obtained were very well within the range of historical vehicle control data. The historical control values were 0.112% ± 0.088% for the vehicle control, ranging from 0.000% to 0.450% for the individual mean group values. Moreover, no dose response relationship was observed.

40 mg/kg b.w. cyclophosphamide administered orally was used as positive control which showed a substantial increase of induced micronucleus frequency.

Table 5.8.1-25: Summary of micronucleus test results and historical control data

Test Group	Dose (mg/kg bw)	sampling time	mean MN per 6000 PCE	SD MN per 6000 PCE	mean MN (%)	SD MN (%)	Range (per 6000 PCE)		Ratio PCE /total Ery	PCE % ratio Vehicle
							min	max		
Vehicle	0	24	2.8	1.4	0.07	0.03	2	5	0.601	100.00
Dose 1	500	24	7.7*	4.1	0.19	0.10	3	15	0.612	101.86
Dose 2	1000	24	3.4	2.7	0.09	0.06	1	8	0.607	101.03
Dose 3	2000	24	5.0*	1.8	0.12	0.05	3	8	0.616	102.53
Positive	40	24	82.8*	21.9	2.07	0.55	63	120	0.598	99.53
Vehicle	0	48	4.1	1.1	0.10	0.03	3	5	0.635	100.00
Dose 3	2000	48	5.9	2.0	0.15	0.05	4	9	0.629	99.06
Vehicle# (Historical control)	-	-	-	-	0.112	0.088	0	27	-	-
CPP# (Historical control)	-	-	-	-	2.337	0.988	24	396	-	-

*: statistically significant ($p<0.05$); #: the historical database is based on the evaluation of the micronucleated cells per 2000 PCE per animal. The original values for the number of MN in the individual animals (0-9 (vehicle) or 8-132 (CPP)) was adapted in the table as MN per 6000 cells.

III. CONCLUSION

In conclusion, it can be stated that during the study described and under the experimental conditions reported, the test item 3-phenoxybenzoic acid (metabolite of BAS 310 I, Alpha-Cypermethrin) **did not induce micronuclei** as determined by the micronucleus test in the bone marrow cells of the mouse.

Report: CA 5.8.1/39
Huckle K.R. et al., 1981a
Species differences in the metabolism of 3-phenoxybenzoic acid
CY-905-031
Guidelines: none
GLP: no

Executive Summary

In this study the metabolism of ^{14}C-M310I011 has been studied in ten mammalian and one avian species in comparison with that of benzoic acid. Animals were given the radiolabeled acids (as their Na-salts) either po (intubation for rodents, gelatin capsule for others) or by ip injection. The dose levels used were 0.1, 1, 10, and 100 mg/kg bw for M310I011 or 10 mg/kg bw for benzoic acid. M310I011 exhibits wide species diversity in its metabolism, unlike benzoic acid, of which benzoylglycine (hippuric acid) is the major urinary metabolite in all species studied. With M310I011, glycine conjugation is the major route of metabolism in three species (sheep, cat, and gerbil), whereas in the mouse the taurine conjugate is the principal metabolite. The ferret eliminates similar amounts of each of these metabolites, whereas the glycylvaline dipeptide conjugate is the major metabolite isolated from the excreta of the mallard duck. Conversely, glucuronic acid conjugates of M310I011 and its 4´-hydroxy derivative (4´-HO-PBA) are the major urinary metabolites in the marmorset, rabbit, guinea pig, and hamster; the rat appears unique in eliminating the O-sulfate conjugate of 4´-HO-PBA as the principal urinary metabolite. In most cases, where amino acid conjugates are the major excretory products, the proportions of hydroxylated metabolites present are minimal. The pattern of metabolism of M310I011 does not significantly vary with dose or route (po or ip) in the sheep, gerbil, or mouse. When administered 4´-HO-PBA, the gerbil and mouse eliminate principally glucuronide and sulfate conjugates rather than amino acid conjugate, which are only minor components (<10% of the dose) in each case. This implies that hydroxylation is a primary metabolic event in determining the eventual fate of M310I011 in many species.

(BASF DocID CY-905-031)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

a) Radiolabelled test material:	^{14}C-3-Phenoxybenzoic acid; Label: [Phenyl-U]-^{14}C
Description:	not reported
Chemical name (IUPAC):	3-Phenoxybenzoic acid
Lot/Batch #:	not reported
Purity:	Radiochemical purity: 99.8% Specific activity: 31 µCi/mg

b)	**Radiolabelled test material**:	3-(4-hydroxyphenoxy)-[^{14}C]-benzoic acid; Label: [Phenyl-U]-^{14}C
	Description:	not reported
	Lot/Batch #:	not reported
	Purity:	Radiochemical purity: 99.6% Specific activity: 17.4 µCi/mg
c)	**Radiolabelled test material**:	^{14}C-benzoic acid; Label: [carboxy]-^{14}C
	Description:	not reported
	Lot/Batch #:	not reported
	Purity:	Radiochemical purity: >99.7% Specific activity: 465 µCi/mg

2. Test animals

Several species as indicated in Table 5.8.1-26 were investigated in the study.

Table 5.8.1-26: Supplementary data for animals used in the metabolism studies

Species			**Animals used**				
Common name	**Strain**	**Species**	**No./Sex**		**Weight**	**Radioactivity administered/animal**	**Diet**
			3PBA	**Benzoic acid**		**µCi**	
Rat	Wistar	Rattus norvegicus	4 m	3 m	250 ± 50 g	10	b
Marmorset		Calithrix jaccus	2 m	-	300 g	10	c
Hamster	Syrian	Mesocricetus auratus	3 f	3 f	150 ± 40 g	7	b
Guinea Pig	Dunkin-Hartley	Cavia porcellus	3 f	3 f	550 ± 50 g	10	d
Rabbit	New Zealand White	Oryctolagus cuniculus	4 f	3 f	3.5 ± 0.5 kg	20	e
Sheep	Welsh Mountain	Ovis ovis	2 f	2 f	21, 28 kg	20	f
Ferret		Mustela furo	4 f	3 f	700 ± 100 g	20	g
Cat	Mongrel	Felis cattus	1 m	1 m	3.2, 3.2 kg	20	g
Gerbil		Meriones libycus	3 m	3 f	60 ± 20 g	5	b
Mouse	CF1	Mus musculus	6 m	-	25 ± 3 g	2	b
Mallard Duck		Anas platyrhynchos	4 m	-	1.1 ± 0.1 kg	20	i

a: Mean weight, together with weight range or individual weights; b: 41B, Labsure Animal Diets, Rank Hovis McDougall, Poole, Dorset; c: Coopers Mazuri Primate Diet, B.P. Nutrition (UK), Stepfield, Witham, Essex; d: RGP, Labsure (see above); e: R14, Labsure (see above); f: Ewe and Lamb Feed, H.C. Styles (Bewdley) worcs.; g: Pedigree Chum, Pedigree Petfoods, Melton Mowbray, Leic.; i: Duck Starter Pellets, SCATS, Sutton Valence, Maidstone, Kent.

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: not reported

2. Animal assignment and treatment:
Animals were given the radiolabeled acids (as their Na-salts) either po (intubation for rodents, gelatin capsule for others) or by ip injection. The dose levels used were 0.1, 1, 10, and 100 mg/kg bw for M310I011 or 10 mg/kg bw for benzoic acid. For the duration of the experiments (up to 72 h), animals were housed individually (except for mice, 2 animals per cage) in appropriately sized all-glass metabolism cages (mini and standard size Metabowls for rodents; Jencons Ltd., Hemel Hempstead, Herts.) or in metal metabolism cages designed to permit the separate collection of urine and faeces.

3. Sample collection and investigations:
Samples of excreta from each cage were collected daily at room temperature and assayed as soon as possible after collection.
Where appropriate, faecal samples were extracted three times with a methanol/water (1:1, v/v) mixture by homogenization with an Ultra-Turrax homogenizer (half-maximal speed for 2 min). The homogenate (50% v/v) was centrifuged for 30 min at 4°C and 4000 g, and the supernatant fluid was analyzed by TLC. Samples (20-50 µL) of faecal supernatant and urine samples (5-30 µL) were analysed by LSC.
For separation and quantification of radiolabeled metabolites, urine (5-20 µL) was subjected to one-dimensional TLC in solvents A, B, and C, and in two dimensions in solvents A (first dimension) x B (second dimension) and BxC, followed by autoradiography and LSC. All urine samples were filtered (0.22 µm) before TLC.

II. RESULTS AND DISCUSSION

1. Elimination of radioactivity
The urinary excretion profiles for animals administered the radiolabeled carboxylic acids (10 mg/kg bw) are shown in Table 5.8.1-27. For each acid, the urine was the major route of elimination of radioactivity in all species. In most cases, 60-100% of the radioactivity administered was rapidly excreted in the urine within 24 h of dosing. The only animals in which the urinary excretion of radioactivity was relatively slow were the ferret and the cat, which excreted only 35% and 24%, respectively, of M310I011 and its metabolites within 24 h, and 45% and 57% respectively, during 3 days. Conversely, benzoic acid and its metabolites were rapidly eliminated by the cat and ferret, as they were in all other species studied. The mallard duck was also relatively slow to eliminate M310I011 and its metabolites, with 45% of the dose detected in the excreta in 24 h.

Faecal elimination appeared, in the majority of cases, to be a minor route of elimination for all compounds, accounting for <5% of the radioactivity aliminated in 72 h. The ferret, however, eliminated 11.6% of the administered radioactivity over a 72-h period in the faeces after treatment with M310I011. In case of benzoic acid, faecal elimination in this species was only 3.2% over 3 days.For those species given M310I011 over a range of dose levels (mouse, gerbil, and sheep) and by differing routes of administration (mouse and sheep), the urine remained the major route of elimination. The most significant difference observed was that in the sheep, which eliminated radioactivity more rapidly via this route after po dosing (98% in 24 h) than after ip administration (80% after 24 h). When 4´-HO-PBA (10 mg/kg bw, ip) was administered to the mouse and gerbil, more rapid urinary excretion of radioactivity was observed than that seen after administration of M310I011 under similar conditions.

Table 5.8.1-27: Urinary excretion of radioactivity following administration of ^{14}C-labeled M310I011 and benzoic acid to various species.

Species	Excretion after administration of carboxylic acid (10 mg/kg bw, ip) [% of administered dose]	
	M310I011	Benzoic Acid
Rat	68.7±5.1	88.0±1.7
Marmorset	73.3±64.7[a]	
Hamster	85.1±8.7	52.6±3.1
Guinea pig	87.8±2.4	70.8±3.7
Rabbit	67.6±1.0	78.6±9.7
Sheep	81.0, 78.4	83.5, 86.3
Ferret	34.8±5.4	53.9±2.5
Cat	24.3	80.7
Gerbil	65.1±4.0 (88.4±7.7)[b]	59.6±10.3
Mouse	67.1±8.0 (97.9±2.2)[b]	
Mallard duck	44.6±3.8 [a,c]	

a: po dose; b: ^{14}C excreted after administration of 4´-HO-P[^{14}C]BA; c: methanol-soluble fraction

2. Identification of the urinary metabolites of M310I011

Metabolite I (M310I011) was separately isolated from the urine of several species (rabbit, guinea pig, gerbil, and marmoset) hamster, marmoset, and rabbit urine by extraction with diethyl ether/ethanol (3:1, v/v) at pH 2.

Metabolite II (M310I025, 4´-HO-PBA) was isolated from hamster, marmoset, and rabbit urine by extraction with diethyl ether/ethanol.

Metabolite III (M310I010) was present in all urine samples collected, and was particularly prominent in those from cat, sheep, ferret, and gerbil. It was stable to enzymic treatment. The zone did not react with ninhydrin until concentrated HCl was added and the chromatogram was heated to 110°C. As chromatography was not effective to identify the standard substance, physical methods were applied for its characterization.

Metabolite IV (PBAglyval) was detected only in the methanol-soluble fraction of the excreta from the mallard duck, in which it was the principal metabolite.

Metabolite V (PBAgluc) was the major metabolite in several species (rabbit, hamster, guinea pig, marmoset, and mouse). It was partially (40%) hydrolyzed after treatment with β-glucuronidase, the radiolabeled aglycone being chromatographically indistinguishable from M310I011 standard, which was confirmed by EI-mass spectrum.
Metabolites VI and VII (4´-HO-PBAgluc and 4´-O-gluc-PBA):

Metabolite VIII (unknown) was the most ubiquitous unidentified metabolite, being particularly prominent in the marmorset and mouse. In urine samples from all species it had very similar chromatographic properties to those of 4´-$HOSO_2$OPBA. It was present in the urine from animals dosed with 4´-HO-PBA and, in the mouse exhibited a similar variation with dose as 4´-$HOSO_2$OPBA after administration of PBA. This suggests that this metabolite is a sulfate ester, perhaps of a dihydroxylated derivative of M310I011, which upon cleavage with sulfatase is not sufficiently stable to be characterized by TLC.
Metabolite IX (M310I014, 4´-$HOSO_2$OPBA) was the major metabolite in the rat and appeared in samples from most species. After treatment with sulfatase it was completely hydrolyzed, the aglycone being indistinguishable from metabolite II (M310I025, 4´-HO-PBA).
Metabolite X (PBAtau) was the major metabolite in the mouse and was also present, in smaller amounts, in the ferret and marmoset.

Table 5.8.1-28: Metabolic profile of urine samples from animals administered [^{14}C] M310I011 (10 mg/kg bw)

Met. No.	Identity	Rat	Marmor-set[a]	Hamster	Guinea pig	Rabbit	Sheep	Ferret	Cat	Gerbil	Mouse[b]	Mallard duck
I	M310I011	6.1±1.1	3.2, 3.1	21.9±2.6	19.7±2.3	37.5±2.6	2.7, 2.8	12.6±7.0	16.5	38.3±2.1	7.4±0.4	3.9±1.9[a,c]
II	M310I025, 4´-HO-PBA	3.1±1.6	3.6, 2.6	18.8±1.2	1.1±0.2	7.9±0.9	8.4, 8.7	2.1±0.8	-	1.5±0.1	3.0±0.9	1.7±0.6
III	M310I010	2.5±1.5	3.5, 2.3	2.1±0.9	2.7±0.9	2.1±0.4	87.0, 89.9	34.8±5.2	70.8	30.8±1.1	1.0±0.1	3.0±1.7
IV	PBAglyval	-	-	-	-	-	-	-	-	-	-	24.4±6.1
V	PBAgluc	5.1±1.3	25.6, 27.8	18.1±2.1	34.7±4.4	17.8±1.1	-	3.7±1.4	4.1	3.5±0.7	18.7±1.7	3.1±0.8
VI	4´-HO-PBAgluc	4.1±1.7	29.5, 28.4	15.5±1.9	15.8±2.7	16.3±1.8	-	-	-	2.3±0.3	6.7±1.5	1.6±0.4
VII	4´-Ogluc-PBA											
VIII	Unidentified	4.2±1.7	17.4, 14.5	3.4±0.2	6.6±1.1	2.6±1.0	-	9.2±4.6	-	4.9±1.3	12.2±1.8	8.6±4.1
IX	M310I014, 4´-$HOSO_2O$-PBA	63.6±0.9	3.7, 4.2	6.5±3.1	3.4±0.7	2.8±0.1	-	7.8±5.5	-	1.8±0.2	7.8±1.9	5.9±1.6
X	PBAtau	-	5.4, 5.3	2.6±0.6	-	-	-	19.3±5.1	2.9	1.4±0.9	36.5±3.4	-

a: po administration; b: estimate (two mice per cage); c: methanol-soluble fraction of excreta from mallard duck

3. Identification of the urinary metabolites of benzoic acid

The metabolic profiles obtained were very similar in comparison with those derived from M310I011, but were qualitatively similar between species. Benzoylglycine (hippuric acid) was the major metabolite in all species studied. Similarly, unchanged benzoic acid was detected in all species but generally constituted <10% dose. The most complex metabolic profile was that observed in the ferret where benzoyl glucuronide was a major urinary metabolite in addition to hippuric acid. Furthermore, two minor metabolites of unknown identity were detected; these were not present in samples from any other species. Benzoylglucuronide was detected in small quantities in the rat, hamster, and guinea pig, but was absnt from other species studies.

4. Identification of urinary metabolites of (M310I025, 4´-HO-PBA) in the mouse and gerbil

In the mouse and gerbil M310I025 is principally eliminated unchanged, together with some M310I014 (4´-$HOSO_2$O-PBA) and the glucuronic acid derivatives of 4´-HO-PBA. This is in markled contrast to the pattern seen with M310I011, for which amino acid conjugates are the major urinary metabolites detected in these species (M310I010 in gerbil and PBAtau in mouse). Minor urinary components (<10%) were detected in gerbil and mouse. The chromatographic mobilities suggest that unidentified XI and XII are the glycine and taurine conjugates of 4´-HO-PBA. Due to extensive degradation following derivatisation no characterisation by means of mass spectrometry was possible.

5. The effect of variation of dose on the metabolism of M310I011 in various species

^{14}C-labeled PBA was administered ip to sheep, mice, and gerbils over a 1000-fold dose range: 0.1, 1, 10, and 100 mg/kg bw. In all three species at these doses, the intraspecies metabolic profiles obtained were qualitatively similar. Thus, the respective amino acid conjugates (M310I010 in gerbil and sheep; PBAtau in mouse) were the major urinary metabolites detected. The sheep and gerbil exhibited little quantitative variation in metabolism with dose; in the sheep M310I010 accounted for 72, 73, 86, and 77% of the administered dose eliminated in 24 h at 0.1, 1, 10, and 100 mg/kg bw, respectively, and in the gerbil corresponding values were 22, 17, 20, and 21%. In the mouse, the taurine conjugate was 20, 21, and 25% of the dose at 0.1, 1, and 10 mg/kg bw, respectively, whereas at 100 mg/kg bw 30% were observed. Of the other reactions detected, glucuronide acid conjugation in the gerbil appeared to decrease as the dose was increased. There was a concomitant increase in unconjugated M310I011: 7.6, 9.1, 25, and 12% of the dose at 0.1, 1, 10, and 100 mg/kg bw, respectively. This possible saturation of the glucuronide pathway may also be seen to small extent in the mouse (26, 27, 17, and 22%), with a concomitant increase in free M310I011 (2.1, 2.1, 5.0, 8.3%). (M310I014, 4´-$HOSO_2$PBA and the unidentified metabolite VIII were present at minor products of M310I011 in the gerbil and mouse at 0.1, 1, and 100 mg/kg bw, as at 10 mg/kg bw. Variations in dose did not appear to appreciably affect the extent of hydroxylation of M310I011 in sheep, gerbil, or mouse.

Table 5.8.1-29: Hydroxylation of M310I011 (10 mg/kg bw) in various species

Species	Major urinary conjugate type	M310I025 4´-HO-PBA	Glucuronic acid conjugates of 4´-HO-PBA	M310I014 4´-$HOSO_2O$-PBA	Total hydroxylation
Rat	Sulfate	2.1	2.8	43.7	48.6
Hamster	Glucuronide	16.0	13.2	5.5	34.7
Marmorset		2.6, 1.7	21.6, 18.4	2.7, 2.7	26.9, 22.8
Rabbit		5.3	11.0	1.9	18.2
Guinea pig		1.0	13.9	3.0	17.9
Mouse	Taurine	2.0	1.1	5.2	8.3
Mallard duck[a]	Glycylvaline	0.8	0.7	2.6	4.1
Sheep	Glycine	6.8, 6.8	/	/	6.8, 6.8
Gerbil		1.0	1.5	1.2	3.7
Ferret[b]		0.8	/	2.7	3.5
Cat		/	/	/	/

6. The effect of route of administration on the metabolism of M310I011 in mouse and sheep

As with the dose-level studies, the metabolic profile of sheep urine remained very similar when M310I011 (10 mg/kg bw) was administered to these animals either by ip or po routes. In each case, M310I010 was the major metabolite (88% and 90% ip and po, respectively) with similar minor amounts of M310I025 (4´-HO-PBA) and unchanged M310I011 being detected. In the mouse, however, at 10 mg/kg bw hydroxylation appeared more extensive after po dosing than ip injection. Amounts of urinary M310I025 increased from 2% (ip) to 13.2% (po) of the dose; amounts of free M310I011 eliminated are also increased 4-fold. PBAtau, however, remains the principal urinary metabolite detected after administration by both routes. It is noteworthy that the radioactivity eliminated in the urine following po dosing was 88%, but was 67% afer ip injection.

7. Discussion

M310I011 is rapidly absorbed and in the majority of cases it undergoes extensive metabolism and is excreted predominantly via the urine; faecal elimination is a minor excretory pathway in most species. The major metabolic reactions include direct conjugation of the carboxyl group with amino acids (glycine in sheep, gerbil, cat, and ferret; taurine in mouse and ferret, a dipeptide, glycylvaline, in the mallard duck) or with glucuronic acid. Oxidation occurs extensively in the rat and, to lesser extents, in some other species. The phenolic compound produced (M310I025) is subsequently conjugated with sulfate or glucuronic acid. Thus, M310I014 forms the major metabolite in the rat, and the ether and ester glucuronides of 4´-HO-PBA (generally accompagnied by glucuronide of M310I011) are formed in the marmorset, hamster, guinea pig, and rabbit. Other hyhydroxylsomers are formed in some species, but are quantitatively, together with other unidentified metabolites, of minor significance. Variations in dose or route did not appreciably affect the pattern of metabolism of M310I011.

Benzoic acid shows very little interspecies variation. It was excreted rapidly in the urine mainly as hippuric acid, except in the ferret (benzoylglucuronide).

M310I025 is mainly metabolized by sulfation and glucuronidation, acting rather as a phenol than a carboxylic acid. Appreciable amounts of unchanged M310I025 are eliminated in the gerbil and mouse. Furthermore, M310I025 is eliminated more rapidly in the urine of both the gerbil and mouse than is M310I011.

III. CONCLUSION

In this study the species differences in metabolism of M310I011 were investigated. It has been shown that M310I011 is rapidly absorbed and in the majority of cases it undergoes extensive metabolism and is excreted predominantly via the urine. The major metabolic reactions include direct conjugation of the carboxyl group with amino acids or with glucuronic acid. Oxidation occurs extensively in the rat and, to lesser extents, in some other species.

Report: CA 5.8.1/40
[redacted] 2000a
CL 206128: Acute oral toxicity study in mice
AL-460-099
Guidelines: EPA 870.1100, OECD 401, JMAFF 59 NohSan No 4200
GLP: yes
(certified by Ministry of Agriculture, Forestry and Fisheries of Japan, Japan)

Executive Summary

Single doses of 1021, 1429, and 2000 mg/kg bw of M310I011 preparations in corn oil were given to groups of 5 male and 5 female animals each by gavage. Animals were observed for 14 days. No mortality occurred in the control and 1021 mg/kg bw dose groups. Each 1/5 males and 3/5 females were found dead in the 1429 and 2000 mg/kg bw dose groups, respectively. Accordingly, the oral LD_{50} was found to be greater than 2000 mg/kg bw for males and 1511 mg/kg bw for females, respectively:

Rat, oral: $LD_{50} > 2000$ mg/kg bw (males),
$LD_{50} = 1511$ mg/kg bw (females)

Clinical signs noted were prone position, lateral position, restlessness, stupor, sedation, decrease in or loss of spontaneous motor activity, convulsions, ptosis, and soiled fur in the external genital region in both males and females. The mean body weights of the administration groups increased throughout the study period. In the animals that died during the study red color and/or edema of the small intestine and red/black contents in the small intestine in both males and females and black spots in the glandular stomach and soiled fur in the external genital region in females were noted. No macroscopic pathologic abnormalities were noted in the animals examined at the end of the observation period.
Under the conditions of this study the median lethal dose of M310I011 after oral administration was found to be greater than 2000 mg/kg bw in males and 1511 mg/kg bw in females, respectively.

DocID (AL-460-099)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: CL 206128 (3-phenoxybenzoic acid)
Description: Powder/white
Lot/Batch #: AC11303-61
Purity/content: 99%

2. Vehicle: Corn oil

3. Test animals:

Species:	Mouse
Strain:	Crj:CD-1
Sex:	male/female
Age:	6 weeks (at administration of test substance)
Weight at dosing (mean):	25-30 g (males), 20-25 g (females)
Source:	Atsugi Breeding Center, Charles River Japan Inc., Shimofurusawa, Atsugi-shi, Kanagawa, Japan
Acclimation period:	8 days
Diet:	Pellet diet MF (Oriental Yeast Co., Ltd., Azusawa, Itabashi-ku, Tokyo), ad libitum
Water:	Well water, sand and charcoal filtered, sterilited with sodium hypochlorite, ad libitum
Housing:	Five animals per aluminium cage with wire-mesh floor (Tokiwa Kagakukikai Co., Ltd., Ueno, Taito-ku, Tokyo)
Environmental conditions:	
Temperature:	24 °C
Humidity:	55%
Air changes:	at least 10 times per hour
Photo period:	Alternating 12-hour light and dark cycles, artificial light from 7 am to 7 pm

B. STUDY DESIGN AND METHODS

1. Dates of work: 13-Oct-1999 - 27-Oct-1999 (Experimental period)

2. Animal assignment and treatment:

Single doses of 1021, 1429, and 2000 mg/kg bw of M310I011 preparations in corn oil were given to groups of 5 male and 5 female animals each by gavage, which were fasted for 2-3 hours before and 3 hours after administration. Clinical signs and symptoms were recorded 30 min, 1, 3, and 6 hours after administration and afterwards at least once daily for the individual animals up to 14 days post-administration. Individual body weights were determined shortly before administration, weekly thereafter and at the end of the study, or when the animal was found dead. The animals were sacrificed after ether anesthesia and subjected to necropsy including gross pathological examination on the last day of the observation period or as soon as possible after death in case of animals that died before.

II. RESULTS AND DISCUSSION

A. MORTALITY

No mortality occurred in the control and 1021 mg/kg bw dose groups. Each 1/5 males and 3/5 females were found dead in the 1429 and 2000 mg/kg bw dose groups, respectively. Deaths occurred at 3 hours (males) and from 3 hours to 1 day (females) after administration.

B. CLINICAL OBSERVATIONS

Clinical signs noted were prone position, lateral position, restlessness, stupor, sedation, decrease in or loss of spontaneous motor activity, convulsions, ptosis, and soiled fur in the external genital region in both males and females. Additional findings included wetted fur in the external genital region in males and tremor, bradypnea, hypothermia, and lacrimation in females. These signs began to appear from 30 min after administration in both males and females and disappeared by day 5 and 2 in surviving males and females, respectively. No clinical signs were observed in the vehicle control animals.

C. BODY WEIGHT

The mean body weights of the administration groups increased throughout the study period.

D. NECROPSY

In the animals that died during the study red color and/or edema of the small intestine and red/black contents in the small intestine in both males and females and black spots in the glandular stomach and soiled fur in the external genital region in females were noted. No macroscopic abnormalities were observed in the surviving animals.

III. CONCLUSION

Under the conditions of this study, the oral LD_{50} for M310I011 in mice was determined to be greater than 2000 mg/kg bw for males and 1511 mg/kg bw for females.

Report: CA 5.8.1/41
[redacted] 1979a
A neurotoxicity study on the pyrethroid metabolite 3-Phenoxybenzoic acid (3-PBA)
CY-470-007
Guidelines: none
GLP: no

Executive Summary

M310I011 (Batch: 19027-6; Purity: 99%) was administered orally by gavage to groups of 8 male and 8 female rats at concentrations of 0, 25, 77 and 375 mg/kg bw/day over a period of 7 days. The positive control substance Cypermethrin was applied at 150 mg/kg bw/day. No clinical signs of intoxication were seen in any of the three groups dosed with M310I011, although mortality did occur with one female death in each dose group and one male death in the high dose group. Slight weight loss was observed in the high dose group during the dosing period. Normal weight gain was observed in all groups after cessation of dosing.
No increase in beta-glucuronidase and beta-galactosidase activities were observed in either the distal or proximal sections of the sciatic/posterior tibial nerve or in the trigeminal ganglia of the groups of rats dosed with M310I011. Groups treated with 25 mg/kg bw showed a statistically significant lower beta-galactosidase activity in the trigeminal ganglia. This decrease in activity however was marginal and was only seen in the females. Moreover this decrease was not seen in any of the rats given higher doses of M310I011.
Rats treated with cypermethrin showed a significant increase in beta-glucuronidase and beta-galactosidase activities. These enzyme increases were largest and most reproducible in the distal section of the sciatic/posterior tibial nerve and in the trigeminal ganglia.
Therefore, under the conditions of the present study the metabolite M310I011 is unlikely to be responsible for the enzymatic changes seen in the sciatic/posterior tibial nerve with cypermethrin.
(Doc ID CY-470-007)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: 3-Phenoxybenzoic acid
Description: not available
Lot/Batch #: 19027-6
Purity: 99%
Stability of test compound: The test substance was stable if kept cool and in the dark.

2. Vehicle and/or positive control: DMSO and Cypermethrin (WL 43467)

3. Test animals:

Species:	Rat
Strain:	Wistar
Sex:	Male and female
Age:	10-12 weeks
Weight at day 1 after dosing:	♂: approximately 93 – 101 g ♀: approximately 96 – 101 g
Source:	Shell Toxicology Laboratory (Tunstall), Sittingbourne, UK
Acclimation period:	no data
Diet:	no data
Water:	no data
Housing:	Single housing
Environmental conditions:	
Temperature:	18 - 24 °C
Humidity:	no data
Air changes:	no data
Photo period:	no data

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: September 1979

2. Animal assignment and treatment:

M310I011 was administered orally by gavage to groups of 8 male and 8 female rats at concentrations of 0, 25 (low dose), 77 (mid dose) and 375 mg/kg bw/day (high dose) over a period of 7 days.

3. Test substance preparation:

M310I011 was applied as a 10% (w/v) solution in DMSO.

4. Statistics:

Means and standard deviations of each test group were calculated for several parameters. Further statistical analyses were performed but no further details were given.

C. METHODS

1. Observations:
A check for moribund and dead animals as well as for clinical signs of toxicity was made before the start of the administration period and at frequent intervals after dosing.

2. Body weight:
Body weight was determined before the start of the administration period and at frequent intervals after dosing.

3. Enzyme determination:
Three weeks after the commencement of dosing the rats were killed by cervical dislocation. The right and left sciatic nerves were dissected as far as the distal phalangeal branche of the posterior tibial nerve. Each dissected sciatic/posterior tibial nerve was then dissected into a “proximal” and “distal” half. The right and left trigeminal ganglia were dissected free after removal of the brain.
Each tissue sample was weighed and homogenized in a volume of ice-cold 0.1 M acetate buffer (pH 4.5; containing 0.1% v/v Triton X-100) calculated to yield a 1% w/v homogenate.
Beta-galactosidase activity was determined using methylumbelliferyl-galactoside as substrate in the enzyme assay using fluorescence measurement for quantitation.
Similarly beta-glucuronidase activity was determined using methylumbelliferyl-glucuronide as substrate.

II. RESULTS AND DISCUSSION

A. TEST SUBSTANCE ANALYSES
No details given

B. OBSERVATIONS

1. Clinical signs of toxicity
No clinical signs were observed throughout the study after treatment with M310I011.
After treatment with Cypermethrin ataxia, abnormal gait, hyperexcitability, and tremor were observed.

2. Mortality
In every group dosed with M310I011 one female rat was found dead. One male rat of the high dose group was found dead. Mortalities occurred between day 2 and day 5.
After treatment with Cypermethrin 2 females were found dead. Mortalities occurred between day 2 and day 8.

C. BODY WEIGHT AND BODY WEIGHT GAIN

No weight gain was observed in any dose group including the control group during the dosing period. After dosing rats of all groups gained weight normally. During dosing weight loss was observed in the 375 mg/kg bw group and in the females of the 77 mg/kg bw group.

D. ENZYME ACTIVITY

No increase in beta-glucuronidase and beta-galactosidase activities were observed in either the distal or proximal sections of the sciatic/posterior tibial nerve or in the trigeminal ganglia of the groups of rats dosed with M310I011. Groups treated with 25 mg/kg bw showed a statistically significant lower beta-galactosidase activity in the trigeminal ganglia. This decrease in activity, however was marginal and was only seen in the females. Moreover this decrease was not seen in any of the rats given higher doses of M310I011.
Rats treated with Cypermethrin showed a significant increase in beta-glucuronidase and beta-galactosidase activities. These enzyme increases were largest and most reproducible in the distal section of the sciatic/posterior tibial nerve and in the trigeminal ganglia.

III. CONCLUSIONS

The administration of Cyphermethrin produces the expected neurochemical changes in the rat peripheral nervous system. The metabolite M310I011 did not induce the biochemical changes that were observed with Cypermethrin, and is therefore unlikely responsible for the neurotoxic properties.

Supplemental information from open literature about toxicokinetics of pyrethroid metabolits in males and female rats was evaluated in the DAR or lambda-cyhalothrin [see Ueyama et al., 2010; discussed in Draft Renewal Assessement Report of Lambda-cyhalothrin or RMS Sweden of February 2013, page 125-129] indicating that elimination and distribution of M310I011 shows no gender-related differences in rats.

Information from the open literature about the endocrine investigations with M310I011 (PBAcid) is summarized in the Table 5.8.1-30 below. The Literature is however described in more detail in the chapter CA 5.8.3.

In vitro studies on endocrine activity:
M310I011 has been reported to show some weak estrogenic effects on mRNA expression level for estrogen receptor and pS2 at approx. 1000-fold higher concentrations than beta-E2 (Jin et al., 2010; [see CA 5.8.3/17 2010/1232199]). However, an activation of an estrogen receptor-dependent gene-expression was consistently not seen in several transactivation assays, but weak anti-estrogenic potential was repeatedly demonstrated in yeast and mammalian systems. While the RIC20 was found in the nanomolar range, a significant inhibition was only seen in the µ-molar concentration range that is not considered relevant for the invivo situation (IC_{50} of 10-100 µM). E-Screen assays in MCF-7 cells showed also very weak estrogen like effect by M310I011 by inducing proliferation at 0.1 µM about 1.3-fold above control, but this was not reproduced by Laffin et al., 2010, [see CA 5.8.3/16 2010/1232196]. An induction of proliferation of MCF-7 cells is not directly attributable to an estrogenic effect but might mirror other stimulis and therefore this study is not considered relevant but rather supportive.
M310I011 was consistently not active as androgen receptor agonist in yeast and in mammalian cells. An anti-androgenic potential was not seen in yeast cells and in mammalian cells the transactivation of human androgen receptor via DHT was reduced about 20% at 1.21 mM. Therefore this finding is rather considered unrelevant for further consideration.
No acitivty was seen of M310I011 on the thyroid receptor dependent genexpression. Some anti-TH activity is calculated with an RIC20 in the µ-molar range (4.76E-06M) but the complete measure did not show any significant inhibition up to 10 µM. As studies with cypermethrins in general did not reveal any thyroid toxicity, this weak potency found for one of the main metabolites is not considered of importance as it is not reflected in any in vivo study.

In vivo studies:
Laffin et al., 2010 [see CA 5.8.3/16 2010/1232196] evaluated the estrogenic activity in vivo using the uterotrophic assay in Sprague-Dawley rats. Ovariectomized animals were orally gavaged at 1, 5, and 10 mg/kg bw of M310I011 dissolved in corn oil.once daily for 3 days. No effect on uterine wet weigth or body weight was found. In addition no effect on organ weights, onset of puberty and sexual maturation was found in a pubertal assay in Sprague-Dawley rats at 0, 1, 5 or 10 mg/kg bw/day administered from weaning (PND 22) until detection of the onset of puberty by vaginal opening.

Considering all available data on endocrine potential, M310I011 is not considered to be estrogenic, anti-estrogenic, androgenic or anti-androgenic active at concentrations relevant for the in vivo situation. A significant effect on the thyroid receptor was not seen, and the slight antagonistic trend on the thyroidreceptor at around 10 µM is not considered a relevant concentration range in the invivo situation and in addition it is not reflected by the toxicity profile of the active ingredient alpha-cypermethrin.

Table 5.8.1-30: Endocrine activity studies with M310I011

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference Doc ID
In vitro assays				
ER-dependent regulation of mRNA expression	Estrogen Receptor (ERα) mRNA levels in MCF-7	Estrogenic active RIE: 24% at 1µM	E2: ↓(at 1nM) RIE set to 100%	Jin et al., 2010 2010/1232199
ER-dependent regulation of mRNA expression	pS2 mRNA levels in MCF-7	Estrogenic active RIE: 28.5% at 1µM	E2: 4-fold ↑ (1 nM) RIE set to 100%	Jin et al., 2010 2010/1232199
ER-activation	hER transactivation in Yeast (lacZ)	ER-agonist: Not active (up to 1E-3M) ER-antagonist: Active LOIC: 1.25±0.7E-05 M IC50: 6.5±2.5E-05 M	Agonist: E2: EC_{50}: 2.1±0.4E-10 M Antagonist: 4HO-TAM: IC_{50}: 2.8±0.7E-6 M	Tyler et al., 2000 2000/1024078
ER-activation	hERα transactivation in Yeast (lacZ)	ER-agonist: Not active	Agonist: E2: EC_{50}: 0.35±0.16E-9M	McCarthy et al., 2006 2006/1051135
ER-activation	hERα transactivation in CV-1 cells (Luciferase)	ER-agonist: Not active (up to 1E-4M) ER-antagonist: IC50:approx. 1E-04M	Agonist: E2: EC_{50}: 7.72E-09 M Antagonist: ICI182780 IC_{100}: 10E-6 M	Sun et al., 2014 2014/1242697
	rERα transactivation in CV-1 cells (Luciferase)	ER-agonist: Not active (up to 1E-4M) ER-antagonist: Not given		
ER-activation	hER transactivation in CV-1 cells (Luciferase)	ER-agonist: Not active ER-antagonist: Active RIC20: 8.84E-08 M IC50: approx. 1E-05M	Agonistic: 17β-E2: EC50: 3.7E-9 M (REC: 100% = 1E-9M 17β-E2)	Du et al., 2010 2010/1232195
ER-activation	hER transactivation in MCF-7 cells (Luciferase)	ER-agonist: Not active (up to 10 µM)	Agonist: E2 (10 nM):2.3-fold induction	Laffin et al., 2010 2010/1232196
ER-activation	hER transactivation in MCF-7 cells (Luciferase)	ER-agonist: Not active	Agonist: E2 EC20: 2.4 E-8M	Tange et al., 2000 2000/1024078
ER-dependent proliferation	E-Screen (MCF-7)	Proliferative effect: not active (up to 10 µM)	E2: 3-fold ↑ at 1 nM	Laffin et al., 2010 2010/1232196
ER-dependent proliferation	E-Screen (MCF-7)	Proliferative effect: active at 0.1 µM PE: 1.32 RPE: 28.6%	E2: 1 nM PE: 2.12 RPE: 100%	Jin et al., 2010 2010/1232199

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference Doc ID
AR-activation	hAR transactivation in Yeast (lacZ)	AR-agonist: Not active AR-antagonist: Not active	Agonist: DHT: EC_{50}: 9.7±0.3E-10M Antagonist: Flutamide: IC_{50}: 6.8±0.7E-6 M	Tyler et al., 2000 2000/1024078
AR-activation	hAR transactivation in CHO cells (Luciferase)	AR-agonist: not tested AR-antagonist: Not active	Antagonist: Flutamide: IC_{20}: 0.16E-6 M IC_{50}: 0.62E-6M	Tange et al., 2014 2014/1242695
AR-activation	hAR transactivation in MDA-kb2 cells (Luciferase)	AR-agonist: Not active AR-antagonist: weakly active RIC_{20}: >1E-5 M	Agonist: DHT: EC50: 3.99E-10 M (REC: 100% = >1E-8M DHT) Antagonistic: n.a.	Du et al., 2010 2010/1232195
AR-activation	hAR transactivation in CV-1 cells (Luciferase)	AR-agonist: Not active AR-antagonist: Weakly active RIC_{20}: 1.21E-03M	Agonist: DHT: EC50: 3.99E-10 M Antagonistic: Flutamid: IC_{50}:1E-6M Nilutamide: IC_{50}:1E-6M	Sun et al., 2007 2007/1070385
TRβ-activation	TRβ transactivation in CV-1 cells (Luciferase)	TR-agonist: Not active TR-antagonist: active RIC_{20}: 4.76E-6 M	Agonist: T3: EC50: 2.94E-9 M (REC: 100% = >1E-7M T3) Antagonistic: n.a.	Du et al., 2010 2010/1232195
In vivo				
Uterus weight	Uterotrophic assay in SD rats	Ostrogenic acivity: Not active	Agonist: EE 0.6µg/kg bw Uterine wet weight: 5x ↑	Laffin et al., 2010 2010/1232196
Age at VO, Age at first diestrus	Pubertal female assay in SD rats	Ostrogenic acivity: Not active	No control	Laffin et al., 2010 2010/1232196

PE: Proliferative effect; E2: 17 beta-Estradiol; EE: ethinyl estradiol; 4HO-TAM: 4-Hydroxy-tamoxifen; RPE: relative proliferative effect calculated as the ratio (PE-1) of the test chemical over (PE-1) of E2 (x 100); RIE: The relative inductive efficiency (RIE) is the ratio between the maximal up-regulation of pS2 expression level by the test compound to that of E2 (x100) or the relative inhibitory effciency of down-regulation of ERalpha expression level by the test compound compared to that of E2 (x 100); EC50/IC50: Effect concentration /Inhibitor concentration at which 50% increase /decrease is found compared to control; LOIC: Lowest observed inhibitory concentration

Toxicological evaluation of M310I011

The QSAR evaluation of M310I011 is of good reliability and by weight of evidence there was no alert for genotoxicity.

In an Ames-test conducted [see CA 5.8.1/36 AL-435-010] M310I011 was not genotoxic without or with metabolic activation. An in vitro chromosome aberration test in P53 deficient V79 cells [see CA 5.8.1/37 2014/1168736] showed a positive response without metabolic activtion but without a clear dose-response relationship. With metabolic activation the isolated finding in the mid dose group was not reproduced by the repeat experiment and was furthermore without dose-response relationship and thus not considered indicative of a positive genotoxic response. The subsequently conducted in vivo Micronucleus test was negative [see CA 5.8.1/38 2015/1001101]. In conclusion M310I011 was not considered to be genotoxic by weight of evidence

The available systemic toxicity studies indicate a moderate short-term toxicity and give no indication that M310I011 is responsible for the neurotoxic properties known for cypermethrins. The acute toxicity is mice is low with an LD_{50} greater than 2000 mg/kg bw for males and 1511 mg/kg bw for females. Information in rat is derived from the 7-day neurotoxicity mode of action study and from the apical endocrine studies in female rats, while the neurotoxicity study indicates some toxicity down to the lowest tested dose of 25 mg/kg bw/day in particular females but without a dose response, the 3-day gavage administration in the uterotrophic assay did not give any evidence for systemic toxicity up to the highest dose tested i.e. 10 mg/kg bw/day. With regard to the pubertal assay there was also no indication for systemic toxicity up to the highest tested dose of 10 mg/kg bw/day when applied from weaning (PND up to the onset of puberty).

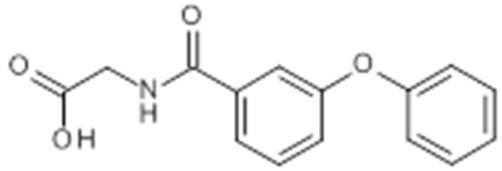
Literature data indicate that M310I011 has no relevant endocrine activity on the estrogenic, androgenic or thyroid system.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I011 is by weight of evidence considered to be not genotoxic. The available toxicological database does not provide any concern for M310I011. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the relevant TTC-threshold. In conclusion M310I011 is considered to be **not toxicologically relevant**.

M310I010 (other denominators: Reg. No. 4108084, 3PBA glycine, WL 46194, CL 117585, N-(3-phenoxybenzoyl)glycine)

M310I010 is the glycine conjugate of M310I011. It is determined as a rat, livestock (goat) and crop (wheat) metabolite.



a. Toxicological evaluation of M310I010

Being the glycin-conjugate of M310I011, toxicological evalution of M310I010 is based on the unconjugated analogon. Conjugation of metabolites with amino acids are according to the EFSA Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment [EFSA Journal 2012;10(07):2799] considered to probably not cause higher toxicity of the metabolites. Moreover it has been shown in several mammalian species that M310I010 is the major metabolite of M310I011 and thus it can be considered to be intrinsically tested in the studies conducted with M310I011.

For the non-conjugated structural analogue M310I011 results from an Ames test an Micronucleus test in vitro and a Micronucleus test in vivo are available [see CA 5.8.1/36 AL-435-010], [see CA 5.8.1/37 2014/1168736] and [see CA 5.8.1/38 2015/1001101]. In the Ames-test M310I011 was not genotoxic without or with metabolic activation. The in vitro chromosome aberration test in P53 deficient V79 cells [see CA 5.8.1/37 2014/1168736] showed a positive response without metabolic activtion but without a clear dose-response relationship. With metabolic activation the isolated finding in the mid dose group was not reproduced by the repeat experiment and was furthermor without dose-response relationship and thus not considered indicative of a positive genotoxic response. The subsequently conducted in vivo Micronucleus test showed negative results [see CA 5.8.1/38 2015/1001101]. In conclusion the glycin-conjugate M310I010 is not considered to be genotoxic by weight of evidence based on the studies conducted with M310I011. The available systemic toxicity studies for M310I011 indicate a moderate acute toxicity and give no indication that M310I011 respectively its analogon M310I010 are responsible for the neurochemical changes known for cypermethrins. Therefore, based on the negative in vitro data regarding mutagenicity and clastogenicity of the structural analogue M310I011, the glycin-conjugate M310I010 is considered to be not genotoxic.

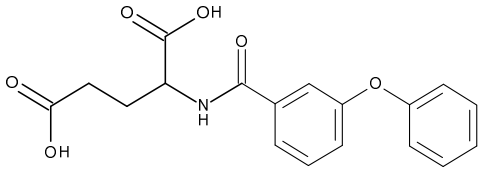
The available systemic toxicity studies for M310I011 indicate a moderate acute toxicity and give no indication that M310I011 respectively its analogon M310I010 are responsible for the neurochemical changes known for cypermethrins.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I010 based on the information available for M310I011 is by weight of evidence considered to be not genotoxic. The available toxicological database does not provide any concern for M310I010. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the relevant TTC-threshold. In conclusion, M310I010 is considered to be **not toxicologically relevant**.

M310I026 (other denominators: Reg. No. 4110960, 3-phenoxy-benzoyl glutamic acid)

M310I026 is the glutamate conjugate of M310I011. It is determined as a crop (wheat) metabolite.



a. Toxicological evaluation of M310I026

Being the glutamate-conjugate of M310I011, toxicological evalution of M310I026 is based on the unconjugated analogon. Conjugation of metabolites with amino acids are according to the EFSA Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment [EFSA Journal 2012;10(07):2799] considered to probably not cause higher toxicity of the metabolites.

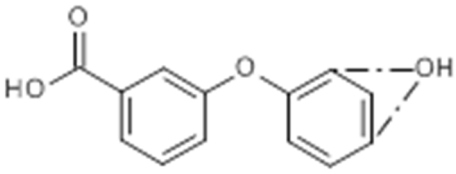
For the non-conjugated structural analogue M310I011 results from an Ames test an Micronucleus test in vitro and a Micronucleus test in vivo are available [see CA 5.8.1/36 AL-435-010], [see CA 5.8.1/37 2014/1168736] and [see CA 5.8.1/38 2015/1001101]. In the Ames-test M310I011 was not genotoxic without or with metabolic activation. The in vitro chromosome aberration test in P53 deficient V79 cells [see CA 5.8.1/37 2014/1168736] showed a positive response without metabolic activtion but without a clear dose-response relationship. With metabolic activation the isolated finding in the mid dose group was not reproduced by the repeat experiment and was furthermor without dose-response relationship and thus not considered indicative of a positive genotoxic response. The subsequently conducted in vivo Micronucleus test showed negative results [see CA 5.8.1/38 2015/1001101]. In conclusion the glutamate-conjugate M310I026 is not considered to be genotoxic by weight of evidence based on the studies conducted with M310I011.

The available systemic toxicity studies for M310I011 indicate a moderate acute toxicity and give no indication that M310I011 respectively its analogon M310I026 are responsible for the neurochemical changes known for cypermethrins.

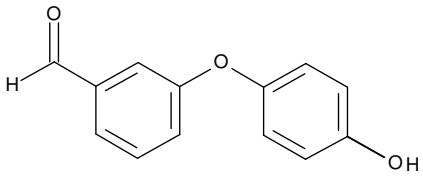
With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

M310I026 based on the information available for M310I011 is by weight of evidence considered to be not genotoxic. The available toxicological database does not provide any concern for M310I026. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the relevant TTC-threshold. In conclusion, M310I026 is considered to be **not toxicologically relevant**.

M310I013 (other denominators: CL 213336, WL 46114) and M310I025 (other denominators: Reg.Nr.4110493)



M310I013



M310I025

M310I013 is a metabolite found in rat. M310I025 is the para-hydroxylated derivative and is a metabolite identified in wheat and cabbage. Given the experimental methods available at study conduct of the cabbage and the wheat study the actual position of the hydroxylation group could not be specified. However, taking together the knowledge on metabolic pathways in crops (wheat, cabbage) and live-stock animals and mammalian species [see structure of M310I017 No. 2 above, and the extensive data on metabolism of M310I011 in mammalian species No. 16 above] it is likely that the actual position of the hydroxylation group is the para-position and thus M310I013 as determined in rat is identical to M310I025 as determined in wheat and cabbage.

a. QSAR Predicitions on M310I013 and M310I025

OASIS TIMES (V.2.27.15.146; Mutagenicity S-9 activated v09.09) [see molecule 12 of reports [see KCA 5.8.1/1 2014/1289314] and [see KCA 5.8.1/2 2014/1289315]]
There were **no** Ames mutagenicity alerts for M310I013 or in-silico generated metabolites and no structural alerts were reported. The same holds true for in-vitro chromosome aberration. In all cases the structures were out of domain.

VEGA: Mutagenicity model (CAESAR, version 2.1.9) [see molecule 12 of report [see KCA 5.8.1/5 2014/1289317]]
M310I013 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules had positive experimental data. The other four metabolites were predicted 'non-mutagen', which was experimentally confirmed. The concordance of the total underlying database was moderate (0.668) and thus is not very robust. Both the positive (similarity 0.844 to 0.854) and the negative molecules (similarity 0.846 to 0.861) have a reasonably similar structure to M310I013. Therefore, the chemical space does adequately cover the structure of M310I013.

VEGA: Mutagenicity model (SarPy; version 1.0.6-DEV) [see molecule 12 of report [see KCA 5.8.1/5 2014/1289317]]

M310I013 could be out of model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. The same metabolites as reported in the CAESAR model were also used in the SarPy model.

VEGA: Mutagenicity model (TOXTREE; version 1.0.0-DEV) [see molecule 12 of report [see KCA 5.8.1/5 2014/1289317]]
M310I013 could be out of the model applicability domain. The prediction is **'non-mutagen'** with no specific structural alerts. Two of the six most similar molecules had positive experimental data. The concordance of the total underlying database was low (0.496) and thus is not very robust. All six molecules (similarity 0.798 to 0.821) have a reasonably similar structure to M310I013. Therefore, the chemical space does adequately cover the structure of M310I013.

Conclusion on QSAR evaluations
No conclusive structural alert was identified for M310I013/M310I025.

b. Toxicological data on M310I013 and its structural analog M310I025

M310I013/M310I025 are reported in the literature to show neither estrogenic nor anti-androgenic activity.

Table 5.8.1-31: Endocrine activity studies with M310I013/M310I025

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference Doc ID
In vitro data were identical for both metabolites				
ER-activation	hER transactivation in MCF-7 cells (Luciferase)	ER-agonist: not active ER-antagonist: Not tested	Agonist: E2 EC20: 2.4 E-8M	Tange et al., 2000 2000/1024078
AR-activation	hAR transactivation in CHO cells (Luciferase)	AR-agonist: not tested AR-antagonist: not active	Antagonist: Flutamide: IC_{20}: 0.16E-6 M IC_{50}: 0.62E-6M	

c. Toxicological evaluation of M310I013/M310I025

The available systemic toxicity studies for M310I011 indicate a moderate acute toxicity and give no indication that M310I011 respectively its analogon M310I013/M310I025 are responsible for the neurotoxic properties known for cypermethrins.

The QSAR evaluation of M310I025 the para-hydroylated moiety is of moderate reliability and by weight of evidence there was no conclusive alert for genotoxicity.
M310I013/M310I025 is the ring-hydroxylated analogon of M310I011. Hydroxylation of the ring system without any cleavage of the ring is often identified as probably not causing higher toxicity of metabolites (EFSA, 2012; 10(07):2799).

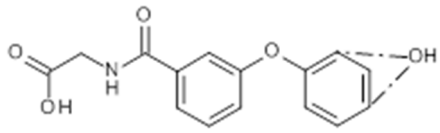
For the structural analogon M310I011 results from an Ames test an Micronucleus test in vitro and a Micronucleus test in vivo are available [see CA 5.8.1/36 AL-435-010], [see CA 5.8.1/37 2014/1168736] and [see CA 5.8.1/38 2015/1001101]. In the Ames-test M310I011 was not genotoxic without or with metabolic activation. The in vitro chromosome aberration test in P53 deficient V79 cells [see CA 5.8.1/37 2014/1168736] showed a positive response without metabolic activtion but without a clear dose-response relationship. With metabolic activation the isolated finding in the mid dose group was not reproduced by the repeat experiment and was furthermor without dose-response relationship and thus not considered indicative of a positive genotoxic response. The subsequently conducted in vivo Micronucleus test was negative [see CA 5.8.1/38 2015/1001101]. In conclusion the structural analogon M310I013/M310I025 was not considered to be genotoxic by weight of evidence based on the studies conducted with M310I011. Therefore, based on the negative in vivo data regarding mutagenicity and clastogenicity of the structural analogue M310I011 in combination with the negative prediction for M310I013 in the various QSAR models, M310I013 is considered to be not genotoxic.

M310I013/M310I025 showed no estrogenic or anti-androgenic properties. Further investigations are not considered relevant.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. However, the acute estimates of M310I025 will slightly exceed the TTC level of 5 µg/kg bw/day [see table section MCA 6.9]. However, the available data on M0310I011 indicate a moderate acute toxicity only [see section 5.8.1 No. 16 above]. The short-term NOAEL determined in the apical endocrine modulation studies was 10 mg/kg bw/day and thus by magnitude above the TTC-threshold without indicating any relevant toxicity.

M310I013/M310I025 based on the information available for M310I011 is by weight of evidence considered to be not genotoxic. The available toxicological database does not provide any concern for M310I025. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is slightly exceeding the relevant TTC-threshold but available toxicity data do not indicate a relevant concern for acute toxicity. In conclusion, M310I013/M310I025 are considered to be **not toxicologically relevant**.

M310I012



M310I012 is a metabolite found in rats and plants (wheat). M310I012 is the glycin-conjugate of M310I013/M310I025 respectively. As discsussed above taking together the knowledge on metabolic pathways in other crops (wheat) live-stock animals and mammalian species [see structure of M310I017 No. 2 above, and data on metabolism of M310I011 No. 16 above] it is likely that the actual position of the hydroxylation group is the para-position.

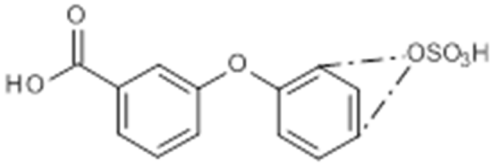
a. Toxicological evaluation of M310I012

Being a rat metabolite M310I012 is intrinsically covered by the toxicological testing of Alpha-cypermethrin. As a glycin-conjugate thereof toxicological evalution of M310I012 is based on the unconjugated analogon M310I013/M310I025. Conjugation of metabolites with amino acids are according to the EFSA Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment [EFSA Journal 2012;10(07):2799] considered to probably not cause higher toxicity of the metabolites.

With regard to chronic consumer exposure the TTC concept for a presumably neurotoxic, non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC-threshold of 0.3 µg/kg bw/day for a presumably neurotoxic, non-genotoxic Cramer-class 3 compound. The estimated acute exposure level is also below the TTC-threshold of 5 µg/kg bw/day.

Based on the information available for M310I011 M310I012 is by weight of evidence is considered to be not genotoxic. The available toxicological database does not provide any concern for M310I026. In addition M310I012 as a rat metabolite is intrinsically covered by the toxicologically testing of alpha-cypermethrin. The estimated chronic exposure is below the TTC-threshold for presumably neurotoxic, non-genotoxic compounds. The estimated acute exposure is also below the relevant TTC-threshold.. In conclusion, M310I012 is considered to be **not toxicologically relevant**.

M310I014



M310I014 is a metabolite found in the rat. M310I014 is the sulate-conjugate of M310I013.

a. Toxicological evaluation of M310I014

Being a rat metabolite M310I014 is intrinsically covered by the toxicological testing of Alpha-cypermethrin.

III. Toxicological evaluation of group D: Phenoxybenzyl alcohol, aldehyde and benzoic acid and conjugates, hydroxylated Phenoxybenzoic acid and conjugates

The group Members of this Group this group include several metabolites identified in mammals and thus also in the rat, namely M310I011 (phenoxybenzoic acid), M310I010 (glycine-conjugate therof), M310I013 (hydroxylated phenoxybenzoic acid), M310I014 (sulfate conjugate) and M310I012 (glycine-conjugate) thereof.

Toxicological data are available for M310I024 (3-phenoxy-benzyl-alcohol), M310I018 (3-phenoxy-benzaldehyd) and M310I011 (3-phenoxybenzoic acid). This database comprises genotoxicity data for all three metabolites considered by weight of evidence not to be genotoxic. The metabolism data reported for M310I011 in several mammalian species including rat and mouse indicate that the hydroxylation in the para-position of the the terminal phenoxyring is the major metabolic route in rats and thus the identified hydroxylation products M310H012, M310I013 and M310I027 are all assumed to follow this pathway (hydroxylation at the para-position) although the final structure could not be defined in all cases.
Moreover the acute toxicity studies available for M310I024 and M310I018 indicate group members to be less toxic than the parent molecule alpha-cypermethrin. A subchronic 90-day rat study conducted with the aldehyde M310I018 indicated the liver and kidney as a target organs characterized by an increased metabolic activity. A NOEL of 50 mg/kg bw/day was determined in this study. In a neurotoxicity study conducted with M310I011 in comparison to cypermethrin effects on the nerval system were investigated. M310I011 is unlikely to be responsible for the enzymatic changes seen in the sciatic/posterior tibial nerve with cypermethrin.
With regard to potential endocrine activity literature data are available on M310I011, M310I24, M310I018. Literature data indicate that M310I011 has no relevant endocrine activity on the estrogenic, androgenic or thyroid system. M310I024 has no relevant activity on the estrogenic or androgenic system. M310I013 and its related structure M310I027 showed no estrogenic or anti-androgenic properties.
Based on this datapackage of relevant group members and the relation to the mammalian metabolism of alpha-cypermethrin **the metabolites of the group D are by weight of evidence not considered to be genotoxic or neurotoxic, do not bear a relevant endocrine activity and are not considered to be toxicological relevant.**

3-Hydroxybenzoic acid

I. Definition of group E: 3-Hydroxybenzoic acid

The "group E" comprises of a single molecule M310I019 i.e. 3-hydroxybenzoic acid

M310I019 (3-Hydroxybenzoic acid)

Evaluation of group E members

M310I019 (other denominators: CAS-No. 99-06-9, EC-No. 202-726-5, m-Hydroxy benzoic acid, 3-Hydroxy benzoic acid, FL-no 08.132)
M310I019 is identified as a food flavor. As such it was evaluated by EFSA [EFSA Journal 2012; 10(12):2994]. M310I019 is reported to be a known metabolite in microbial metabolism. Some investigations on pharmacological and nutritional benefitial effects are reported for M310I019 as a metabolite.

a. Toxicological data on M310I019

The EPA ACToR data-warehouse [http://actor.epa.gov/actor/GenericChemical?casrn=99-06-9] reports based on the information of NLM TOXNET Toxicology an acute oral LD_{50} of 2000 mg/kg bw in mice [reference: Quarterly Journal of Pharmacy & Pharmacology. Vol. 19, Pg. 483, 1946]. For rats an LD_{50} of 3700 mg/kg is reported [reference: Bollettino Chimico Farmaceutico. Vol. 112, Pg. 53, 1973]. In the UMD list of Acute Toxins, Teratogens, Carcinogens or Mutagens it is reported that M310I019 meets the University of Maryland definition of a Teratogen for the purpose of the Chemical Hygiene Plan as it was listed as a teratogen in the "Dangerous Properties of Industrial Materials", 7th Ed., by N. Irving Sax and Richard J. Lewis, however the underlying data could not be identified. There is some additional limited evidence published that M310I019 is not teratogenic [see CA 5.8.1/42 1972/1000521 below].

M310I019 was evaluated by EFSA as a food flavour [EFSA Journal 2012; 10(12):2994]. The group evaluation 20 comprised benzyl alcohols, benzaldehydes, a related acetal, benzoic acids, and related esters from chemical groups 23 and 30. M310I019 was assigned to the subgroup 2 (hydroxy- and alkoxy-ringsubstituted benzyl derivatives). M310I019 was assigned to structural Cramer class I and a threshold of concern (based on the TTC concept) of 1800 µg/person/day i.e. 30 µg/kg bw/day was derived. With regard to M310I019 no specific toxicological data are reported instead M310I019 was evaluated based on the grouping approach. Toxicological evaluation by EFSA based on grouping approach: By weight of evidence no safety concern with respect to genotoxicity was identified. Acute toxicity, repeated dose toxicity data and developmental / reproductive toxicity data of group candidate and supporting substances were consistent with the group evaluation and with the conclusion based on the TTC concept.

Some on pharmacological and nutritional benefitial effects are reported for M310I019 as a metabolite.

Report: CA 5.8.1/42
[redacted] 1972b
Biochemical mechanisms of salicylate teratology in the rat
1972/1000521
Guidelines: none
GLP: no

In this non-guideline prenatal toxicity study with limited exposure and limited parameters investigated M310I019 (m-Hydroxybenzoic acid) was investigated in comparison to salicylates and their other presumed derivatives. Groups of adult female Spreague-Dawley rats, aged 100 days at weaning, weighing 190 – 220 g were used. The animals were fed with Purina laboratory chow and water ad libitum. They were paired overnight with male rats of the same strain. 10 female animals were treated subcutaneously with m-Hydroxybenzoic acid at a dose equivalent to 380 mg/kg acetyl-salicylic acid on day 9 of gestation. The concurrent control of 15 female rats was treated with the vehicle i.e. deionized water. The source and purity of the administered compounds is not reported in the publication but indicated to be referenced in the underlying author's dissertation. The dose was administered in two equal portions with 2 hour interval. Animals were housed until day 20 of gestation, when they were sacrificed and the fetuses were removed. Uterus was inspected for resorptions and fetal death and external congenital malformations were reported. The mean fetus weight, the number of alive fetuses and the number and percentage of resorptions were in the same range as the control [see Table 5.8.1-32 below]. In contrast to the in parallel investigated acetyl-salicylic acid and salicylic acid no external malformations were reported for the fetuses from dams treated with M310I019.

Table 5.8.1-32: Results of non-guideline teratogenicity study with M310I019

Treatment group	No of dams	Dam mortality	Implan-tations	Mean fetus weight	No. of fetuses (alive)	Resorptions		Malformed fetus	
			Total No.	(g)	Total No.	Total No.	% of total implan-tations	Total No.	% of total living fetuses
Control	15	0	172	3.89 ± 0.44	169	3	1.7	0	0
Acetyl-Salicylic acid	10	0	114	**3.02 ± 0.70****	**93**	**21**	**18.4**	0	0
Salicylic acid	17	**1**	178	**3.09 ± 0.93****	**95**	**83**	**46.6**	0	0
M310I019	10	0	123	3.96 ± 0.43	120	3	2.4	0	0

** significantly different from control $p < 0.01$
Bold figures indicate treatment related changes

The study is considered to be of limited value due to significant differences from guideline teratogenicity studies. In particular the duration of exposure is insufficient and the parameters investigated are limited.

Classification of study: supplemental information

Information with regard to the endocrine potential of M310I019 was found in the open literature. A summary table of the available literature is provided in Table 5.8.1-33 below. The literature is however described in more detail in the chapter CA 5.8.3.

In vitro studies:

Considering all available data on endocrine potential and on aromatase inhibition, M310I019 is considered to display neither estrogenic activity in vitro nor inhibitory effects on aromatase activity in vitro.

Table 5.8.1-33: Endocrine activity studies with M310I019

Endpoint	Test system with endpoint	Endocrine activity	Positive control	Reference Doc ID
In vitro assays				
E-screen	MCF-7 cells (proliferation)	ER-agonist: Not active ER-antagonist: Not determined	Agonist: E2: EC50: 12E-3 M Antagonist:4HO-TAM: IC_{50}: 2.8±0.7E-6 M	van Meeuwen et al., 2008 2008/1102356
Aromatase inhibition	in vitro microsomal conversion of 1-β-^{3}H-androstenedion	Aromatase inhibition: Not active	4-OH-androstendion IC_{100}: 1E-6 M	van Meeuwen et al., 2008 2008/1102356

b. Toxicological evaluation of M310I019

M310I019 was evaluated by EFSA in the context of food-flavour ingredients [see Scientific Opinion on flavouring group evaluation, Revision 4: Benzylalcohols, benzaldehydes, a related acetal, benzoic acids, and related esters from chemical groups 23 and 30, EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (CEF); http://www.efsa.europa.eu/de/efsajournal/doc/2994.pdf]. Evaluated as a Cramer-class 1 compound a threshold fo concern of 1800 µg/person/day was defined for the so-called FL-no. 08.132 based on a grouping approach. The available toxicological information for M310I019 gives no indication of a toxicological concern.

With regard to consumer exposure the TTC concept for a non-genotoxic compound was applied. The estimated exposure levels - back-calculated from worst-case exposure levels of alpha-cypermethrin for operators and consumers based on 100 % utilization of the reference value 0.02 mg/kg bw – would be below the TTC for a of 0.3 µg/kg bw/day for a presumably neurotoxic , non-genotoxic Cramer-class 3 compound and thus of course below the much higher threshold defined by EFSA for M310I019 being a Cramer-class 1 compound evaluated as a food flavor.
In conclusion M310I019 is considered to be **not toxicologically relevant**.

Toxicological evaluation of group E: 3-Hydroxybenzoic acid

Based on the grouping approach evaluated by EFSA as a food flavour **3-hydroxybenzoic acid is by weight of evidence not considered to be genotoxic or neurotoxic, does not bear a relevant endocrine activity and is not considered to be more toxic than the parent molecule.**

CA 5.8.2 Supplementary studies on the active substance or related substances

Studies evaluated in the draft monograph of the Rapporteur Member State Belgium September 1999: A mechanistic neurotoxicity study with oral administration in rats was presented in the first DAR as supplementary study but was used in the list of Endpoints under point Neurotoxicity/Delayed neurotoxicity and is therefore shifted into chapter CA 5.7. Furthermore literature data were discussed within the ECCO Peer review meeting - Full Report on alpha-cypermethrin [see KCA 5.8.2/1 AL-901-031] to conclude that no further studies on developmental neurotoxicity are necessary. These data are also shifted for a better understanding into chapter CA 5.7, too.

The endpoint "Other toxicological studies" during the last Annex I listing of Alpha-Cypermethrin was therefore as follows:

Other toxicological studies	No data- not required

Studies submitted in this AIR 3 dossier (Part I: peer-reviewed studies from other DARs):
In the DAR (2008) of zeta-cypermetrhin two crucial studies are presented that demonstrate the continuous exposure of pups via milk. One study is focusing on placental and lactational transfer, the other one is focusing on lactational transfer of dietary administered zeta-cypermethrin. Based on the similarity of properties a lactational transfer is thereby proven for alpha-cypermethrin, too. Based on these data an estimation of pup exposure via milk is calculated in CA 5.7 in order to demonstrate that lactational exposure of pups is at a sufficient degree to be used in human risk assessment.

Table 5.8.2-1: Summary of already peer-reviewed supplementary studies

Study	Test substance information	Endpoints			Reference
		NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Effect	
Dietary placental and lactation transfer, rats, 50, 125, 300 ppm GD 6-20 or GD6-LD21	Zeta-cypermethrin: batch No. PL03-0427; 81.8%			Zeta-cypermethrin was detected in maternal and fetal plasma	Copied from DAR zeta-cypermethrin (2006) : see CA 5.8.2/5 Doc ID 2006/7013990 [redacted]2005 a
Dietary exposure from GD 6 to LD17, rats, 125, 375 ppm and transfer in milk	Zeta-cypermethrin: batch No. PL03-0427; 81.8%			Dietary administration permits transfer from food to milk	Copied from DAR zeta-cypermethrin (2006): see CA 5.8.2/5 Doc ID 2006/7013990 [redacted] 2004 and amended by the EFSA conclusion with regard to R64, which was considered not relevant

Studies submitted in this AIR 3 dossier (Part II: not peer-reviewed studies):
Upon request of US EPA, an immunotoxicity study in male rats was conducted investigating the primary T-cell dependent IgM antibody response as assessed by the anti-Sheep red blood cell (SRBC) IgM ELISA and additionally by weight changes of spleen and thymus. The respective study is listed in Table 5.8.2-2. The immunotoxicity study did not reveal a specific effect on the T-cell dependent immune response or on immune-system related organs. Additionally, a 14-day range finding study was performed to set the dose levels for the subsequent immunotoxicity studies. This study is not listed in Table 5.8.2-2as the results are not end-point relevant.

For submission in Japan a general pharmacology study has been conducted investigating the effect of alpha-cypermethrin on vital functions of animals of different species. The respective study is listed in Table 5.8.2-2. The study showed the typical acute toxicity via action on the CNS and the cardiovascular system at sublethal doses. Furthermore a study to investigate the potential use of methocarbamol, mephenesin, or halothane as antidots on acute toxicity in rats has been performed. These studies are not mandatory in the EU and are therefore only summarized roughly.

In addition, literature data were taken into account. However, these data are judged to represent supplemental information only and do not provide relevant endpoints for human riskassessment.

Table 5.8.2-2: Summary of newly submitted not peer reviewed supplementary studies

Study	Test substance information	Endpoints			Reference
		NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Effect	
28day Immunotoxicity study, Rat (Wistar, ♂), Diet 0-50-150-450 ppm 0-4-12-34 mg/kg bw	alpha-cypermethrin, batch COD 000595, purity: 99.2%	34 (systemic) 34 (immunotox)	> 34	No effect	Doc ID 2010/1057357 see CA 5.8.2/2
General pharmacological study Rat (SD, ♂), Mice (ICR, ♂&♀) Guinea Pigs (Hartley, ♂) Rabbits(Kbs:JW, ♂)	alpha-cypermethrin,batch 351, purity 93.2% Aqueous solution, DMSO			alpha-cypermethrin induces acute toxicity via action on CNS and Cardiovascular system at sublethal doses	Doc ID AL-452-001 see CA 5.8.2/3
Effects of antidotes on acute toxicity Rat (SD), Gavage (corn oil)	alpha-cypermethrin125 mg/kg bw 351, purity 93.2%			Neither methocarbamol, mephenesin, nor halothane are effective antidots	Doc ID AL-452-002 see CA 5.8.2/4

Literature data			
Study	**Test substance information**	**Reported effects**	**Reference**
Induction of CYP-mRNA Luciferase-Assay for PXR and CAR activation N-in-one cell culture assay for CYP activity	Among others: alpha-cypermethrin (purity: not given) cypermethrin (purity: not given) 10, 50 µM	alpha-cypermethrin and cypermethrin show the same enzyme induction pattern, induced both CAR and PXR to the same extent. CYP activity was only screened.	Abass et al., 2012 2012/1367022 see CA 5.8.2/6
CYP inhibition was determined in the N-in-one cell culture assay and in the single substrate assay	Among others: alpha-cypermethrin (purity: not given) cypermethrin (purity: not given) 1, 5, 25, 50, 100 µM	Both alpha-cypermethrin and cypermethrin show IC_{50} values in a similar higher µM concentration range	Abass et al., 2013 2013/1416883 see CA 5.8.2/7
Direct pup dosing from PND 6 or 8-15 Rat (Wistar, ♂), 0-1.49 mg/kg bw Gavage in corn oil (5 ml/kg)	cypermethrin (cis/trans:63/37; purity: 92.4%)	At 1.49 mg/kg bw no clinical effects and no BW effects were induced Questionable results on: ↑MA(PND35) ↓DOPA ↑HVA Oxidative changes in striatum and erythrocytes, GPx and GSH level	Nasuti et al., 2007 2007/1070386 see CA 5.8.2/8
In vitro cell differentiation of neuronal and glial cells lines	Among others: Alpha-cypermethrin or cypermethrin (purity: not given) 1 and 10 µM	No effect on neurite outgrowth	Flaskos et al., 2007 2007/1070387 see CA 5.8.2/10
In vitro impairment of the transient outward potassium current (I_A)	alpha-cypermethrin (purity: no data) theta-cypermethrin (purity: no data) 1, 10, and 100 nM	Both alpha-and theta-cypermethrin modulate I_A, but show differences in the modulation	Tian et al., 2008 2008/1102217 see CA 5.8.2/11
In vitro impairment of the delayed rectifier potassium current (I_K)	alpha-cypermethrin (purity: no data) theta-cypermethrin (purity: no data) 1, 10, and 100 nM	Both alpha-and theta-cypermethrin modulate I_K in a comparable way	Tian et al., 2009 2009/1130985 see CA 5.8.2/12
Motor function in male Long-Evans rats (age: 55-57 days)	Cypermethrin (Cis/trans: 48.7/51.3; purity: 88%); 0.1– 120 mg/kg bw; gavage in corn oil (1 mL/kg)	ED30 of 10.7±1.34 mg/kg: Threshold level of 4.3±1.14 mg/kg bw	Wolansky et al., 2006 2006/1051134 see CA 5.8.2/13
Hydrolytic and oxidative metabolism in rat and human hepatic microsomes	Among others: cypermethrin (cis/trans ratio 49/51, purity: > 98%) 0.1, 1, 5, 10, 20, or 50 µM	Cypermethrin metabolism in hepatic microsomes: Rat 85% oxidative vs. 15% hydrolytic metabolism Human 100 % hydrolytic metabolism	Scollon et al., 2009 2009/1130987 see CA 5.8.2/14
Esterase distribution in small intestine, liver and serum Metabolic degradation of alpha-cypermethrin via rat serum CE	Among others: Alpha-cypermethrin (99% pure, mixture of isomers)	Humans express esterase activity in small intestine (hCE2) and in the liver (hCE1 & 2) but not in blood serum Rats express esterase activity in all three tissues. CE activity for selected pyrethroids showed lower activity in rats than in humans. Alpha-cypermethrin was poorly metabolized by rat serum CE	Crow et al., 2007 2007/1070525 see CA 5.8.2/15

Thus, the conclusion for relevant endpoints for the current re-registration is drawn as follows:

Other toxicological studies (SANCO/11802 data point 5.8)

Supplementary studies on the active substance	**Immunotoxicity** No evidence for specific immunotoxicity No classification required
	Placental and lactational transfer demonstrated for alpha-cypermethrin via zeta-cypermethrin No classification required

Report: CA 5.8.2/2
[redacted] 2010a
BAS 310 I (Alpha-Cypermethrin) - Immunotoxicity study in male Wistar rats - Administration via the diet for 4 weeks
2010/1057357
Guidelines: EPA 870.7800
GLP: yes
(certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

BAS 310 I (batch COD – 000595, purity 99.2%) was administered via the diet to male Wistar rats at dietary dose levels of 0, 50, 150 and 450 ppm, respective 0, 3.99, 11.83 and 34.34 mg/kg bw/day for 4 weeks. Cyclophosphamide monohydrate (4.5 mg/kg bw/day) was used as a positive control. SRBC IgM antibody titers were similar between control and BAS 310 I treated animals. No immunopathological findings as well as general systemic toxicity findings were observed after treatment of animals with BAS 310 I.
The administration of the positive control Cyclophosphamide monohydrate (4.5 mg/kg bw/d) led to effects indicative of immunotoxicity, i.e. reduced absolute and relative spleen and thymus weight. Furthermore, SRBC IgM antibody titers were significantly lower compared to the control group. In addition, an impaired body weight development was observed after treatment of animals with cyclophosphamide.

Based on the results of the study the NOAEL was identified at 450 ppm (34 mg/kg bw/day) for general toxicity as well as for immunotoxicity for BAS 310 I.

(DocID 2010/1057357)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: BAS 310 I
Description: solid, white
Lot/Batch #: COD – 000595
Purity: 99.2%
Stability of test compound: The test substance was stable over the study period (Expiry date 01.06.2013).

2. Positive control: Cyclophosphamide Monohydrate (CAS No.: 6055-19-2)
Description: solid, white
Lot/Batch #: 1362353
Purity: 100%
Stability of test compound: The test substance was stable over the study period (Expiry date 08.10.2010).

3. Test animals:

Species:	Rat
Strain:	Crl:WI (Han)
Sex:	Male
Age:	36 ± 1 days at delivery; 42 ± 1 at the beginning of the administration period
Weight at dosing:	168.64 – 171.01 g
Source:	Charles River, Sulzfeld, Germany
Acclimation period:	6 days
Diet:	Kliba maintenance diet rat/mouse Provimi Kliba SA, Kaiseraugst, Switzerland, ad libitum
Water:	Tap water ad libitum
Housing:	Group housing (4 animals per cage) in H-Temp (PSU, TECNIPLAST, Hohenpeißenberg, Germany) cages; floor area about 2065 cm².
Environmental conditions:	
Temperature:	20 - 24°C
Humidity:	30 - 70%
Air changes:	Fully air-conditioned rooms; 15 of air-changes per hour
Photo period:	12 h light / 12 h dark (06:00 - 18:00 / 18:00 - 06:00)

B. STUDY DESIGN AND METHODS

The study was conducted at BASF SE, Experimental Toxicology and Ecology, 67056 Ludwigshafen, Germany.

1. In life dates: 09-Feb-2010 - 16-Mar-2010)
(Dates of experimental work:09-Feb-2010 to 07-Sep-2010)

2. Animal assignment and treatment:
BAS 310 I was administered to groups of 8 male rats at dietary concentrations of 0, 50, 150, and 450 ppm for 4 weeks via the diet. Cyclophosphamide monohydrate (4.5 mg/kg bw/d) was administered by gavage as a solution in drinking water.
All animals were immunized 6 days before blood sampling and necropsy using 0.5 ml sheep red blood cells ($4x10^8$ SRBC/ml) administered intraperitoneally. The animals were assigned to the treatment groups by means of a computer generated randomization lists based on body weights.

3. Test substance preparation and analysis:
Previous tests indicated that BAS 310 I was stable in the diet at room temperature for at least 30 days. The stability of Cyclophosphamide Monohydrate (positive control substance) in the vehicle (drinking water) over a period of 32 days (in the freezer) was proven before the start of the study

Homogeneity analyses were performed at all concentrations at the start of the study (see Table 5.8.2-3). Samples also served for concentration control. From a strict chemical viewpoint, a homogeneity assay was not necessary because the compound is completely soluble in water at the concentration being dosed.

Table 5.8.2-3: Analysis of diet preparations for homogeneity and test-item content

Dose level [ppm]	Sampling	Concentration [ppm] Mean ± SD	Relative standard deviation [%]	Mean / of nominal concentration
50 ppm	23.01.2011	50.2 ± 2.3	4.6	100.4
150 ppm	23.01.2011	159.1 ± 5.8	3.6	106.1
450 ppm	23.01.2011	433.7 ± 14.1	3.2	96.4

Relative standard deviations of the homogeneity samples in the range of 3.2 to 4.6% indicate the homogenous distribution of BAS 310 I in the diet preparations. The actual test-item concentrations were in the range of 96.4 to 106.1% of the nominal concentrations.
For Cyclophosphamide monohydrate the 98.4 % of the nominal concentration was achieved.

4. Statistics:
Means and standard deviations of each test group were calculated for several parameters. Further statistical analyses were performed according to following tables:

Statistics of clinical examinations

Parameter	Statistical test
Body weight, body weight gains (control and test substance treatment)	A comparison of each group with the control group was performed using DUNNETT's test (two-sided) for the hypothesis of equal means
Body weight, body weight gains (control and Cyclophosphamide monohydrate treatment)	A comparison of the dose group with the control group was performed using the t-test (two-sided) for the hypothesis of equal means

Statistics of clinical pathology

Parameter	Statistical test
Clinical pathology parameters	Non-parametric one-way analysis using KRUSKAL-WALLIS test (two-sided). If the resulting p-value was equal or less than 0.05, a pair-wise comparison of each dose group with the control group was performed using Wilcoxon-test (two-sided) for the equal medians

Statistics of pathology

Parameter	Statistical test
Organ weights	Non-parametric one-way analysis using KRUSKALWALLIS test (two-sided). If the resulting p-value was equal or less than 0.05, a pairwise comparison of each dose group with the control group was performed using the WILCOXON test for the hypothesis of equal medians

C. METHODS

1. Observations:
The animals were examined for evident signs of toxicity or mortality twice daily on working days and once daily on weekends and public holidays.

The clinical condition of the test animals were recorded individually.
Detailed clinical observations of all animals were performed in a standard arena prior to the administration period and weekly thereafter. The standard arena had a size of 50 x 37.5 cm with walls of 25 cm height. If applicable the findings were ranked according to the degree of severity. The following parameters were examined:

1. abnormal behavior during handling
2. fur
3. skin
4. posture
5. salivation
6. respiration
7. activity/arousal level
8. tremors
9. convulsions
10. abnormal movements
11. impairment of gait
12. lacrimation
13. palpebral closure
14. exophthalmus
15. feces (appearance/consistency)
16. urine
17. pupil size

2. Body weight:
The body weight of the animals was determined before the start of the administration period in order to randomize the animals, at the start of the exposure period and thereafter twice weekly. The difference between the body weight on the respective day of weighing and the body weight on day 0 was calculated as body weight change.

3. Food consumption, food efficiency and compound intake:
Group food consumption was determined weekly (as representative value over 1 day) for each cage. The average food consumption was used to estimate the mean food consumption in grams per animal and day.
Water consumption was observed daily by visual inspection of the water bottles for any overt changes in volume.

The mean daily intake of test substance (group means) was calculated based upon individual values for body weight and food consumption.

$$\text{Substance intake for day x} = \frac{FC_x \times C}{BW_x}$$

BW_x = body weight on study day x [g]; FC_x= mean daily food consumption on study day x [g]; C = concentration in the food on study day x [mg/day]

4. Immuntoxicological examinations:
Primary T-cell dependent antibody response (anti-SRBC IgM ELISA) was performed on two dilutions (1:64 and 1:128)

4. Sacrifice and pathology:
All animals were sacrificed by decapitation under isoflurane anesthesia. The exsanguinated animals were necropsied and assessed by gross pathology. The following organs were sampled, weighed and examined histopathologically:

Pathology:
The following organs were collected (column C), weighed (W) and examined histopathologically (H, ✓: all groups, #: control and top dose).

C	W	H		C	W	H		C	W	H	
			Adrenals				lachrymal glands				seminal vesicles
			Aorta				larynx				skin
			Brain				liver				spinal cord (3 levels)[@]
			bone marrow[§]				lung	✓	✓		spleen
			Caecum				lymph nodes[#]				sternum w. marrow
			Colon				mammary gland (♀)				stomach (fore- & glandular-)
			Duodenum				muscle, skeletal				testes
			Epididymides				nerve, peripheral (sciatic n.)	✓	✓		thymus
			Esophagus				nose/nasal cavity[$]				thyroid/parathyroid
			Eyes				ovaries and oviduct				trachea
			femur (with joint)				pancreas				urinary bladder
			gall bladder				Payer's patches				uterus
✓			gross lesions				pituitary				vagina
			Heart				pharynx				
			Ileum				prostate		✓		body (anesthethized)
			Jejunum				rectum				
			Kidneys				salivary glands*				

[§] from femur; [#] mandibular and mesenteric; [@] cervical, thoracic, lumbar); *submandibular and sublingual, [$] histopath at level III;

II. RESULTS AND DISCUSSION

A. OBSERVATIONS

1. Clinical signs of toxicity

No test substance-related clinical signs were observed in all animals treated with BAS 310 I and in animals which received Cyclophosphamide monohydrate as positive control.

2. Mortality

No mortality was observed throughout the study.

B. BODY WEIGHT AND BODY WEIGHT GAIN

No significant changes were observed in all animals tested with BAS 310 I. During the administration period the body weight change of the test group 4 (positive control) was significantly lower from study day 7 to study day 28 (maximum of 13% less on study day 24) [see Figure 5.8.2-1:and Table 5.8.2-4:].

Figure 5.8.2-1: Body weight and body weight development of male rats administered BAS 310 I for 4 weeks

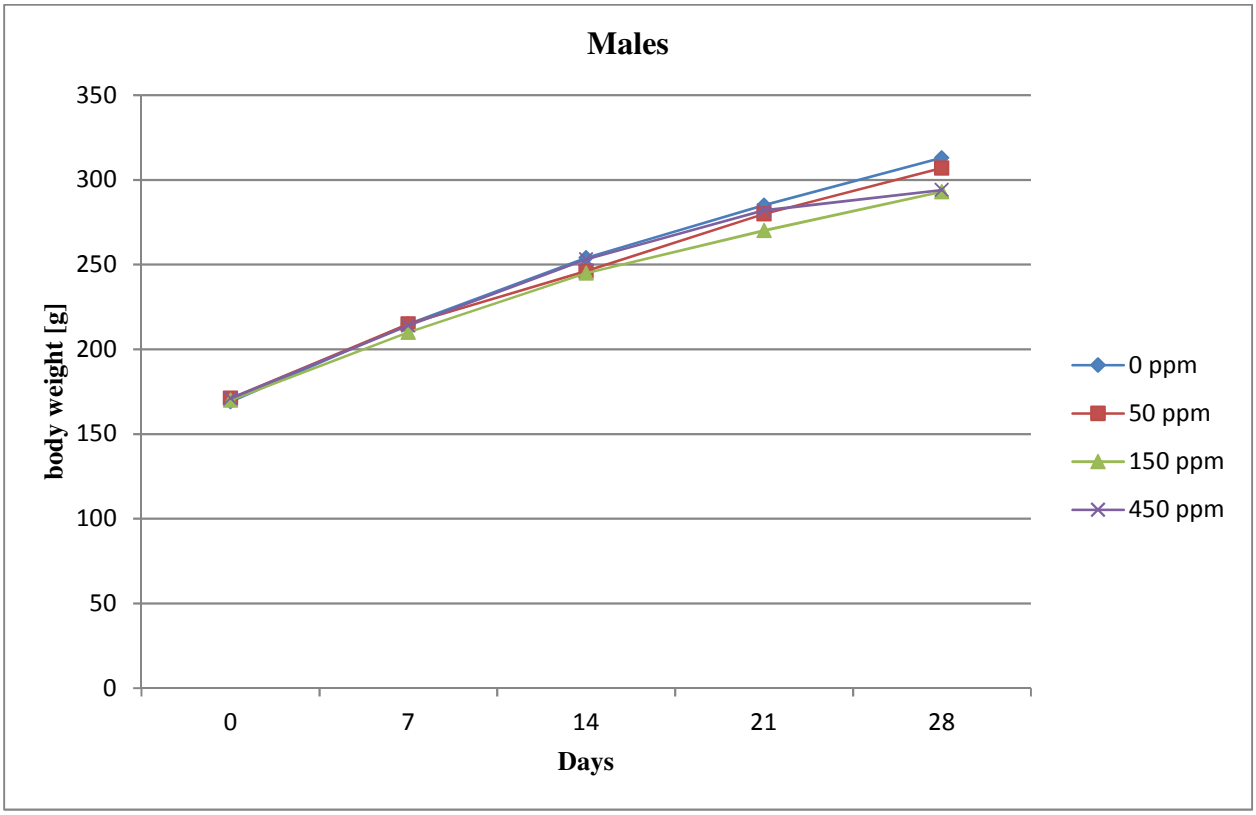


Table 5.8.2-4: Body weight and food consumption data of rats administered BAS 310 I for 4 weeks

	Males				
Dose level [ppm]	0	50	150	450	Positive control
Body weight [g]					
- Day 0	168.64	171.01	169.67	170.91	170.02
- Day 28	312.65	307.19	292.70	304.84	296.38
% (compared to control)		-.175	-6.38	-2.50	-5.21
Overall body weight gain [g]	144.01	136.18	123.03	133.92	126.35*
% (compared to control)		-5.44	-14.57	-7.00	-12.26
Food consumption [g/day]					
- average week 1-4[§]	21.21	20.52	19.77	19.74	20.10
% (compared to control)		96.77	93.20	93.06	94.77

* $p \leq 0.05$ (Dunnett's test, two sided); [$] calculated from the weekly means

C. FOOD CONSUMPTION AND COMPOUND INTAKE

No effects on food consumption and food efficiency were observed.
The mean daily test substance intake was 3.99, 11.83, and 34.34 mg/kg bw/day at dietary dose levels of 50, 150 and 450 ppm, respectively.

D. BLOOD ANALYSIS

1. Primary T-cell dependent antibody response (anti-SRBC IgM ELISA)
Six days after immunization no difference between control and substance treated animals was observed. SRBC titers were significantly lower in animals treated with Cyclophosphamide monohydrate [see Table 5.8.2-5].

Table 5.8.2-5: Blood analysis findings of rats administered BAS 310 I for 4 weeks

Sex	Females								
Dose [ppm]	Values					% (compared to control)			
	0	50	150	450	Pos. control	50	150	450	Pos. control
SRBC [LU/l]	4.14	3.15	4.31	4.52	0.84*	76.09	104.10	109.18	20.29*

* $p \leq 0.05$ (Kruskal-Wallis and Wilcoxon-test, two sided)

E. NECROPSY

1. Organ weight

Terminal body weights (absolute and relative) were not significantly affected by treatment with BAS 310 I or Cyclophosphamide [see Table 5.8.2-6].

Mean absolute and relative weights of organs were comparable between control and BAS 310 I treated animals. Cyclophosphamide treated animals revealed a significant decrease of spleen and thymus weights.

Table 5.8.2-6: Terminal body and organ weights of rats administered BAS 310 I for 4 weeks

Sex	Females								
Dose [ppm]	Values					% (compared to control)			
	0	50	150	450	Pos. control	50	150	450	Pos. control
Terminal body weight [g]	289.7	284.8	269.9	285.1	274.8	98.31	93.17	98.41	94.86
Spleen abs. [g]	0.603	0.590	0.513	0.556	0.384**	97.84	85.07	92.21	63.68**
rel. [%]	0.208	0.207	0.190	0.195	0.140**	99.52	91.79	102.63	71.79**
Thymus abs. [mg]	504.4	460.6	401.9	456.9	265.1**	91.32	87.26	113.68	58.02**
rel. [%]	0.174	0.162	0.147	0.158	0.096**	93.10	90.74	107.48	60.76**

** $p \leq 0.01$; Wilcoxon test, two sided

2. Gross and histopathology

No gross lesions were observed. In the absence of gross lesions no histopathological investigations were carried out.

III. CONCLUSIONS

Under the conditions of the study BAS 310 I did not reveal any signs of immunotoxicity when administered via the diet over a period of 4 weeks to male Wistar rats. The NOAEL for the immunotoxicologically relevant endpoints was set to 450 ppm (34 mg/kg bw/day), the highest dose tested. The oral administration of the positive control substance cyclophosphamide monohydrate (4.5 mg/kg bw/day) led to severe findings indicative of immunotoxicity. This was represented by significantly lower SRBC IgM antibody titres as well as reduced spleen and thymus weights. Thus, assay sensitivity was verified in the present immunotoxicity study performed in male Wistar rats.

Report: CA 5.8.2/3
[redacted] 2000a
Alphacypermethrin: General pharmacological study
AL-452-001
Guidelines: JMAFF 59 NohSan No 4200
GLP: yes
(certified by Ministry of Agriculture, Forestry and Fisheries of Japan, Japan)

Executive Summary

Alphacypermethrin (batch 351, purity 93.2%) was tested in a general pharmacological study in mice, rats, guinea pigs and rabbits. Clinical signs of toxicology, effects on the autonomic and central nervous system, effects on respiration and circulation, effects on digestive organs, effects on skeletal muscles and renal function were observed during the study.
Excitatory toxic signs typical for cyano pyrethroids were observed at 40 mg/kg or higher in mice and rats. Intravenous injection of the test substance to male rabbits at 12 mg/kg elicited cardiorespiratory changes and death. Oral administration at sub-lethal doses produced a decrease in transport activity in the small intestine in mice, and a decrease in body weight and temperature as well as an increase in urinary glucose. The present in vitro experiments revealed an increase in both muscle and nerve stimulated contraction of the diaphragm of rats and an increase in spontaneous motility of the ileum, and a decrease in agonist-induced contraction of the organ in the guinea pig. No distinct effects on hexobarbital sleeping time in male mice or on grip strength in male rats were observed.

(DocID 2000/7000975)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alphacypermethrin
Description: solid (crystalline), yellowish white
Lot/Batch #: 351
Purity: 93.2%

2. Test animals:
Species: Mice
Strain: ICR
Sex: Male and female
Age: 6 weeks (at arrival)
Source: Atsugi Breeding Center of Charles River Japan Inc., Kanagawa
Acclimation period: at least 1 week

Diet:	Certified diet MF, Oriental Yeast Co., Ltd., Tokyo, ad libitum
Water:	Tap water (with sodium hypochlorite, UV exposed), ad libitum
Housing:	Group housing (3 mice/sex and dose for ckinical observations; 8 male mice/dose for the other tests) in stainless steel cages
Species:	Rats
Strain:	Sprague-Dawley
Sex:	Male
Age:	6 weeks (at arrival)
Source:	Tsukuba Breeding Center of Charles River Japan Inc., Ibaraki
Acclimation period:	at least 1 week
Diet:	Certified diet MF, Oriental Yeast Co., Ltd., Tokyo, ad libitum
Water:	Tap water (with sodium hypochlorite, UV exposed), ad libitum
Housing:	Group housing (5 rats) in stainless steel cages
Species:	Guinea pig
Strain:	Hartley
Sex:	Male
Age:	6 weeks (at arrival)
Source:	Tsukuba Breeding Center of Charles River Japan Inc., Ibaraki
Acclimation period:	at least 1 week
Diet:	Certified diet LRC4, Oriental Yeast Co., Ltd., Tokyo, ad libitum
Water:	Tap water (with sodium hypochlorite, UV exposed), ad libitum
Housing:	Group housing (5 guinea pigs) in aluminum cages
Species:	Rabbits
Strain:	Kbs:JW
Sex:	Male
Age:	7-8 weeks (at arrival)
Weight at dosing:	1.52 – 1.84 kg
Source:	Kitayama Labes Co., Ltd., Nagano
Acclimation period:	at least 1 week
Diet:	Certified diet LRC4, Oriental Yeast Co., Ltd., Tokyo, ad libitum
Water:	Tap water (with sodium hypochlorite, UV exposed), ad libitum
Housing:	Individually in aluminum cages

Environmental conditions:	
Temperature:	22°C
Humidity:	55%
Air changes:	>10 air-changes per hour
Photo period:	12 h light / 12 h dark (07:00 - 19:00 / 19:00 - 07:00)

B. STUDY DESIGN AND METHODS

The study was conducted at the Institute of Environmental Toxicology, Ibaraki, Japan.

1. In life dates: 17-Aug-1999 - 27-Sep-1999)
(Dates of experimental work: 30-Aug-1999 to 16-Mar-2000)

2. Animal assignment and treatment:
Mice were treated with 2.6, 6.4, 16, 40, 100, and 250 mg/kg for assessment of general toxicity. For hexobarbital sleeping time measurements male mice were treated with 2.6, 6.4, 16, 40, and 100 mg/kg bw.
Transport activity of the small intestine in male mice was determined after treatment with 1.0, 2.6, 6.4, 16, 40, and 100 mg/kg.

Rats were treated with 16, 40, 100, 250, and 625 mg/kg test substance.
For assessment of cardiorespiratory parameters male rabbits were treated with 0.75, 3, and 12 mg/kg.
The solution containing the test substance was administered in a volume of 10 mL/kg to mice and rats, and 1 mL/kg to rabbits.

For assessment of agonist-induced contractions of the ileum in male guinea pigs the test substance was applied at concentrations of 0.1, 1, 10, and 100 µg/mL.

For assessment of electrical stimulation-induced contraction of diaphragm in male rats the test substance was applied at concentrations of 0.1, 1, 10, and 100 µg/mL.

In the in vitro experiments the maximum concentration emulsible in Krebs-Ringer solution containing Tween 80 was 100 µg/mL.

3. Test substance preparation:
Since the test substance is insoluble in water it was suspended in 1% Tween 80 aqueous solution for oral application to mice and rats. For intravenous injection to rabbits the test substance was dissolved in dimethylsulfoxide (DMSO, 15% final concentration) and then emulsified by mixing with physiological saline containing Tween 80 (5%). The suspension and emulsion were prepared immediately before each experiment.

4. Statistics:
The data were statistically analyzed by Dunnett´s or Steel´s multiple comparison test. Differences between groups were considered to be statistically significant at the $p \leq 0.05$ level.

C. METHODS

1. Effects on central nervous system:
Clinical signs of male and female mice were observed immediately before and 1 h, 3h, 6 h, 1 day, 2 days, 3 days, and 7 days after oral administration of the test substance (3 animals/dose).
Hexobarbital sleeping time in male mice was studied 1 h after oral administration (8 animals/dose). The hypnosis was induced by subcutaneous injection of hexobarbital (100 mg/kg) 1 h after the administration of the test substance. The time between disappearance and reappearance of righting reflex was measured.
Clinical signs of toxicity and body temperature in male rats were observed 1 day before and 1 h, 3h, 6 h, 1 day, 2 days, 3 days, and 7 days after oral administration of the test substance (5 animals/dose).

2. Effects on respiration and circulation
Cardiorespiratory function in male rabbits was evaluated after intravenous injection at cumulative doses of 0.75, 3, and 13 mg/kg every one hour (4 animals/dose). For assessment of respiratory function a thermistor type respiration sensor was fixed to a cannula inserted into a cut opening of the trachea. Relative change of respiratory pattern was recorded with 0.3 s of time constant. Blood pressure was measured using a polyethylene tubing, which was inserted into the femoral artery about 10 cm with the opposite of the tubing being connected to a high pressure transducer. Values of systolic, diastolic, and mean blood pressure were measured with a digital meter. The electrodes for recording the ECG were fixed on a right forelimb and right and left hindlimbs. Heart rate was measured with a digital counter.

3. Effects on autonomic nervous system
Pupil size in male rats was observed 1 day before and 1 h, 3h, 6 h, 1 day, 2 days, 3 days, and 7 days after oral administration of the test substance (5 animals/dose).
The effects on the phrenic nerve-diaphragm of male rats was evaluated in vitro by applying the test substance at concentrations of 0.1, 1, 10, and 100 µg/mL (4 animals/dose). Four preparations of the isolated phrenic nerve diaphragm were used to study the effects of the test substance on the muscle contraction induced by direct or indirect stimulation. The test substance was cumulatively applied every 5 min to the medium in the Magnus tube. The contraction was expressed as % calculated with reference to the value recorded immediately before the vehicle. Bipolar electrodes for direct and indirect stimulation were located on the diaphragm and on the phrenic nerve, respectively, and the preparation was suspended in the Magnus tube filled with aerated Krebs-Ringer´s solution. Direct stimulation (8 msec) and indirect stimulation (0.05 msec) at supramaximal voltage were applied alternately with an interval of 5 s with an electric stimulator. Contraction was recorded isometrically.

4. Effects on digestive organs
Transport activity of the small intestine in male mice was studied 1 h after oral administration of the test substance (8 animals/dose). The mice were deprived of food for about 16 h before the experiment. Charcoal suspension was administered orally in a volume of 10 mL/kg 1 h after the administration of the test substance. Mice were euthanized with ether 30 min after the administration of the charcoal and their intestines were isolated. The mobility of the charcoal (%) was calculated by comparing the length of charcoal transported in the small intestine with the total length. Charcoal powder was uniformly suspended in 10% acacia aqueous solution to give a concentration of 10% (w/v).

The effects on the ileum of male guinea pigs was evaluated in vitro by applying the test substance at concentrations of 0.1, 1, 10, and 100 µg/mL (4 animals/dose). Four preparations of the isolated ileum were used to study the effects of the test substance on the spontaneous motility and agonist-induced contraction. After confirming contraction induced by acetylcholine (50 ng/mL, 30 s) effects of the test substance on spontaneous motility were observed for 5 min in the presence of the test substance. Thereafter, contraction was induced by acetylcholine (50 ng/mL), histamine (50 ng/mL), or high potassium concentration (50 mM). The contraction was expressed as % calculated with reference to the respective mean values of two consecutive values recorded immediately before the vehicle.

5. Effects on skeletal muscle
Grip strength in male rats were observed 1 day before and 1 h, 3h, 6 h, 1 day, 2 days, 3 days, and 7 days after oral administration of the test substance (5 animals/dose).

6. Effects on renal function
Urine of male rats was collected to evaluate renal function during 3 h collection period after loading physiological saline, which was initiated 1 h following the oral administration of the test substance (5 animals/dose). Effects on urine volume, urinary electrolyte excretion, osmolality, pH, occult blood, protein, ketone, and glucose were examined.

II. RESULTS AND DISCUSSION

A. Effects on central nervous system

1. Male and female mice (toxicological signs and body weights)
In male mice one animal of the 40 and 100 mg/kg group and all animals of the 250 mg/kg dose group died within 6 h after the administration. In female mice two animals of the 250 mg/kg group died within 3 h after administration. Abnormal signs observed in the survived mice disappeared within 2 days. No distinct abnormalities were observed for male and female mice at 16 mg/kg or less. No distinct changes were observed for body weights at the doses up to 250 mg/kg.
Nonspecific abnormal signs including both excitatory and inhibitory signs were observed at 40 mg/kg upwards. The abnormal signs consisted of a decrease in alertness and visual placing, manifestation of passivity and thrashing, a decrease in spontaneous activity and reactivity, exaggerated touch and startle response, manifestation of Straub tail, tremors, and clonic convulsions, prostrate, staggering gait, a decrease in righting reflex, limb tone, grip strength, body tone, ipsilateral flexor reflex (IFR), manifestation of salivation, hypothermia, whitish skin, and a decrease in respiratory rate.

2. Male rats (toxicological signs, body temperature, and body weights)
One animal of the 625 mg/kg group died within 2 days after administration. Abnormal signs observed in the survived rats disappeared within 7 days. No distinct abnormalities were observed at 16 mg/kg or less. Body weights were significantly decreased 1 day through 3 days at the highest dose. No distinct changes were observed for body weights at the doses up to 250 mg/kg.
Oral administration of the test substance dose-dependently produced excitatory abnormal signs at 40 mg/kg or more. The abnormal signs consisted of manifestation of thrashing, manifestation of Straub tail, clonic convulsions, prostrate, staggering gait, salivation, urination, shivering, and whitish skin.
Body temperature was transiently decreased 1 and 3 h after administration of 100 mg/kg or more. At 250 and 625 mg/kg increased values of 2 and/or 3 days were observed, but seemed not to be related to the administration of the test substance.

3. Male mice (hexobarbital sleeping time)
No distinct changes were observed for the sleeping time up to 100 mg/kg, although 6/8 animals of the 100 mg/kg group died 30 min through 3.5 h after the injection of hexobarbital.

B. Effects on Respiration and circulation

1. Male rabbits (heart rate, blood pressure, electrocardiogram, respiration)
Injection of vehicle (5% Tween 80 aqueous solution containing 15% DMSO) produced a decrease in blood pressure during the last half of injection and thereafter, a slight increase in blood pressure that lasted for a few minutes. Injection of 3 mg/kg test substance elicited bursts in electrocardiogram related to convulsions and slightly increased systolic blood pressure. Injection of 12 mg/kg test substance elicited remarkable increase in blood pressure and tidal volume a few minutes after injection. Thereafter, low level of blood pressure continued during several ten minutes before death. All animals died at that dose. Cardiovascular collapse and decreased respiratory rate occurred immediately before death. No changes related to the test substance were observed at the dose of 0.75 mg/kg.

C. Effects on autonomic nervous system

1. Male rabbits (pupil size)
Mean pupil size of the 250 mg/kg group was slightly but significantly increased 3 h after administration. As this effect was only observed in some animals and was not observed in the other dose groups or at the other time points this was not considered to be treatment related.

D. Effects on digestive organs

1. Male mice (transport activity of the small intestine)
Transport activity of the small intestine was slightly decreased at the doses of 40 mg/kg or more. The mobility of the 100 mg/kg group decreased to 70% of the control group. Changes related to the test substance were not observed at the doses of 16 mg/kg or less. Statistical significance is noted for the value of the 2.6 mg/kg group. However, it seems to be accidental because significant changes were not observed at the higher two successive doses.

2. Male guinea pigs (effects on isolated ileum)
Application of the test substance at concentrations of 10 µg/mL or more increased motility. The increase observed at 100 µg/mL was still observed after washing out the test substance. The changes at 1 µg/mL were marginal and no distinct changes were observed at lower concentrations. Application of the test substance inhibited histamine-induced contraction to about 90% of pre-application at 10 µg/mL or more. Application of the test substance also inhibited high potassium-induced contraction to about 80% at 1 µg/mL or more. Acetylcholine-induced contraction was not significantly affected up to concentrations of 100 µg/mL.

E. Effects on skeletal Muscle

1. Male rats (effects on grip strength and on isolated phrenic nerve-diaphragm preparation)
No test substance related changes of grip strength were observed at doses up to 625 mg/kg. Occasional changes at 100 and 250 mg/kg were considered to be not treatment-related due as the values were close to the pre-administration values and the control values were small.
Treatment with 1 µg/mL or more increased both direct (muscle) stimulation-induced and indirect (nerve) stimulation-induced contraction. Direct contraction was increased more strongly than the indirect contraction (50% vs. 10-20%). No effects were observed at concentrations of 0.1 µg/mL.

F. Effects on renal function

1. Male rats (urinalysis)
After administration of the test substance at 100 mg/kg an increased glucose concentration was observed in the urine. No effect on glucose concentration was observed at 40 mg/kg or less. No changes of the other parameters were observed at the doses up to 625 mg/kg. Although no statistical significance was noted in the 625 mg/kg group, 3/4 survived animals in the group exhibited severe and extreme scores on glucose. The high incidence suggests that urine glucose of the group is increased by the test substance. On the other hand statistical significance in urinary K and Cl excretions of the 100 and 250 mg/kg groups seems not to be related to the administration of the test substance, because the values of the 625 mg/kg group are larger than those of the lower groups but were not statistically significant.

III. CONCLUSIONS

Alphacypermethrin (batch 351, purity 93.2%) was tested in a general pharmacological study in mice, rats, guinea pigs and rabbits. The test substance produced acute toxicities through its action on the nervous and cardiorespiratory systems at sublethal doses close to lethal doses.

Report: CA 5.8.2/4
[redacted] 2000b
Alphacypermethrin: Effects of antidotes on acute toxicity
AL-452-002
Guidelines: <none>
GLP: yes
(certified by Ministry of Agriculture, Forestry and Fisheries of Japan, Japan)
Remark: This study was not submitted within the first Annex I inclusion. As this study is supplemental, the data are only shortly summarized.

Executive Summary

In 1999 an antidote study with methocarbamol, mephenesin and halothane was conducted in each 10 male rats to investigate their possible use as antidotes in acute oral intoxications. Alpha-cypermethrin (Batch 351, purity 93.2%) was dissolved in corn oil and orally administered at a dose of 125 mg/kg bw to 6 week old male Sprague-Dawley rats. Methocarbamol (100 mg/kg) and mephenesin (100 mg/kg) were subcutaneously treated immediately after, and 4, 8, 12, 16, 20, and 24 hours after the administration of alpha-cypermethrin. Halothane was continuously exposed to the animals in a chamber at a concentration less than 1% up to 24 hours after the administration. The animals were observed 4, 8, 12, 16, 20, 24, 28, 48, 72, 96, 120, 144, and 168h after the administration of the test substance. As control group served 10 animals treated with saline after the intoxication with alpha-cypermethrin.
The saline –treated group produced salivation, convulsions and 6 out of 10 rats died within 48 hours. Methocarbamol and mephenesin did not show any protective effects on the incidence or latency of the toxic parameters, halothane protected the animals from the acute toxic signs (salivation and convulsions) although it had no protective effects on the lethality.

Classification of the study: Supplemental information

Additional toxicological information found in public literature

Taken from the DAR of Zeta-cypermethrin:

Report: CA 5.8.2/5
Anonymous, 2006a
Draft Assessment Report (DAR) Zeta-Cypermethrin - Volume 1 to volume 3 - Initial risk assessment provided by the RMS Belgium of the third stage (part A) of the review programme referred to in article 8(2) of council directive 91/414/EEC
2006/7013990

Dietary placental transfer and lactation transfer of zetacypermethrin at 50, 125, 300 ppm in rats from GD 6-20, or GD6-LD21 [redacted] 2005a)

And

Measurement of zetacypermethrin in milk following dietary administration at 125 and 375 ppm from GD 6 to LD 17 [redacted] 2004)

Guidelines: none (not applicable: open literature)
GLP: no

Dietary placental transfer and lactation transfer of zetacypermethrin at 50, 125, 300 ppm in rats from GD 6-20, or GD6-LD21 ([redacted] 2005a)

Executive Summary

The placental and lactational transfer of zetacypermethrin in rats was evaluated in this developmental toxicity study. Test diet containing zetacypermethrin was offered to 6 mated female rats per group from GD 6 through GD 20 (placental transfer animals) or lactation day 21 (lactational transfer animals) at 0, 50, 125, 300 ppm for placental transfer and at 125 ppm for lactational transfer. No mortality and no clinical signs of toxicity were observed. No effect on body weight or food consumtion was observed. At the scheduled necropsy on gestation day 20, 2 females were non gravid. Mean number of pups born, percent males at birth and live litter size were within the historical control data ranges in the placental transfer group. The mean litter percentage of post-implantation loss, live litter size, early and late resorptions at the different dose levels were similar to the control group. The higher percentage of early resorptions at 300 ppm was below the maximum mean value in the laboratory historical control data for definitive studies. In the lactation transfer group no effects on gestation length or the process of parturition were noted in the maternal animals of this group. Furthermore no abnormalities were found regarding mortality, body weight or food consumption. Postnatal survival was within the range of historical control data at most time points. From PND 4 (post selection) to 7 postnatal survival was slightly lower than the historical control data, but was considered a spurious finding. Plasma analyses showed that maternal and fetal plasma are accurate indices of internal exposure to zetacypermethrin during gestation and lactation.

I. MATERIAL AND METHODS

A. MATERIALS

1 ***Test Material:***

Description:	*Zetacypermethrin*
Lot/Batch no.:	*PL03-0427*
Purity:	*81.8%*
Stability:	*Stability in diet was verified for 15 days*

2 ***Dosing:***

Dose levels:	*0, 50, 125, 300 ppm (in diet)*
Dosing period:	*GD 6 – GD 20 or LD 21*

3 ***Test animals***

Species:	*Rat*
Strain:	*Crl:CD(SD)*
Sex:	*Female (6/group)*
Acclimation period:	*yes (length not specified)*

B. STUDY DESIGN AND METHODS

After an acclimation period untreated females were paired with untreated male rats. Test diet containing zetacypermethrin was offered to 6 F0 female rats per group from GD 6 through GD 20 (placental transfer animals) or lactation day 21 (lactational transfer animals) at 0, 50, 125, 300 ppm for placental transfer and at 125 ppm for lactational transfer. A control group was included for placental transfer.
Placental transfer: females were necropsied on GD 20; blood and brain samples were collected from 6 females/group. Blood and brain samples were collected from fetuses (pooled within litters). Samples were analyzed for zetacypermethrin.
Lactational transfer: all females were allowed to deliver. F1 pups were potentially exposed to zetacypermethrin in utero, through nursing during lactation and via direct consumption late in lactation. Blood was collected from all dams on LD 5. Blood and brain samples were collected and pooled from F1 culled pups on post natal day 5. F0 females were necropsied on LD21; blood and brain samples were collected from all dams. Blood and brain samples were collected from 1 weanling/sex/litter on post natal day 21. F0 and F1 samples were analyzed for zetacypermethrin. Samples were analyzed using a gas chromatograph with a µECD and capillary column. Stability in diet of zetacypermethrin was observed for 15 days at room temperature.

II. RESULTS AND DISCUSSION

A MATERNAL OBSERVATIONS (PLACENTAL PHASE)

The following observations were performed in the placental transfer phase with dose groups 0, 50, 125, and 300 ppm.
Mortality: all females survived until necropsy day 20.
Clinical signs: no compound-related clinical signs were noted.
Body weight: main bw gain were slightly decreased at 300 ppm but did not affect the body weight.
Food consumption: was not affected.
Parturition: at the scheduled necropsy on gestation day 20, 2 females were non gravid.
Histopathology:
GD 20 laparohysterectomy (placental transfer group): Mean number of pups born, % males at birth and live litter size were within the historical control data ranges.
The mean litter percentage of postimplantation loss, live litter size, early and late resorptions at the different dose levels were similar to the control group. The higher % of early resorptions at 300 ppm was below the maximum mean value in the laboratory historical control data for definitive studies.

B MATERNAL OBSERVATIONS (LACTATIONAL PHASE)

The following observations were performed in the lactational transfer phase with dose groups 0 and 125 ppm.
Mortality: All females delivered offspring and survived to the scheduled necropsy on lactation day 21.
Body weight: was not affected.
Food consumption: was elevated as is commonly seen in nursing rats.
No effects on gestation length or the process of parturition were noted in this group.

C LITTER DATA F1

Postnatal survival from PND 4 (post selection) to 7 was slightly lower than the historical control data. This effect was not seen in the developmental neurotoxicity study. Therefore, the effect on PND 4-7 survival in this study is considered a spurious finding and not treatment-related. In addition, postnatal survival on PND 0, 0-1, 1-4 (preselection), 7-14, 14-21, birth to PND 4 (preselection), and PND 4-21 ((postselection), was within the range of historical control data. Mortality was not considered to be compound-related due to the small sample size (6 dams/group).

Table 5.8.2-7: Gestational and lactational parameters after zetacypermethrin dietary exposure.

Endpoints/dose	*0*	*50*	*125*	*300 ppm*	*Historical control data*
Compound consumption (mg/kg bw/d)					
Gestation:	*0.0*	*4.1*	*10.0*	*23.6*	
Lactation:			*20.6*		
No. females on study	*6*	*6*	*6*	*6*	*1755*
Not pregnant GD 20			*1*	*1*	
Gravid with viable pup	*6*	*6*	*5*	*5*	*1583*
Body weight GD 20 (g)	*397*	*399*	*417*	*394*	
Bw changes D0-20 (g)	*142*	*143*	*157*	*139*	
Food consumption D0-20 gestation(g/animal/d)	*25*	*25*	*25*	*25*	
Food consumption 1-21 lactation (g/animal/d)			*56*		
Laparohysterectomy data:					
F1 litter mean					
Viable fetuses PND 0	*14.8*	*14.7*	*16*	*14*	*14.2*
Dead fetuses	*0*	*0*	*0*	*0*	
Preimplantation loss	*2.0*	*2.8*	*2.8*	*3.0*	
Early resorptions (%)	*0.5(3.2%)*	*0.5*	*0.0*	*1.2(7.2%)*	*(8.6%)*
Late resorptions	*0.2*	*0.0*	*0.0*	*0.0*	
Implantation sites	*15.5*	*15.2*	*16*	*15.2*	*15.5*
Corpora lutea	*17.5*	*18*	*18.8*	*18.2*	
Litter survival PND 4-7:			*92.6%*		*95.8%*
Zetacypermethrin in plasma(ppm):					
Gestation day 20					
Dams	*0.11±0.04*	*0.31±r0.1*	*0.35±0.15*	*0.57±0.24*	
Pups	*0.08±0.02*	*0.12±0.01*	*0.11±0.02*	*0.14±0.04*	
Lactational day 5/21					
Dams D5/D21			*0.50±0.13/ 0.57±0.12*		
Pups D5/D21			*0.26±0.08/ 0.23±0.11*		

D TOXICOKINETIC ANALYSIS IN BLOOD

Gestation day 20:
Dams: the results of gestation day 20 control group samples probably reflect interference from an unidentified source (2 large early-eluting peaks). The levels found in the maternal plasma samples for the test-article groups show the expected increase in plasma with the dose.

Fetal plasma: low levels were detected in fetal plasma samples. Again, the control group was contamined with an unidentified source (2 large early eluting peaks). Mean concentration of zetacypermethrin in fetal plasma were lower than those in maternal plasma at all dose levels on GD 20 suggesting a minimal transplacental transfer.

Lactational day 5 and 21:
Plasma levels in F0 females were similar for lactational day 5 and 21 but higher than those measured on GD 20 reflecting higher lactational compound consumption and a higher plasma level for offsprings on both lactation day 5 and 21.

III. CONCLUSION

The present results show that maternal and fetal plasma are accurate indices of internal exposure to zetacypermethrin during gestation and lactation. RMS (Belgium) agreed that the increased incidence of early resorptions seen at top dose is not compound-related because these effects were not seen in the next reported study performed with somewhat higher doses. The historical control data are not clearly reported in this study making the use of these data difficult.

Taken from the DAR of Zeta-cypermethrin:

[see KCA 5.8.2/5 2006/7013990]

Measurement of zetacypermethrin in milk following dietary administration at 125 and 375 ppm from GD 6 to LD 17 [redacted] 2004)

Executive Summary

The dietary transfer of zetacypermethrin into the milk of female rats and the effect of the in utero and post-natal exposure through milk on the offspring was evaluated in this study. Groups of 15 mated female rats (Crl:CD (SD) IGS BR) were exposed to zetacypermethrin at dietary dose levels of 125 or 375 ppm from gestation day 6 through lactation day 17.
No mortality was observed. Impaired use of left hind limb was seen in 3 females at top dose. Slight reduction of body weight gain during gestation days 6-9 and lactation days 1-4 and lower body weights on lactation days 4-17 were observed in the 375 ppm group. Food consumption was also lowered during gestation days 6-9 and during lactation days 1-17 in the high dose group. Gestation length and parturition were unaffected by the compound. Mean offspring weights were lowered at 375 ppm throughout the post natal period and in the 125 ppm group during PN day 11-17. These effects were considered to be compound-related. Reduced litter weight changes were observed at 125 and 375 ppm. No other abnormalities were observed. Analysis of zetacypermethrin in milk samples show that zetacypermethrin was detected in the milk of nursing pups following dietary administration to the mothers. Levels were dose-related and time-related with a plateau on lactation day 11.
The results of this study indicate that dietary administration to mothers permits to have transfer from food to milk and to quantify exposure of pups via milk.

I. MATERIAL AND METHODS

A. MATERIALS

1 ***Test Material:***

Description:	*Zetacypermethrin*
Lot/Batch no.:	*PL03-0427*
Purity:	*81.8%*
Stability:	*Stability and homogeneity were confirmed*

2 ***Dosing:***

Dose levels:	*0, 125, 375 ppm (in diet)*
Dosing period:	*GD 6 – LD 17*

3 ***Test animals***

Species:	*Rat*
Strain:	*Crl:CD(SD) IGS BR*
Sex:	*Female (15/test group; 10/control group)*

B. STUDY DESIGN AND METHODS

15 mated female rats (Crl:CD (SD) IGS BR) per group received zetacypermethrin in their diet from gestation day 6 through lactation day 17 at 125 or 375 ppm. The control group contained 10 females. Rats were observed twice daily. Animals were allowed to deliver and rear their offspring to lactation day 17. The offsprings were exposed to zetacypermethrin in utero and through nursing during lactation. On PND 4, litters were culled to 8 pups/litter. All F1 pups were observed daily. On lactation day 4, 11 and 17, milk samples were collected from each dam for the determination of zetacypermethrin. Whole brain samples were also collected from pups on PND 4, 11 and 17 for potential future analysis. All surviving F0 females and surviving F1 pups were euthanized and discarded 17 days postpartum. The test article formulation was found to be homogeneous and contained the amounts specified.

II. RESULTS AND DISCUSSION

A. OBSERVATIONS

MATERNAL OBSERVATIONS
Mortality: there were no mortalities in the study. Postnatal survival was unaffected at the different doses.
Clinical signs: impaired use of left hind limb was seen in 3 females at top dose.
Body weight: while maternal body weight was not affected, mean body weight gain was lowered at 375 ppm during gestation days 6-9 and body weights were lower on lactation days 4-17. Main bw gain was lower during lactation days 1-4.
Food consumption: was lowered during gestation days 6-9 and during lactation days1-17 at top dose.

LITTER DATA F1
The mean number of pups born, live litter size and percent of males/litter at birth were unaffected by treatment. The number of F1 pups found dead and missing, as well as the general condition of all F1 pups were unaffected by maternal exposure.
Mean offspring weights were lowered at 375 ppm throughout the post natal period and in the 125 ppm group during PN day 11-17. These effects were considered to be compound-related. Reduced litter weight changes were observed at 125 and 375 ppm.

Table 5.8.2-8: Dietary administration of zetacypermethrin from GD6 to LD17.

Endpoints/dose	*0*	*125 ppm*	*375 ppm*
Compound ingestion during gestation Day 6-20 (mg/kg bw/d)	*0*	*8.59*	*24.38*
Compound ingestion during lactation Day 1-17 (mg/kg bw/d)	*0*	*16.71*	*47.73*
No females on study/gravid	*10/10*	*15/15*	*15/15*
Clinical signs:			

Impaired use left hindlimb during lactation					*3 rats, days 13-17, day 14 and day 12-17*	
Bw lactation day 4					*↓2%*	
Bw lactation day 11					*↓8%*	
Bw lactation day 17					*↓6%*	
Bw changes						
GD 6-9					*↓71%*	
LD 1-4					*↓52%*	
Food consumption during gestation (g/animal/day) Day 6-9					*↓29%*	
Food consumption during lactation (g/animal/day) Day 1-17					*↓9%*	
Pup data:						
Mortality	*2*		*3*		*7*	
Missing/cannibalized	*0*		*1*		*1*	
Litter weight						
PND 1 (g)	*7.2*	*6.9*	*7.3*	*7.1*	*6.9*	*6.5*
PND 4	*9.5*	*9.1*			*↓8%*	*↓11%*
PND 7	*15.7*	*14.8*			*↓12%*	*↓11%*
PND 11	*24.6*	*23.1*	*(↓4%)*	*(↓3%)*	*↓17%*	*↓15%*
PND 14	*32.7*	*31.4*	*(↓6%)*	*(↓10%)*	*↓19%*	*↓18%*
PND 17	*39.4*	*37.6*	*(↓6%)*	*(↓7%)*	*↓17%*	*↓17%*
Litter weight changes (g)						
PND 1-4	*2.3*	*2.3*	*2.3*	*2.1*	*1.7**	*1.6**
PND 4-7	*6.2*	*5.7*	*6.3*	*5.8*	*5.2**	*5*
PND 7-11	*8.9*	*8.3*	*7.8**	*7.3*	*6.5**	*6.4**
PND 11-14	*8*	*8.1*	*6.8*	*6.7*	*6.2*	*6.0*
PND 14-17	*6.7*	*6.2*	*6.6*	*6.0*	*6.1*	*5.7*
Mean zetacypermethrin in milk (ppm)						
Day 4			*0.58±1.09*		*7.86±4.91*	
Day 11			*3.9±1.6*		*10.46±2.87*	
Day 17			*3.17±1.21*		*11.55±2.12*	

** statistically significantly different from control; () not statistically different from control*

B ANALYSIS OF ZETACYPERMETHRIN IN MILK

Analysis of zetacypermethrin in milk samples show that zetacypermethrin was detected in the milk of nursing pups following dietary administration to the mothers. Levels were dose-related and time-related with a plateau on lactation day 11. No residues were found in control group animals. Gestation length and parturition were unaffected by the compound.

III. CONCLUSION

The results of this study indicate that dietary administration to mothers permits to have transfer from food to milk and to quantify exposure of pups via milk.

Report: CA 5.8.2/6
Abass K. et al., 2012a
Characterization of human cytochrome P450 induction by pesticides
2012/1367022
Guidelines: <none>
GLP: no
(certified by <none>)

Executive Summary of Literature

The authors investigated several pyrethroids (alpha-cypermethrin (highest purity available: not given), cypermethrin (cis/trans ratio not given, highest purity available: not given), deltamethrin, fenvalerate and lambda cyhalothrin together with representatives of organophosphorus, carbamates and other pesticides on their property to induce cytochrome P450 enzymes. A HepG2 transfection system was used to investigate the activation of human PXR via the dual-Luciferase Reporter Assay System (Promega). Activation of mouse and human CAR was investigated in C3A hepatoma cells with a Gal4-responsive luciferase reporter. Induction of CYP-mRNA was investigated by c-DNA synthesis and subsequent real time PCR and in addition enzyme activities of CYPs were investigated in the N-in-one cell culture assay. All pesticides were tested at 10 and 50 µM. These concentrations did not induce significant cytotoxicity, measured as Lactate dehydrogenase release (50 µM alphacypermethrin: 2.4%; cypermethrin: 3.0%). Both alpha-cypermethrin and cypermethrin pyrethroids induced the human nuclear receptor CAR and PXR to the same extent (see Figure 1 of the publication below), induced mRNA release of CYP2A6 (alpha-cypermethrin > cypermethrin), 2B6 and 3A4 (alpha-cypermethrin < cypermethrin), and lead to 3-4 fold induction for CYP2B6 and CYP 3A4 in HepaRG cells. This enzyme induction is in agreement with a CYP3A11 induction in mouse primary hepatocytes. Pyrethroids were rather ineffective on CYP1A2 mRNA release and enzyme induction.

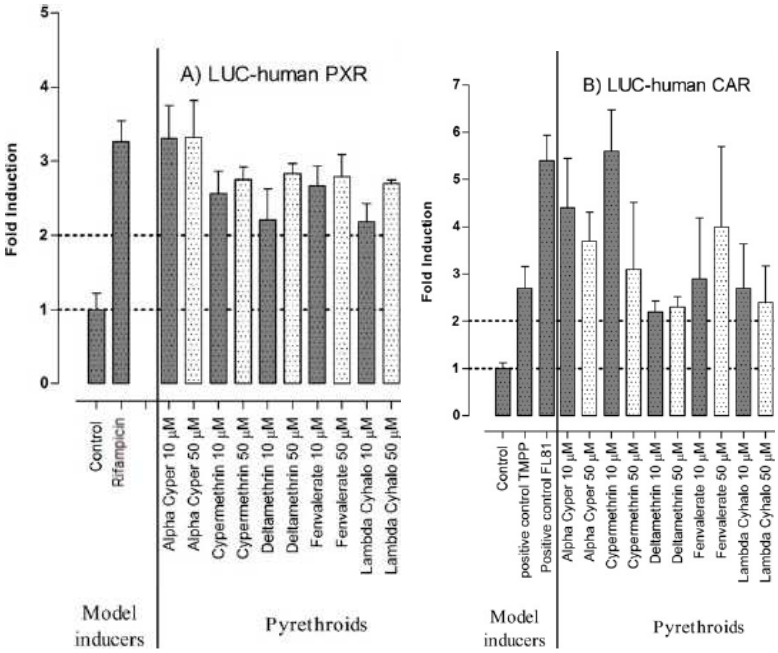


Figure 1 of the publication: Activation of human A) PXR and B) CAR by model inducers and pesticides. Nuclear receptor activation was measured by luciferase reporter gene activity. Results are expressed as mean ± SD of at least three independent experiments.

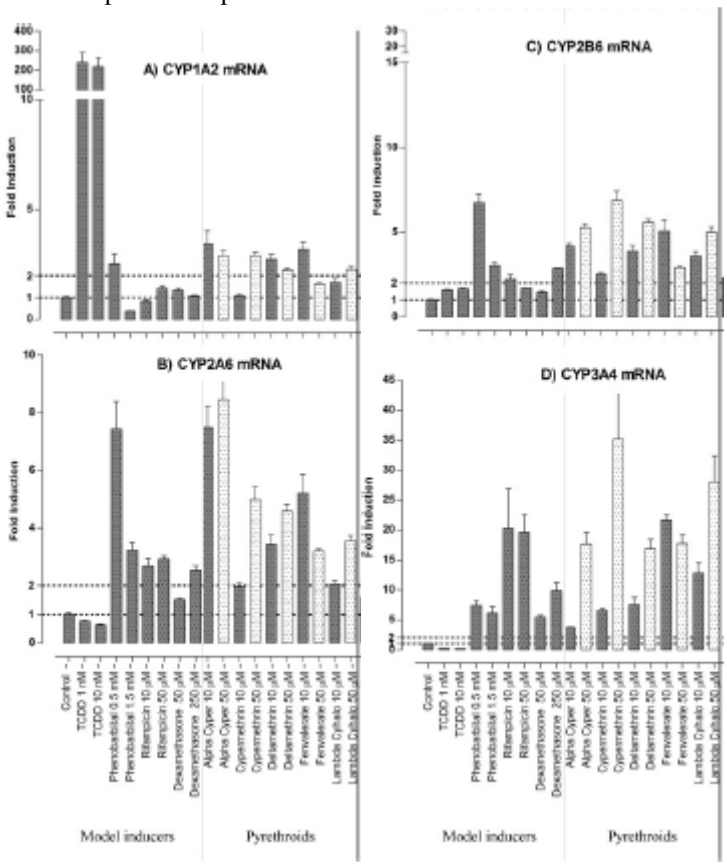


Figure 2 of the publication: Cyp mRNA levels in human HepaRG cell line after 24 h exposure to the test compounds. Total RNAs were isolated and subjected to RT-PCR. The mRNA levels are expressed as relative to the respective control cell level (normalized to 1) and the data are expressed as mean ± SD of six replicates.

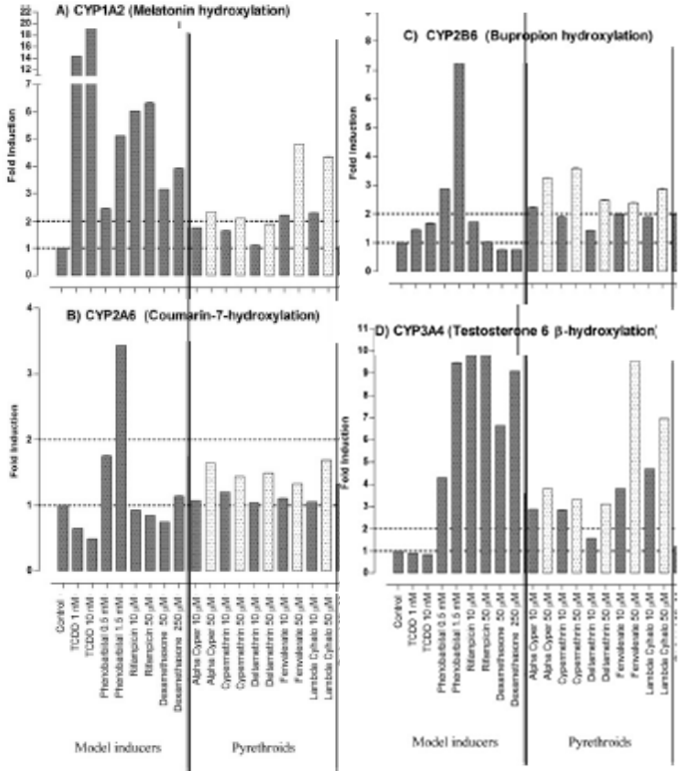


Figure 3 of the publication: A) CYP1A2, B) CYP2A6, C)CYP2B6, D) CYP3A4 enzyme activities measured by N-in-one assay. The enzyme activity levels are expressed as relative to the respective control cell level (normalized to 1) and the data are expressed as mean ± SD of three replicates.

Conclusion of the author: Pyrethroids moderately induced CYP2A6 and CYP2B6 mRNAs, and strongly upregulated CYP3A4 mRNA in HepaRG cells. In contrast to the human nuclear receptor the mouse CAR seems to be much less responsive to the pesticides than the human receptor.

Conclusion of the applicant: The used in vitro test system N-in-one for determination of enzyme acivity is considered as screening assay only (taking into consideration the following report Abass et al., 2013 Doc ID 2013/1416883 in which discrepancies to the single substrate assay were shown). The described data however confirm similarity of alphacypermethrin and cypermethrin with regard to mRNA induction. CYP P450 enzymes are not the main enzymes relevant for metabolic degradation of cypermethrins in humans, therefore, and based on the fact that the liver is not the primary target organ, these data are considered as supplemental information to support the bridging approach but are not considered relevant for risk assessment.

Classification of study: Supplemental information

Report: CA 5.8.2/7
Abass K.,Pelkonen O., 2012a
The inhibition of major human hepatic cytochrome P450 enzymes by 18 pesticides: Comparison of the N-in-one and single substrate approaches
2013/1416883
Guidelines: <none>
GLP: no

Executive Summary of Literature

The authors investigated the potential inhibitory interactions of a selection of widely used pesticides (among those alpha-cypermethrin and cypermethrin, deltamethrin, fenvalerate and lambda-cyhlothrin) by the N-in-one assay in human hepatic microsomes (from liver samples obtained from the University Hospital of Oulu) and compared this with a single substrate assay. The inhibitory interactions of tested compounds were investigated at 1, 5, 25, 50, and 100 µM. The respective pesticides were preincubated with the microsomal protein and the substrate for 2 min before the addition of NADPH initiated the reaction. After 20 min the reaction was stopped, protein fraction was removed and supernatant was analysed via LC/MS-MS. Reduced substrate degradation was interpreted as inhibition based on CYP interaction with the pesticide.
The data for alpha-cypermethrin and cypermethrin were extracted from the Supplementary data in the online version, at http://dx.doi.org/10.1016/j.tiv.2012.05.003 and are presented below in Table 1 Supplementary of the publication (see below).

Table 1 Supplementary. IC_{50} values (µM) of the studied pyrethroid pesticides on different human P450 enzymes using pooled human hepatic microsomes as measured by the N-in-one[1] and single substrate[2] approaches.

	λ-Cyhalothrin		α-Cypermethrin		Cypermethrin		Deltamethrin		Fenvalerate	
CYP isoform	N-in-one	Single	N-in-one	Single	N-in-one	Single	N-in-one	Single	N-in-one	Single
CYP1A2	315.7	315.0	361.2	319.7	593.3	960.0	386.4	334.6	980.0	979.0
CYP 2A6	297.1	193.8	241.3	195.3	235.3	196.8	226.0	138.1	618.8	145.8
CYP 2B6	389.4	200.6	233.1	197.5	225.0	235.0	118.8	143.5	537.5	405.4
CYP 2C8	863.0	945.0	328.0	215.0	271.6	178.6	405.3	484.0	332.0	441.3
CYP 2C9	302.4	273.1	890.4	1595	1043	1198	196.1	336.9	165.7	380.5
CYP2C19	260.2	273.1	679.1	595.1	1771	1198	176.2	336.9	173.9	380.5
CYP 2D6	252.7	3.1	405.0	537.0	1198	1289	261.0	3.3	121.0	3.1
CYP2E1	422.5	1172	4.0	1115	290.3	543.0	139.4	1649	18.4	1771
CYP3A4[3]	1294	3.1	871.0	1613	1543	70.0	1649	42.9	422.0	19.3
CYP3A4[4]	106.5	3.9	3.4	148.0	191.0	249.0	115.4	18.6	13.4	74.3

[1] Each pesticide was added at final concentrations of 1, 5, 25, 50 and100 µMto the incubation mixture for the IC_{50} values determination. The IC_{50} values were determined from three technical replicates from the same human microsomal poolby linear regression analysis from the plot of the logarithm of inhibitor concentration versus percentage of the activity remaining after inhibition using MicroCal Origin 6.0 (MicroCal Software. Inc.. Northampton. MA). The enzyme activities in the presence of inhibitors were compared with the control incubations (incubations with the solvent only). IC_{50} values higher than 100 µM were extrapolated based on the linear regression analysis obtained from tested concentrations.
[2] IC_{50} values (µM) measured by single substrate assays were obtained from our previous study (Abass et al. 2009) for comparison between the assays.
[3] 1-OH-MDZ
[4] SO_2-OME

Alpha-cypermethrin inhibited CYP2E1 and CYP3A4 with IC50 values of 4.0 and 3.4 µM in the N-in-one assay. This enzyme inhibition was not reproduced in the single substrate approach.

Conclusion of the author: The N-in-one screening assay seems useful and reliable for most CYP activities when a comprehensive and quick evaluation of potential interactions with CYPs is needed.

Conclusion of the applicant: The enzyme inhibition with alpha-cypermethrin seen in the N-in-one screening assay with CYP2E1 and CYP3A4 is highly questionable because it was not reproducible in the single substrate assay. In the single substrate assay enzyme inhibition is at a compareable range between alpha-cypermethrin and cypermethrin.

Classification of study: Supplementary information

Report: CA 5.8.2/8
Nasuti C. et al., 2006a
Dopaminergic system modulation, behavioral changes, and oxidative stress after neonatal administration of Pyrethroids
2007/1070386
Guidelines: none
GLP: no

Executive Summary of Literature

The authors investigated the effects of cypermethrin (cis/trans:63/37%; purity: 92.4%,) and permethrin (cis/trans:75/25%, purity: 94%) on the behavior of rats after neonatal exposure (postnatal day (PND) 6 or 8 to PND 15) via gavage in corn oil. Furthermore, the level of oxidative stress and the effects on the dopaminergic system were determined. Each group consisted of 10 dams, each with 4 male pups treated with vehicle alone, 1.49 cypermethrin or 34.05 mg/kg bw permethrin dissolved in corn oil by oral gavage (5 mL/kg bw). The doses correspond to 1/10 of the LD_{50} values. Litter weight was recorded at PND1, PND7, PND14 and PND21.
Neurotransmitter status and degradation (dopamin, 3,4-dihydroxyphenylacetic acid (DOPA) and homovanillic acid (HVA)) in striatum was investigated in 10 animals per group (PND35).
Open field studies (number of ambulatory episodes, number of rears, number of grooming movements, number of entries into the center) were conducted at PND21 and PND35 with 10 rats per group.
Antioxidant activities (GPx and SOD activities) in plasma were investigated in the blood of six rats per group (PND35). Protein oxidation (determination of carbonyl group content) and lipid peroxidation (oxidation index) was determined in the striatum and in erythrocytes. GSH content was determined in the cells from striatum and the respiratory burst in monocytes was determined using Lucigenin as an indicator of superoxide anion radicals.

Results:
Rats treated with cypermethrin or permethrin showed no signs of pyrethroids poisoning or gross behavioral abnormalities throughout the experimental period and no effect on body weight was observed, when comparing control and treated groups (see Figure 1 of the publication, below).

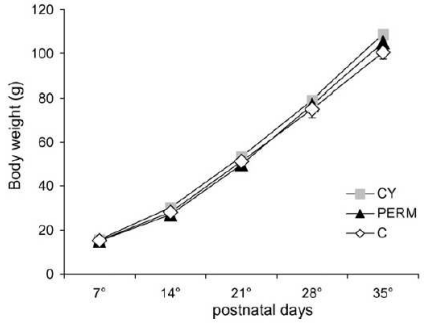


Fig. 1. Body weight in CY-treated (■), PERM-treated (▲) and control rats (◊) during 35 postnatal days. Data are presented as mean ± S.D.
*$P < 0.05$ vs. control (C).

In the open field observation no significant modifications were observed on PND21 (see Fig. 2 of the publication below), whereas on PND35 pyrethroid treatment increased the spontaneous locomotor activity of rats in comparison with the control group (see Fig. 3 of the publication below). In addition a significant increase of rearing was observed after treatment with cypermethrin on PND35. With regard to anxiety-like behavior, the cypermethrin-treated rats had more entries and time spent in the center than control animals.

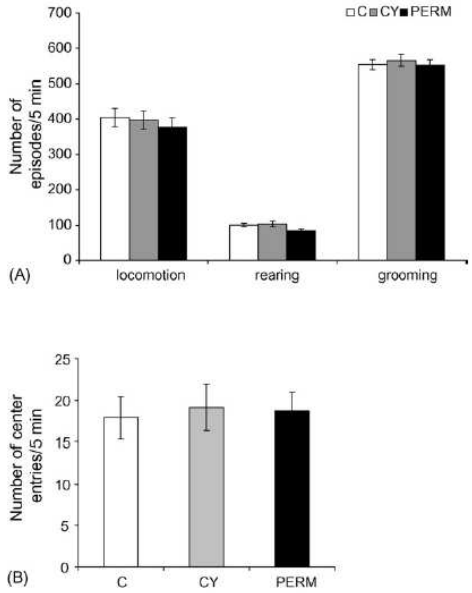


Fig. 2. Effects of neonatal cypermethrin (CY) and permethrin (PERM) exposure on locomotion, rearing and grooming (A) and anxiety test (B) in rats observed at 21 days of age. Data are expressed as mean ± S.D. for 10 rats ($n = 10$L). $^{*}P < 0.05$ vs. control (C).

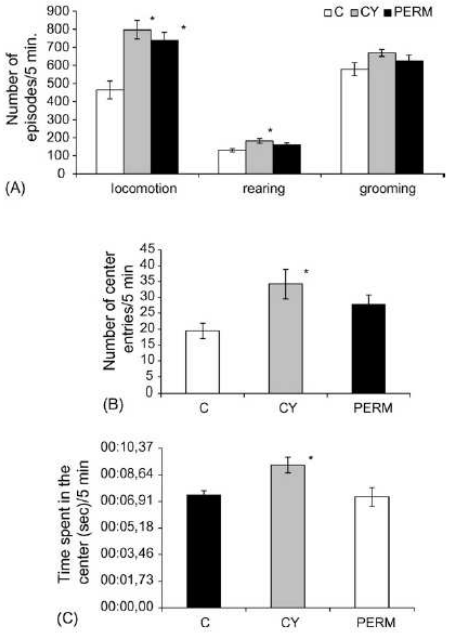


Fig. 3. Effects of neonatal cypermethrin (CY) and permethrin (PERM) exposure on locomotion, rearing and grooming (A) and anxiety test (B and C) in rats observed at 35 days of age. Data are expressed as mean ± S.D. for 10 rats ($n = 10$L). $^{*}P < 0.05$ vs. control (C).

Monoamine levels: On PND 35 pyrethroid-treated animals exhibited lower striatal dopamine levels. No significant changes were observed in DOPAC levels, while HVA concentration increased significantly in rats treated with cypermethrin (26±5.2 pmol/mg tissue) and permethrin (24±4.2 pmol/mg tissue) compared to the control animals (17±1.6 pmol/mg tissue).

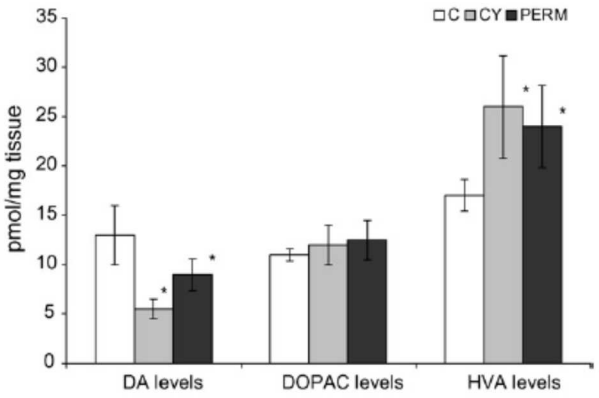


Fig. 4. Levels of dopamine (DA), 3,4-dihydroxyphenylacetic acid (DOPAC), and 3-methoxy-4-hydroxyphenylacetic acid (HVA) in striatum cell of rats from control group (□), CY-treated (▨) and PERM-treated (■) groups. Data are presented as mean ± S.D. for four rats. Measurements were performed in triplicate for each sample. $^{*}P < 0.05$ vs. control (C).

Plasmatic superoxide dismutase (SOD) and gluthathione peroxidase (GPx) activities: Pyrethroid-treatment led to a decrease of plasmatic GPx activity on PND 35 of approximately 15%. No change was measured in SOD activity (see Fiure 5 of publication below).

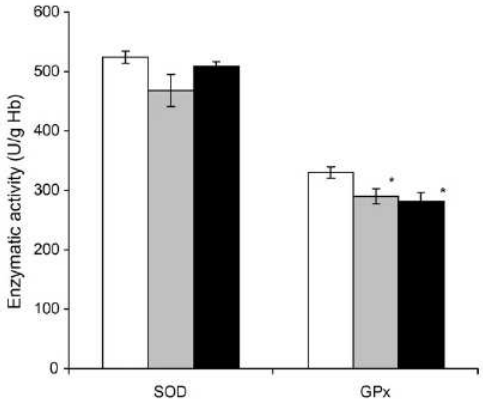


Fig. 5. SOD and GPx blood levels measured in rats, at 35 days of age, from control group (□), CY-treated (▨) and PERM-treated (■) groups. Data are presented as mean ± S.D. for six rats. $^{*}P < 0.05$ vs. control (C).

Protein oxidation (measured as Carbonyl groups) and lipidperoxidation (measured as Oxidation index) in striatum and erythrocytes: A significant increase of protein oxidation in striatum was observed in both pyrethroids-treated groups, whereas no changes in the lipidperoxidation (see Fig. 6 of the publication below) were seen. In erythrocytes no protein oxidation was found but significant increased lipidperoxidation (see Fig. 9 of the publication, below).
Effect on plasma membrane fluidity in striatum and erythrocytes: No physicochemical change of plasma membrane phospholipid fluidity was seen in striatum or in erythrocytes.

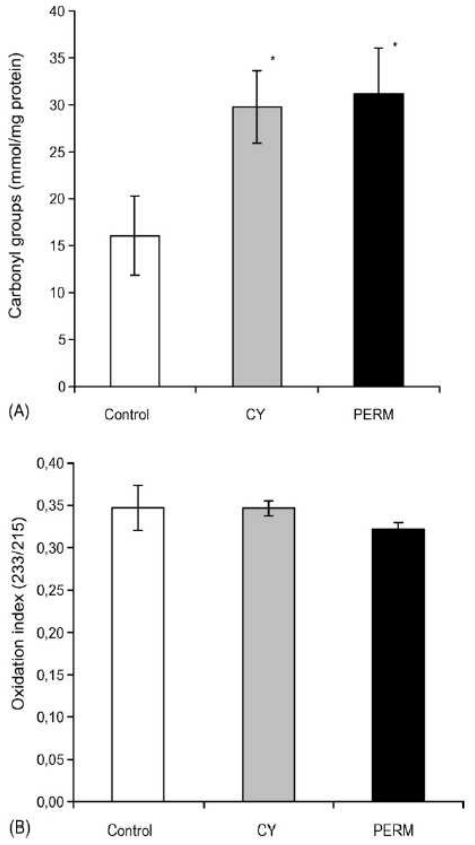


Fig. 6. Effect of pyrethoids on protein carbonyl formation (A) and oxidation index measured on lipids extracted (B) in striatum cells of rats from control group (□), CY-treated ( ) and PERM-treated (■) groups. The results are indicated as mean values ± S.D. for six rats. *$P < 0.05$ vs. control.

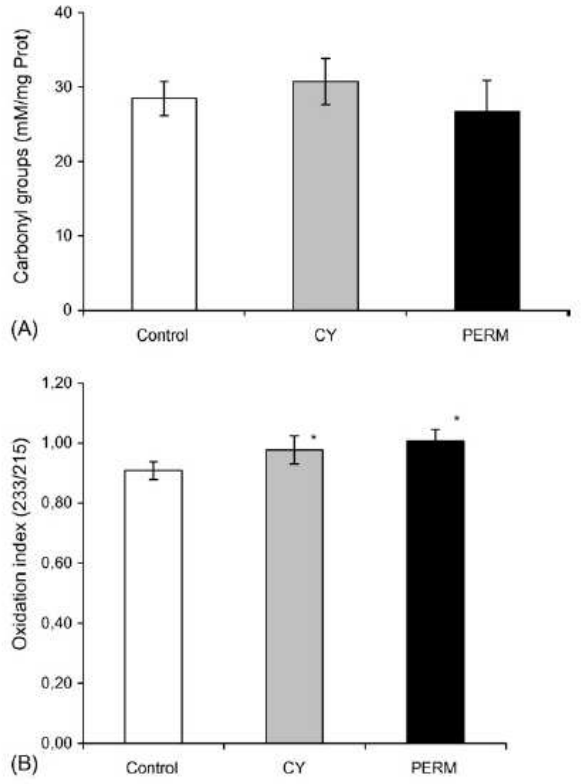


Fig. 9. Effect of pyrethoids on protein carbonyl formation (A) and oxidation index measured on lipids extracted (B) in erythrocytes of rats from control group (□), CY-treated ( ) and PERM-treated (■) groups. The results are indicated as mean values ± S.D. for six rats. *$P < 0.05$ vs. control.

GSH levels in striatum cells (see Fig. 8 of the publication below): Data indicate increased levels in the cypermethrin-treated animals and decreased levels of GSH in the permethrin-treated animals.
Furthermore the respiratory burst behavior in monocytes from rats after activation with PMA showed a decreased superoxide anion production in the monocytes of rats treated with cypermethrin and permethrin (see Fig. 10 of the publication, below).

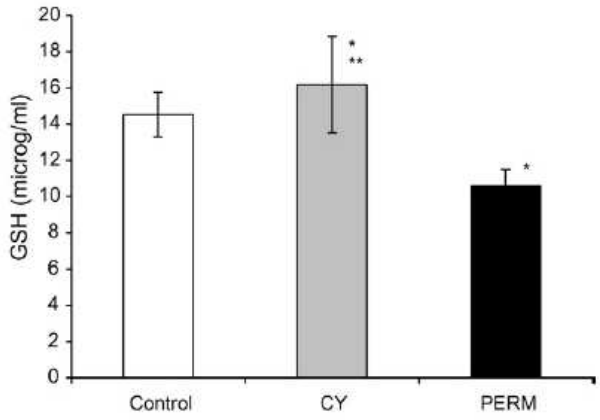


Fig. 8. GSH level in striatum cells of rats from control group (□), CY-treated (▒) and PERM-treated (■) groups. Data represent the means ± S.D. for six rats. $^{*}P<0.05$ vs. control, $^{**}P<0.05$ vs. PERM. For experimental details, see Section 2.

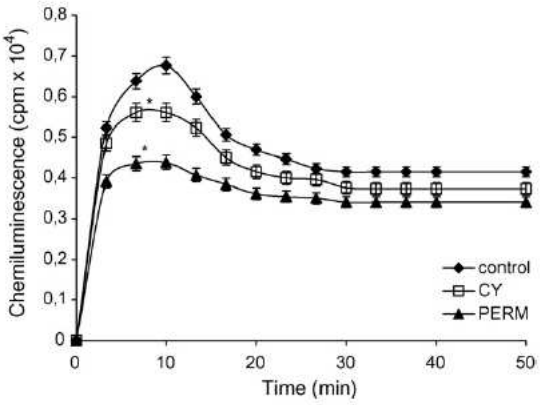


Fig. 10. Time course of lucigenin-amplified chemiluminescence of monocytes in rats from control group (♦), CY-treated (□), and PERM-treated (▲) groups. Monocytes were stimulated with 3×10^{-4} mol/L phorbol myristate acetate. Chemiluminescence was measured as counts per minute (cpm). The results are indicated as mean values ± S.D. for six rats. $^{*}P<0.05$ vs. control.

Conclusion from the author: The increased locomotion and rearing on PND 35 indicates dopaminergic impairment; the lack of increased locomotion on PND 21 indicates possible behavioral alterations by pyrethroids that are not observed directly but later in the life of the animal. From the two hypothesis presented to explain lower dopamin levels in striatum after cypermethrin or permethrin treatment (increased DA turnover and reduced DA biosynthesis) the data suggest an increased catabolic pathway of DA. Accelerated DA turnover induces formation of reactive oxygen species. This is discussed to be in line with reduced blood glutathione peroxidase levels and increased protein oxidation in striatum. No change in lipid peroxidation in striatum indicates that brain antioxidants can partially protect from free readicals produced by DA degradation. GSH levels, as representative for brain antioxidants, were found to be increased for cypermethrin but reduced for permethrin. The authors attributed this to increased hydrophilicity of cypermethrin. A further difference to previous studies was seen with regard to erythrocyte plasma fluidity which shows alterations on adult rats but not in rat pups, although both showed alteration in lipid peroxidation as only damage assessed in erythrocytes. That protein oxidation is observed in striatum but not in erythrocytes is discussed as result of different distribution of pyrethroids in brain and blood and by DA oxidation products that bind to proteins. An immunosuppressive effect evaluated as reduced superoxide anion production in rat monocytes was seen for cypermethrin and permethrin and is suggested to be based on altered signal transduction or NADPH oxidase compex formation. In conclusion the data indicate that neonatal exposure to pyrethroids during the critical period of growth has long-term effects on the biochemistry and behavior of male rats.

Comment from the applicant: The publication shows reporting deficiencies, for example with regard to unclear treatment period (PND 6-15 or PND 8-15, both is mentioned), with regard to number of samples and replicates (some measurements are not quantified), and with regard to clinical observations (interval and the timespan of observation is not mentioned). Furthermore the work shows some methodical deficiencies as the data are produced with cypermethrin of low purity, and with only one dose which hampers differentiation between incidental and treatment related effects, furthermore it is produced with litters of only one sex, which might induce a systemic confounder. MA was performed twice and only over a very short period of time (2 min adaptation, 5 min recording) which is not adequate to demonstrate intra-session habituation. Therefore the significant changes of MA assessment on PND 35 are of questionable relevance, furthermore the data are not discussed in consideration of historic control data. Repeated maternal separation which is needed for direct pup dosing is considered to be a systemic confounder for increased motoractivity and increased number of center entries for Wistar rats [see KCA 5.8.2/9 2015/1001752]. Furthermore some results suggest inconsistencies of the findings so that the reliability of the data is questioned (see figure 8 of the publication: GSH level in striatum shows opposing trends for cypermethrin and permethrin, and for cypermethrin the effect is hardly significant compared to control (although otherwise discussed). Measurement of DA and its metabolites in striatum are expressed in terms of picomoles per gram of tissue, although weighing of striatum is not mentioned in the report.
The immunosuppressive effect described in the study is not confirmed in the Immunotoxicity study performed with alpha-cypermethrin (see CA 5.8.2/2).

However, these data indicate that cypermethrin application to pups from PND6/8 onwards without preexposure of dams does not lead to clinical signs of neurotoxicity at a dose level of 1.4 mg/kg bw. Taking the purity and isomeric composition into account, this indicates that alpha-cypermethrin at a level of 0.44 mg/kg bw is not considered to induce neurotoxicity.
This is taken up in the chapter CA 5.7 when discussing developmental neurotoxicity.

In conclusion, based on the above discussed deficiencies this study is of insufficient quality, furthermore based on the unrealistic and hazard driven exposure technique this study is not considered relevant for human risk assessment.

Classification of study: Supplemental information

Report: CA 5.8.2/10
Flaskos J. et al., 2006a
The effects of Diazinon and Cypermethrin on the differentiation of neuronal and glial cell lines
2007/1070387
Guidelines: none
GLP: no

Executive Summary of Literature

The authors investigated the effects of cypermethrin (in the chapter material and methods alpha-cypermethrin is mentioned, purity: no data) and diazinon (purity: no data) on the differentiation of neuronal (mouse N2a neuroblastoma) and glial (rat C6 glioma) cell lines. Cell differentiation was either induced by incubation in serum-free medium containing 0.3 mM dibutyryl cAMP (N2a cells) or by replacing cAMP with 2 mM sodium butyric acid (C6 cells). Cell differentiation was monitored for 24 h in the presence or absence of test substances (1 and 10 µM). Cells differentiated in the presence and absence of pesticide were fixed, stained with Coomassie Brilliant blue dye, and the outgrowth of axon-like processes in N2a cells and process outgrowth in C6 cells were determined. Cell growth and viability was assessed with the MTT assay and the acetylcholinesterase activity was determined by the method of Ellmann, adapted for a microtiter plate format.
Conclusion of the author: Cypermethrin had no significant effect on process outgrowth in either differentiating neuronal cell line.
Conclusion of the applicant: Both substance (cypermethrin or alpha-cypermethrin) and purity is not clear. Therefore the result is not further considered, however under the conditions of the study no direct effect on neurite outgrowth was seen.

Classification of study: Not further considered

Report: CA 5.8.2/11
Tian Y-T et al., 2007a
Effects of alpha- and theta-Cypermethrin insecticide on transient outward potassium current in rat hippocampal CA3 neurons
2008/1102217
Guidelines: none
GLP: no

Executive Summary of Literature

The authors investigated the effects of alpha-cypermethrin (purity: no data) and theta-cypermethrin (purity: no data) on the transient outward potassium (I_A) channels in freshly dissociated hippocampal CA3 neurons of rat, using whole-cell patch clamp technique. Cypermethrins were first dissolved in DMSO and then in extracellular solution. Cypermethrins were used at concentrations of 1, 10, and 100 nM. The peak amplitudes of I_A were increased about 5%, 9%, and 24% after incubation with alpha-cypermethrin in a dose-dependent manner. In contrast, incubation with theta-cypermethrin decreased the amplitudes of I_A by about 12%, 22%, and 54%, respectively. For analysis of the current–voltage (I–V) relationship, neurons were held at -70 mV. I_A currents were obtained by depolarizing steps from a command potential of -60 to +90 mV at 10 mV steps. Upon the application of alpha-cypermethrin, the amplitudes of I_A currents were enhanced differently at different membrane potential, but the amplitudes of I_A currents at different membrane potential were reduced by theta-cypermethrin (see Fig. 1 of the publication below).

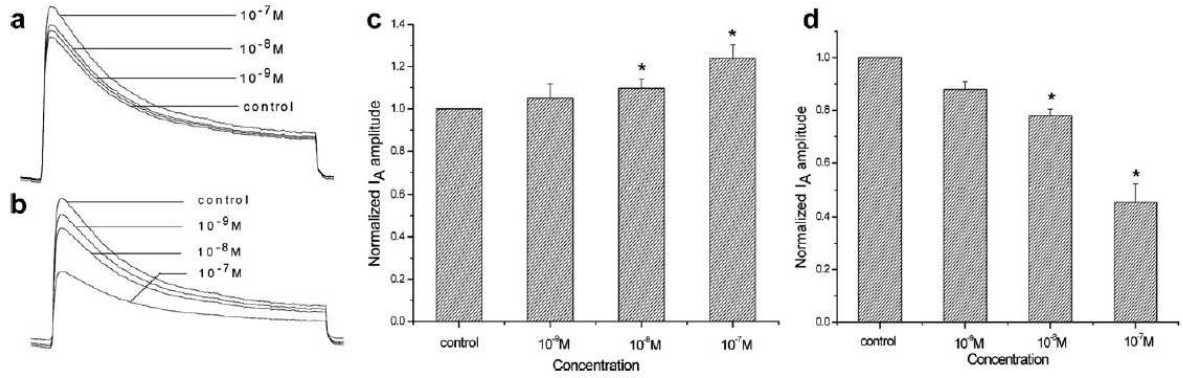


Fig. 1. Dose-dependent effects of alpha-cypermethrin and theta-cypermethrin on I_A. (a) I_A valves were increased at different concentrations of alpha-cypermethrin. (b) I_A valves were decreased at different concentrations of theta-cypermethrin. (c) Dose-dependent effects of alpha-cypermethrin on I_A ($n = 6$, $^*P < 0.05$ *vs* control). (d) Dose-dependent effects of theta-cypermethrin on I_A ($n = 6$, $^*P < 0.05$ *vs* control).

The membrane potential at half-activation of I_A activation curve was significantly shifted to the negative potentials at 10 and 100 nM alpha-cypermethrin and theta-cypermethrin. The membrane potential at half-activation of I_A inactivation curve was significantly shifted to the negative potential at all theta-cypermethrin concentrations but not with alpha-cypermethrin. Theta-cypermethrin and lower alpha-cypermethrin concentrations (1 and 10 nM) had no marked effect on the recovery of I_A from inactivation. Incubation with 100 nM alpha-cypermethrin prolonged the time constants for the recovery from inactivation kinetics of I_A.

Conclusion of the author: These results suggest that alpha-cypermethrin and theta-cypermethrin exhibit different effects on the transient outward potassium channels but are both able to modulate potassium channels in CNS.
Conclusion of the applicant: The report lack information on purity of used substances. However, the data indicate that cis 2-Isomers of cypermethrin (alpha-cypermethrin) and Trans 4 isomers of cypermethrin (Theta-cypermethrin) both act on the outward potassium channel. This is considered as a further proof for the Bridging rationale between cypermethrins. How far these in vitro data are relevant for the invivo situation is unclear.

Classification of study: Supplementary information

Report: CA 5.8.2/12
Tian Y-T et al., 2009a
Effect of alpha-Cypermethrin and theta-Cypermethrin on delayed rectifier potassium currents in rat hippocampal neurons
2009/1130985
Guidelines: none
GLP: no

Executive Summary of Literature

The authors investigated the effects of alpha-cypermethrin (purity: no data) and theta-cypermethrin (purity: no data) on the delayed rectifier potassium current (I_K) in hippocampal neurons of rat, using whole-cell patch clamp technique. Cypermethrins were first dissolved in DMSO and then diluted directly in artificial cerebrospinal fluid. Cypermethrins were used at concentrations of 1, 10, and 100 nM and the final DMSO concentration was <0.1%. Measurements for data analyses were made only at steady-state amplitudes of I_K. Holding potential was -50 mV. I_K was elicited by applying a single depolarizing voltage pulse to +70 mV following a 120-ms prepulse at -110 mV with a 50-ms interval at -50 mV to inactivate I_A. The application of different concentrations alpha-cypermethrin and theta-cypermethrin produced a dose dependent effect on I_K amplitude. A dose-dependent decrease of 26%, 34%, and 40% by alpha-cypermethrin and of 5%, 27%, and 53% by theta-cypermethrin was observed (see Figure 1 of publication below).

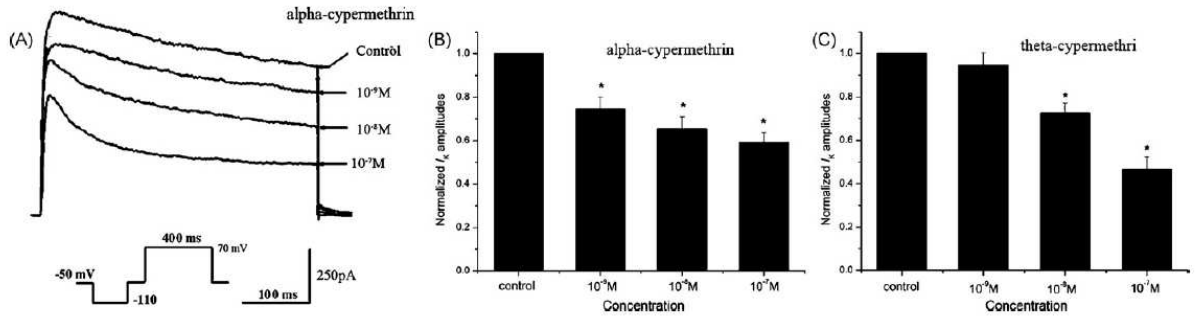


Fig. 1. The effects of cypermethrin on I_K. (A) Original traces of I_K before and after the application of alpha-cypermethrin at different concentrations showing the inhibitory effects of cypermethrin on the steady-state current amplitudes of I_K. Note that various concentrations of cypermethrin were applied to the same neuron. Voltage protocol was exhibited in the inset. (B) Dose-dependent effects of alpha-cypermethrin on I_K ($n = 8$, $^*P < 0.05$ vs control). (C) Dose-dependent effects of theta-cypermethrin on I_K ($n = 8$, $^*P < 0.05$ vs control). Data are presented as means ± S.E.M. ($n = 8$).

The action of alpha-cypermethrin and theta-cypermethrin were quite comparable with regard to reduction of I_K current amplitudes , decrease of slope factor and shift of the membrane potential at half-activation of I_K inactivation curve to the negative potential.
Conclusion of the author: The present study showed that the relative low concentrations of cypermethrin is related to the electrophysiological activities of rat hippocampal CA3 neurons by effects on the voltage gated delayed rectifier K^+ channels. The cypermethrin induced change of K^+ currents in the hippocampal CA3 neurons may contribute to its neurotoxicity by eliciting abnormal neuronal discharge.

Conclusion of the applicant: The report lack information on purity of used alpha-cypermethrin and theta-cypermethrin. However, the data indicate that cis 2-Isomers of cypermethrin (alpha-cypermethrin) and trans 4-isomers of cypermethrin (Theta-cypermethrin) both act in vitro on the delayed rectifier potassium current (I_K) of hippocampal neurons of rat. This is considered as a further proof for the Bridging rationale between cypermethrins. How far these in vitro data are relevant for the invivo situation is unclear.

Classification of study: Supplementary information

Report: CA 5.8.2/13
Wolansky M.J. et al., 2005a
Relative potencies for acute effects of Pyrethroids on motor function in rats
2006/1051134
Guidelines: none
GLP: no

Executive Summary of Literature

The objective of this report was to characterize individual dose-response curves for in vivo motor function of male Long-Evans rats (age: 55-57 days) as a chosen endpoint and calculate relative potencies for the used pyrethroids (cypermethrin, deltamethrin, beta-cyfluthrin, esfenvalerate, lamda-cyhalothrin, fenpropathrin, resmethrin, S-bioallethrin, permethrin, bifenthrin and tefluthrin). Cypermethrin (Cis/trans: 48.7%/51.3%; purity: 88%) was investigated in 8-18 animals (unclear individual number of animals for each pyrethroid) at 6 different doses between 0.1 – 120 mg/kg bw in corn oil by gavage (dosing volume 1 mL/kg). Motor activity was measured for 1 h using figure-eight mazes, each consisting of a series of interconnected alleys converging on a central arena and covered with transparent acrylic plastic. Testing was performed at the time of peak effects (1.5 h for cypermethrin). No signs of excessive toxicity were observed in the animals treated with cypermethrin. All tested pyrethroids induced dose-dependent decreases in motor activity. ED_{30} values (dose at which reduction of 30% total motor activity in figure-eight maze compared to the corresponding vehicle-treated control group was observed) ranged from 1.2 to 292 mg/kg bw. Cypermethrin showed an ED_{30} of 10.7±1.34 mg/kg bw. The Threshold dose (defined as the highest no-effect dose level at which treated rats would respond with 100% control performance) was calculated to be 4.3±1.14 mg/kg bw.

Comment from the applicant: This publication has been discussed in the RAR of Lambda-Cyhalothrin and was mentioned to lack detailed data as number of animals per dose and detailed results per dose level. The RMS (Sweden) concluded that the study is considered as supporting data only.
Furthermore EPA used this study in the assessment of alpha-cypermethrin as the most robust data set for extrapolating risk for the entity of cypermethrins after conducting a benchmark dose analysis (BMD). The argumentation was that the design of the Wolansky study with measured motor activity at the time of peak effect after exposure to several doses (6 for cypermethrin) in at least 8 animals /group minimize variability and increase the confidence in the body mass dose estimates. Moreover, each pyrethroid was evaluated by the same scientist thus decreased some of the variability associated with neurobehavioral measures.

After BMD analysis the acute dietary risk assessment was performed using the $BMDL_{1SD}$ value of 7.16 mg/kg bw, based on decreased locomotor activity at the BMD_{1SD} value of 11.20 mg/kg bw. BMD_{1SD} is the central estimate of the dose that results in decreased motor activity compared to control animals based upon a 1 standard deviation using Benchmark Dose Analysis, whereas $BMDL_{1SD}$ is the 95% lower confidence limit of the central estimate. EPA further concluded that there is no increase in hazard from repeated exposures to the cypermethrins; therefore, the acute dietary exposure assessment is protective of chronic dietary exposures since acute exposure levels are higher than chronic exposure levels. Accordingly, a dietary exposure assessment for the purpose of assessing chronic dietary risk was not conducted.
This study was used in the context of this submission to demonstrate similarity of potencies within the group of cypermethrins.

Classification of study: Supplementary information

Report: CA 5.8.2/14
Scollon E.J. et al., 2008a
In vitro metabolism of Pyrethroid pesticides by rat and human hepatic microsomes and cytochrome P450 isoforms
2009/1130987
Guidelines: none
GLP: no

Executive Summary of Literature

The objective of this study was to determine the intrinsic clearance (CL_{int}), a measurement of metabolic rate of several Type I and II pyrethroids in rat and human hepatic microsomes; to assess the relative role of oxidative and hydrolytic pathways of these pyrethroids; to investigate the potential interaction of diastereomers of permethrin at the metabolic level; and to screen rat and human P_{450} isoforms for their ability to metabolize pyrethroids. Amongst others, cypermethrin (cis/trans ratio 49/51, purity: > 98%) was used in the experiments.
3 pools of rat microsomes were prepared from 18 male Long-Evans rats (age: 70 days, weight: 275-299 g). Pooled human adult liver microsomes were purchased from CellzDirect (mixed gender pool of 15), Cedra (mixed gender pool of 15), and XenoTech (mixed gender pool of 50). Furthermore supersomes containing specific rat or human P_{450} isoforms were used. Estimates of K_m and V_{max} were determined in rat hepatic microsomes for bifenthrin, S-bioallethrin, bioresmethrin, cis-permethrin, trans-permethrn, and a 40:60 cis/trans permethrin mixture. This determination was done to ensure pyrethroids concentrations in the parent depletion assays described below were << K_m. The assays were conducted in duplicate for each of the three pools of microsomes by measuring the disappearance of parent compounds over 10 min. Assays were initiated by the addition of pyrethroid in 25 µL of methanol to a 3.5-ml incubation mixture containing NADPH at time 0 to attain final pyrethroid concentrations of 0.1, 1, 5, 10, 20, or 50 µM. At time 0, 2, 6, 8, and 10 min, 250 µL aliquots were removed from the mixture for analytical determination. This assay revealed general K_m values between 3.7 and 31.1 µM for the different pyrethroids. Therefore determination of metabolic rate constants in rat and human microsomes were than determined at a concentration of 0.5µM (except for S-bioallethrin: 1µM).
Assays to assess the microsomal hydrolysis of the pyrethroids were conducted following the assay principle described above but in the absence of NADPH. Each incubation, with or without NADPH, was conducted in duplicate using the three different pools of microsomes (n = 3).
Result: The calculation of metabolic rate constants and intrinsic hepatic clearance rates of cypermethrin, determined by summation of all the co-occurring isomers, revealed a higher clearance in the rat relative to human hepatic microsomes. Furthermore rat hepatic microsomes show mainly oxidative (85%) and to a minor extend hydrolytic (15%) cleavage. In contrast human hepatic microsomes show mainly hydrolytic cleavage of cypermethrin (see Table from the publication, below).

CL_{int} and percentage oxidative and hydrolytic metabolism of Type II pyrethroids from rat and human hepatic microsomes

Pyrethroid	Isomer	Species	CL_{int}	Percentage Oxidative	Percentage Hydrolytic
Cypermethrin	49% cis	Rat	284 ± 17	85	15
	51% trans	Human	104 ± 63	N.S.[b]	100

N.S., no significant difference.
[b] The clearance values in the presence or absence of NADPH were not significantly different as assessed by an unpaired t test, $p < 0.05$.

Of the P450 isoforms examined, rat CYP1A1, CYP2C6, CYP2C11, CPY3A1, and CYP3A2 and human CYP 2C8 and CYP2C19 have the most Cypermethrin-metabolizing activity.
Conclusion of the author: Cypermethrin metabolism in rat is mainly oxidative, whereas in human hepatic micorsomes mainly hydrolytic cleavage is observed.
Classification of study: Supplementary information

Report: CA 5.8.2/15
Crow J.A. et al., 2007a
Hydrolysis of pyrethroids by human and rat tissues: Examination of intestinal, liver and serum carboxylesterase
2007/1070525
Guidelines: none
GLP: no

Executive summary

In this study the distribution and activities of esterases that catalyze pyrethroid metabolism have been investigated in vitro using several human and rat tissues, including small intestine, liver and serum. Alpha-cypermethrin (99% pure, mixture of isomers) was used together with other pyrethroids. The pyrethroid hydrolysis reactions in human and rat tissue microsomes and cytosols were performed after pre-incubation of pyrethroid substrate with buffer and addition of subcellular fraction at a final protein concentration of 0.5 mg/ml. After 15 or 30 min incubations, a time in which product formation is linear, the reactions were quenched and the hydrolysis products were quantified by HPLC. The different subcellular fractions with the CE protein fractions were either purified or purchased.
Result:
Intestine: The major esterase in human intestine is carboxylesterase 2 (hCE2) located in the microsomal fraction. This fraction was able to effectively hydrolyse trans-permethrin, but not bioresmethrin or deltamethrin. Alpha-cypermethrin was not investigated. Rat intestinal fraction (with CE activity located both in the microsomal and cytosolic fraction) was 4-5 times less active than human intestine carboxylesterases.
Liver: hCE1 and hCE2 were clearly present in both liver fractions. A comparison of hydrolytic kinetic parameters for rat and human hepatic microsomes and cytosol toward trans-permethrin is shown in Table 1 of the publication (below).

Table 1
Kinetic parameters of *trans*-permethrin hydrolysis catalyzed by human and rat hepatic subcellular fractions (microsomes and cytosols)[a]

Species (fraction)	Trans-permethrin		
	K_m (μM)	V_{max} (nmol/min/mg)	CL_{int}[b] (nmol/min/mg/mM)
Rat (microsomes)	19.2±3.2	1.3±0.3	67.7
Rat (cytosol)	24.6	0.56	22.8
Human (microsomes[c])	20.7±3.0	1.1±0.1	53.1
Human (cytosol[d])	3.4±0.8	0.47±0.02	138.2

[a] Rat microsomes and cytosol (pooled) were prepared from another study (Ross et al., 2006). Values (±SE) were obtained by non-linear regression of kinetic plots. The microsomal kinetic data are from Ross et al. (2006). The cytosol kinetic data are from this study.
[b] $CL_{int} = V_{max} / K_m$.
[c] Pooled sample (n=18 individuals).
[d] Pooled sample (n=20 individuals).

The data demonstrate that microsomal degradation is very similar in rat and human, but for cytosol the Km value is much lower in human fraction than in the rat fraction and the intrinsic clearance (CL_{int}) is higher.
Serum: Rat serum possesses 4% of the total hydrolytic capacity in the rat, while human serum did not show any hydrolyzing capacity. The capacity to hydrolyse alpha-cypermethrin was investigated in rat serum at a quite high pyrethroid substrate concentration of 50 µM and specific activity was quite low (<1nmol/min/mL serum)
Conclusion of the author: While the hepatic CEs undoubtedly contribute the bulk of pyrethroid hydrolysis, the major difference between rats and humans when attempting to extrapolate animal studies of pyrethroid exposure is the possibility in rats to hydrolyse pyrethroids via serum CEs.
Conclusion of the applicant: The rather poor metabolic capacity of alpha-cypermethrin in rat serum is not considered a relevant degradation process.

CA 5.8.3 Endocrine disrupting properties

Studies evaluated in the draft monograph of rapporteur member state Belgium of Sep. 1999:
The evaluation of potential endocrine disruption was not a data-requirement at the time of Annex I inclusion of alpha-cypermethrin. However, the regulatory data package of alpha-cypermethrin neither indicates a potential to affect the estrogen or androgen system nor the pituitary-thyroid axis.

The available repeat dose toxicology studies, used to support the pesticide registration for alpha-cypermethrin, showed almost always adverse effects limited to lowered body weight and food consumption, clinical signs of neurotoxicity, and signs of skin irritation. Liver weight increases were frequently seen but were suggested to be the result of an adaptive response due to microsomal enzyme induction. Testes weight changes were seen at doses above the LOAEL in combination with reduced body weight and were, without histopathological correlate, suggested to reflect body weight changes. Increased relative adrenal weight changes were only observed once, in the interim sacrifice in the 78 week mice study, also here as result of reduced body weight. No study revealed any treatment-related histopathological findings in adrenals, epididymides, ovaries, pituitary, prostate, seminal vesicles, thyroid with parathyroids, uterus with cervix, vagina and testes. It is worth to be mentioned as especially the 90 day studies in mice (AL-425-006) and rat (AL-425-007) were histopathologically investigated 1993 and 1994 by [redacted] author of numerous papers on male rodent reproductive pathology. In addition there was no evidence for hormonally induced carcinogenicity in life time studies in rats and mice with cypermethrin and alpha-cypermethrin.

Fertility studies with alpha-cypermethrin (Gharda study), even if poorly documented as stated in the monograph, did not show any adverse effect on parturition, reproduction and lactation response. There was no difference in the total number of pups born to control and treated P0, P1 and P2 mothers. Fertility index and litter size was unaffected. Based on reported “delayed pregnancy in this study in the mid and high dose”, it was concluded that the NOAEL of this study is 5 mg/kg bw/day. In the discussions during the ECCO Meetings the RMS stated that the delayed pregnancy finding was a mis-interpretation. Therefore the overall NOAEL for reproductive properties for Alpha-cypermethrin was > 20 mg/kg bw/d in the Review Report 2004 and it was referred to the cypermethrin dietary multigeneration study which reassures the lack of reproductive effects.
The multigeneration study with cypermethrin listed in the monograph showed lower litter sizes and weights and lower pup weights at parental toxic doses mainly for the first generation at 500 ppm. There were no increases in these effects over the three generations. Fertility was not affected at any dose in any generation. At 10 and 100 ppm there were no adverse effects on parental rats and their litters of any generation. Histopathological investigations in the F2 generation and F3B weanlings showed no pathological changes in ovaries, testes and prostate/uterus, thyroid and parathyroid, pituitary, and adrenals. Based on missing effects on fertility in both studies further studies on sperm parameters were not considered to be required.

In summary, taken the complete set of toxicology studies with alpha-cypermethrin, supplemented with existing cypermethrin studies, there is no indication for human relevant endocrine related effects.

Studies submitted in this AIR 3 dossier (not yet peer-reviewed):

The literature search revealed a huge dataset on studies investigating endocrine related effects of cypermethrin, beta-cypermethrin and alpha-cypermethrin as well as metabolites thereof.

Based on the weight of evidence, alpha-cypermethrin is not predicted to have androgenic or anti-androgenic activity:
Cypermethrin has been repeatedly tested to have no androgenic activity *in vitro*. Although there is evidence for weak anti-androgenic activity *in vitro* for cypermethrin, this is not seen with beta-cypermethrin. Based on the isomeric composition of cypermethrin (8 isomers), beta-cypermethrin (4 isomers of cypermethrin) and alpha-cypermethrin (2 isomers (only cis-2-isomers) of beta-cypermethrin), alpha-cypermethrin is considered more comparable with beta-cypermethrin and therefore is not predicted to have androgenic or anti-androgenic activity in vitro. There is evidence that the anti-androgenic activity of cypermethrin is attributable largely to metabolites. As alpha-cypermethrin consists entirely of *cis*-isomers, which are less susceptible to metabolism, similar activity is not predicted. Mechanistic data indicate that the effects of cypermethrin may not be due to a direct interaction with the androgen receptor.
In vivo, Wu et al., (2008) and Zhang et al (2008) report a week positive response in a Hershberger assay with cypermethrin but not for beta-cypermethrin. Also here, based on the isomeric composition of cypermethrin, beta-cypermethrin and alpha-cypermethrin, a positive response in a Hershberger assay is not predicted for alpha-cypermethrin. However, a number of studies report effects on the testes (weight changes, histopathology and changes in histoarchitecture), spermatogenesis, circulating levels of testosterone, LH and FSH and effects on other tissues of the male reproductive tract. The majority of these studies are limited in some way by the use of inappropriate histopathological techniques or diagnosis; dosing with commercial formulations containing high levels of organic solvent; the use of high dose levels causing significant general toxicity or the absence of information on general toxicity; or the use of intraperitoneal dosing. Nevertheless findings cannot be entirely discounted based on the consistency of responses reported. However, effects do not appear to be characteristic of an endocrine mode of action, but are considered more likely to represent general non-specific testicular toxicity due to oxidative stress. Effects are seen at relatively high dose levels of cypermethrin and beta-cypermethrin in a 15day adult intact rat assay and are associated with general systemic toxicity.
The question remains, if the non-specific testicular toxicity as well as the impairment of sperm parameters is already induced by alpha-cypermethrin, which in general needs to be dosed lower based on the higher neurotoxicity. This was addressed in a separate study with alpha-cypermethrin, which is summarized in detail below. Based on these data, alpha-cypermethrin has proven to induce no effect on reproductive organ weights and histopathology in male rat reproductive organs up to the highest tolerable dose in this assay. In addition no effects on sperm parameters were shown (sperm motility, sperm number and morphology were unaffected). This is thereby the proof that the cis-2 isomers of alpha-cypermethrin alone are not sufficient to induce reproductive organ toxicity even under the worst case scenario of repeated bolus administration.

There is limited evidence for the oestrogenicity(less anti-oestrogenicity) of cypermethrin *in vitro*, and also the evidence for oestrogenicity of alpha-cypermethrin in vitro is restricted to two cell proliferation assays (E-Screen assay) of low comparability showing some proliferative effect. A potential induction of proliferation of MCF-7 cells is not directly attributable to an estrogenic effect; therefore the relevance of this information is questionable. Also here, studies with metabolites indicate that estrogenic effects may be due to metabolites generated in vitro, rather than to the parent substance. Furthermore the respective highly potent metabolites were not formed in vivo with alpha-cypermethrin and furthermore the lower metabolic breakdown of alpha-cypermethrin reduces the potential of activation via metabolism. Anyway, *in vivo*, no positive uterotrophic effect is noted for alpha-cypermethrin in rats and in mice reported by Liang et al., 2005.

Only one study investigated the thyroid receptor interaction of Cypermethrin and its metabolites 3-PBA and DCVA and showed only weak or even no antagonistic effects in the 10 µM dose range, therefore the impact is considered of minor relevance for the human risk assessment.

The aromatase and steroidogenesis assays showed no convincing evidence for an effect of cypermethrin. In vitro-assays with relatively high concentrations of questionable physiological relevance indicated a potential direct effect of cypermethrin but not of M310I011 (3-PBA) on sperm motility.

In order to address potential effects on sperm parameters in a proven sensitive study (positive for cypermethrin and beta-cypermethrin), a 15 day adult male rat assay was performed. The dose levels were chosen in a way that no significant systemic toxicity was induced but that dose levels used in the cypermethrin and beta-cypermethrin were reflected with regard to the underlying alpha-cypermethrin amount. Thereby the study design is considered appropriate to get the information if cis-2 isomers are responsible for the formerly described effects on sperm parameters. This study together with the respective range finder is summarized in detail within the chapter CA 5.8.3/50 and below in short.

Table 5.8.3-1: Summary of not yet reviewed alpha-cypermethrin screening studies

Study	Dosages (mg/kg bw/ day)	NOAEL (mg/kg bw/day)	Main adverse effect	Reference
15-day adult male rat assay Wistar rat Vehicle: Corn oil	M: 2.0, 3.5, 6.6	M: 6.6	none	[redacted] 2014 2014/1289319 RF & 2014/1275120 see CA 5.8.3/50 & 51

Overall there is no convincing evidence for a potential of endocrine activity of alpha-cypermethrin in *vivo* up to dose levels not also causing significant systemic toxicity. A potential non-specific testicular toxicity has been investigated for alpha-cypermethrin in a test system which showed positive outcome for beta-cypermethrin and cypermethrin. This study showed no effects on sex organ weight parameters, no effect on sperm parameters and no indication of any effect on Androgen receptor density. Thereby it is appropriate to conclude that alpha-cypermethrin is neither endocrine active nor does it show testicular toxicity up to maximal tolerable dose levels.

Based on weight of evidence from the available studies and the literature data, the following input in the Annex I listing of alpha-cypermethrin is considered reasonable.

Other toxicological studies (SANCO/11802 data point 5.8)	
Endocrine disrupting properties	No endocrine effects on the oestrogen, androgen or thyroid hormone system.

For convenience of the reviewer brief summaries of the respective peer-reviewed studies used from the monograph as well as summaries from the literature data are provided below. Furthermore the 15 day adult male rat assay is described in detail.

Overview of already peer reviewed studies

RAT studies (see Table 5.8.3-2)

5 Week Feeding Study with WL 85871 (Alphacypermethrin) in rats ([redacted] 1982; SBGR.81.212; DocID: AL-420-003; see CA 5.3)

Alphacypermethrin was given to groups of 10 male and 10 female Wistar rats for 35 days at dietary concentrations of 25, 100, 200, 400, and 800 ppm. The NOAEL was determined to be 200 ppm on the basis of clinical signs of neurotoxicity, effects on body weight and food consumption, and hematology at 400 ppm. Mortality occurred at 800 ppm. Testes weights adjusted for initial body weight were reduced in the highest dose but showed no difference to control after adjustment for terminal body weight, suggesting they reflected bodyweight differences. In female rats no endocrine related organs were investigated. Macroscopic and microscopic investigations were done on testes, seminal vesicles, prostate and epididymides. Two animals in the 800 ppm dose group with general thin or small appearance showed small seminal vesicles (#035, 042), one in addition small epididymides and prostate (#035). In absence of any histopathological changes in the respective organs these changes were considered to be not specific treatment related effects. Therefore, it is concluded that alpha-cypermethrin did not induce substance-related endocrine effects at any dose.

6-Week range finding feeding study in the rat [redacted] 1993; SBTR.93.002; DocID: AL-420-006 see CA 5.3)

Alphacypermethrin was given to groups of 5 male and 5 female Crl: CD BR rats for 42 days at dietary concentrations of 50, 200, 800, and 1200 ppm. Rats of both sexes fed 1200 ppm and males fed 800 ppm were sacrificed during weeks 2 to 4 because of severe treatment related clinical signs. The NOAEL was determined to be 200 pm on the basis of severe clinical signs (hunched posture, piloerection, unkempt appearance), clinical signs of neurotoxicity (high stepping gait, splayed gait, hypersensitivity) and decreased body weights at 800 ppm. Neither absolute nor relative adrenal, testes or ovaries weights were changed.

There were no macroscopic or microscopic treatment-related findings in endocrine organs including uterus. Therefore, it is concluded that alpha-cypermethrin did not induce substance-related endocrine effects at any dose.

A 90-day feeding study in rats ([redacted] 1982; SBGR.81.293; DocID: AL-425-003 see CA 5.3)

Alphacypermethrin was administered to groups of 20 male and 20 female Wistar rats for 90 days at dietary concentrations of 20, 60, 180, and 540 ppm. The NOAEL was 180 ppm, based on decreased body weights and clinical signs of neurotoxicity at 540 ppm. Absolute testes weights were unchanged although relative testes weights in the highest dose group at 540 ppm were increased. Since the terminal bodyweights were reduced in this group about 8.6%, this change is considered to be a direct effect of growth retardation and not a true biological effect on the organ. Macroscopically there were two males in the high dose with reddened testes, seminal vesicles appeared normal. Histopathology of testes revealed no lesions in 18 of 20 animals, one animal with bilateral interstitial oedema and one with interstitial congestion in the high dose. Seminal vesicles were unremarkable in all treated animals. 4 females showed small cysts of no specific kind in ovaries at the highest dose. No other organs showed treatment-related macroscopic or microscopic effects. Based on this study alpha-cypermethrin did not induce substance-related endocrine effects at any dose.

A 90-day feeding study in rats ([redacted] 1994; SBTR.93.050; DocID: AL-425-007, see CA 5.7/6)

Alphacypermethrin was administered to groups of 15 male and 15 female Crd:CD (Sprague Dawley) BR rats for 90 days at dietary concentrations of 50, 250 and 500 ppm. The NOAEL was 50 ppm, based on decreased body weights and changes in hematology and clinical chemistry at 250 ppm. Absolute testes weights were unchanged although relative testes weights in the highest dose group were increased. Since the terminal bodyweights were reduced in this group (-8.8 %), the changes in relative weights of the testes were considered to reflect the bodyweight changes. This was supported by the microscopic investigations on epididymides, prostate, seminal vesicles and testes, which did not reveal substance-related effects at any dose. Furthermore, no substance related effects were seen in adrenals, thyroid, or female sex organs (ovaries and uterus). Based on this study alpha-cypermethrin did not induce substance-related endocrine effects at any dose.

2 year feeding study of WL43467 in rats ([redacted] 978; TLGR.0189.78; DocID CY-427-001 / addenda: [redacted] 1979; DocID CY-427-002 / [redacted] 1981; DocID CY-427-003 / [redacted] 1985; DocID CY-427-004 see CA 5.5)

This study was performed with Cypermethrin which contains approximately 25% alpha-cypermethrin. The rat cancer study has been used worldwide to fulfil the regulatory requirement for a cancer study in a second rodent species.

Several different groups of male and female Wistar rats received dietary concentrations of 1, 10, 100, and 1000 ppm Cypermethrin. A control group of 48 animals per sex received untreated diet. Groups of rats were scheduled for necropsy after 6 months (6/sex treated, 12/sex control), 12 months (6/sex treated, 12/sex control), 18 months (12/sex treated, 24/sex control) and 2 years (24/sex treated, 48/sex control) of treatment. No significant differences in survival were seen between treatment and control groups. There were no treatment-related effects on hematology, clinical chemistry and organ weights. Absolute testes weights were reduced in the 6 month interim high dose group but not in the 12, 18 or 24 month male rats, and without histological evidence of damage, the effect is not considered to be of toxicological significance. Macroscopic and histopathological examination did not indicate any toxicologically significant lesions in any organ or other effects that were considered related to treatment. The NOAEL was 100 ppm, based on decreased body weights in both sexes at 1000 ppm. No substance-related endocrine effects were observed at any dose.

Oral (gavage) rat developmental toxicity (teratogenicity) study [redacted] 1994; SLN/2/92; DocID: AL-432-002 see CA 5.6)

The effects of Alphacypermethrin on the pregnancy and embryonic or fetal development of the rat were investigated at 0, 3, 9, 18/15 and 15 mg/kg/day administered in corn oil by gavage from day 6 to day 15 post mating. Administration of Alphacypermethrin via gavage at 15 or 18 mg/kg/day to pregnant rats during fetal organogenesis elicited maternal toxicity characterized by changes in clinical conditions and reduction in food consumption and body weight gain during the dosing period. Further, a slight reduction in fetal weights was observed at these dose levels and was attributed to the observed maternal toxicity. The NOAEL for maternal toxic effects was 9 mg/kg/day, based upon clinical signs and reduced body weights at 15 mg/kg/day. The NOAEL for embryotoxic effects was also 9 mg/kg/day, based upon reduced fetal weights at 15 mg/kg/day. No substance-related endocrine effects were observed at any dose.

Toxicity studies on the insecticide WL 43467 (Cypermethrin): A three generation reproduction study in rats [redacted] 1978; TLGR.0188.78; DocID CY-430-001, and addenda [redacted] 1979; DocID CY-430-002 / [redacted] 1979; DocID CY-430-003 / [redacted] 1979; DocID CY-430-004 / [redacted] 1985; DocID CY-430-005)

This study was performed with Cypermethrin, which contains approximately 25% Alphacypermethrin. The rat reproduction and development study with Cypermethrin has been used worldwide to fulfil the regulatory requirement for a multi-generation fertility and development study for Alphacypermethrin.

Groups of approximately 30 Wistar rats per sex in each generation received Cypermethrin at dietary concentrations of 0, 10, 100, or 500 ppm throughout the entire experimental period up to weaning of F3 litters (pre-mating, mating, gestation and rearing). Two litters were produced from each parental generation.

Significantly reduced body weights and food consumption were observed in males and females of the parental generation at the 500 ppm level prior to mating. Minor reproductive effects were recorded at 500 ppm, comprising lower litter sizes and weights and lower pup weights, mainly for the F0-generation. At 10 and 100 ppm, no treatment-related effects on parents or litters at any generation were observed.

Macroscopic and microscopic investigations were performed on ovaries, testes and prostate/uterus, thyroid and parathyroid, pituitary, and adrenals of rats of F0, F1 and F2 parents as well as on F3B weanlings. No compound-related gross or microscopic pathological findings were observed. Especially the endocrine organs of the F3B generation at 500 ppm showed no abnormality in all examined pups. The NOAEL for parental animals was 100 ppm, based upon reduced food intake and body weight at 500 ppm during the pre-mating period. The NOAEL for reproduction and developmental effects was also 100 ppm, based upon reduced litter size at birth and reduced mean pup weights on day 21 for F1b females and F3B males. No substance-related endocrine effects were observed at any dose.

MICE studies (see Table Table 5.8.3-3)

Alphacypermethrin: preliminary toxicity study by dietary administration to CD-1 mice for four weeks [redacted] 1993; Report No.: 92/0346; DocID: AL-420-005) see CA 5.3

Groups of eight male and eight female CD-1 mice received Alphacypermethrin via the diet, at concentrations of 200, 400, 800, 1200, or 1600 ppm for 29 days. The NOAEL was 400 ppm, based on reduced body weight gains in females and reduced terminal body weight in males and females receiving 800 ppm. Absolute and relative weights of adrenal, testes or uterus were unchanged up to the highest dose of 1600 ppm. Macroscopic or microscopic evaluations on endocrine organs were not performed.

Alphacypermethrin preliminary toxicity study by dietary administration to CD-1 mice for 13 weeks [redacted] 1984; Report No.: 92/SHL009/0849; DocID: AL-425-006) see CA 5.3

Alphacypermethrin was fed to groups of 12 male and 12 female CD-1 mice at dietary concentrations of 50, 250, or 1000 ppm in the diet. The NOAEL was 50 ppm, based on decreased body weight gains, skin crusts, and increased aspartate aminotransferase activity at 250 ppm. At 1000 ppm relative testes weights were increased about 28 %, whereas the abs. weight was unaffected, suggesting this reflects bodyweight reduction (-23%). Other endocrine organs as adrenals and uterus were unaffected. Macroscopic and microscopic evaluations on adrenals, epididymides, ovaries, pituitary, prostate, testes, thyroid with parathyroid, and uterus revealed no substance-related endocrine effects.

Alphacypermethrin: Oncogenicity study by dietary administration to CD1 mice ([redacted] 1996; Report No.: 95/SHL010/0596; DocID AL-428-002, and amendment: [redacted] 1999; DocID AL-428-003) see CA 5.5

Groups of 72 male and 72 female CD-1 mice received diets containing 0, 30, 100, and 300 ppm of Alphacypermethrin. Interim sacrifices of 20 males and 20 females of each group were made after 52 weeks. The remainder of the animals were sacrificed after 78 weeks of treatment. The NOAEL was 30 ppm, based on decreased body weight and changes in appearance (ungroomed coats). Rel. adrenals weights in the interim group males showed some increase which was based on missing effect on absolute weights and without histopathological findings considered to reflect body weight changes. No inter-group differences in organ weights for absolute and relative testes were noted. Macroscopic and microscopic evaluations were performed on adrenals, epididymides, ovaries, pituitary, prostate, seminal vesicles, testes, thyroid, uterus and vagina. . There were no macroscopic changes in animals killed after 52 and 78 weeks of treatment which were considered to be related to treatment. No treatment-related neoplastic or non-neoplastic findings were observed during histopathological examination. Therefore, no substance-related endocrine effects were observed at any dose.

DOG studies

13 week oral dietary toxicity study in dogs ([redacted] 1984; Report No.: 3197; DocID: AL-425-005) see CA 5.3

Groups of 4 male and 4 female Beagle dogs received diets containing 0, 30 and 90 ppm of Alphacypermethrin while a fourth group comprised of 6 males and 6 females received 270 ppm test substance for up to 13 weeks. The NOAEL was 90 ppm, based upon clinical signs of neurotoxicity at 270 ppm. Organ weights were taken for adrenals, ovaries, pituitary, prostate/uterus, testes and thyroids. Histopathological examination of the listed tissues was undertaken for all the animals on the study and did not reveal any changes attritubed to alpha-cypermethrin. Lesions recorded like for example immature testes were considered to fall within the range of background pathology commonly seen in 6-12 month old Beagle dogs. It was concluded, that no substance-related endocrine effects were observed at any dose.

52 week oral (dietary) toxicity study in dogs ([redacted] 1995; Report No.: 11110; DocID: AL-427-001) see CA 5.3

Groups of 4 male and 4 female Beagle dogs were dosed orally via the diet, 7 days/week, for 52 consecutive weeks at concentrations of 0, 60, 120, or 240 ppm Alphacypermethrin. The NOAEL was 60 ppm, based on clinical signs of skin irritation in one female receiving 120 ppm Alphacypermethrin. Organ weights were taken for adrenals, ovaries, pituitary, prostate/uterus, testes and epididymides and thyroids with parathyroids. Histopathological examination of the listed tissues was undertaken for all the animals on the study. An incidental change was high uterus weights in females receiving 120 pppm. following absolute ($P<0.05$) and covariance analysis ($P< 0.001$). However, no dose-response relationship was evident. Moreover, because dogs may be in different stages of oestrus at the time of sacrifice, uterine weights may show normal biological variation. No substance-related endocrine effects were observed at any dose.

Table 5.8.3-2: Repeated dose studies in rats with organ weight investigations /histopathology performed on endocrine organs

Doc ID Study type Strain Pathologist Date	AL-420-003 5 week RF Wistar rats [redacted] Dezember 81	AL-420-006 6 week RF Crl:CD BR rats [redacted] Dezember 93	AL-425-003 90day rat SPF albino rats [redacted] August 82	AL-425-007 90day rat Crd:CD(SD)BR [redacted] April 94 (not submitted)	CY-427-001 (& addenda) 24month Wistar rats [redacted] Dezember 78
Administration	diet	diet	diet	diet	diet
Doses ppm	0-25-100-200-400-800 ppm	0-50-200-800 (♀only)	0-20-60-180-540 ppm	0-50-250-500 ppm	0-1-10-100-1000 ppm
NOAEL LOAEL	200 ppm 400 ppm	200 ppm 800 ppm	180 ppm 540 ppm	50 ppm 250 ppm	100 ppm 1000 ppm
Endocrine organ weights measured	Testes	Adrenals, testes, ovaries,	Testes	Adrenals, testes, ovaries	Testes
Histopathology performed on:	Epididymis, seminal vesicles, prostate, testes	Adrenals, ovaries, testes, uterus	Adrenals, epididymides, ovaries, pituitary, prostate, seminal vesicles, testes, thyroid, uterus	Adrenals, epididymis, ovaries, pituitary, prostate, seminal vesicles, teste, thyroid, uterus	Adrenals, ovaries, pituitary, prostate (seminal vesicles stored for reference), testes, thyroid/parathyroid, uterus,
Male					
Dose with sign. weight effects on male endocrine organs	Control 800 ppm	none	Control 540 ppm	Control 500 ppm	Control 1000 ppm
Mean body weight [% to control]	-26%		458.40 419.3** [-8.6%]	485.00 442* [-8.8%]	(6m) 514 476*** -8%
Testes abs.	3.17 2.972**		3.49 3.55	3.41 3.63	3.40 3.07*
Testes rel.	3.118 3.128		3.46 3.6*	3.39 3.7*	n.s.
Female					
Dose with sign. weight effects on female endocrine organs	n.a.	none	n.a.	none	n.a.

n.a. = not applicable, *P<0.05; **P<0.01

Table 5.8.3-3: Repeated dose studies in mice with organ weight investigations /histopathology performed on endocrine organs

Doc ID Study type Strain Pathologist Date	AL-420-005 28 day mice CD-1 Mice [redacted] October 1992	AL-425-006 90 day mice CD-1 Mice [redacted] Februar 93	AL-428-002 & AL-428-003 78 weeks CD-1 Mice [redacted] May, 1996	
Administration	Diet	diet	diet	
Doses ppm	0-200-400-800-1200-1600 ppm	0-50-250-1000 ppm	0-30-100-300 ppm	
NOAEL LOAEL	400 ppm 800 ppm	50 ppm 250 ppm	30 ppm 100 ppm	
Endocrine organ weights measured	Adrenals, testes, uterus with cervix	Adrenals, testes, uterus with cervix	Adrenals, testes, uterus with cervix	
Histopathology performed on:	Only preserved: adrenals, epididymis, ovaries, pituitary, prostate, seminal vesicles, testes, thyroid with parathyroid, uterus with cervix	Adrenals, epididymis, ovaries, pituitary, prostate, seminal vesicles (only preserved), testes, thyroid with parathyroids, uterus with cervix	Adrenals, epididymis, ovaries, pituitary, prostate, seminal vesicles, testes, thyroid with parathyroids, uterus with cervix, vagina	
Male				
Dose with sign. weight effects on male endocrine organs Mean body weight [% to control] Adrenals abs. Adrenals rel. Testes abs. Testes rel.	None	control 1000 ppm 44.5 34.1** [-23%] 0.250 0.248 0.563 0.72**	control 300 ppm (week 52) 54.1 44.9** [-17%] (week 52) 0.004 0.005 0.0076 0.0125* n.s. n.s.	
Female				
Dose with sign. weight effects on female endocrine organs	None	none	none	

n.a. = not applicable, *P<0.05; **P<0.01

Literature data

Several publications with cypermethrin, beta-cypermethrin and alpha-cypermethrin and their common metabolites are related to endocrine effects and are discussed in the following. Based on the fact that alpha-cypermethrin is more related to beta-cypermethrin than cypermethrin, the literature focused on studies with beta-cypermethrin and publications with a potential to give insight into the Mode of action. Findings for common metabolites are discussed in this chapter as well, however the overall evaluation of metabolites is done in chapter CA 5.8.1.

The literature data are splitted up in the following sub-sections and in case of literature data dealing with different test systems the literature is placed in each section to inform about the outcome of the respective investigations. Whereever a study is splitted up it is indicated by a header that cross-references to the first citation.

- Androgen receptor assays *in vitro*
- Oestrogen receptor assays in vitro
- Aromatase and steroidogenesis assays in vitro
- Thyroid receptor assays in vitro

- Studies in male animals in vivo
  - Investigations with Cypermethrin
  - Investigations with beta-Cypermethrin
  - Investigations with alpha-Cypermethrin
  - Investigations with metabolites
- Studies in female animals in vivo
  - Investigations with Cypermethrin
  - Investigations with alpha-Cypermethrin
  - Investigations with metabolites
- Other studies
- Not yet peer reviewed studies

Each single chapter ends with a summary.

Androgen receptor assays *in vitro*

Report: CA 5.8.3/1
Kojima H. et al., 2004a
Screening for estrogen and androgen receptor activities in 200 pesticides by in vitro reporter gene assays using Chinese hamster ovary cells
2004/1040394
Guidelines: none
GLP: no

The authors tested a large number of pesticides for agonist and antagonist activity against the human oestrogen receptors α- and β- and the human androgen receptor in stably transfected CHO cells. The authors report that there was no evidence of androgenic or anti-androgenic activity for cypermethrin.

Classification of study: supplemental information

Report: CA 5.8.3/2
Tyler C.R. et al., 1999a
Metabolism and environmental degradation of pyrethroid insecticides produce compounds with endocrine activities
2000/1024078
Guidelines: none
GLP: no

The authors investigate interaction with the human androgen receptors in yeast cell reporter gene assays using a series of pyrethroids (including cypermethrin 'mixed isomers', 95.8% pure) and the common pyrethroid degradation products including M310I024, M310I011 as well as M310I001. Results were compared to the reference compounds 4-OH-tamoxifen and flutamide. Anti-oestrogenic and anti-androgenic effects were measured through co-incubation with 17β-oestrodiol or DHT, respectively. Cypermethrin showed no androgenic activity (very weak androgen activity is noted only at very high concentrations), but did show some evidence of weak anti-androgenic activity at high concentrations (LOEC = 10^{-3} M). The common metabolites M310I024 (PBAlc) was not active as androgen receptor agonist in yeast, but showed some anti-androgenic potential in the µ-molar range. M310I011 (PBA) and M310I001 (DCVA) showed neither androgenic nor anti-androgenic activity in yeast cells.

Classification of study: supplemental information

Report: CA 5.8.3/3
Vinggaard A.M. et al., 2007a
Screening of 397 chemicals and development of a quantitative structure - Activity relationship model for androgen receptor antagonism
2008/1102136
Guidelines: none
GLP: no

As part of a large screening study, the anti-androgenic activity of cypermethrin (no purity given) was investigated in a transfected CHO-K1 cell line reporter gene assay. Cells were exposed for 20 hours to concentrations of 1, 3, 10 and 30 µM and results expressed as the IC25 relative to the response seen with 0.1 nM the androgen R1881 (set as 100%). Cypermethrin was considered to be 'positive' in this assay (i.e. it showed 25% inhibition of the response to 0.1 nM of R1881 at a non-cytotoxic concentration of ≤10 µM and was assigned to Category 5, the least potent of 5 categories, based on an IC25 value of between 3 and 10 µM.

Classification of study: supplemental information

Report: CA 5.8.3/4
Xu L.-C. et al., 2007a
Evaluation of androgen receptor transcriptional activities of some pesticides in vitro
2008/1102137
Guidelines: none
GLP: no

The authors investigated human androgen receptor transcriptional activation in the stably-transfected monkey CV-1 cell line for a number of pesticides, including cypermethrin (>98% pure). Anti-androgenic activity was also investigated using the reference compound DHT. Cypermethrin is reported to have no androgenic activity and only weak anti-androgenic activity; activity was reported at all of the concentrations investigated (10^{-7}, 10^{-6}, 10^{-5} M) and the IC50 is reported to be 6.8 x10^{-5} M.

Classification of study: supplemental information

Report: CA 5.8.3/5
Zhang J. et al., 2008a
The antiandrogenic activity of pyrethroid pesticides Cyfluthrin and beta-Cyfluthrin
2008/1102138
Guidelines: none
GLP: no

Zhang *et al* investigated the anti-androgenic activity of cyfluthrin and beta-cyfluthrin (no purities given) *in vitro* and *in vivo*; comparison is also made to cypermethrin and beta-cypermethrin. *In vitro* anti-androgenic activity was assessed using a transcriptional activity assay in the stably transfected MDA-kb2 cell line and the reference compound DHT. In vivo anti-androgenic activity was investigated in the Hershberger assay with Testosterone propionate as androgen. Anti-androgenic activity is reported for cypermethrin but not for beta-cypermethrin at the single concentration investigated of 10^{-5} M in vitro. Weak anti-androgen activity was also found in vivo for Cypermethrin at 50 mg/kg bw, but not for beta-cypermethrin.

Classification of study: supplemental information

Report: CA 5.8.3/6
Wu W. et al., 2008a
Antiandrogenic effects of Cypermethrin and beta-Cypermethrin
2008/1102139
Guidelines: none
GLP: no

The anti-androgenic properties of cypermethrin and beta-cypermethrin (no purities given) were investigated *in vivo* in castrated male SD rats using a Hershberger assay using cypermethrin dose levels of 7, 21 or 63 mg/kg bw/d and beta-cypermethrin dose levels of 6, 18 and 54 mg/kg bw/d. In vitro the substances were investigated in a transcriptional activation assay based on MDA-kb2 cells at concentrations from 10^{-5} – 10^{-8} mol/L. Cypermethrin showed anti-androgenic activity *in vitro* at concentrations of 10^{-6} and 10^{-5} M; no effects were seen with beta-cypermethrin at any concentration. *In vivo*, administration of cypermethrin was shown to affect androgen-dependent organ weight at the mid- and high dose level groups. No effects were seen with beta-cypermethrin. The authors therefore conclude that cypermethrin is a weak anti-androgen *in vitro* and *in vivo*, but that beta-cypermethrin showed no such activity.

Classification of study: supplemental information

Report: CA 5.8.3/7
Du G. e al., 2010a
Assessing hormone receptor activities of pyrethroid insecticides and their metabolites in reporter gene assays
2010/1232195
Guidelines: none
GLP: no

Cypermethrin (purity: 92%) and its metabolites M310I011 (99% pure) and DCVA (99% pure) were assessed for activity in androgen receptor (stably transfected MDA-kb2 cells) reporter gene assays at non-cytotoxic concentrations of between 10^{-9} M and 10^{-5} M. There was no evidence of any androgenic activity. M310I011 showed slight (but not significant) anti-androgenic activity (RIC_{20} >10^{-5} M); DCVA did not show any anti-androgenic activity; cypermethrin showed significant anti-androgenic activity at concentrations of 10^{-6} and 10^{-5} M (RIC_{20} =1.64 x10^{-6} M).

Classification of study: supplemental information

Report: CA 5.8.3/8
Sun H. et al., 2006a
Antiandrogenic activity of Pyrethroid pesticides and their metabolite in reporter gene assay
2007/1070385
Guidelines: none
GLP: no

The androgenic and anti-androgenic effects of cypermethrin (>99% pure) and the pyrethroid metabolite M310I011 (>99% pure) were investigated in a human androgen receptor mediated reporter gene assay using the CV-1 cell line. Cells were exposed to non-cytotoxic concentrations of cypermethrin or M310I011 of 10^{-11} to 10^{-5} M in the absence or presence of DHT for 24 hours. No evidence of androgenic activity was seen in this study; however both cypermethrin and M310I011 showed anti-androgenic activity (i.e. inhibition of the response to DHT). IC_{20} values of 0.42 mM and 1.21 mM are reported for cypermethrin and M310I011, respectively. The authors conclude that cypermethrin and M310I011 show comparable anti-androgenic activity in this assay.

Classification of study: supplemental information

Report: CA 5.8.3/9
Tange S. et al., 2014a
In vitro metabolism of cis- and trans-Permethrin by rat liver microsomes, and its effect on estrogenic and anti-androgenic activities
2014/1242695
Guidelines: none
GLP: no

The authors report that permethrin showed weak anti-androgenic activity (IC20 =51.73 µM) in transfected CHO cells. Trans-permethrin showed stronger anti-androgen activity (IC20 =8.58µM). Cis-permethrin showed a comparably weak anti-androgenic activity (IC20 =21.91 µM). Incubation with liver microsomal fraction enhanced the anti-androgenic activity of permethrin and trans-permethrin, but did not have an effect on cis-permethrin. Following incubation with liver microsomal fraction, the metabolites of trans-permethrin were identified as phenoxybenzoic alcohol (M310I024) (quantitatively most important), phenoxybenzoic aldehyde (M301I018), phenoxybenzoic acid (M310I011) and 4-OH phenoxybenzoic alcohol. The metabolism of cis-permethrin was much less extensive; however the additional metabolite 4-OH cis-permethrin was identified. Incubation of permethrin showed intermediate results, indicating that the majority of metabolites originated from the *trans*-isomers. Of the metabolites, M310I024 (98% pure) showed greater anti-androgenic activity (IC_{20} =1.15 µM). M310I018 (97% pure) showed the strongest anti-androgenic activity (IC_{20} =0.17 µM). M310I011 (98% pure) showed no anti-androgenic activity. 4-OH *cis*-permethrin showed comparable anti-androgenic activity to *cis*-permethrin (IC_{20} =23.83 µM). 4-OH phenoxybenzoic alcohol (>99% pure) showed no anti-androgenic activity. 4-OH phenoxybenzoic acid (M310I025; >98% pure) showed no anti-androgenic activity. 2-OH phenoxybenzoic alcohol (>98% pure) showed anti-androgenic activity (IC_{20} =4.89 µM). 2-OH phenoxybenxoic acid showed no anti-androgenic activity.

Summary of responses and effects of metabolism (Tange *et al*, 2014)

	Anti-androgenic activity IC_{20} (µM)	Effect of metabolism on activity
Permethrin	51.73	↑
cis-permethrin	21.91	-
trans-permethrin	8.58	↑
M310I024	1.15	
M310I018	0.17	
M310I011	-	
4-OH *cis*-permethrin	23.83	
4-OH phenoxybenzoic alcohol		
4-OH phenoxybenzoic acid (M310I025)	-	
2-OH phenoxybenzoic alcohol	4.89	
2-OH phenoxybenzoic acid	-	

The authors conclude that the metabolism of permethrin and trans-permethrin results in increased anti-androgenic activity; however no effect is seen with *cis*-permethrin. This finding is notable with regard to alpha-cypermethrin, which consists only of *cis*-isomers of cypermethrin.

Classification of study: supplemental information

Report: CA 5.8.3/10
Kjeldsen L.S. et al., 2013a
Currently used pesticides and their mixtures affect the function of sex hormone receptors and aromatase enzyme activity
2013/1417281
Guidelines: none
GLP: no

Several test compounds, including Cypermethrin (>95% pure) were tested on their androgenic/anti-androgenic potential in a transactivation assay in the concentration range of 10^{-10} to 10^{-5} M. The androgen receptor transactivation assay was performed in CHO-K1 cells that were transiently co-transfected with the MMTV-LUC reporter vector and the human AR expression plasmid pSVARO. The authors reported that cypermethrin showed no androgenic effects, but showed a slight tendency to antagonize the R1881- or DHT-induced (25 pM) androgen receptor transactivity at the highest test concentration, although not statistically significant.

Classification of study: supplemental information

Report: CA 5.8.3/11
Ait-Aissa S. et al., 2010a
Anti-androgenic activities of environmental pesticides in the MDA-kb2 reporter cell line
2010/1232202
Guidelines: none
GLP: no

Several test compounds, including cypermethrin (no purity given) were tested on their androgenic/anti-androgenic potential in a transactivation assay. The androgen receptor transactivation assay was performed in MDA-kb2 cells that were stably transfected with the MMTV-LUC reporter vector. In these cells this promoter is up-regulated by two endogenous nuclear receptors, AR and GR. Cypermethrin was tested in the range of 0.01 to 10 µM in solvent alone (DMSO) or in the presence of 0.1 nM DHT for the determination of androgenic or anti-androgenic effects, respectively. Cypermethrin was not cytotoxic up to concentrations of 10 µM. No androgenic or anti-androgenic effect was reported for cypermethrin in the applied in vitro model using MDA-kb2 cells.

Classification of study: supplemental information

Overview of androgen receptor assays

Available assays are relatively consistent in identifying cypermethrin as a weak anti-androgen at relatively high concentrations which may not be of direct relevance to the human risk assessment. Similar activity is not reported for beta-cypermethrin, indicating that the activity is associated with specific isomers. There is no evidence of any androgenic activity, neither for cypermethrin nor beta-cypermethrin.

No androgenic activity was found in any test system for the common metabolites M310I011, M310I018, M310I024, M310I001 and for the 4-hydroxylated PBA metabolite M310I025.

Anti-androgenic activity was reported for M310I024 (IC_{20}=1.15µM) and even stronger for M310I018 (IC_{20}=0.17 µM), but no relevant activity was found for the most dominant metabolite M310I011 (reported activity in the millimolar range not considered of any relevance) or its in position 4 hydroxylated derivative M310I025 nor for M310I001.

The US EPA assessment cautions that none of the studies directly assesses binding to the androgen receptor. In general, transcriptional activation assays and receptor binding assays are both needed to increase the level of confidence and minimise the potential for false negative responses. Transcriptional activation assays do not measure receptor binding and involve post-binding events; therefore, they are not a direct substitute for receptor binding assays. Additional mechanistic studies (summarised below) indicate that the anti-androgenic effects of cypermethrin are not due to a direct interaction with the receptor.

The differences in response reported for cypermethrin and beta-cypermethrin is notable and indicates that the anti-androgenic effects of cypermethrin (8 isomers, for isomeric composition of cypermethrins see Document N5, Bridging rational) are associated with specific isomers. The absence of anti-androgenic effects for beta-cypermethrin (4 isomers of cypermethrin, cis-2 and trans-4) would indicate a similar lack of effects for alpha-cypermethrin (2 isomers of beta-cypermethrin, only cis-2). The study of Tange *et al* (2014) performed with permethrin, *cis*-permethrin and *trans*-permethrin shows greater anti-androgenic activity with *trans*-permethrin compared to permethrin and *cis*-permethrin; and furthermore shows an increase in anti-androgenic potential following the metabolism of permethrin and *trans*-permethrin but not following the metabolism of *cis*-permethrin. This effect is attributable to the much less extensive metabolism of the *cis*-isomers and an apparent association of anti-androgenic activity with permethrin metabolites common to other pyrethroids. These observations are pertinent for cypermethrin and alpha-cypermethrin due to the common metabolites and the fact that alpha-cypermethrin consists solely of *cis*-isomers. The difference in response between cypermethrin and beta-cypermethrin may be explained by the lower proportion of *trans*-isomers in beta-cypermethrin.

Therefore it is concluded that weight of evidence indicate that alpha-cypermethrin is not at risk to be androgenic and as well is not at risk to be anti-androgenic or to induce anti-androgenic response via metabolic degradation. This is in line with in vivo findings.

Table 5.8.3-4: Summary of androgen receptor assays

Study	Finding for parent	Comment and data for metabolites
Kojima *et al* (2004)	Cypermethrin: Negative (androgenic) Negative (anti-androgenic)	hAR transactivation in mammalian cells
Tyler *et al* (2000)	Cypermethrin: Negative (androgenic): Positive (anti-androgenic): - weak response (10^{-3} M),	hAR transactivation in Yeast: Negative (androgenic): - M310I024, M310I011, M310I001; Positive (anti-androgenic): - M310I024: weak: IC50: 37 µM, but Negative (anti-androgenic): - M310I011 & M310I001
Vingaard *et al* (2008)	Cypermethrin: Positive (anti-androgenic): - weak response: IC25: 3-10 µM	hAR transactivation in mammalian cells
Xu *et al* (2008)	Cypermethrin: Negative (androgenic) Positive (anti-androgenic): - weak response: IC50: 6.8 x10^{-5} M	hAR transactivation in mammalian cells
Zhang *et al* (2008)	Cypermethrin: Positive (anti-androgenic) - Weak response: 10^{-5} M Beta-cypermethrin: Negative (anti-androgenic)	hAR transactivation in mammalian cells
Wu *et al* (2008)	Cypermethrin: Positive (anti-androgenic) - 10^{-6} and 10^{-5} M Beta-cypermethrin: Negative (anti-androgenic)	hAR transactivation in mammalian cells
Du *et al* (2010)	Cypermethrin: Positive (anti-androgenic) - 10^{-6} and 10^{-5} M	hAR-transactivation in mammalian cells: Negative (androgenic): - M310I011, M310I001 Negative (anti-androgenic): - M310I001 Positive (anti-androgenic): - M310I011:Weak response (RIC20 > 10µM)
Sun *et al* (2007)	Cypermethrin: Negative (androgenic) Positive (anti-androgenic): - weak response IC20: 0.42 mM	hAR-transactivation in mammalian cells: Negative (androgenic): - M310I011 Positive (anti-androgenic): - M310I011: very weak response: IC20: 1.21 mM;
Tange *et al* (2014)	Trans-Permethrin > Cis-Permethrin > Permethrin: Positive (anti-androgenic):	hAR-transactivation in mammalian cells: Positive (anti-androgenic): - M310I024: weak response IC_{20} 1,15 µM; - M310I018: IC_{20} 0.17µM;

Study	Finding for parent	Comment and data for metabolites
	- IC_{20} 8.58 µM, 21.91 µM, 51,73 µM, respectively	Negative: (anti-androgenic): - M310I011
Kjeldsen *et al* (2013)	Cypermethrin Negative (androgenic) Negative (anti-androgenic): - unsignificant effect at 10 µM	hAR-transactivation in mammalian cells
Ait-Aissa *et al* (2010)	Cypermethrin (tested up to 10 µM): Negative (androgenic) Negative (anti-androgenic)	hAR-transactivation in mammalian cells

Oestrogen receptor assays *in vitro*

Kojima *et al* (2004) see CA 5.8.3/1 2004/1040394
Screening for Estrogen and Androgen Receptor Activities in 200 Pesticides by *In Vitro* Reporter Gene Assays Using Chinese Hamster Ovary Cells
Environmental Health Perspectives 112(5):524-531

The authors tested a large number of pesticides for agonist and antagonist activity against human oestrogen α- and β- receptors in stably transfected CHO-K1 cells. Cypermethrin (>95% pure) was tested at concentrations between 10^{-8} M to 10^{-5} M and is reported to show activity in the ER-α transactivation assay (REC20 =8.1 x10^{-6} M). The US EPA considers that this study does not provide sufficient information to permit independent review; specifically, the absence of solubility and cytotoxicity data are cited as limiting factors.

Classification of study: supplemental information

Tyler *et al* (2000) see CA 5.8.3/2 2000/1024078
Metabolism and environmental degradation of pyrethroid insecticides produce compounds with endocrine activities
Environmental Toxicology & Chemistry 19(4):801-809

The authors investigated interaction with the human oestrogen receptor in a yeast cell reporter gene assay using a series of pyrethroids (including cypermethrin) and the common pyrethroid degradation products M310I024 and M310I011. Results were compared to the reference compound 4-OH-tamoxifen. Anti-oestrogenic effects were measured through co-incubation with 17β-oestrodiol. Cypermethrin (95.8% pure) is stated to have showed no oestrogenic activity (data not shown) but is, however, reported to have anti-oestrogenic activity (LOIC = 7±3 x10^{-4} M), 1000-10000 times less potent than 4-OH-tamoxifen. Of the seven pyrethroids tested, four pyrethroids showed anti-oestrogenic activity; cypermethrin was the least potent. The pyrethroid metabolite 3-phenoxybenzylalcohol showed weak oestrogenic activity (10^5 times less potent than 17β-oestrodiol). The metabolite M310I011 also showed anti-oestrogenic activity (10^2-10^3 times less potent than 4-OH-tamoxifen). The authors conclude that the hormone receptor interaction seen in this assay with various pyrethroids may in fact be due to their metabolites, which are stated to be generated by yeast esterase activity.

Classification of study: supplemental information

Du *et al* (2010) see CA 5.8.3/7 2010/1232195
Assessing Hormone Receptor Activities of Pyrethroid Insecticides and Their Metabolites in Reporter Gene Assays
Toxicological Sciences 116(1):58-66

Cypermethrin (92% pure) and its metabolites M310I011 (3-PBA, 99% pure) and DCCA (99% pure) were assessed for activity in oestrogen receptor transfected CV-1 cells using a reporter gene assay at non-cytotoxic concentrations of between 10^{-9} M and 10^{-5} M. There was no evidence of any oestrogenic activity for any substance investigated. 3-PBA showed significant anti-oestrogenic activity at concentrations of 10^{-6} and 10^{-5} M (RIC_{20} =8.84 x10^{-8} M); DCCA showed significant anti-oestrogenic activity at concentrations of ≥10^{-8} M (RIC_{20} =7.23 x10^{-9} M); cypermethrin did not show any anti-oestrogenic activity.

Classification of study: supplemental information

Report: CA 5.8.3/12
Chen H. et al., 2001a
Estrogenicity of organophosphorus and pyrethroid pesticides
2002/1027197
Guidelines: none
GLP: no

The authors investigated the oestrogenic activity of a number of pesticides, including cypermethrin (>90% pure), in three *in vitro* screening assays. A competitive binding assay with ^{3}H-oestradiol was performed using rat uterine cytosolic oestrogen receptors at concentrations between 10^{-12} and 10^{-6} M. Oestrogen receptor activation was assessed using the E-screen (induction of proliferation of oestrogen-sensitive MCF-7 cells) at concentrations between 10^{-11} and 10^{-6} M. Transcriptional activation of the oestrogen receptor was also measured using a pS2 mRNA assay in the MCF-7 cell line at concentrations between 10^{-10} and 10^{-6} M. The MCF-7 assay (E-screen) showed cypermethrin (purity: >90%) -induced proliferation; values were statistically significant compared to controls at a concentration of 10^{-8} M only and not at higher concentrations. The maximum fold proliferation is reported to be relatively low (1.46 at 10^{-8} M). The proliferation induced by cypermethrin was blocked by co-incubation with the oestrogen receptor antagonist ICI 182 780, leading to the authors' conclusion that cypermethrin acts as a partial oestrogen receptor agonist. Cypermethrin was also shown to competitively inhibit the binding of ^{3}H-oestradiol to rat uterine cytosolic oestrogen receptors at the higher concentrations tested (≥10^{-6} M); an IC_{50} of 0.562 mM was calculated. The maximum level of inhibition was approximately 50% and the relative potency was about 500,000 times lower than oestradiol. Expression of pS2 mRNA in MCF-7 cells was also significantly increased (2.01-fold) following incubation with cypermethrin at the highest investigated concentration of 10^{-6} M. The US EPA assessment considers that this study satisfies the requirement for an oestrogen receptor binding assay.

Classification of study: supplemental information

Report: CA 5.8.3/13
Kim I.Y. et al., 2003a
Assessing estrogenic activity of pyrethroid insecticides using in vitro combination assays
2004/1040395
Guidelines: none
GLP: no

The authors investigated the oestrogenicity/anti-oestrogenicity of several pyrethroids, amongst them cypermethrin (98% pure) *in vitro* in MCF-7 BUS cells (E-screen). Oestrogen receptor levels and PS2 mRNA levels were also assessed. A competitive binding assay (with 3H-oestradiol) was performed using ovariectomised rat uterine cytosol.
No oestrogenic-like effect of cypermethrin (10^{-5} M) was seen following treatment for 6 days in the E-Screen assay. There was no evidence of cytotoxicity. No anti-oestrogenic effect was seen with 10^{-6} M cypermethrin in MCF-7 cells simultaneously exposed to ICI 182 780. No evidence for the inhibition of binding for ^{3}H-oestradiol to the oestrogen receptor was seen in uterine cytosol prepared from ovariectomised rats. Oestrogen receptor levels and PS2 mRNA levels were not assessed with cypermethrin in the absence of any indication of oestrogenic activity. This assay does not therefore provide any evidence for oestrogen receptor binding or activation for cypermethrin (in contrast to one of the other pesticides tested); however the reliability of the assay is limited by poor reporting. The USEPA assessment considers that the study is limited due to the absence of replicate data and the use of a single concentration.

Classification of study: supplemental information

Report: CA 5.8.3/14
Lemaire G. et al., 2006a
Activation of alpha- and beta-estrogen receptors by persistent pesticides in reporter cell lines
2006/1051139
Guidelines: none
GLP: no

A range of pesticides (including cypermethrin, >95% pure) were screened for transcriptional activation of the oestrogen α- and β-receptors using the stably transfected HeLa cell line. Cypermethrin was tested at a single concentration of 10 µM. In the absence of any indication of a positive response in HeLa ERα or HeLa ERβ, further concentrations were not tested. US EPA raises concerns over the validity of this assay as only a single concentration was tested, increasing the likelihood of a false negative response.

Classification of study: supplemental information

Report: CA 5.8.3/15
Liang L. et al., 2005a
A study on the estrogenic effects of alpha- and theta-Cypermethrin
2005/1043340
Guidelines: none
GLP: no

A positive result is reported for alpha-cypermethrin (at the single tested concentration of 10^{-8} M, purity not given) and theta-cypermethrin (at the single tested concentration of 10^{-8} M, purity not given) in the MCF-7 cell proliferation assay; the level of induced proliferation was greater for alpha-cypermethrin than for theta-cypermethrin, and was in fact greater than that induced by the positive control (10^{-8} M β-oestradiol). Some inhibition of proliferation was induced by the solvent control (0.1% ethanol). The results of this study therefore indicate oestrogenic activity for alpha-cypermethrin and theta-cypermethrin; however the level of response seen indicates that the study is of questionable reliability.

Classification of study: supplemental information

Report: CA 5.8.3/16
Laffin B. et al., 2009a
The pyrethroid metabolites 3-phenoxybenzoic acid and 3-phenoxybenzyl alcohol do not exhibit estrogenic activity in the MCF-7 human breast carcinoma cell line or Sprague-Dawley rats
2010/1232196
Guidelines: none
GLP: no

The authors investigated the oestrogenicity of the pyrethroid metabolites M310I011 and M310I024 *in vitro*. MCF-7 cells were exposed to concentrations of 1 nM, 10 nM or 10 µM of each metabolite for 3 or 6 days in the absence or presence of ICI 182,780 to investigate proliferation stimulus. In a reporter gene assay, ERE3-lucriferase transfected cells were exposed to metabolite concentrations of 1 nM, 10 nM, 1 µM or 10 µM for 24 hours in the absence or presence of ICI 182,780. No effects of either metabolite were seen on MCF-7 cell proliferation or reporter gene expression.

Classification of study: supplemental information

Report: CA 5.8.3/17
Jin M. et al., 2009a
Estrogenic activities of two synthetic pyrethroids and their metabolites
2010/1232199
Guidelines: none
GLP: no

The oestrogenic activity of cypermethrin (unclear substance: the abstract states that beta-cypermethrin was investigated, 98% pure), and its metabolites M310I011 (PBA, 98% pure), M310I024 (PBAlc, 98% pure) and M310I018 (PBAld, ≥97% pure) were investigated *in vitro* using the MCF-7 cell proliferation assay. Cells were exposed to cypermethrin or its metabolites at concentrations between 10^{-9} to 10^{-5} M for six days. Levels of pS2 (oestrogen-inducible) mRNA and ER-α mRNA were measured using PCR in cells exposed to 10^{-6} M for 2 days. Significantly increased cell proliferation was seen in cells exposed to cypermethrin at concentrations of 10^{-8}, 10^{-7} and 10^{-6} M, with a maximal effect (1.63 times controls) seen at 10^{-7} M. Significantly increased cell proliferation was seen following exposure to M310I011 at a concentration of 10^{-7} M only; however this effect was marginal (1.32 times controls). Significantly increased cell proliferation was seen in cells exposed to M310I024 at concentrations of 10^{-9}, 10^{-8}, 10^{-7} and 10^{-6} M, with a maximal effect (1.7 times controls) seen at 10^{-7} M – a relatively low concentration and only 10-100 times lower than reported for oestradiol. No significant effects on cell proliferation were seen at any concentration of M310I018. Increased expression of pS2 mRNA was seen in response to cypermethrin (2.15 times the control) and all metabolites; the greatest response was seen with M310I024 (3.45 times). A lower response was seen with cypermethrin and relatively weak responses were seen with M310I011 (1.85 times) and 3-phenoxybenzaldehyde (1.37 times). Reduced expression of ER-α mRNA was seen following exposure to cypermethrin and M310I024; a marginal effect was seen with M310I011 (-12.0%) and no effect was seen with M310I018 (PBAld) (-3.7%). The results of this study show oestrogenic activity *in vitro* with cypermethrin and indicate that this may be increased by initial metabolism to M310I024 but may also reduced by subsequent metabolism to the aldehyde and acid derivatives. A notable feature of this study is the production of non-linear dose-response curves for all of the substances investigated. The concentrations investigated do not indicate significant cytotoxicity and may therefore reflect a technical problem with the assay.

Classification of study: supplemental information

Report: CA 5.8.3/18
Wang Q. et al., 2011a
Estrogenicity activity assessment and mechanism exploration on Alpha-Cypermethrin in vitro
2011/1296734
Guidelines: none
GLP: no

The effects of alpha-cypermethrin (commercial grade, no purity given) were assessed in the MCF-7 cell line; cells were exposed to concentrations of 10^{-7}, 10^{-8} or 10^{-9} M for five days. Levels of ERα mRNA were also measured using PCR. A significant increase in cell proliferation was seen at all alpha-cypermethrin concentrations; the response was maximal (1.38 times the control value) at the highest concentration tested however the blank control value was also significantly higher (1.16 times the solvent control). Findings therefore indicate either solvent toxicity or that value for alpha-cypermethrin at concentrations of 10^{-8} M (response 1.13 times the solvent control) or 10^{-9} M (1.19 times the solvent control) is within the background range. Exposure to alpha-cypermethrinwas also shown to slightly up-regulate the relative expression of ERα mRNA at concentrations of 10^{-9} M (1.39 times the control response), 10^{-8} M (1.45 times) and at 10^{-7} M (1.61 times). Values for ERα mRNA expression are calculated using β-actin as an internal control according to the $2^{-\Delta\Delta Ct}$ method.

Classification of study: supplemental information

Report: CA 5.8.3/19
McCarthy A.R. et al., 2005a
Estrogenicity of Pyrethroid insecticide metabolites
2006/1051135
Guidelines: none
GLP: no

The authors investigated the oestrogenic activity of cypermethrin and its metabolites 3-(4-hydroxy-3-phenoxy)benzoic acid, N-3-(phenoxybenzoyl)glycine, M310I024, 3-phenoxybenzaldehyde and M310I011 (no purities given) using a recombinant yeast assay expressing the human oestrogen α-receptor. Weak oestrogenic activity is reported for M310I024 (EC_{50} =6.67 µM), 3-phenoxybenzaldehyde (EC_{50} =4.80 µM) and 3-(4-hydroxy-3-phenoxy)benzyl alcohol (EC_{50} =6.75 µM). The relative potencies of these metabolites were similar and ranged from 5-7 $x10^{-5}$ times less than oestradiol.

Classification of study: supplemental information

Tange *et al* (2014) see CA 5.8.3/9 2014/1242695
In vitro metabolism of cis- and trans-permethrin by rat liver microsomes and its effect on estrogenic and anti-androgenic activities
Environmental Toxicology & Pharmacology 37 (2014) 996-1005

The authors report that permethrin showed weak oestrogenic activity (EC_{20} =18.82 µM) in transfected MCF-7 cells. Trans-permethrin also showed weak oestrogenic activity (EC_{20} =25.10 µM). In contrast, cis-permethrin showed no oestrogenic activity. The incubation of permethrin with liver microsomal fraction enhanced the oestrogenic activity. A marked enhancement of oestrogenic activity was apparent for cis-permethrin, but no effect was seen with trans-permethrin. Following incubation with liver microsomal fraction, the metabolites of trans-permethrin were identified as phenoxybenzoic alcohol (quantitatively most important), phenoxybenzoic aldehyde, phenoxybenzoic acid and 4-OH phenoxybenzoic alcohol. The metabolism of cis-permethrin was much less extensive; however the additional metabolite 4-OH cis-permethrin was identified. Incubation of permethrin showed intermediate results, indicating that the majority of metabolites originated from the trans-isomers. Of the metabolites, phenoxybenzoic alcohol (98% pure) showed comparable oestrogenic activity (EC_{20} =19.97 µM) to permethrin. Phenoxybenzoic aldehyde (97% pure) showed no oestrogenic activity. Phenoxybenzoic acid (98% pure) showed no oestrogenic activity. 4-OH cis-permethrin showed oestrogenic activity (EC_{20} =10.77 µM), in contrast to cis-permethrin. 4-OH phenoxybenzoic alcohol (>99% pure) showed oestrogenic activity (EC_{20} =2.48 µM). 4-OH phenoxybenzoic acid (>98% pure) showed no oestrogenic activity. 2-OH phenoxybenzoic alcohol (>98% pure) showed no oestrogenic activity. 2-OH phenoxybenzoic acid showed no oestrogenic activity.

Summary of responses and effects of metabolism (Tange *et al*, 2014)

	Oestrogenic activity EC20 (µM)	Effect of metabolism on activity
Permethrin	18.82	↑
cis-permethrin	-	↑↑
trans-permethrin	25.10	-
Phenoxybenzoic alcohol (M310I024)	19.97	
Phenoxybenzoic aldehyde (M310I018)	-	
Phenoxybenzoic acid (M310I011)	-	
4-OH *cis*-permethrin	10.77	
4-OH phenoxybenzoic alcohol	2.48	
4-OH phenoxybenzoic arid (M310I025)	-	
2-OH phenoxybenzoic alcohol	-	
2-OH phenoxybenzoic acid	-	

The authors conclude that metabolism of permethrin results in enhanced oestrogenicity due to the production of 4-OH *cis*-permethrin, phenoxybenzoic alcohol and 4-OH phenoxybenzoic alcohol. Metabolism of *cis*-permethrin similarly results in enhanced oestrogenicity; however no effect is seen with *trans*-permethrin. No effect is seen with *cis*-permethrin; this finding is of relevance to alpha-cypermethrinas this consists of the *cis*-isomers only.

Classification of study: supplemental information

Report: CA 5.8.3/20
Meeuwen J.A. van et al., 2008a
Aromatase inhibiting and combined estrogenic effects of parabens and estrogenic effects of other additives in cosmetics
2008/1102356
Guidelines: none
GLP: no

Several test compounds, including M310I019 (3-hydroxy benzoic acid; no purity given) were tested on their estrogenic potential in the E-screen using MCF-7 cells. The test substances were tested in the range of 1 nM to 10 mM. The authors report that none of the tested hydroxylated benzoic acid isomers did induce cell proliferation in MCF-7 cells and therefore no EC_{50} values could be derived and thus no estrogenic equivalency factor could be calculated. In conclusion, M310I019 showed no estrogenic potential in the E-screen under the conditions applied.

Classification of study: supplemental information

Kjeldsen *et al* (2013) see CA 5.8.3/10 2013/1417281
Currently used pesticides and their mixtures affect the function of sex hormone receptors and aromatase enzyme activity
Toxicology and Applied Pharmacology 272 (2013) 453-464

Several test compounds, including Cypermethrin (>95% pure) were tested on their estrogenic/anti-estrogenic in a transactivation assay. The estrogen receptor transactivation assay was performed in MVLN cells that were derived from MCF-7 cells carrying an estrogen response element luciferase reporter vector. The authors reported that cypermethrin slightly but statistically significantly induced estrogen receptor-mediated gene expression at 1-10 µM with a maximum effect of 113% (at 10 µM) of the solvent control. No EC_{50} could be determined since no concentration-response curve could be established. Cytotoxicity was observed at concentrations of ≥5 µM. No anti-estrogenic activity, determined in the presence of 25 pM 17β-estradiol, was observed.

Classification of study: supplemental information

Report: CA 5.8.3/21
Sun H. et al., 2013a
Pyrethroid and their metabolite, 3-phenoxybenzoic acid showed similar (anti)estrogenic activity in human and rat estrogen receptor alpha-mediated reporter gene assays
2014/1242697
Guidelines: none
GLP: no

Cypermethrin (92% pure) and its metabolite M310I011 (3-PBA, 99% pure) were assessed for activity in human or rat estrogen receptor transfected CV-1 cells using a luciferase reporter assay. No cytotoxicity was observed up to 100 µM of the test substance concentrations as determined by MTT assay in former experiments, microscopic examination, and determination of expression of co-transfected Renilla luciferase in the actual experiments. Cypermethrin showed an estrogenic activity in both, the hER and rER assay. The maximum induction (%) was 26% and 34% in the hER and rER assay, when compared to 100 nM E2. EC_{50} values were 0.37 µM and 6.78 µM in the hER and rER assay, respectively. PBA showed no estrogenic activity up to concentrations of 100 µM in the hER and rER assays. No anti-estrogenic effects were observed for cypermethrin in both assays. PBA (100 µM) significantly reduced the activity of 1 nM E2 to 55% in the hER assay, indicating that PBA possesses an anti-estrogenic potential.

Classification of study: supplemental information

Overview of oestrogen receptor assays

The available assays are inconsistent for cypermethrin; reporting negative, weak positive and positive responses in oestrogen receptor binding and activation assays. The US EPA identify the study of Chen et al (2011) as meeting the requirement for an oestrogen receptor binding assay but do not consider the studies adequate to meet the requirement for an oestrogen receptor transcriptional activation assay.
For alpha-cypermethrin only two MCF-7 proliferation assays are published: Liang *et al*, 2005 reports a marked positive response but is of questionable reliability; a further study (Wang *et al*, 2011, same lab and same method than Liang et al, 2005) reports a weaker proliferative response, but the study is also of limited reliability. A potential induction of proliferation of MCF-7 cells is not directly attributable to an estrogenic effect; therefore the relevance of this information is questionable.
Studies with metabolites indicate that estrogenic effects of rather weak potency may be due to the metabolite M310I024 rather than due to the parent substance. For M310I018 only some estrogenic activity was reported on mRNA level or in yeast cells, but not in a transactivation assay in mammalian cells, and no relevant estrogenic activity was reported for M310I011 and M310I019, M310I001 or M310I025. Worth to mention are results from Tange *et al* (2014) reporting oestrogenic activity in vitro for permethrin and *trans*-permethrin, but not for *cis*-permethrin. Metabolic degradation is shown to increase the oestrogenic response to permethrin but not to *trans*-permethrin, and also results in a positive response for *cis*-permethrin. Data indicate that estrogenic findings of permethrin are mainly due to the metabolite 4-OH phenoxybenzoic alcohol and 4-OH *cis*-permethrin; however these metabolites are both not found in any in vivo metabolic pathway of alpha-cypermethrin nor in plants and are therefore not considered relevant in the endocrine evaluation of alpha-cypermethrin. However, the difference in the proliferative response of MCF-7 cells seen with cypermethrin (weak, inconsistent oestrogenicity) and with alpha-cypermethrin (only two studies but both reporting positive responses) may be attributable to the metabolism in vitro.
Furthermore, weak anti-estrogenic activity is reported for M310I001 and M310I011 in yeast cells and moderate activity in two transactivation assays, both in CV-1 cells. According to these studies M310I001 is slightly more potent than M310I011, which showed relevant anti-estrogenic activity at physiologically rather unrelevant higher µ-molar concentrations.

Weight of evidence for cypermethrin does not indicate estrogenic effects and are considered valid for alpha-cypermethrin, too. Observed proliferative effect induces by alpha-cypermethrin might be due to in vitro generated metabolites, however these metabolites are not conisered relevant in vivo. An estrogenic effect via M310I024 is considered not relevant based on the slow metabolic release and the further detoxification of this metabolite via Phase II conjugation.
Weight of evidence indicate no anti-estrogenic activity for cypermethrin and is considered valid for alpha-cypermethrin, too. Furthermore the weak anti-estrogenic effects observed for M310I001 and M310I011 are considered less relevant for alpha-cypermethrin based on the slow metabolic release and the further detoxification of this metabolites via Phase II conjugation.

Table 5.8.3-5: Summary of oestrogen receptor assays

Study	Finding for parent	Comment and data for metabolites
Kojima *et al* (2004)	Cypermethrin: Positive (oestrogenic): - ERα: REC20: 8.1 x10^{-6} M - ERβ: not active Negative (anti-oestrogenic)	hER transactivation in mammalian cells
Tyler *et al* (2000)	Cypermethrin: Negative (oestrogenic) Positive (anti-oestrogenic) - Very weak response LOIC: 7E-04M	hER transactivation in Yeast Positive (oestrogenic): - M310I024 (EC_{50}: 20 µM) Negative (oestrogenic): - M310I011, M310I001, Negative (anti-oestrogenic): - M310I024 Positive (anti-oestrogenic): - M310I011: weak response LOIC: 1.25±0.7E-05 M - M310I001: very weak response LOIC: 6±4E-05 M
Du *et al* (2010)	Cypermethrin: Negative (oestrogenic) Negative(anti-oestrogenic)	hER transactivation in mammalian CV-1 cells, Negative (oestrogenic): - M310I011, M310I001 Positive (anti-oestrogenic) - M310I011: RIC_{20}: 8.84E-8 M; IC_{50} ~ 10µM - M310I001: RIC_{20}: 7.23E-9 M; IC_{50} ~ 1µM
Chen *et al* (2011)	Cypermethrin: Positive (oestrogenic) 1.46 fold proliferation increase at 0.01µM but Positive (anti-oestrogenic) competitive binding inhibition at 0.56 µM	ER-dependent regulation of mRNA E-Screen (MCF-7) rER (uterus) competitive binding assay Result possibly unreliable; authors conclude that cypermethrin is a partial agonist
Kim *et al* (2004)	Cypermethrin Negative (oestrogenic) Negative (anti-oestrogenic)	ER-dependent regulation of protein rER (uterus) competitive binding E-Screen (MCF-7) Limited reporting
Lemaire *et al* (2006)	Cypermethrin Negative (oestrogenic)	hER transactivation in mammalian cells Single concentration tested (10 µM)
Liang *et al* (2005)	Alpha-cypermethrin Positive (oestrogenic) - Strong response (10^{-8} M) Theta-cypermethrin Positive (oestrogenic) - Strong response (10^{-8} M)	E-Screen (MCF-7) Proliferative response is greater than seen with the positive control estradiol: questionable reliability

Study	Finding for parent	Comment and data for metabolites
Laffin *et al* (2010)		E-Screen (MCF-7) hER transactivation assay Negative (oestrogenic): - M310I011, M310I024: 1nM-10µM
Jin *et al* (2010)	Cypermethrin Positive (oestrogenic) 1.63 proliferation increase 2.15fold pS2 mRNA induction RIE : 68.7%	E-Screen (MCF-7) Positive (oestrogenic): - M310I011: 1.32 fold - M310I024: 1.7 fold Negative (oestrogenic): - M310I018 ER mRNA in MCF-7 & pS2 mRNA levels in MCF-7: Positive (oestrogenic): - M310I011: 1.85 fold pS2 induction & RIE:24% - M310I018: 1.37 fold pS2 induction & RIE: 7.3% - M310I024: 3.45 fold pS2 induction & RIE: 88.7% Study of questionable reliability.
Wang *et al* (2011)	Alpha-cypermethrin Positive (oestrogenic): - weak response	E-Screen (MCF-7) and ERα mRNA in MCF-7 Study may not be reliable.
McCarthy *et al* (2010)		hERα transactivation in Yeast Positive (oestrogenic) - M310I024: weak EC_{50}: 6.7±3.1xE-6M - M310I018: weak EC_{50}: 4.8±3.4xE-6M Negative (oestrogenic) - M310I011 & M310I025;
Tange *et al* (2014)	Permethrin, trans-permethrin cis-permethrin Positive (oestrogenic) - Permethrin: weak response, EC20:18.82 µM - trans-permethrin: weak response EC20: 25.1 µM Negative (oestrogenic) - cis-permethrin	hER transactivation in MCF-7 Positive (oestrogenic): - M310I024, weak response EC20:19.97 µM Negative (oestrogenic): - M310I018: not active up to 100 µM - M310I011: not active
van Meeuwen *et al* (2008)		E-Screen (MCF-7) Negative (oestrogenic) - M310I019

Study	Finding for parent	Comment and data for metabolites
Kjeldsen *et al* (2013)	Cypermethrin: Positive (oestrogenic) - weak response: 1.13fold of solvent control at 10 µM Negative (anti-oestrogenic)	ER transactivation assay in MCF-7 Weak positive response reported at cytotoxic concentrations, no dose-response effects
Sun *et al* (2014)	Cypermethrin: Positive (oestrogenic) - hER: EC50: 0.37µM - rER: EC50: 6.78µM - Negative (anti-oestrogenic)	hER and rER transactivation assay in CV-1 cells Negative (oestrogenic) - M310I011 (in both systems) Positive (anti-oestrogenic) - M310I011: weak response: IC_{50}(hER) approx. 100 µM

RIE: Relative inhibitory efficiency is the ratio between the maximal down-regulation of Era expression level by the test compound to that of E2(x100), IC50: Concentration of 50% response inhibition.

Aromatase and steroidogenesis assays in vitro

Report: CA 5.8.3/22
Laville N. et al., 2006a
Modulation of aromatase activity and mRNA by various selected pesticides in the human choriocarcinoma JEG-3 cell line
2006/1051133
Guidelines: none
GLP: no

The effects of a number of pesticides including cypermethrin (no purity given) on aromatase were assessed in the human JEG-3 cell line (tritium release assay). No inhibition of aromatase activity was seen at a concentration of 10 µM cypermethrin. The authors report that cypermethrin increases aromatase activity; values are not reported, but from the graphical data induction of approximately 1.4x, 1.5x and 1.6x the control was seen at concentrations of 1, 3, and 10 µM, respectively. The induction of aromatase activity was seen at concentrations of ≥3 µM after exposure for 24 hours (but not following exposure for 2 hours), and attained statistical significance only at the highest concentration tested of 10 µM. A marginal increase in the expression of *cyp19* mRNA (1.4 times) was not statistically significant compared to controls. The authors conclude that cypermethrin induces aromatase activity but (based on the response seen only at 24 hours), consider that this is not through transcriptional activation. It is speculated that some effect of cypermethrin on post-transcriptional regulation may be involved. The US EPA have reservations about the reliability of this study but consider that this may be acceptable if replicate data are provided.

Classification of study: supplemental information

Report: CA 5.8.3/23
Morinaga H. et al., 2004a
A benzimidazole fungicide, Benomyl, and its metabolite, Carbendazim, induce aromatase activity in a human granulose-like tumor cell line (KGN)
2004/1040397
Guidelines: none
GLP: no

In this study, a number of pesticides and other chemicals were screened for effects on catalytic aromatase activity (assessed as the conversion of androstenedione to oestrone) using the steroidogenic KGN cell line, which exhibits a relatively high level of aromatase activity. The single concentration of 10^{-5} M cypermethrin (no purity given) investigated was shown to slightly (but significantly) inhibit aromatase activity relative to controls, following incubation for 30 hours. There was no concurrent assessment of cytotoxicity; however significant cytotoxicity is not reported at this concentration in other *in vitro* assays. Further investigations were not performed. The US EPA review raises concerns over the reliability of this study (potential for false negative or false positive results) due to the use of a single concentration.

Classification of study: supplemental information

Report: CA 5.8.3/24
Taxvig C. et al., 2013a
In vitro - in vivo correlations for endocrine activity of a mixture of currently used pesticides
2013/1416882
Guidelines: none
GLP: no

The authors investigated the effects of various pesticides including cypermethrin (no purity given), either individually or in various combinations in the *in vitro* H295R cell steroidogenesis assay (exposure for 48 hours at concentrations between 1.6-100 µM). Cypermethrin at concentrations of 3.13, 6.25 and 12.5 µM was found to significantly increase oestradiol production. No significant effects were seen at concentrations of 25 or 100 µM and a marginal (but statistically significant) reduction in oestradiol production was seen at 50 µM. Cypermethrin at concentrations of 6.25 and 12.5 µM was found to significantly increase progesterone production; no effects were seen at concentrations of ≥25 µM. No effects of cypermethrin at any concentration were seen on testosterone production. All cypermethrin concentrations are reported to be non-cytotoxic.

Classification of study: supplemental information

van Meeuwen *et al* (2008) see CA 5.8.3/20 2008/1102356
Aromatase inhibiting and combined estrogenic effects of parabens and estrogenic effects of other additives in cosmetics
Toxicology and Applied Pharmacology 230 (2008) 372-382

Several test compounds, including M310I019 (3-hydroxy benzoic acid; no purity given) were tested on their effect on aromatase activity in vitro with microsomes from human placenta samples. M310I019 was tested in triplicates in 24-well plates in the range of 100 nM to 100 µM. For determination of aromatase activity 15 µg protein in 250 µL final volume was used together with 1β-^{3}H-androstenedion (357 nM). The authors reported that none of the hydroxylated benzoic acid isomers did inhibit aromatase statistically significant. In conclusion, M310I019 did not inhibit aromatase activity in microsomes derived from human placental tissue.

Classification of study: supplemental information

Overview of aromatase and steroidogenesis assays

Laville *et al* (2006) note the discrepancy between the results of their study with cypermethrin which reported weak aromatase induction and the study of Morinaga et al (2004) which reports weak aromatase inhibition with cypermethrin. The authors speculate that this discrepancy may be due to different regulation of aromatase expression between different cell types. Taxvig *et al* (2013) report a significant increase in the production of oestradiol in H295R cells exposed to cypermethrin, which is consistent with the findings of Laville *et al* (2006); however findings are inconsistent and only apparent at intermediate concentrations. The authors also report increased progesterone production at intermediate concentrations. Based on the results of these studies, there is no convincing evidence for an effect of cypermethrin on aromatase activity or steroidogenesis. No aromatase inhibiting effect was reported for the metabolite M310I019.

Table 5.8.3-6: Summary of aromatase and steroidogenesis assays

Study	Finding	Comment
Laville *et al* (2006)	Weak aromatase induction by cypermetrhrin	Response may be due to post-transcriptional regulation
Morinaga *et al* (2004)	Weak aromatase inhibition by cypermethrin	Single concentration assessed
Taxvig *et al* (2013)	Inconsistent effect on steroidogenesis	Increased oestradiol and progesterone synthesis seen at intermediate concentrations
van Meeuwen *et al* (2008)	Negative for M310I019	No aromatase-inhibiting effect reported for 3-hydroxy benzoic acid

Thyroid receptor assays in vitro

Du *et al* (2010) see CA 5.8.3/7 2010/1232195
Assessing Hormone Receptor Activities of Pyrethroid Insecticides and Their Metabolites in Reporter Gene Assays
Toxicological Sciences 116(1):58-66

Cypermethrin (92% pure) and its metabolites M310I011 (99% pure) and M310I001 (in report: DCCA; 99% pure) were assessed for activity in thyroid hormone receptor (stably transfected CV-1 cells) reporter gene assays at non-cytotoxic concentrations of between 10-9 M and 10-5 M. For agonistic activity tests, the CV-1 cells were exposed to various concentrations of tested chemicals, various concentrations of T3 (10-11 to 10-6 M) (positive control), or 0.1% DMSO (negative control). Cypermethrin showed significant but only slight antagonistic activity just reaching the relative inhibitory concentration of 20% to control at 10-5 µM (RIC20= 8.31 x10-6 M) and M310I011 showed a similar antagonistic activity without significance. M310I001 did not show any activity.

Classification of study: supplemental information

Overview of Thyroid receptor assays

Du *et al* [see CA 5.8.3/7 2010/1232195] investigated the potential of Cypermethrin, 3-PBA and DCCA to suppress T3-binding to the thyroid receptor. Among the investigated pyrethroids, cypermethrin and its metabolite were the least potent antagonists, which hardly or even barely reached RIC20 levels at 10^{-5}M. DCCA showed no activity.

Based on the results of this study, showing no agonistic and only weak antagonistic effects in the 10 µM dose range for cypermethrin, and less activity for the metabolites M310I011 and M310I001, the impact is considered of rather minor relevance for the human risk assessment of alpha-cypermethrin.

Table 5.8.3-7: Summary of Thyroid receptor interaction

Study	Finding for parent	Comment and findings for metabolites
Du *et al* (2010)	Cypermethrin: Negative (TR-Agonist) Positive (TR-Antagonist) - Weak response: RIC20: 8.31 x 10^{-6}M No response up to 10^{-6}M	TRβ-transactivation in CV-1 cells: Negative (TR-Agonist): - M310I011; M310I001 Negative (TR-Antagonist) - M310I001 Positive (TR-Antagonist) - M310I011, RIC20:4.76 x 10^{-6} M (not significant) No effect up to 10^{-6} molar

RIC20: Concentration oft he tested cheicals showing 20% reduction in the activity of 5x10^{-9}M T3 via TRβ

Studies in male animals *in vivo*

Investigations with Cypermethrin

Rat - Hershberger assays

Wu *et al* (2008) see CA 5.8.3/6 2008/1102139
Anti-androgenic properties of cypermethrin and β-cypermethrin
Chinese journal of industrial hygiene and occupational diseases 26(4)

The anti-androgenic properties of cypermethrin (no purity given) and beta-cypermethrin (no purity given) were investigated *in vivo* in castrated male SD rats using a Hershberger assay using cypermethrin dose levels of 7, 21 or 63 mg/kg bw/d and beta-cypermethrin dose levels of 6, 18 and 54 mg/kg bw/d. In vitro the substances were investigated in a transcriptional activation assay based on MDA-kb2 cells at concentrations from 10^{-5} – 10^{-8} mol/L. Cypermethrin showed anti-androgenic activity *in vitro* at concentrations of 10^{-6} and 10^{-5} M; no effects were seen with beta-cypermethrin at any concentration. *In vivo*, administration of cypermethrin was shown to affect androgen-dependent organ weight at the mid- and high dose level groups. No effects were seen with beta-cypermethrin. The authors therefore conclude that cypermethrin is a weak anti-androgen *in vitro* and *in vivo*, but that beta-cypermethrin showed no such activity.

Classification of study: supplemental information

Zhang *et al* (2008) see CA 5.8.3/5 2008/1102138
The antiandrogenic activity of pyrethroid pesticides cyfluthrin and β-cyfluthrin
Reproductive Toxicology 25: 491–496

Zhang *et al* (same lab than Wu et al., 2008) investigated the anti-androgenic activity of cyfluthrin and beta-cyfluthrin (no purities given) *in vitro* and *in vivo*; and obviously reproduced the findings of Wu et al., 2008 with cypermethrin and beta-cypermethrin. *In vivo*, anti-androgenic activity was assessed in a Hershberger assay. No effects were seen with beta-cypermethrin; however the administration of cypermethrin at 50 mg/kg bw/d is reported to result in significantly lower mean weights of the seminal vesicles, ventral and dorsolateral prostate in castrated rats treated with testosterone propionate. No effects were noted in mean weights of the LABC, Cowper's gland or glans penis. The US EPA assessment considers this study to show positive anti-androgenic effects for cypermethrin.

Classification of study: supplemental information

Report: CA 5.8.3/25
Tong J.-W. et al., 2009a
Anti-androgenic activity of Cypermethrin and Fenpropathrin in Hershberger assays
2009/1130983
Guidelines: none
GLP: no

In this study, groups of six young castrated male SD rats were administered subcutaneous injections of testosterone propionate and cypermethrin (in peanut oil; 94.1% pure) at dose levels of 0, 45, 90 or 180 mg/kg bw/d on seven consecutive days. A positive control group was administered the anti-androgen flutamide. Rats were sacrificed at 24 hours following administration of the final dose and organ weights recorded. Signs of toxicity were observed at the highest dose level; bodyweight gain was slightly (~20%) reduced in all treated groups. Absolute weights of the seminal vesicles, dorsal and ventral prostate and *levator ani* muscle were not significantly affected by treatment with cypermethrin. Serum levels of testosterone, FSH and LH were unaffected by treatment. Responses to the positive control confirmed the sensitivity of this assay. There is no evidence of anti-androgenic activity under the conditions of this assay.

Classification of study: supplemental information

Other studies

Rat - 15 day adult male rat assay

Report: CA 5.8.3/26
Hu J.-x. et al., 2011a
Toxic effects of Cypermethrin on the male reproductive system: with emphasis on the androgen receptor
2011/1296731
Guidelines: none
GLP: no

Cypermethrin (commercially sourced; 97.53% pure) was assessed in a 15-day study in groups of 12-intact adult male SD rats administered gavage doses of 0, 6.25, 12.5, 25 or 50 mg/kg bw/d. Following sacrifice, weights of the testes, epididymides, seminal vesicles and prostate were recorded. One testis from each rat was used for sperm counts; the other testis was used for histopathological assessment (following fixation in Bouin's), electron microscopy and immunohistochemical analysis of the androgen receptor. Plasma levels of testosterone, FSH and LH were measured using radioimmunoassay; FSH and LH were measured using commercial kits for human hormones and are therefore of questionable relevance. Reduced weight gain was seen in rats administered cypermethrin at dose levels of ≥12.5 mg/kg bw/d; bodyweight stasis/slight weight loss over the last few days of the study is noted in rats administered 25 mg/kg bw/d but not at 50 mg/kg bw/d. Bodyweight values are not reported but are presented graphically; based on the graphical data, terminal bodyweights are estimated to be approximately 100%, 94%, 90% and 90% of controls at dose levels of 6.25, 12.5, 25 and 50 mg/kg bw/d respectively. Overall weight gains of 100%, 67%, 45% and 45% are estimated. Mean absolute prostate weight was significantly lower than controls at dose levels of 25 mg/kg bw/d (76.2% of controls) and at 50 mg/kg bw/d (78.3%); mean absolute weights of the testes, epididymides and seminal vesicles were not significantly different to controls. Relative organ weights are not reported but relative prostate weights are estimated to be approximately 85% and 88% of the control value at 25 and 50 mg/kg bw/d respectively. Daily sperm production was significantly reduced in rats administered 50 mg/kg bw/d. Histopathological assessment of the testes showed a dose-related and statistically significant increase in the proportion of seminiferous tubules showing degeneration, in all treated groups. At dose levels of 6.25 and 12.5 mg/kg bw/d, slight tubular distortion, lumen changes and reduced spermatid numbers were observed. More marked findings, including seminiferous tubular atrophy were observed at 25 and 50 mg/kg bw/d. Seminiferous tubule numbers were reduced at 6.25 and 12.5 mg/kg bw/d only. A dose-related decrease in tubular circumference was seen in all treated groups. Ultrastructurally, various changes were observed including swollen mitochondria and disruption of gap junctions. Immunohistochemistry showed significantly reduced androgen receptor levels in the Sertoli cells, Leydig cells and peritubular myoid cells at dose levels of ≥12.5 mg/kg bw/d. Significantly reduced serum testosterone levels associated with significantly increased levels of FSH and LH were seen at 50 mg/kg bw/d.
The authors conclude that cypermethrin acts as an anti-androgen in the rat, reducing androgen receptor expression. Reduced expression of the androgen receptor in Sertoli cells adversely affects spermatogenesis. Reduced expression of the androgen receptor in Leydig cells adversely affects spermatogenesis and also reduces the level of testosterone synthesis.

Remark from the applicant: The study claims that testosterone is decreased, but the expected histological findings are missing which are normally associated with reduced testosterone (retention of spermatids, apoptosis of pachytene spermatocytes and round spermatids). These findings were obviously not observed in this study and their absence was not discussed. The FSH and LH levels were detected with a human detection kit and are therefore less convincing, too. Histopathological pictures are not convincing (necrosis, atrophy and distortion is not visible).
It is notable that effects on prostate weight were not reproduced in a comparable study performed by the same laboratory at a slightly higher dose level of 60 mg/kg bw/d that did not affect bodyweights (Li *et al*; 2013). The body weights of the used animals are unusually low for animals of this strain at that age. The study is limited due to methodological defects in histopathology and RIA.

Classification of study: supplemental information

A comparable assay in the same lab was performed by Li et al (2013):

Report:	CA 5.8.3/27 Fang L.Y. et al., 2012a Effects of Cypermethrin on male reproductive system in adult rats 2013/1416880
Guidelines:	none
GLP:	no

In this study, groups of 12 male SD rats were administered cypermethrin (98% pure) by gavage at dose levels of 0, 7.5, 15, 3 or 60 mg/kg bw/d on 15 consecutive days. Rats were sacrificed within 2 hours of the final dose. No signs of toxicity were observed; bodyweights were unaffected by treatment. Absolute weights of the testes, epididymides, seminal vesicles and prostate were, in contrast to Hu et al., 2010, unaffected by treatment. Daily sperm production decreased with a dose-response relationship in all treated groups; values attained statistical significance at 30 and 60 mg/kg bw/d. Morphological assessment showed effects on seminiferous tubule diameter in all treated groups. Histopathology revealed slight distortion of the tubules at dose levels of 7.5 and 15 mg/kg bw/d; more severe effects including tubular atrophy, reduced numbers of germ cells, Sertoli cells and Leydig cells were observed at 30 and 60 mg/kg bw/d. Serum testosterone levels were reduced in all treated groups; significantly at 30 and 60 mg/kg bw/d. FSH levels were increased in all treated groups, significantly at 60 mg/kg bw/d only. A trend to higher LH levels is seen in all treated groups; however values do not attain statistical significance in any group.

Remark from the applicant: The study claims that testosterone is decreased, but the expected histological findings are missing which are normally associated with reduced testosterone (retention of spermatids, apoptosis of pachytene spermatocytes and round spermatids). These findings were obviously not observed in this study and their absence was not discussed. The FSH and LH levels were detected with a human detection kit and are therefore less convincing, too. Histopathological pictures are not convincing (necrosis, atrophy and distortion is not visible).

Classification of study: supplemental information

Rat – 30 day

Report: CA 5.8.3/28
Grewal K.K. et al., 2010a
Toxic impacts of Cypermethrin behavior and histology of certain tissues of albino rats
2010/1232204
Guidelines: none
GLP: no

Groups of male and female rats (strain unspecified) were gavaged with cypermethrin (source and purity unspecified), in arachis oil at dose levels of 5 or 20 mg/kg bw/d for 30 days. The highest dose level was sufficient to cause a low rate of mortality; signs of toxicity were observed at both dose levels. Body weights are not reported. No gross changes were apparent in the testes; however testes are reported to have appeared smaller in treated rats. A significantly lower relative testis weight (56% of controls) was seen at the highest dose level. Histopathology is reported to have revealed extensive damage to the seminiferous tubules and alterations to the histoarchitecture; however the methods used (formalin fixation) and interpretation are less than optimal. The highest dose level also produced a marked reduction in spermatogenesis. This study is considered to be of limited reliability.

Classification of study: not further considered

Rat – 45 day

Report: CA 5.8.3/29
Joshi S.C. et al., 2010a
Evaluation of reproductive and developmental toxicity of Cypermethrin in male albino rats
2011/1296732
Guidelines: none
GLP: no

Groups of six male rats (strain not reported) were gavaged with cypermethrin (technical grade; purity unspecified) at dose levels of 0, 50, 75 or 100 mg/kg bw/d for 45 consecutive days. At termination, the testes were assessed histopathologically (following fixation in Bouin's) and were additionally assessed for various biochemical parameters. Sperm motility was also assessed. Treated males were also mated with untreated females; however the exact timing of this procedure is unclear. In a separate experiment, mated female rats were gavaged with cypermethrin at dose levels of 0 or 100 mg/kg bw/d on Gestation Days 6-17. Absolute weights of the testes, epididymides, ventral prostate and seminal vesicles were significantly decreased in all treated groups; effects on the epididymides and prostate exhibit a dose-response relationship. Values are presented graphically and with apparently incorrect units. Bodyweight effects and relative organ weights are not reported; relative organ weights cannot be calculated and the effects reported on absolute organ weights cannot be interpreted. Based on findings from other studies (e.g. Hu *et al*, 2011, reduced bodyweight gain or weight loss would be expected at all dose levels used in this study. Histopathology revealed reduced size of the seminiferous tubules, the complete arrest of spermatogenesis, increased intertubular space and ruptured interstitial cells at cypermethrin dose levels of 50 and 75 mg/kg bw/d. More severe effects including reduced numbers of Leydig and Sertoli cells were seen at 100 mg/kg bw/d. Biochemical analysis of the testes revealed significantly reduced levels of glycogen and sialic acid; significantly increased levels of protein, cholesterol, acid phosphatase and alkaline phosphatase in all treated groups and with a dose-response relationship. Effects on glycogen content were particularly marked, with reductions of 76%, 88% and 92% relative to controls seen at dose levels of 50, 75 and 100 mg/kg bw/d, respectively. Levels of serum testosterone, LH and FSH were significantly reduced in all treated groups in a treatment-related manner. Testicular and epididymal sperm counts were significantly reduced by treatment in all groups; caudal sperm motility was also significantly reduced in all treated groups. Findings correlate with reduced fertility levels of 50%, 20% and 0% in the respective treated groups, compared to 100% in the controls. Reduced weight gain is reported in treated females; however values are not presented. Treatment with cypermethrin is stated to result in reduced implantation numbers, high foetal loss, reduced litter size and reduced foetal size and weight; however no values are presented. The presented absolute weight data for male reproductive organs and hormone measurements are scientifically unsufficient for interpretation without data about body weight and details on systemic toxicity. The value of the histopathological investigations in this study is limited as documented in Figures showing shrinkage artefacts by the use of Bouin's as a fixative and the apparent comparison of findings in seminiferous tubules of different stages. More generally, the study is limited by poor reporting (specifically the lack of any information on systemic toxicity or bodyweight data) and the use of relatively high dose levels.

Classification of study: not further considered

Rats - 12-week

Report: CA 5.8.3/30
Elbetieha A. et al., 2001a
Evaluation of the toxic potentials of Cypermethrin pesticide on some reproductive and fertility parameters in the male rats
2001/1031943
Guidelines: none
GLP: no

Groups of male SD rats (approximately three months old) were administered cypermethrin (as the commercial product 'Cypermethrin 10') in the drinking water at dose levels stated to be equivalent to 0, 13.15, 18.93 or 39.66 mg/day (calculated to be equivalent to approximately 45, 63 and 132 mg/kg bw/d, based on an initial bodyweight of 300 g). Dose levels are reported to be equivalent to 2.5%, 5% and 10% of the LD_{50} (which is reported to be 4123 mg/kg bw in water). Males were treated for 12 weeks and then mated (1:2) with untreated females. Rats were cohabited for 10 days, after which point males were sacrificed; females were sacrificed after a further 10 days. Weight loss was reported for males in the intermediate dose group (~10% of the initial bodyweight); more marked weight loss (~16%) was seen at the highest dose level. Weight gain in the low dose group (~10 g) was comparable to controls. Effects on fertility (reduced numbers of pregnant females, implantation sites and viable foetuses; increased resorptions) were seen in all treated groups. Absolute weights of the testes, seminal vesicles and preputial gland were significantly increased in treated groups; sperm counts and daily sperm production were significantly reduced. Microscopically, the testes of treated rats were found to have congested blood vessels, haemorrhage around the semiferous tubules and increased connective tissue between tubules. Testicular morphology was also altered in exposed rats. At the highest dose level, serum levels of testosterone, FSH and LH were all significantly lower than controls. The reliability of this study is limited by the use of a commercial product containing relatively high levels of organic solvent and other co-formulants. Furthermore this report shows a high degree of additional methodological deficits, missing of relevant data in regard to general toxicity, mistakes in data presentation and questionable interpretation. **This study has been already discussed in the Zeta-Cypermetrhin DAR (2008) and was classified as questionable.**

Classification of study: not further considered

Rat – GD 7-21

Taxvig *et al* (2013) see CA 5.8.3/24 2013/1416882
In vitro - in vivo correlations for endocrine activity of a mixture of currently used pesticides
Toxicology & Applied Pharmacology 272: 757-766

In the *in vivo* phase of this study, groups of 12 female mated Wistar rats were gavaged with mixtures of 3 (Mix3) or "3 plus additional 2" (Mix5) pesticides (including cypermethrin as one of Mix3 and Mix5) on Gestation Days 7-21. Mixtures included compounds administered in equal proportions at single compound dose levels of 1, 3 or 10 mg/kg bw/d. No effects were seen on maternal bodyweight or adjusted bodyweight for Mix3. Plasma hormone levels in dams treated with Mix3 were unaffected for estradiol, progesterone, T3 and T4 at GD21. Litter parameters were unaffected by treatment and anogenital distance was comparable in all groups. Testosterone levels in placentas were unaffected in the Mix3 up to the highest dose as well as testiculare hormone levels in pups on GD21. No histopathological changes of testes were seen. Cypermethrin and its metabolite M310I011 were detected in the amniotic fluid from rats of all treated groups. The authors conclude that this study provides evidence that all single pesticides are able to affect steroidogenesis in vitro, but only the Mix5 exerted endocrine activity in vivo in dams and female fetuses, but that this is due to terbuthylazine. In conclusion, doses up to 10 mg/kg bw cypermethrin were not able to affect endocrine system in Wistar rats in vivo.

Classification of study: supplemental information

Mouse – PND 0-21

Report: CA 5.8.3/31
Wang H. et al., 2009a
Maternal Cypermethrin exposure during lactation impairs testicular development and spermatogenesis in male mouse offspring
2010/1232193
Guidelines: none
GLP: no

Groups of maternal female ICR mice were administered cypermethrin (Sigma) by gavage (in corn oil) at dose levels of 0 or 25 mg/kg bw/d from PND 0-21. Twelve male pups from six litters per group were sacrificed at PND 21 or PND 70. There were no signs of maternal toxicity; bodyweights were unaffected by treatment. Pup weights are not shown. Testes weights in pups (sacrificed at PND 21) from treated female mice were significantly lower than controls. Histopathology of the testes (limited methodological details presented) is reported to show a marked reduction in the layers of spermatogenic cells and an increased seminiferous tubule internal diameter in the treated group. Immunostaining did not show any difference in the numbers of Leydig cells between the treated and control groups; no significant differences in the number of apoptotic cells (investigated using TUNEL) were seen. Serum testosterone levels were markedly lower in the offspring of treated mice and testicular testosterone levels were also significantly lower; however the relevance of measuring serum testosterone levels in very young animals is questionabledue to the fluctuations of testosterone levels with regard to the prenatal surge of testosterone at around day 5, then very low levels of this hormone until puberty (day 35) starting to increase again at around day 25 (descend of testis). Levels of testicular cytochrome P450scc mRNA and protein were significantly lower in the offspring of treated mice. Testicular StAR, 17beta-HSD and $P450_{17\alpha}$ mRNA levels were slightly lower in the offspring of treated mice; however no significant effect was seen on levels of the corresponding proteins. In adult males (sacrificed at PND 70), maternal treatment with cypermethrin was shown to have a slight but significant negative effect on testes weight and epididymal sperm count (although findings are not normalised for epididymal weight and therefore the result is questionable). Histopathology of the testes showed disturbed spermatogenesis and an increase in the internal diameter of the seminiferous tubules. No significant effects were seen on serum testosterone levels or on testicular mRNA or protein levels of cytochrome P450scc mRNA, StAR, 17β-HSD and $P450_{17\alpha}$. Following mating with untreated females, no differences in fertility or litter parameters were seen between the males from treated or untreated mothers. The authors conclude that exposure to cypermethrin during lactation causes temporary endocrine disruption but permanent effects on testicular development and spermatogenesis in male offspring. The study is considered to be of limited value due to significant methodological flaws.

Classification of study: not further considered

Mouse – PND 21-42 study

Report: CA 5.8.3/32
Jin Y. et al., 2011a
Cypermethrin exposure during puberty induces oxidative stress and endocrine disruption in male mice
2011/1296733
Guidelines: none
GLP: no

Groups of young male ICR mice were orally administered cypermethrin at dose levels of 0, 5, 10 and 20 mg/kg bw/d (assumed to be by gavage) from PND 21-42 and sacrificed on the last day of treatment. At termination, the livers were removed, homogenised and assessed for markers of oxidative status. The expression of genes encoding antioxidant proteins was also assessed. General toxicity is not reported. Slight, but significant increases in superoxide dismutase, glutathione peroxidase and catalase activities were seen at the highest dose level only; malondialdehyde levels were unaffected. Total antioxidant capacity was reduced in all treated groups; significantly at 10 and 20 mg/kg bw/d. Hepatic superoxide dismutase and glutathione peroxidase mRNA levels were significantly increased at dose levels of 10 and 20 mg/kg bw/d; levels of catalase mRNA were unaffected by treatment. Hepatic HMG-CoA reductase levels were unaffected by treatment; however HMG-CoA synthase levels were markedly reduced. Testicular StAR mRNA levels were significantly reduced at 20 mg/kg bw/d. Testicular levels of P450scc and P450 17β-HSD were similar in all groups; however levels of P450 17α were significantly reduced at dose levels of 10 and 20 mg/kg bw/d. Serum testosterone level was slightly (but significantly) lower at 20 mg/kg bw/d. The study therefore indicates that endocrine effects are related to oxidative stress following the administration of cypermethrin to young male mice; however it is notable that effects were seen only in the presence of hepatic effects.

Classification of study: supplemental information

Mouse – PND 35-70 study

Report: CA 5.8.3/33
Wang H. et al., 2009b
Cypermethrin exposure during puberty disrupts testosterone synthesis via downregulating StAR in mouse testes
2010/1232194
Guidelines: none
GLP: no

Groups of male CD-1 mice (group size not reported) were gavaged with cypermethrin (sourced from Sigma) in corn oil at a dose level of 25 mg/kg bw/d from PND 35-70. No bodyweight effects were seen; signs of toxicity are not reported. Weights of the testes and epididymides were unaffected by treatment; however epididymal sperm count (not normalised to epididymal weight) was significantly reduced in treated mice. Treated mice showed changes to the architecture of the seminal vesicles and an increase in the number of apoptotic (TUNEL-positive) cells. Serum and testicular levels of testosterone were significantly lower in treated mice. Levels of testicular StAR protein and cytochrome $P450_{17\alpha}$ were significantly lower in cypermethrin-treated mice; levels of P450scc and 17β-HSD were unaffected by treatment. Immunostaining did not show any difference in the numbers of Leydig cells between control and treated mice.

Classification of study: supplemental information

Mouse – 5 day i.p.

Report: CA 5.8.3/34
Kumar S. et al., 2003a
Demonstration of sperm head shape abnormality and clastogenic potential of Cypermethrin
2004/1040396
Guidelines: none
GLP: no

Groups of male Swiss mice were administered cypermethrin (no further details provided) by intraperitoneal injection (in 0.15% DMSO) at dose levels of 0, 30, 60 and 90 mg/kg bw/d for five days. Animals were sacrificed on the 35th day, the testes weighed and caudal epididymal sperm visually assessed for abnormalities following staining with eosin-Y. Reduced weight gain was seen in mice administered 60 and 90 mg/kg bw/d cypermethrin. No significant effects of treatment were seen on testes weight. The proportion of abnormal sperm (various abnormalities of head shape) was significantly increased in mice at toxic dose levels of 60 and 90 mg/kg bw/d from 1.13% to approximately 2.5-3%. The relevance of this study is questionable due to the use of intraperitoneal dosing at comparatively high dose levels.

Classification of study: not further considered

Mouse – 14 days

Report: CA 5.8.3/35
Shaikh T.M.,Elfayoumi R.I., 2013a
Protictive effects of vitamine E against testicular enzymes toxicity induced by Cypermethrin in mice
2013/1417280
Guidelines: none
GLP: no

Groups of ten male MF1 mice were administered cypermethrin (as an unspecified 10% commercial product) by gavage in corn oil at dose levels of 0 or 2.8 mg/kg bw/d for 14 days. Additional groups were administered Vitamin E (100 mg/kg bw/d) or cypermethrin and Vitamin E. Testis weights were recorded and the levels of testicular enzymes were assessed following homogenisation. General toxicity is not reported. Mean absolute testis weight was significantly reduced in the group administered cypermethrin (to 77% of the control value); significant protection was seen in the group administered cypermethrin and Vitamin E (90% of the control value). Testicular homogenates from cypermethrin-treated mice showed significantly lower activities of AST, ALT, Acid Phosphatase (AcP), ALP and a markedly higher level of LDH. Co-administration of Vitamin E was shown to afford considerable protection against the effects of cypermethrin. The toxicological significance of these biochemical effects is unclear and do not appear to be directly relevant to an endocrine MoA. The reliability of the study is further limited through the use of a commercial product (likely to contain high levels of organic solvent and other co-formulants) and in the absence of any reporting of general toxicity. More generally, the study is poorly reported and the graphical data presented indicate an unusually low level of variation.

Classification of study: not further considered

Mouse – 34 days

Report: CA 5.8.3/36
Rodriguez H. et al., 2008a
Cypermethrin effects on the adult mice seminal glands
2009/1130984
Guidelines: none
GLP: no

Groups of 15 male CF1 mice were administered cypermethrin (92.5% purity; in vegetable oil) by a single intraperitoneal injection at a dose levels of 0 or 485 mg/kg bw. Three mice per group were sacrificed at time points of up to 34 days following treatment and the 'seminal glands' subjected to morphometric and histopathological assessment. In treated mice, significantly increased epithelial cell height was seen at 24 hours, with recovery at later time points. Cellular proliferation (measured by immunostaining for the marker protein Ki-67) was markedly increased at 24 hours only. Mast cell recruitment was significantly increased at 24 hours and continued to increase at subsequent time points. The endpoints morphometric analysis of epithelium height in the stretched and folded seminal glands and mast cell quantification are no validated endpoints and their interpretation is impossible without knowledge of the usual variation of these parameters. The toxicological relevance of the findings of this study is unclear, particularly given the use of a high intraperitoneal dose level and the small group size.

Classification of study: not further considered

Mouse – 6 / 12 week treatment with recovery

Report: CA 5.8.3/37
Al-Hamdani N.M.H.,Yajurvedi H.N., 2010a
Cypermethrin reversibly alters sperm count without altering fertility in mice
2010/1232192
Guidelines: none
GLP: no

Groups of 5 male Swiss mice were gavaged with cypermethrin (in water; as the commercial product Superkiller 10% EC) at dose levels of 1.38, 2.76 or 5.52 mg/kg bw/d on alternate days for 6 or 12 weeks, with or without a 6-week recovery period. Additional groups of treated males were mated with untreated females, following treatment for 6 weeks. General toxicity is not reported. Sperm counts were reduced in all treated groups, but effects were shown to be largely reversible after 6 weeks treatment withdrawal. The numbers of abnormal sperm were similarly significantly higher in all treated groups and were largely reversible after a further 6 weeks without treatment. No significant effects were seen on litter size, but litter weights were significantly lower in all treated groups. The reliability of this study is limited by the use of a commercial product containing relatively high levels of organic solvent and other co-formulants.

Classification of study: not further considered

Report: CA 5.8.3/38
Al-Hamdani N.M.H.,Narasinhachary Y.H., 2011a
Endocrine disruptive action of Cypermethrin in male mice
2011/1296735
Guidelines: none
GLP: no

Groups of 5 male Swiss mice were gavaged with cypermethrin (in water; as the commercial product Superkiller 10% EC) at dose levels of 1.38, 2.76 or 5.52 mg/kg bw/d on alternate days for 6 or 12 weeks with or without a 6-week recovery period. Bodyweight effects were seen in all treated groups, with weight loss observed at Weeks 8 and 12. Relative weights of the testes, epididymides, vas deferens, seminal vesicles and prostate were lower in all treated groups and generally showed a dose-response relationship. Some evidence for recovery was seen; however some organ weights were lower following the recovery period. Histopathology of the testes of treated mice revealed reduced spermatogenesis, degenerate spermatids, exfoliated spermatocytes, congested blood vessels and vacuolation of the seminiferous tubules. Recovery was seen at the low dose level but not at the highest dose level. Testicular architecture was also affected by treatment. Marked reductions in serum testosterone levels were seen in all treated groups; some evidence of recovery was seen at low and mid-dose levels following the 6-week treatment and at the low dose level following treatment for 12 weeks.

Classification of study: not further considered

Rabbit – 71 day i.p. administration

Report: CA 5.8.3/39
Ahmad L. et al., 2012a
Toxico-pathological effects of Cypermethrin upon male reproductive system in rabbits
2012/1367122
Guidelines: none
GLP: no

Adult male NZW rabbits 'procured from the local market' were administered cypermethrin technical (92% purity) in mustard oil by intraperitoneal injection at dose levels of 0, 50, 100 or 150 mg/kg bw at weekly intervals for ten weeks. Rabbits (2/group) were terminated at weekly intervals. The testes and epididymides were weighed and subject to histopathology. Sperm head counts were made from homogenised testes or epididymides. Serum testosterone concentrations were also measured. Signs of toxicity were observed in cypermethrin-treated groups; weight gain was reduced in all treated groups in a dose-related manner. Weights of the testes and epididymides were variable, but were higher in treated groups compared to controls. Grossly, the testes of treated animals appeared to be swollen. Microscopically, treated groups showed reduced spermatogenesis and the proliferation of connective tissue. Spermatogenic gain cells and apoptotic bodies were also seen at the higher dose levels. Testicular findings showed a time- and dose-response relationship. Sperm head counts were reduced in all treated groups; values often attained statistical significance. Significantly reduced serum testosterone concentrations were seen in all treated groups. The reliability of this study is limited by the use of intraperitoneal dosing, high dose levels, small group size and the potential confounding influence of systemic toxicity.

Classification of study: not further considered

Rabbit – 12 week

Report: CA 5.8.3/40
Yousef M.I. et al., 2002a
Protective role of Isoflavones against the toxic effect of Cypermethrin on semen quality and testosterone levels of rabbits
2003/1034259
Guidelines: none
GLP: no

Groups of six male NZW rabbits were administered cypermethrin (as a commercial 25% EC product) at dose levels of 0 or 24 mg/kg bw/d on alternate days for 12 weeks. Additional groups were administered isoflavones (genistein / daidzein mixture) at 2 mg/kg bw/d, or a combination of cypermethrin and isoflavones. Rabbits administered cypermethrin were of lower bodyweight at the end of the study period; however initial bodyweights are not reported. Food consumption was also lower in this group. Relative testes and epididymal weights were significantly lower in the cypermethrin-treated group; serum testosterone concentration was also significantly lower. Co-administration of isoflavones demonstrated a protective effect. Treatment with cypermethrin resulted in reduced ejaculate volume, increased reaction time; reduced sperm output, sperm concentration and motility and an increase in the proportions of abnormal and dead sperm. Co-administration of isoflavones again demonstrated a protective effect. The authors attribute the protective effects of isoflavone administration to its role as an antioxidant; however this conclusion is not considered to be particularly robust in the absence of consistent data for the role of isoflavones as antioxidants and also considering their documented hormonal effects. There is no measurement of endpoints directly related to oxidative stress in this study and reporting of other effects is limited by the absence of any indication of variability (results presented as graphs without error bars). Treatment with isoflavones alone in this study is also noted to have some beneficial effects on sperm parameters, which is somewhat surprising given the well documented oestrogenic effects of genistein. The study is considered to be of limited reliability.

Classification of study: supplemental information

Investigations with beta-cypermethrin

Rat - 15 day adult male rat assay

Report: CA 5.8.3/41
Liu L. et al., 2010a
Effects of Beta-Cypermethrin on male rat reproductive system
2010/1232198
Guidelines: none
GLP: no

Groups of 10 intact male SD rats were gavaged with beta-cypermethrin (>95% pure) at dose levels of 0, 15 or 30 mg/kg bw/d for 15 consecutive days. Signs of toxicity were not reported; weight gain was unaffected by treatment. Mean relative weights of the testes, epididymides and seminal vesicles were unaffected by treatment. Sperm head count (reflecting spermatogenic cell death) and thereof calculated daily sperm production were significantly reduced in both treated groups. Immunohistochemical analysis of androgen receptor levels revealed significantly reduced levels in both treated groups, with a dose-response relationship evident.
Remark from the applicant: Histopathology of the testes in this study was performed following formalin fixation, which may result in the production of artefacts including tubular shrinkage and chromatin condensation; immunohistochemial staining of Androgen receptor was not done in stage-aware fashion thereby the different expression patterns of stage V-VI and stage IX versus stage VII-VIII sertoli cells/seminiferous tubule are not considered. Assessments are therefore considered to be of limited reliability.

Classification of study: supplemental information

Mice – 35 days

Report: CA 5.8.3/42
Wang X.-Z. et al., 2009a
Beta-Cypermethrin impairs reproductive function in male mice by inducing oxidative stress
2009/1130986
Guidelines: none
GLP: no

Groups of ten male Kunbai mice were administered beta-cypermethrin (>99% pure) by gavage (in corn oil) at dose levels of 1, 10 or 20 mg/kg bw/d for 35 days. Reduced weight gain was seen in all treated groups; absolute weights of the testes, epididymides, seminal vesicles and prostate were reduced in treated groups. The co-administration of Vitamin E (20 mg/kg bw/d) with beta-cypermethrin (20 mg/kg bw/d) was shown to afford protection against bodyweight and organ weight effects. Reduced sperm counts and sperm viability were seen in treated groups; sperm motility was decreased and acrosome deformity was significantly increased at the highest dose level. A protective effect of Vitamin E was also seen on sperm parameters. At the low dose level, a decrease in the number of Leydig cells was observed. At the highest dose level, a marked effect was seen and spermatids were nearly completely absent from the lumen of the seminiferous tubules. Epididymal sperm were also nearly completely absent in this group. The effects on sperm count were prevented by co-administration of Vitamin E. Administration of beta-cypermethrin was also shown to cause ultrastructural effects in Leydig cells; a protective effect of Vitamin E was seen. The higher dose levels of beta-cypermethrin were shown to increase testicular malondialdehyde levels, levels of NO were also increased; these effects were reversed by Vitamin E. Total and inducible NO-synthase levels were increased in all treated groups; a protective effect of Vitamin E was apparent. Administration of beta-cypermethrin at the two higher dose levels was also shown to reduce the activities of catalase, glutathione peroxidase and SOD. The authors therefore conclude that the observed effects of beta-cypermethrin on the testes are due to oxidative stress.

Classification of study: supplemental information

Report: CA 5.8.3/43
Yulan Z. et al., 2009a
Beta-Cypermethrin induces Sertoli cells dedifferentiation by oxidative stress
2010/1232200
Guidelines: none
GLP: no

Male mice were gavaged with beta-cypermethrin (>99% pure) at dose levels of 0 or 20 mg/kg bw/d with or without Vitamin E (15 mg/kg bw/d) for 35 days. After termination, the testes were subjected to immunohistochemical analysis or measurement of total antioxidant capacity. General toxicity is not documented. In testes from the control group, PCNA staining was seen in nuclei from spermatogonia at the base of the seminiferous tubules. Staining was not detected in Leydig, Sertoli or spermatogenic cells. In mice treated with beta-cypermethrin, PCNA staining of spermatogonia was significantly reduced, but staining was seen in the Sertoli cells. In mice administered beta-cypermethrin and Vitamin E, PCNA expression in the Sertoli cells was reduced. In testes from the control group, Connexin 43 (Gap Junction alpha-1 protein; a marker of differentiation) staining was seen mainly in the in Leydig cells, but also in Sertoli cells and spermatocytes. No staining was observed in spermatids or sperm. In mice treated with beta-cypermethrin, Leydig cell staining was significantly reduced, as was staining in Sertoli cells and spermatocytes. This effect was not seen in the testes of mice administered beta-cypermethrin and Vitamin E. Total Antioxidant Capacity was markedly reduced in the testes of mice administered beta-cypermethrin; this effect was almost completely abolished by co-treatment with Vitamin E. The authors conclude that beta-cypermethrin administration to mice causes Sertoli cell de-differentiation and proliferation as a result of induced oxidative stress. The accuracy of this report is less than optimal: Methods are described (immunohistochemical detection of substance P which was definitely not performed. Based on the fact that similar endpoints (total oxidative capacity and the effect of vitamin E) are discussed in detail elsewhere (see Wang et al., 2009), this report is not further considered.

Classification of study: not further considered

Investigations with alpha-cypermethrin

Rat – 30 day

Manna *et al* (2004) see CA 5.3.1/1 2004/1040514
Repeated dose toxicity of alfa-cypermethrin in rats
J. Vet. Sci 5(3): 241-245

Adult Wistar rats (5/sex) were gavaged with alpha-cypermethrin (in DMSO; >99% pure) at dose levels of 0 or 14.5 mg/kg bw/d (equivalent to 10% of the LD50) for 30 days. Signs of toxicity are not reported; however clinical chemistry analyses reported significant effects of treatment including elevated ALP, AST, ALT and LDH activity; haematology revealed effects including a marked reduction in erythrocyte count. Necropsy revealed severe gastrointestinal haemorrhage. Significantly lower liver CAT and SOD activity were reported; liver malondialdehyde level was significantly reduced. The glutathione level was slightly lower; glycogen and cytochrome P450 levels were significantly lower. The pattern of induced enzymes nevertheless strongly suggests oxidative metabolism. Interestingly the GSH level is still unaffected in this study, which indicates a partially unimpaired anti-oxidative system in the liver (see copied tables from the publication).

Table 2. Effects of α-CP on certain biochemical parameters in serum and blood of rats after daily oral administration at 14.5 mg/kg for 30 days (Values are mean ± SE, n = 10)

Parameters	Control	α-CP treated
ALP activity (IU/L)	78.03 ± 2.58	161.53 ± 6.60*
AST activity (IU/L)	59.45 ± 3.52	72.00 ± 4.97*
ALT activity (IU/L)	12.00 ± 1.43	26.50 ± 1.67*
LDH activity (IU/L)	49.41 ± 2.58	64.80 ± 2.01*
TP (gm/dl)	8.12 ± .022	6.41 ± 0.17*
ALB (gm/dl)	4.53 ± 0.29	4.50 ± 0.26
GLB (gm/dl)	3.81 ± 0.21	2.16 ± 0.49*
Blood Glucose mmol/L	3.70 ± 0.48	6.22 ± 0.85*

*$p < 0.05$ in comparison with control
ALP: Alkaline Phosphatase, AST: Aspartate transaminase, ALT: Alanine transaminase; LDH: Lactate dehydrogenase, TP: Total protein; ALB: Albumin; GLB: Globulin.

Table 3. Effects of α-CP on certain biochemical parameters in liver of rats after daily oral administration at 14.5 mg/kg for 30 days (Values are mean ± SE, n = 10)

Parameters	Control	α-CP treated
CAT activity (U/mg protein)	0.39 ± 0.04	0.07 ± 0.01*
SOD (U/mg protein)	0.48 ± 0.02	0.13 ± 0.01*
MDA (nmol/mg protein)	0.24 ± 0.02	2.85 ± 0.18*
GSH (μmol/mg protein)	1.41 ± 0.16	1.30 ± 0.05
Glycogen (mg%)	7.94 ± 0.24	5.15 ± 0.34*
P450 (nmol/mg microsomal protein)	2.91 ± 0.02	2.74 ± 0.04*
b5 (nmol/ mg microsomal protein)	1.16 ± 0.07	1.28 ± 0.05

*$p < 0.05$ in comparison with control
CAT: Catalase, SOD: Superoxide dismutase, MDA: Malondialdehyde, GSH: Reduced glutathione.

Histopathology of the testes (Bouin's used as fixative) is reported to reveal interstitial oedema and vacuolation of the seminiferous tubules, otherwise care should be taken when tissue *pieces* are used for histopathology based on squeezing artefacts of tissues. The reliability of this study is therefore limited by confounding toxicity and sub-optimal histopathological methods.

Classification of study: supplemental information

Mouse – 28 day

Prakash *et al* (2010) see CA 5.3.1/2 2010/1232197
Evaluation of Testicular Toxicity Following Short-term Exposure to Cypermethrin in Albino Mice
Toxicology International 17(1):18-21

Groups of 6 male Swiss mice were gavaged with alpha-cypermethrin (>99% purity; in arachis oil) at dose levels of 0 (controls) or 250 mg/kg bw/d for 14 or 28 days. No bodyweight effects are reported; sign of toxicity are not specifically mentioned but would be expected at this dose level. Weights of the testes, seminal vesicles, prostate gland and epididymides are reported to be increased; however values are not actually reported. AST activity in testicular homogenates was significantly reduced at Day 14 and slightly (but significantly) increased at Day 28. ALT activity was significantly increased, ALP activity was significantly reduced and cholesterol levels were significantly increased. The toxicological significance of these biochemical investigations is unclear. Plasma testosterone levels are stated to be significantly decreased at both time points; however a high level of variation is apparent and the level of significance is not reported. However, the weight increase of testes and accessory glands combined with testosterone decrease are not convincing. Histopathology revealed various testicular changes; however methodological data are deficient and the methods used are known to produce artefacts. TEM also revealed ultrastructural effects in the treated groups but no control data are shown for comparison. The described findings like rupture of cell membrans, increased lipochrome pigment and shrinkage in the nucleus, or decreased cytoplasmic organelles are not seen in the pictures. There is no indication that the histopathological evaluation was performed in a stage aware fashion and therefore it is questionable if tubuli of the same stage were looked at. Overall the study is considered to have significant methodological and reporting deficiencies and additionally used a very high dose level of alpha-cypermethrin likely to have resulted in significant general toxicity.

Classification of study: not further considered

Rabbit – 28 day dermal application

Aksoy et al (2009) see CA 5.3.3/1 2009/1130982
Investigation of potential testicular toxicity of subchronic dermal application of alphacypermethrin in rabbits
J. vet. Pharmacol. Therap. 32 (Suppl. 1), 129–265

In this poster presentation, groups of seven male NZW rabbits were dermally administered alpha-cypermethrin (source/purity not reported; in DMSO) daily at dose levels of 0, 40, 200 or 1000 mg/kg bw/d for 28 days. No deaths occurred; however partial paralysis was apparent in rabbits at the highest dose level. Bodyweights were reported to have been unaffected by treatment. The proportion of abnormal spermatozoa was significantly higher in treated groups and in a dose-dependent manner. Histopathological investigation of the testes revealed severe degeneration including vacuolation between spermatogonial cells, collapsed/shrunken tubules, decreased spermatogonial cell count and intratubular giant cell formation. The severity of the testicular findings was dose-related. The reliability of this reference is very limited due to the lack of methodological data.

Classification of study: not further considered

Investigations with metabolites

Rat – 90 day

Beck L.S. (1991a) and Halliwell W.H. (1991a) see CA 5.8.1/34 1980/1001707 and CA 5.8.1/35 1980/1001708
Histopathologic Evaluation of Tissues and Organs from albino rats in a 90 day subchronic oral dosing study with meta-Phenoxybenzylaldehyde (3-PBAld) provided by Ethyl Corporation

The study is described in detail under section CA 5.8.1/34 and 5.8.1/35 and suffers some weaknesses with regard to unequal dose volume, non GLP etc. Nevertheless it gives some insight into the relevance of the *in vitro* anti-androgenic activity of 3-phenoxybenzaldehyd. Sprague Dawley rats were administered 3-PBAld in corn oil via gavage over a period of 90 days at dose levels of 50, 150, or 300 mg/kg bw/d. Organ weights were measured for liver, kidneys, adrenals, gonads, heart, spleen and brain. In addition to those mentioned organs histopathology was performed on all animals of the control and high dose group on parathyroid gland, pituitary gland, prostate, and thyroid gland as well as uterus. Abs. and rel. organ weights for adrenals, testes and uterus were unaffected and histopathology revealed no effect on endocrine organs (adrenal gland, parathyroid, pituitary gland and thyroid) or the reproductive system (epididymis, prostate, seminal vesicles, testes, cervix, uterus, ovaries, uterine horns, and vagina) that could be attributed to the test substance. The LOAEL was determined at 150 mg/kg bw as based on effects on liver and kidney weights. The results of this 90 day study show that the in vitro found anti-androgenic effect of 3-PBAld is not displayed in vivo.

Classification of study: supplemental information

Overview of studies in male animals *in vivo*

A large number of studies investigating effects in male animals are available for cypermethrin, alpha- or beta-cypermethrin. The results of some of these studies are limited by the use of commercial cypermethrin products which are likely to contain high levels of organic solvent components. A number of studies investigating testicular histopathology are also limited by the use of non-current terminology, the absence of staging assessment and tissue processing methods including fixation in Bouin's fluid or formalin, which may result in the production of artefacts. A number of studies are also limited through the use of non-physiological (intraperitoneal) dosing routes, the use of excessively high dose levels and/or the lack of reporting of general systemic toxic effects.
Wu et al., (2008) and Zhang *et al* (2008) report a positive response in a Hershberger assay with cypermethrin in SD rats at relatively high oral dose levels of 63 mg/kg bw (no positive response at 7 and only prostate weight reduced at 21 mg/kg bw) and 50 mg/kg bw/d (the only dose level investigated); a negative result is reported for beta-cypermethrin at 6, 18, 54 mg/kg bw by Wu et al., 2008 and at 50 mg/kg bw by Zhang et al., 2008. Findings are consistent with the results of studies *in vitro* which report weak anti-androgenic effects for cypermethrin but not for beta-cypermethrin, possibly due to metabolites. A negative result for reproductive organ weight changes is reported for cypermethrin by Tong *et al* (2009) at higher dose levels (up to 180 mg/kg bw/d); however this study used subcutaneous dosing. The US EPA assessment considers the Zhang *et al* (2008) study to be sufficiently robust to meet the data requirement for a Hershberger assay. Organ weight effects are also noted by a number of other authors. Weights of androgen-sensitive organs and tissues (prostate, testis, seminal vesicle, epididymides) are generally reported to have been reduced by treatment; For example Hu et al., 2011, reported significant organ weight effects on seminal vesicles and prostate at 50 mg/kg bw, however 2 years later the same lab published under Li *et al* (2013) a compareable study with higher dose levels but no effects on organ weights in intact rats administered cypermethrin at gavage dose levels up to 60 mg/kg bw/d (i.e. a dose level higher than that assessed in the Hershberger assay of Zhang *et al* (2008) and similar to that used in the Hershberger of Wu et al., 2008). Elbetieha *et al* (2001) report significantly increased organ (testes and seminal vesicle) weights in rats administered a commercial product containing 10% cypermethrin; however it is possible that other components of the formulation contribute to this effect. Based on *in vitro* data showing a difference in anti-androgenic activity between cypermethrin (active) and beta-cypermethrin (inactive) and the likely role of metabolism in the anti-androgenic potential of cypermethrin, it is considered unlikely that alpha-cypermethrin would show anti-androgenic effects in the Hershberger assay. This is supported by the positive response to cypermethrin but not to the same dose level of beta-cypermethrin reported by Wu et al., (2008) and Zhang *et al* (2008) and also by the 15 day adult rat male assay by Liu *et al* (2010) with beta-cypermethrin. Data indicate that the weak anti-androgenic activity of cypermethrin (and other pyrethroids) is associated with metabolites. The greater level of metabolism of *trans*-isomers compared to *cis*-isomers might explains the lower activity of beta-cypermethrin (which contains a lower proportion of *trans*-isomers) and would also predict an absence of activity for alpha-cypermethrin (which consists entirely of *cis2*-isomers). An anti-androgenic effect however was not reproducible under in vivo circumstances with the most potent in vitro metabolite 3-phenoxybenzaldehyde.

Testicular histopathology is noted in a number of studies; however the reliability of a number of studies is questioned due to the use of tissue fixation and processing methods known to produce artefacts such as tubular shrinkage and chromatin condensation; in the respect it is notable that changes to tubular histoarchitecture are reported by a number of authors. The study of Grewal *et al* (2010) is not considered to be reliable in the absence of any documentation of systemic toxicity and due to poor fixation techniques. The study of Wang *et al* (2010b) uses Bouin's fixation which can induced shrinkage artefacts and the histopathological interpretation is less than optimal. It is notable in this study that sperm effects were associated with oxidative stress and bodyweight effects, which would indicate specific organ toxicity rather than effects mediated through an endocrine MoA. This conclusion is further supported by the protective effect of Vitamin E.

Highly interesting are the 15 day intact male rat studies of Hu *et al* (2011) and Li *et al* (2013) which appear to be generally reliable. Although the studies are not entirely consistent with regard to reproductive organ weight effects (reported at the dose level of 25 and 50 mg/kg bw/d by Hu *et al*, 2011 but not at the highest dose level of 60 mg/kg bw/d by Li *et al*), both studies report effects on the testes (histopathology and reduced sperm production) associated with a reduction in serum testosterone levels. Reduced sperm production after substance administration in corn oil was seen with cypermethrin from doses of 30 mg/kg bw/day onwards, with beta-cypermethrin from 15 mg/kg bw onwards. Under the hypothetical assumption that sperm toxicity is attributed to the cis2-Isomers, this would imply an effect with alpha-cypermethrin at 6.6 mg/kg bw (22% of the cypermethrin) or 6 mg/kg bw (40% of beta-cypermethrin).
The 71day i.p. rabbit study of Ahmad *et al* (2012) is of very limited value to the use of a non-physiological route of administration (intraperitoneal injection), the high dose levels used and the lack of reporting of systemic toxicity, and the very small group size. The rabbit study of Yousef *et al* (2011) is of limited value due to the use of a commercial product. While the study reports protection against the testicular effects by co-administration of an isoflavone mixture, there is no direct evidence for any effects on oxidative status. Isoflavones such as genistein are additionally known to have hormonal effects. The rabbit study of Aksoy *et al* (2009) using dermal administration of alpha-cypermethrin is a poster abstract and, as such, contains only very limited methodological information. The mouse pubertal study of Jin *et al* (2011) reports the down-regulation of genes involved in cholesterol transport and testosterone synthesis, resulting in reduced levels of circulating testosterone. Findings are associated with oxidative stress and, additionally, the liver appears to be more sensitive than the testes. Effects in the testes may therefore be secondary to other effects and do not represent an endocrine MoA. The study of Wang *et al* (2010a) is not considered to be reliable due to serious deficiencies in methodology and reporting. While this study suggests that a reduction in circulating testosterone may be due to reduced expression of testicular StAR and $P450_{17\alpha}$ mRNA, results are not entirely consistent with other similar studies. Jin *et al* (2011) also report the down-regulation of testicular StAR and $P450_{17\alpha}$ mRNA, but Wang *et al* (2010b) report the down-regulation of testicular StAR and $P450_{scc}$ mRNA. The study of Rodriguez *et al* (2009) is severely limited due to the use of a very high intraperitoneal dose. Liu *et al* (2010) report a reduction in androgen receptor expression as responsible for reduced spermatogenesis in mice exposed to beta-cypermethrin. The study is limited in terms of methodology due to the use of formalin fixation (which is known to induce artefacts); immunochemical staining for the androgen receptor was not standardised according to stage. Joshi *et al* (2011) used Bouin's as a fixative and testicular histopathology was not standardised for stages. The study is additionally not considered to be reliable in the absence of

any information on systemic toxicity or bodyweight; based on data from other studies, general toxicity would be predicted at the dose levels used. Elbetieha *et al* (2010) used a commercialformulation of cypermethrin; the results of this study are therefore of limited relevance. The studies of Al-Hamdani & Narasinhachary (2010, 2011) are not considered to be reliable due to the use of a commercial formulation of cypermethrin. The study of Prakash *et al* (2010) has significant methodological deficiencies including the use of Bouin's as a fixative and the absence of information on stage standardisation for the histopathological assessment. The dose level used in this study (250 mg/kg bw/d) is very high and would be predicted to result in significant general toxicity, although findings are not discussed. Biochemical investigation of homogenised testes shows significant differences in the activities in some enzymes (ALT, AST, ALP); however the toxicological significance of this finding is unclear. The study is not considered to be sufficiently reliable. Manna *et al* (2004) report histopathology associated with effects of alpha-cypermethrin on oxidative status in the testes; however this would appear to a non-specific effect as findings were associated with general toxicity and effects on hepatic oxidative status. The study of Kumar *et al* (2004) reports the induction of sperm abnormalities by cypermethrin, but is of limited relevance due to the use of a relatively high intraperitoneal dose. Zhou *et al* (2010) report the induction of oxidative stress in mice administered beta-cypermethrin, with significant protection given by co-administration of Vitamin E. The study is somewhat limited by the lack of discussion of general toxicity; however findings are consistent with those of Wang *et al* (2009) in indicating that testicular toxicity for beta-cypermethrin is secondary to oxidative stress rather than due to an endocrine MoA. Al Shaikh & Elfayoumi (2013) similarly report a protective effect of Vitamin E against reduced testis weight and biochemical changes induced by cypermethrin in the mouse. The study was performed with a commercial product and is therefore of limited reliability; however the consistency with other studies is notable. As with other studies, the value of Kumar *et al* (2004) is also limited due to the use of a non-physiological route of administration (intraperitoneal injection).

Testicular effects are reported for cypermethrin, beta-cypermethrin and alpha-cypermethrin; and this is in contrast to the different responses seen in the Hershberger assays, where Cypermethrin but not beta-cypermethrin induced reproductive organ weight changes. The nature of the histopathological findings, an association with general toxicity (including oxidative stress) and a number of studies demonstrating protective effects of anti-oxidants are clearly inconsistent with an endocrine MoA. Findings represent a more general toxic mechanism, and one that is not specific to the testes or male reproductive tract. While findings apparently consistent with an endocrine mode of action such as reduced circulating testosterone are reported by a number of authors, this effect could equally be induced by direct toxicity to the testes. Other findings such as increased LH / FSH levels and reduced weights of reproductive tract organs would also occur as a secondary effect of testicular toxicity. This direct effect on the testes is associated with general systemic toxicity (where reported) and is seen only at dose levels likely to result in general toxicity.

In order to investigate the potential to induce testicular toxicity /sperm toxicity with alpha-cypermethrin, a separate study has been performed and is further presented under CA 5.8.3/51.

Table 5.8.3-8: Summary of studies in male animals *in vivo*

Study	Result	Dose level	Comment
Wu et al., (2008)	Positive Hershberger (rat) Negative Hershberger (rat)	(neg. at 7-21) **≥63** mg/kg bw/d Cyp Neg at 6-18-54 mg/kg bw/d beta-Cyp	Same lab than Zhang et al., 2008
Zhang *et al* (2008)	Positive Hershberger (rat) Negative Hershberger (rat)	50 mg/kg bw/d (oral) Cyp 50 mg/kg bw/d (oral) beta-Cyp	Relied on by US EPA
Tong *et al* (2009)	Negative Hershberger (rat)	≤180 mg/kg bw/d (s/c) Cypermethrin	-
Hu *et al* (2011)	Reduced prostate weight Reduced sperm production Testicular histopathology Reduced testosterone, increased LH/FSH Reduced androgen receptor expression	Rat ≤50 mg/kg bw/d (gavage) Cypermethrin	Methodological deficiencies
Grewal *et al* (2010)	Reduced testis weight Testicular histopathology	Rat 5, 20 mg/kg bw/d (gavage)	Methodological deficiencies
Wang *et al* (2009)	Reduced testis, prostate, SV weights Reduced sperm count/mobility	Mouse ≤20 mg/kg bw/d beta-cypermethrin	Induction of oxidative stress; protective effects of Vitamin E
Li *et al* (2013)	No effects on organ weights; reduced sperm production Testicular histopathology Reduced testosterone, increased LH/FSH	Rat ≤60 mg/kg bw/d (gavage) Cypermethrin	
Ahmad *et al* (2012)	Increased testis & epididymal weights Testicular histopathology Reduced testosterone	Rabbit ≤150 mg/kg bw (ip) Cypermethrin	Limited reporting High dose level
Yousef *et al* (2011)	Reduced testes & epididymal weights Reduced testosterone Reduced sperm production	Rabbit 24 mg/kg bw/d (25% EC product) gavage	Protection by isoflavones
Aksoy *et al* (2009)	Testicular histopathology Abnormal sperm	Rabbit ≤1000 mg/kg bw/d (dermal) alpha-cypermethrin	Very limited methodological data

Study	Result	Dose level	Comment
Jin *et al* (2011)	Reduced testosterone Reduced testicular StAR mRNA Reduced testicular P45017α-HSD	Mouse ≤20 mg/kg bw/d oral	Findings associated with oxidative stress in the testes and liver
Wang *et al* (2010)	Testicular histopathology Reduced testosterone Reduced sperm count Reduced StAR protein Reduced P45017α	Mouse 25 mg/kg bw/d oral Cypermethrin	
Rodriguez *et al* (2010)	Seminal gland histopathology	Mouse 485 mg/kg bw ip Cypermethrin	Very high dose level Non-relevant exposure route Findings of unclear significance
Liu *et al* (2010)	Testicular histopathology Reduced sperm count Reduced androgen receptor levels	Rat ≤30 mg/kg bw/d oral beta-cypermethrin	Methodological deficiencies
Joshi *et al* (2011)	Reduced testes, prostate, epididymal and SV weights Testicular histopathology Reduced spermatogenesis Reduced testosterone, LH, FSH Reduced fertility	Rat ≤100 mg/kg bw/d oral Cypermethrin	Methodological deficiencies, poor reporting
Elbetieha *et al* (2001)	Increased testes and SV weights Reduced sperm production Testicular histopathology Reduced fertility Reduced testosterone, LH, FSH	Rat ≤132 mg/kg bw/d oral Cypermethrin 10% EC	Limited reliability
Al-Hamdani & Narasinhachary (2010)	Reduced sperm counts	Mouse ≤5.52 mg/kg bw/d oral Cypermethrin 10% EC	Effects reversible following withdrawal of treatment
Prakash *et al* (2011)	Increased testes, prostate, epididymal and SV weights Reduced testosterone Testicular histopathology	Mouse 250 mg/kg bw/d oral alpha-cypermethrin	Limited relevance due to the high dose level

Study	Result	Dose level	Comment
Manna *et al* (2004)	Testicular histopathology	Rat 14.5 mg/kg bw/d oral alpha-cypermethrin	Limited relevance due to significant systemic toxicity
Kumar *et al* (2004)	Sperm abnormalities	Mouse 90 mg/kg bw/d ip cypermethrin	Limited relevance due to ip dosing
Zhou *et al* (2010)	Oxidative stress	Mouse 20 mg/kg bw/d oral beta-cypermethrin	Protection by Vitamin E
Wang *et al* (2010)	Testicular histopathology Reduced testosterone Reduced spermatogenesis Reduced P450scc	Mouse 25 mg/kg bw/d oral Cypermethrin	Maternal animals dosed: effects seen in offspring
Al Shaikh & Elfayoumi (2013)	Reduced testis weight Biochemical changes	Mouse 2.8 mg/kg bw/d oral Cypermethrin 10% EC	Limited value; protection by Vitamin E

Studies in female animals i*n vivo*

Investigations with cypermethrin

Rat - 28 day

Report: CA 5.8.3/27
Fang L.Y. et al., 2012a
Effects of Cypermethrin on male reproductive system in adult rats
2013/1416880
Guidelines: none
GLP: no

Groups of 8-10 weaned female SD rats (age unspecified) were administered cypermethrin (95% purity) by gavage in 5 ml/kg bw peanut oil at dose levels of 0, 20, 40 or 80 mg/kg bw/d for 28 days. Weight gains in the cypermethrin-treated groups were lower than controls, but without a dose-response relationship. The rate of vaginal opening was increased, in the first week of treatment in the mid and high dose groups, and organ weights (ovarian, uterus and vagina) were increased with a clear dose-response relationship. However the informative value of this study is limited based on reporting deficiencies: Therate of vaginal opening is reported by week only and is no usual parameter and hardly interpretable. Standardized parameter for estrogenic/anti-estrogenic activity like the age at vaginal opening in combination with the body weight at the day of vaginal opening, length of cycle and age at first estrus after vaginal opening, percent of animals cycling and percent of animals cycling regularly are not given, and theage of animals was not specified. Mean and relative ovary, uterus and vagina weights were significantly higher in all treated groups; findings show a clear dose-response relationship.

Table 5.8.3-9: Time of vaginal opening

Group	Week 1	Week 2	Week 3
- control	37.5%	75.0%	100.0%
20 mg/kg bw/d	40.0%	70.0%	100.0%
40 mg/kg bw/d	62.5%	100.0%	100.0%
80 mg/kg bw/d	60.0%	90.0%	100.0%
+ control	100.0%	100.0%	100.0%

The results of this study therefore indicate some estrogenic effect of cypermethrin after dosing over 28 days *in vivo* but the reliability is somehow hampered based on deficits in methods and missing standard parameters.

Classification of study: supplemental information

Rat - 30 day

Grewal *et al* (2010) see CA 5.8.3/28 2010/1232204
Toxic Impacts of Cypermethrin on Behavior and Histology of Certain Tissues of Albino Rats
Toxicology International 17(2):94-98

Groups of male and female rats (strain unspecified) were gavaged with cypermethrin (in arachis oil; source and purity unspecified) at dose levels of 5 or 20 mg/kg bw/d for 30 days. The highest dose level was sufficient to cause a low rate of mortality; signs of toxicity were observed at both dose levels. Body weights are not reported. In female rats a complete loss of ovarian follicular cells and oocytes was seen at the highest dose level; accumulation of fluid was seen in the Graafian follicle. The study is considered to be of limited reliability in the absence of any specific reporting of systemic toxicity.

Classification of study: not futher considered

Rat – GD 7-21

Taxvig *et al* (2013) see CA 5.8.3/24 2013/1416882
In vitro - in vivo correlations for endocrine activity of a mixture of currently used pesticides
Toxicology & Applied Pharmacology 272: 757-766

In the *in vivo* phase of this study, groups of 12 female mated Wistar rats were gavaged with mixtures of 3 (Mix3) or "3 plus additional 2" (Mix5) pesticides (including cypermethrin as one of Mix3 and Mix5) on Gestation Days 7-21. Mixtures included compounds administered in equal proportions at single compound dose levels of 1, 3 or 10 mg/kg bw/d. No effects were seen on maternal bodyweight or adjusted bodyweight for Mix3. Plasma hormone levels in dams treated with Mix3 were unaffected for estradiol, progesterone, T3 and T4 at GD21. Litter parameters were unaffected by treatment and anogenital distance was comparable in all groups. Testosterone levels in placentas were unaffected in the Mix3 up to the highest dose as well as testiculare hormone levels in pup on GD21. No histopathological changes of testes were seen. Cypermethrin and its metabolite M310I011 were detected in the amniotic fluid from rats of all treated groups. The authors conclude that this study provides evidence that all single pesticides are able to affect steroidogenesis in vitro, but only the Mix5 exerted endocrine activity in vivo in dams and female fetuses, but that this is due to terbuthylazine. In conclusion, doses up to 10 mg/kg bw cypermethrin were not able to affect endocrine system in Wistar rats in vivo.

Classification of study: supplemental information

Investigations with alpha-cypermethrin

Rat and Mice – uterotrophic assay

Liang *et al* (2005) see CA 5.8.3/15 2005/1043340
A study on the estrogenic effects of alpha-and theta-cypermethrin
Chin Occup Med. August 2005, Vol 32, No.4, 25-26

In an uterotrophic assay performed in groups of eight ovariectomised female SD rats, no effect on uterus weight was seen in response to subcutaneous injection of alpha-cypermethrin (14 mg/kg bw/d; no purity given) or theta-cypermethrin (520 mg/kg bw/d; no purity given) on five consecutive days. A suitable response to the positive control substance (100 µg/kg bw β-oestradiol) was seen in this assay.
In an uterotrophic assay performed in groups of eight intact female NH mice, no effect on uterus weight was seen in response to subcutaneous injection of alpha-cypermethrin (3 mg/kg bw/d) or theta-cypermethrin (10 mg/kg bw/d) on three consecutive days. A suitable response to the positive control substance (100 µg/kg bw β-oestradiol) was seen in this assay.
The reporting of the study is limited, no data on bodyweight or organweight are given. Taken the results of this study as given, this would indicate no oestrogenic effect of alpha-cypermethrin and theta-cypermethrin after s.c. injection in rats and mice.

Classification of study: supplemental information

Investigations with Metabolites

Rat - pubertal female assay with metabolites

Laffin *et al* (2010) see CA 5.8.3/16 2010/1232196
The pyrethroid metabolites M310I011 and M310I024 do not exhibit estrogenic activity in the MCF-7 human breast carcinoma cell line or Sprague–Dawley rats
Toxicology 267: 39-44

In a pubertal assay, groups of 5-8 immature female rats were gavaged with the pyrethroid metabolites M310I011 and M310I024 (no purities given) at dose levels of 0, 1, 5 or 10 mg/kg bw/d from weaning until detection of the onset of puberty. Rats were subsequently assessed daily for first dioestrus. The authors do not report any statistically significant effect of exposure to either metabolite on uterine wet weight; however slightly higher absolute (1.6 times control) and relative (1.4 times control) uterus weights were seen in rats administered 10 mg/kg bw/d M310I011. No significant effect was seen of either metabolite on the attainment of puberty or sexual maturity.

Classification of study: supplemental information

Summary of studies in female animals *in vivo*

Li *et al* (2008) report a positive response in a female pubertal assay at dose levels of 20, 40 and 80 mg/kg bw/d cypermethrin. A trend to earlier attainment of puberty is seen from 40 mg/kg bw onwards; however the value of the study is limited by reporting and by relatively high doses used which are considered to induce general toxicity. A clear and dose-related increase in ovary, uterus and vaginal weights is seen at all dose levels in this study. The clear response seen in this study is somewhat unexpected given the absence of a clear effect of cypermethrin in oestrogenicity assays *in vitro*; however the response may be attributable to the generation of higher levels of metabolites *in vivo*. The study of Laffin *et al* (2010) indicates a weak oestrogenic effect of the metabolite M310I011; it is notable that the dose levels used in this study are lower than the cypermethrin dose levels used by Li *et al* (2008).
Liang et al (2005) reports negative results in the uterotrophic assays performed in the rat and mouse for alpha-cypermethrin (14 and 3 mg/kg bw/d respectively) and theta-cypermethrin (520 and 10 mg/kg bw/d respectively). This study used administration by subcutaneous injection. The study of Taxvig *et al* (20008) does not show any hormonal change in dams and fetuses related to cypermethrin doses during pregnancy of up to 10 mg/kg bw although either cypermethrin and its metabolite M310I011 are detectable at all dose levels in the amniotic fluid at GD21. Grewal *et al* (2010) does not provide any relevant information.
Although evidence for the oestrogenicity of cypermethrin *in vitro* is inconsistent, based on information on the relative oestrogenic activity of cypermethrin metabolites, the positive response for cypermethrin in vivo reported by Li *et al* (2008) is likely to be due (at least in part) to its metabolites formed after gavage of high doses, like for example via M310I024. Furthermore *in vitro* data indicate that the oestrogenicity of permethrin is increased by the formation of 4-HO cis-permethrin and more relevant by the formation of 4-HO-PBAlc. The metabolism of *cis*-isomers is less than that of *trans*-isomers, and for alpha-cypermethrin no formation of 4-HO cis-permethrin or 4-HO-PBAlc was observed in vivo. In this respect the positive oestrogenic responses with cypermethrin described by Li *et al* (2008) are considered less relevant for alpha-cypermethrin. This is supported by Liang *et al* (2005) based on no uterotrophic effect for alpha-cypermethrin in the rat and mouse respectively. However the dose levels are lower than those used for cypermethrin by Li *et al* (2008) (20-80 mg/kg bw/d), however these may have been limited by the higher toxicity of the cis-isomers.

In conclusion, no endocrine related effects are reported for alpha-cypermethrin or are considered relevant under consideration of the metabolic turnover in females in vivo.

Table 5.8.3-10: Overview of studies in female animals *in vivo*

Study	Result	Dose level	Comment
Li *et al*, 2008	Earlier puberty Increased ovary, uterus & vagina weights	Rat ≥20 mg/kg bw/d oral Cypermethrin	-
Laffin *et al*, 2010	No effects on puberty Slightly increased uterus weight	Rat ≤10 mg/kg bw/d oral 3-PBA	-
Liang *et al*, 2005	No uterotrophic effect	Rat / mouse alpha-cypermethrin 14 / 3 mg/kg bw/d theta-cypermethrin 520 / 10 mg/kg bw/d	-
Taxvig *et al*, 2013	Exposure of fetuses during pregnancy to cypermethrin and 3-PBA, no hormonal changes, no histopathological testis changes	Rat ≤10 mg/kg bw/d (GD7-21)(mixture study)	cypermethrin administered as a mixture component
Grewal *et al*, 2010	Loss of ovarian follicle cells and oocytes	Rat ≤20 mg/kg bw/d oral Cypermethrin	Limited reliability

Other studies

Gene expression and regulation studies

Report: CA 5.8.3/44
Pan C. et al., 2012a
Effects of Cypermethrin on the ligand-independent interaction between androgen receptor and steroid receptor coactivator-1
2012/1367063
Guidelines: none
GLP: no

The authors report the use of a two-hybrid assay to investigate the effects of cypermethrin (99% pure) on the interaction between the androgen receptor N-terminal and C-terminal domains using a CAT reporter gene assay in CV-1 cells. Recruitment of the steroid receptor coactivator-1 (SRC-1) was shown to be mediated primarily by the N-terminus of the androgen receptor. Exposure to cypermethrin at a concentration of 10^{-5} M was shown to significantly inhibit the interaction of androgen receptor activation function-1 (AF-1) on the N-terminal domain and SRC-1. The authors postulate that the interaction with cypermethrin may down-regulate expression of androgen receptor-dependent genes.

Classification of study: supplemental information

Report: CA 5.8.3/45
Pan C. et al., 2013a
Anti-androgen effects of the pyrethroid pesticide Cypermethrin on interactions of androgen receptor with corepressors
2013/1416881
Guidelines: none
GLP: no

In a further study by the same authors using mammalian two-hybrid assays, cypermethrin (995 pure) at a concentration of 10^{-5} M was shown to significantly enhance the interaction between the androgen receptor and its co-repressors SMRT and NCoR. These repressors are known to play key roles in the transcriptional repression of the androgen receptor. The authors propose that this interaction is a novel mechanism for the anti-androgenic activity of cypermethrin.

Classification of study: supplemental information

Report: CA 5.8.3/46
Hu J.-x. et al., 2011b
Anti-androgen effects of Cypermethrin on the amino- and carboxyl-terminal interaction of the androgen receptor
2012/1367064
Guidelines: none
GLP: no

This study investigated the potential of cypermethrin (99% pure) to interact with specific domains (the carboxy-terminal ligand-binding domain and the amino-terminal domain) of the androgen receptor using a mammalian two-hybrid assay. Interaction of these two domains is required for full transcription potential of the receptor and agonists such as DHT are known to induce this interaction. Co-transfected CV-1 cells were exposed to concentrations of cypermethrin for 24 hours alone or in the presence of the androgen agonist DHT. Exposure of cells to cypermethrin at concentrations of 10^{-7} to 10^{-5} M did not induce expression of the CAT reporter gene. Exposure to cypermethrin at a concentration of 10^{-5} M (but not 10^{-6} or 10^{-7} M) in the presence of DHT (concentrations of 10^{-7} M and 10^{-8} M) was shown to significantly reduce the induced expression of the reporter gene. Results are presented graphically and do not appear to be of great magnitude (a relative CAT activity of approximately 6 compared to approximately 7 in controls at both DHT concentrations). Exposure to the known androgen receptor antagonist nilutamide was shown to significantly reduce DHT-induced expression of the reporter gene at the concentrations investigated (10-7 to 10^{-5} M) and to a much greater degree compared to cypermethrin (relative CAT activity of approximately 1.5 compared to approximately 7 in controls at 10^{-8} M DHT). The authors suggest that cypermethrin exerts an antagonistic effect on the androgen receptor through interference with the interaction between the receptor and ligands such as DHT.

Classification of study: supplemental information

Summary: **The results of these studies indicate that the anti-androgenic effect of cypermethrin seen in studies *in vitro* may not be due to direct binding to the androgen receptor. In this respect, it is notable that the studies of androgenicity *in vitro* are transcriptional activation assays and therefore do not directly measure receptor binding.**

Effects on sperm in vitro

Report: CA 5.8.3/47
Song L. et al., 2008a
Effects of Fenvalerate and Cypermethrin on rat sperm motility patterns in vitro as measured by computer-assisted sperm analysis
2008/1102216
Guidelines: none
GLP: no

The effects of cypermethrin (99.05% purity) on rat sperm mobility were assessed *in vitro*. Sperm suspensions were incubated with cypermethrin at concentrations of 1, 4, 16 or 64 µM for 1, 2, or 4 hours and motility assessed using an automated system. Following exposure for 1 hour, sperm curvilinear velocity was unaffected by treatment. Average path velocity was significantly increased at 64 µM; straightline velocity, beat cross frequency, linearity and straightness were all significantly reduced at ≥ 4 µM; the amplitude of lateral head displacement was significantly reduced at 64 µM only. Following exposure for 2 hours, curvilinear velocity and average path velocity were unaffected by treatment. Straightline velocity, beat cross frequency, linearity and straightness were significantly reduced at ≥ 4 µM; linearity was also significantly reduced at 1 µM. The amplitude of lateral head displacement was unaffected by treatment. Following exposure for 4 hours, curvilinear velocity was significantly reduced at ≥16 µM. Average path velocity was unaffected by treatment. Straightline velocity, beat cross frequency, linearity and straightness were significantly reduced at ≥16 µM; the amplitude of lateral head displacement was unaffected by treatment.

Classification of study: supplemental information

Report: CA 5.8.3/48
Yuan C. et al., 2009a
Effects of Permethrin, Cypermethrin and 3-Phenoxybenzoic acid on rat sperm motility in vitro evaluated with computer-assisted sperm analysis
2010/1232201
Guidelines: none
GLP: no

The effects of cypermethrin and its metabolite M310I011 on sperm motility were assessed *in vitro*. Epididymal sperm from male SD rats was exposed to cypermethrin (>99% purity) or M310I011 at concentrations of 1, 4, 16 or 64 µM for 1, 2 or 4 hours; sperm parameters were measured using an automated system. Exposure to cypermethrin concentrations of ≥4 µM for 1 or 2 hours resulted in significantly decreased straight-line velocity, linearity, beat cross frequency and straightness. After exposure for 4 hours, effects were only seen at ≥16 µM. At the highest concentration, a number of samples showed no motile sperm. In contrast to cypermethrin, the metabolite M310I011 was shown not to have any effects on sperm motility parameters.

Classification of study: supplemental information

Summary: Effects on sperm motility *in vitro* are shown by the studies of Song *et al* (2008) and Yuan *et al* (2010) for cypermethrin. Studies used relatively high concentrations of questionable physiological relevance.
However, the effects seen are not indicative of an endocrine MoA and are more likely to be a consequence of the known effects of cypermethrin on sodium channels.

Not yet peer reviewed studies

Report:	CA 5.8.3/49 [redacted] 2014 BAS 310 I (Alphacypermethrin) 15-Day Intact Adult Male Rat Assay Administration by Gavage 2014/1275120
Guidelines:	U.S. EPA Office of Science Coordination and Policy, Washington D.C., U.S.A; Validation of 15-day intact adult male rat assay as a potential screen in the endocrine disruptor screening program tier-1 battery, 29 Aug 2007
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)
Remark:	~~The study report is at the time of dossier submission not finalized with regard to QAU-check and will therefore be delivered at a later stage.~~ The range finder used for dose setting is provided under KCA 5.8.3/51 2014/1289319.

Executive Summary
The aim of this study was to determine the toxicological profile of alpha-cypermethrin including the target organs and the "no observed adverse effect level" in the 15 days adult male rat assay after oral administration by gavage in corn oil. In this type of study special attention is given to selected male reproductive organs and sperm parameters.
The test item was administered to groups of 15 male adult Wistar rats for 15 days at doses of 2.0, 3.5, and 6.6 mg/kg bw/day. No treatment related clinical signs of toxicity and no mortalities were observed. No adverse effects on body weight were observed. Furthermore, no adverse effects on food consumption and body weight gain were observed. When compared to the control group the mean absolute and relative organ weights were not significantly changed. No test item related macroscopic lesions were recorded. Sperm motility and morphology as well as sperm head counts in testes or in cauda epididymidis were not influenced by the test item. Furthermore, no significant differences in androgen receptor staining could be detected. In conclusion, a NOAEL of 6.6 mg/kg bw/day can be derived, as no adverse findings were observed after treatment with 6.6 mg/kg bw/day (highest dose tested).
(DocID 2014/1275120)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:	BAS 310 I (Alphacypermethrin)
Description:	solid / white
Lot/Batch #:	PMAM000622
Purity:	99.2%
Stability of test compound:	The test substance was stable under storage conditions over the test period as guaranteed by the sponsor (expiry date 12 Dec 2015)

2. Vehicle: corn oil (1 mL/kg)

3. Test animals:

Species:	Rat
Strain:	Crl:WI(Han)
Sex:	Male
Age:	70 ± 1 days
Weight at dosing (range):	265.1 - 306.2 g
Source:	Charles River Laboratories, Research Models and Services, Germany GmbH, Sulzfeld, Germany
Acclimation period:	at least 7 days
Diet:	Ground Kliba maintenance diet mouse/rat "GLP", Provimi Kliba SA, Kaiseraugst, Switzerland, ad libitum
Water:	water, ad libitum
Housing:	group housing (maximum 4 per cage) in polysulfonate (H-Temp) cages type 2000P with dust-free wooden bedding
Environmental conditions:	
Temperature:	20 - 24°C
Humidity:	30 - 70%
Air changes:	15 per hour
Photo period:	12 h light / 12 h dark (06:00 - 18:00 / 18:00 - 06:00)

B. STUDY DESIGN AND METHODS

1. Dates of experimental work: 26-Aug-2014 – 13-Jan-2015
(In life dates: 02/03-Sep-2014 (start of administration) to 17/18-Sep-2014 (necropsy)

2. Animal assignment and treatment:

Range finding study

The test item was administered to groups of 3 male Wistar rats for 14 days at dose levels of 0 (vehicle control; test group 0), 10 mg/kg body weight/day (test group 1) and 25 mg/kg body weight/day (test group 2) by gavage. Corn oil (1 ml/kg) served as vehicle.

Main study

The animals were distributed into 4 test groups of each 15 animals. Each test group was subdivided into Section A and B including 8 and 7 animals, respectively. The animals were treated with the test item for 15 days at daily doses of 2, 3.5, 6.6 mg/kg bw or with the vehicle alone with an administration volume of 1 mL/kg bw.

Table 5.8.3-11: Overview on test groups and applied doses

Test group	Dose (mg/kg bw/d)[b]	Concentration (g/100 mL)	No. of animals	Section	Animal No.	Cage No.
0	0	0.0	15	A	1 – 4 5 – 8	1 2
				B	9 – 12 13 – 15	3 4
1	2.0	0.2	15	A	16 – 19 20 – 23	5 6
				B	24 – 27 28 – 30	7 8
2	3.5	0.35	15	A	31 – 34 35 – 38	9 10
				B	39 – 42 43 – 45	11 12
3	6.6	0.66	15	A	46 – 49 50 – 53	13 14
				B	54 – 57 58 – 60	15 16

3. Test substance preparation and analysis:

For the test substance preparations, the specific amount of test substance will be weighed and filled up with Corn Oil Ph. Up to the desired volume, subsequently released manually.
The stability of the test substance in corn oil at room temperature over a period of 7 days had been verified during the study (Project No.: 01Y0265/01Y012). Homogeneity of BAS 310 I (Alphacypermethrin) in the diet was performed in the highest and lowest concentration. Additionally, concentration control was performed in all concentrations at the beginning of the administration period.

4. Statistics:

Means and standard deviations (S.D.) of each test group were calculated for several parameters. In addition, the following statistical analyses were carried out:

Parameter	Statistical test	Markers in the tables	References
Clinical examinations Body weight and body weight change	A comparison of each group with the control group was performed using DUNNETT's test (two-sided) for the hypothesis of equal means	* for $p \leq 0.05$ ** for $p \leq 0.01$	DUNNETT, C.W. (1955): A multiple comparison procedure for comparing several treatments with a control. JASA, Vol. 50, 1096-1121 DUNNETT, C.W. (1964). New tables for multiple comparisons with a control. Biometrics, Vol. 20, 482-491

Feces, rearing, grip strength forelimbs, grip strength hindlimbs, foot-splay test, motor activity	Non-parametric one-way analysis using KRUSKAL-WALLIS test (two-sided). If the resulting p-value was equal or less than 0.05, a pairwise comparison of each dose group with the control group was performed using WILCOXON test (two-sided) for the equal medians	* for $p \leq 0.05$ ** for $p \leq 0.01$	SIEGEL, S. (1956): Non-parametric statistics for the behavioural sciences. McGraw-Hill New York
<u>Pathology</u> Weight parameters Cell count	Non-parametric one-way analysis using KRUSKAL-WALLIS test (two-sided). If the resulting p-value was equal or less than 0.05, a pairwise comparison of each dose group with the control group was performed using WILCOXON-test (two-sided) for the equal medians WILCOXON-test (two-sided)	* for $p \leq 0.05$ ** for $p \leq 0.01$	HETTMANNSPERGER, T.P. (1984): Statistical Inference based on Ranks, John Wiley & Sons New York, 132-140. International Mathematical and Statistical Libraries, Inc., 2500 Park West Tower One, Houston, Texas 77042-3020, USA, nakl-1 - nakl-3 MILLER, R.G. (1981): Simultaneous Statistical Inference, Springer-Verlag New York Inc., 165-167 NIJENHUIS, A. and S.W. WILF (1978): Combinatorial Algorithms, Academic Press, New York, 32-33
<u>Clinical pathology</u> Sperm analysis parameters	Pairwise comparison of each dose group with the control group using the WILCOXON-test (one-sided) with Bonferroni-Holm adjustment for the hypothesis of equal medians; If only control and one dose group are measured, WILCOXON-test (one-sided) without adjustment were used. For the percentage of abnormal sperms (ABNORMAL6_C) values <6% were set to 6% (cut off 6%)	* for $p \leq 0.05$ ** for $p \leq 0.01$	HOLM, S. (1979): A Simple Sequentially Rejective Multiple Test Procedure. Scand. J. Statist. 6, 65-70

C. METHODS

1. Observations:

Range finding study

Body weight was determined before the start of the administration period in order to randomize the animals and on study days 0, 3, 7, 10 and 14. Food consumption and water consumption were determined on study days 3, 7, 10 and 14. The animals were checked daily for any abnormal clinically signs before the administration as well as within 2 hours and within 5 hours after the administration. Because the animals of test group 2 (25 mg/kg bw/d) were sacrificed moribund on study day 0 no food consumption and water consumption could be determined.
The animals were sacrificed using CO_2, necropsied and assessed by gross pathology. Animals which died intercurrently were sacrificed in a moribund condition, were sacrificed without further examinations.

Main study

Mortality

A check for moribund and dead animals was made twice daily from Mondays to Fridays and once daily on Saturdays, Sundays and public holidays. If animals were in a moribund state, they were sacrificed and necropsied.

Clinical signs

During the recovery period the animals were checked once daily for any abnormal clinical signs. Abnormalities and changes were documented for each animal.

Detailed clinical observations

All animals were subjected to detailed clinical observations (DCO) outside their cages once before the beginning of the administration period (day 0) and subsequently once a week. For observation, the rats were removed from their cages and placed in a standard arena (50×37.5×25 cm). The scope of examinations and the scoring of the findings that were observed were based on the current index of findings in PDS ToxData® software and include but were not limited to the following parameters listed:

1. Abnormal behavior in handling
2. Fur
3. Skin
4. Posture
5. Salivation
6. Respiration
7. Activity/arousal level
8. Tremors
9. Convulsions
10. Abnormal movements
11. Gait abnormalities
12. Lacrimation
13. Palpebral closure
14. Exophthalmos
15. Assessment of the feces discharged during the examination (appearance/consistency)
16. Assessment of the urine discharged during the examination
17. Pupil size

2. Body weight and body weight change:
Body weight was determined before the start of the administration period in order to randomize the rats. During the administration period the body weight was determined on study day 0 (start of administration period) and thereafter at weekly intervals. The difference between the body weight on the respective day of weighing and the body weight on day 0 was calculated as body weight change.

3. Food and water consumption:
Food consumption was determined weekly (as representative value over 3 days) and calculated as mean food consumption in grams per rat and day. Drinking water consumption was monitored by daily visual inspection of the water bottles for any changes in volume.

3. Clinical pathology:
Blood samples were taken from fasted animals by puncturing the retrobulbar venous plexus under Isoflurane anesthesia. Blood sampling and examination were carried out in a randomized sequence (the list of randomization instructions will be compiled with a computer).

The following parameters have been examined:

Hormones:
Serum samples were taken and frozen at -80°C for storage. Measurement of FSH, LH and Testosterone should only have been carried out if there was an indication for effects on organ weights and/or sperm parameters. The determination should also have been triggered based upon potential alterations of histopathology.

Sperm parameters:
Immediately after necropsy and organ weight determination the right testis and cauda epididymidis were taken from all male animals.

The following parameters were determined:

Parameter	Unit	Method	References
Sperm motility	%	microscopic evaluation	Slott, Suarez and Perreault, "Rat sperm motility analysis: Methodological considerations" in: "Reproductive Toxicology", Vol 5, pp. 449-458 (1991)
Sperm morphology	%	vital staining with eosin; microscopic evaluation	M.H. Feuston, K.R. Bodnar, S.L. Kerstetter, C.P. Grink, M.J. Belcak and E.J. Singer, "Reproductive
Sperm head count (cauda epididymis)	Mio/g cauda epididymis	microscopic evaluation with MAKLER chamber after homogenization	Toxicity of 2-Methoxyethanol Applied Dermally to Occluded and Nonoccluded Sites in Male Rats" in:
Sperm head count (testis)	Mio/g testis	microscopic evaluation with MAKLER chamber after homogenization	Toxicology and applied Pharmacology 100, 145-161 (1989) (Laboratory modification)

Sperm motility examinations were carried out in a randomized sequence. Sperm head count (testis and cauda epididymis) and morphology was evaluated in all animals.

The right testis and right cauda epididymidis were deep frozen at -20°C until evaluation.

4. Pathology and histopathology:

Necropsy

The animals were sacrificed by decapitation under isoflurane anesthesia. The exsanguinated animals were necropsied and assessed by gross pathology. Animals which die intercurrently or were sacrificied in a moribund state were necropsied as soon as possible after their death and assessed by gross pathology.

Organ weights

The following weights were determined in all animals sacrificed on schedule:

1. Anesthetized animals
2. Adrenal glands
3. Brain
4. Cauda epididymis
5. Epididymides
6. Heart
7. Kidneys
8. Liver
9. Pituitary gland
10. Prostate
11. Seminal vesicles with coagulating glands
12. Spleen
13. Testes
14. Thyroid glands

Organ/tissue fixation

The following organs/tissue specimens were carefully removed, processed histotechnically and examined.
The following organs or tissues were fixed in 4% formaldehyde solution or in modified Davidson´s solution:

1. All gross lesions	26. Ovaries
2. Adrenal glands	27. Oviducts
3. Aorta	28. Pancreas
4. Bone marrow (femur)	29. Parathyroid glands
5. Brain	30. Pharynx
6. Cecum	31. Pituitary gland
7. Coagulating gland	32. Prostate
8. Colon	33. Rectum
9. Duodenum	34. Salivary glands (mandibular and sublingual glands)
10. Epididymis, left	
11. Esophagus	35. Sciatic nerve
12. Extraorbital lacrimal glands	36. Seminal vesicles
13. Eyes with optic nerve (modified Davidson's solution)	37. Skeletal muscle
	38. Skin
14. Femur with knee joint	39. Spinal cord (cervical, thoracic and lumbar cord)
15. Harderian glands	
16. Heart	40. Spleen
17. Ileum	41. Sternum with marrow
18. Jejunum (with Peyer's patches)	42. Stomach (forestomach and glandular stomach)
19. Kidneys	
20. Larynx	43. Testis, left (modified Davidson's solution)
21. Liver	
22. Lungs	44. Thymus
23. Lymph nodes (mesenteric and axillary lymph nodes)	45. Thyroid glands
	46. Trachea
24. Mammary gland (male)	47. Urinary bladder
25. Nose (nasal cavity)	

Histopathology

Fixation was followed by histotechnical and immunohistological processing, examination by light microscopy and assessment of findings according to the table below.

Organs	**Test group**			
	0	**1**	**2**	**3**
1. All gross lesions	A2	A2	A2	A2
2. Adrenal glands	A1			A1
3. Epididymis, left	A1			A1
4. Pituitary gland	A1			A1
5. Prostate	A1			A1
6. Seminal vesicles with coagulating gland	A1			A1
7. Testis, left	A1/I1	I1*	I1*	A1/I1

A = hematoxylin and eosin (H&E) stain
I = immunohistology (androgen receptor)
* = not evaluated
1 = all animals/test group
2 = all animals affected/test group
Prostate served as positive control for androgen receptor staining.

The organs were trimmed according to the "Revised guides for organ sampling and trimming in rats and mice". (Ruehl-Fehlert et al., 2003; Kittel et al., 2004; Morawietz et al., 2004).

A correlation between gross lesions and histopathological findings was attempted. After completion of the histopathological assessment by the study pathologist an internal peer review was performed. Results presented in this report reflect the consensus opinion of the study pathologist and the peer review pathologist.

Immunohistology staining
Sections were dewaxed with xylene and graded ethanol, the slides were then washed and kept in deionized water until antigen retrieval. Antigen retrieval was performed with 0.2 mol boric acid, pH 7.0 for 10 minutes at 120°C in a pressure cooker. Sections were cooled in deionized water. Endogenous peroxidase was then blocked with 6% hydrogen peroxide. Sections were rinsed in deionized water and incubated with the primary antibody (androgen receptor N-20, 1:600; Santa Cruz Biotechnology Inc., Heidelberg, Germany) for 2 hours at room temperature. Sections were then rinsed in wash buffer, and the label enzyme complex (polymerfluid, ZYTOMED Systems GmbH, Berlin, Germany) was applied for 30 minutes. Sections were then rinsed in wash buffer before the chromogen DAB (ZYTOMED Systems GmbH, Berlin, Germany) was applied for 3 minutes. Slides were counterstained with Mayer's hematoxylin and coverslipped with Pertex.
For evaluation and assessment of androgen staining, the immunostained slides of all animals were digitalized using a Hamamatsu NanoZoomer (Slide Scanner). Resulting images were viewed and processed with the Hamamatsu NanoZoomer viewer software (NDP.view.2.3.10).

Evaluation and assessment of testis tubule perimeter as well as number of Sertoli cells positively stained for androgen receptor
Beginning at the top left, the digitalized image was searched for the presence of stage VII, VIII, and IX tubuli, respectively. Tubuli were staged according to the criteria in Russell et al. (1990). Once a tubule of an appropriate stage was found, the perimeter of this tubule was measured using the freehand form feature of the software and exported as a jpg-file. Sertoli cells in this tubule were differentiated and detected on the base of their location within this tubule and their shape and size. Sertoli cells positively stained for androgen receptor (brown DAB staining on nuclei) were counted in each tubule. The number of positive Sertoli cells per tubule perimeter was calculated as positive cells divided by the length of the perimeter. In each image it was attempted to find at least 5 tubuli of each of the 3 stages of interest. This was possible in slides of most animals. On the whole, approximately 100 tubuli per group and stage were evaluated using slides from all 15 animals per test group.

II. RESULTS AND DISCUSSION

RANGE FINDING STUDY

Mortality
All animals of test group 2 (25 mg/kg bw/d) were sacrificed moribund on study day 0 between 2-5 h after administration. No other animal died prematurely or were sacrificed moribund in the present study.

Clinical examinations
Tonic-clonic convulsions, tremors, piloerection, poor general condition, salivation and ploughed nose-first into bedding were observed in all animals of test group 2 (25 mg/kg bw/d). Additionally, nasal discharge and rolling convulsions were observed in one of three animals of test group 2 (25 mg/kg bw/d). The high dose group was thus discontinued as severe grades of tremors and further neuriotoxic effects were observed at the day of dosing between hour 2 and 5.
All animals of test group 1 (10 mg/kg bw/d) showed unsteady gait, piloerection, tremors, poor general condition, salivation and ploughed nose-first into bedding. Additionally, ataxia, splayed limbs, abdominal position, hyperexcitability and excessive grooming was observed in animal No. 6 of test group 1 (10 mg/kg bw). Two animals (No. 5 and 6) did not recover completely from administration at study day 1 and showed poor general condition ahead of the substance administration at study day 2. Body weight loss or stagnation were obvious in all animals of test group 1 (10 mg/kg bw). Therefore the dose was reduced from 10 to 5 mg/kg bw/d from study day 2 onwards. Animal No. 6 of test group 1, which showed ataxia, tremors, splayed hindlimbs, unsteady gait, salivation and poor general state after the first administration of 5 mg/kg bw on study day 2, received no test substance preparation on study days 3, 4 and 5. Animal No. 4 and 5 showed tendency of recovery under administration of 5 mg/kg bw and no test substance-related adverse findings were observed from study day 4 onwards.

Food consumption
No significantly changes of food consumption could be observed. On study day 3 food consumption of test group 1 (10 as well as 5 mg/kg bw/d) was decreased by 25% compared to the control group.

Body weight data
Body weight loss was observed in test group 1 (10 as well as 5 mg/kg bw/d) on study day 3. Body weight change was significantly decreased on study day 10 by 45% compared to the control group.

Table 5.8.3-12: Mean body weight of rats administered alpha-cypermethrin

Sex	Males		
Test-item	Control	Alpha-cypermethrin	
Dose level [mg/kg bw]	0	5/10	25
Mean body weight [g]			
- D 0	288.5	289.0	284.7
- D 3	295.5	280.4	#
Δ% (compared to control)	-	-5.1	-
- D 7	305.7	294.7	#
Δ% (compared to control)	-	-3.6	-
- D10	311.1	301.4	#
Δ% (compared to control)	-	-3.1	-
- D14	319.5	310.6	#
Δ% (compared to control)	-	-2.8	-

#: Group excluded from statistics

Table 5.8.3-13: Mean body weight gain of rats administered alpha-cypermethrin

Sex	Males		
Test-item	Control	Alpha-cypermethrin	
Dose level [mg/kg bw]	0	5/10	25
Mean body weight gain [g]			
- D 0 - 3	7.0	-8.6	
Δ% (compared to control)	-	-222.3	-
- D 0 - 7	17.2	5.7	#
Δ% (compared to control)	-	-67.1	-
- D 0 - 10	22.6	**12.4***	#
Δ% (compared to control)	-	-45.3	-
- D 0 - 14	31.0	21.5	#
Δ% (compared to control)	-	-30.6	-

*: $p \leq 0.05$
#: Group excluded from statistics

Based on the results of the range finding study a dose slightly above 5 mg/kg bw and below 10 mg/kg bw was considered adequate as high dose in the main study.

Cypermethrin and beta-cypermethrin are reported in the open literature to show effects in the 15 day adult male rat assay, which is considered a Level 4 in vivo Screening Assay in the Guidance on Identifying Endocrine Disrupting Effects (ECETOC Technical Report No. 106). The hypothesis is presented that cypermethrin and beta-cypermethrin act as anti-androgens in the rat by reducing androgen receptor expression and thereby adversely affects spermatogenesis and testosterone synthesis (see CA 5.8.3/41 2010/1232198, CA5.8.3/26 2011/1296731 and CA5.8.3/27 2013/1416880). The table Table 5.8.3-14 summarizes the reported effects.
The dose proposal for the main part of the 15day adult rat assay was based on the alpha-cypermethrin equivalent dose considering the CIS-2 portion in cypermethrin and beta-cypermethrin as presented in the bridging statement (see KCA 5.0).

Table 5.8.3-14: Lowest observed effect levels seen in the 15 day rat male assay reported in the open literature and the approximate alpha-cypermethrin portion

Reference	Substance Dose levels purity	Organ weigth	Testicular histology	Sperm-parameters	Hormon (T, LH, FSH)	AR-staining	CIS 2 portion
KCA 5.8.3/26	Cypermethrin 0-6.25-12.5-25-50 Purity: 97.53%	**25**	**6.25**	50	T: 50 FSH: 50 LH: 50	**12.5**	22%
KCA 5.8.3/27	Cypermethrin 0-7.5-15-30-60 Purity: 98%	>60	7.5	**30**	**T: 30** FSH: 60 LH: >60	n.a.	22%
KCA 5.8.3/41	Beta-cypermethrin 0-15-30 Purity: >95%	>30	n.a.	**15**	n.a.	**15**	40%
alpha-cypermethrin equiv. dose based on cypermethrin data		25*0.22= 5.5	6.25*0.22 = 1.4	30*0.22= 6.6	30*0.22 = 6.6	12.5*0.22 = 2.75	
alpha-cypermethrin equiv. dose based on beta-cypermethrin data				15*0.4= 6		15*0.4= 6	

n.a.: not applicable as the parameter was not investigated in the study not applicable as the parameter was not investigated in the study
n.d.: Study did not investigate this parameter
T: Testosterone; LH: Luteinizing hormone; FSH:follicle-stimulating hormone;

Based on this conversion into alpha-cypermethrin equivalent dose it was considered adequate to use 2.0, 3.5 and 6.6 mg/kg bw in the main study because:

a) In case that the portion of alpha-cypermethrin is responsible for the reported effects, one would expect histopathological changes in the testes at a dose level of 1.4 mg/kg bw/day, followed by changes in AR distribution at a dose level of 2.75 or at least 6 mg/kg bw followed by effects on sperm parameters and hormone level at a dose of 6 mg/kg bw or at least 6.6 mg/kg bw
b) it was considered reasonable to take the dose levels 2.0 and 3.5 mg/kg bw/day into account which are currently used for ADI and AOEL reference dose setting.

The change from SD to Wistar rats is not considered to reduce the sensitivity of the study because:

a) the Wistar strain Crl:WI(Han) has shown sensitivity to endocrine related effects in several studies performed in house
b) the basis for recommending SD rats for this type of study as done in the "Integrated summary report for validation of 15-day intact adult male rat assay as a potential screeni in the endocrine disruptor screening program Tier-1 Battery" (online available under http://www.epa.gov/endo/pubs/isr_adultmalerat.pdf) **is not sensitivity** but the availability of this rat strain and its frequent use as animal model of choice for determining general toxicological effects in US-laboratories and the available historical control data
c) according to the "Reviewer`s appendix to the white paper on species/stock/strain in endocrine disruptor assays" published in 2004 by the EPA (online available under http://www.epa.gov/endo/pubs/edt02wdisc.pdf) an unselected Wistar outbred such as the Wistar Hannover which has a moderate litter size and moderate production index might be preferable since it is likely to be more representative of humans that have not been selected long-term for high fecundity. Furthermore, Wistar rats are more sensitive than SD/CD rats to decreases in testes and seminal vesicle weights and testosterone levels by DES, inhibition of sperm count and daily sperm production by BPA, and inhibition of spermatogenesis by lead.

MAIN STUDY

A. TEST SUBSTANCE ANALYSES

Stability analyses
The stability of BAS 310 I (Alphacypermethrin) in corn oil at room temperature for a period of 7 days was proven before the start of the study.

Homogeneity control analyses
Considering the low relative standard deviation in the homogeneity analysis, it can be concluded that BAS 310 I (Alphacypermethrin) was distributed homogeneously in corn oil.

Concentration control analyses
The mean values (low, mid and high doses) of BAS 310 I (Alphacypermethrin) in corn oil were found to be in the range of 90-110% of the nominal concentrations.
These results demonstrated the correctness of the concentrations of BAS 310 I (Alphacypermethrin) in corn oil.

B. OBSERVATIONS

1. Clinical signs of toxicity

No treatment related clinical signs of toxicity were reported.

2. Mortality

No animal died prematurely in the present study.

C. BODY WEIGHT AND BODY WEIGHT GAIN

No test substance-related differences with regard to the mean body weights and body weight change values were noted for male animals of test groups 1-3 (2.0, 3.5 and 6.6 mg/kg bw/day). Body weight increased throughout the study period in any dose group and no significant differences were observed between treated and control group animals. Body weight gains from day 0 - 7 and 0 – 14 were slightly lower in the treated groups without being statistically significant ranging from 2.5% to 12.2%. No dose-dependency of the body weight gain changes was observed and therefore the effects are considered incidental.

Table 5.8.3-15: Mean body weight of rats administered alpha-cypermethrin

Sex	**Males**			
Test-item	Control	Alpha-cypermethrin		
Dose level [mg/kg bw]	0	2.0	3.5	6.6
Mean body weight [g]				
- D 0	291.0	289.6	292.6	290.6
Δ% (compared to control)	-	-0.5	0.5	-0.1
- D 7	313.9	311.9	312.8	311.7
Δ% (compared to control)	-	-0.6	-0.3	-0.7
- D 14	335.0	329.6	331.2	330.7
Δ% (compared to control)	-	-1.6	-1.1	-1.3

Table 5.8.3-16: Mean body weight gain of rats administered alpha-cypermethrin

Sex	**Males**			
Test-item	Control	Alpha-cypermethrin		
Dose level [mg/kg bw]	0	2.0	3.5	6.6
Mean body weight gain [g]				
- D 0 – 7	22.9	22.3	20.2	21.1
Δ% (compared to control)	-	-2.5	-11.5	-7.9
- D 0 - 14	44.0	40.0	38.6	40.1
Δ% (compared to control)	-	-9.1	-12.2	-9.0

D. FOOD CONSUMPTION

No test substance-related, adverse findings were observed. All recorded values were within the biological range typical for this strain of rats.

Table 5.8.3-17: Food consumption of rats administered alpha-cypermethrin

Sex	**Males**			
Test-item	Control	Alpha-cypermethrin		
Dose level [mg/kg bw]	0	2.0	3.5	6.6
Mean food consumption [g]				
- D 0 – 7	23.0	21.8	22.0	21.3
Δ% (compared to control)	-	-5.3	-4.2	-7.2
- D 0 - 14	22.2	20.4	21.2	22.1
Δ% (compared to control)	-	-8.1	-4.5	-0.7

E. NECROPSY

1. Organ weight

When compared to the control group (set to 100%) the mean absolute and relative (to final bw) organ weights were not significantly changed (see Table 5.8.3-18 and Table 5.8.3-19). A slight reduction of absolute and relative prostate and seminal vesicle weight was observed being not statistically significant.

Table 5.8.3-18: Mean absolute organ weights of rats administered alpha-cypermethrin

Sex	**Males**			
Test-item	Control	Alpha-cypermethrin		
Dose level [mg/kg bw]	0	2.0	3.5	6.6
Mean absolute organ weight [g]				
- Prostate	0.757	0.741	0.745	0.676
% (compared to control)	100	98	98	89
- Seminal vesicle	0.939	0.907	0.887	0.872
% (compared to control)	100	97	94	93
- Testes	3.321	3.389	3.423	3.371
% (compared to control)	100	102	103	102

Table 5.8.3-19: Mean relative organ weights of rats administered alpha-cypermethrin

Sex	**Males**			
Test-item	Control	Alpha-cypermethrin		
Dose level [mg/kg bw]	0	2.0	3.5	6.6
Mean relative organ weight [g]				
- Prostate	0.243	0.241	0.242	0.221
% (compared to control)	100	99	100	91
- Seminal vesicle	0.302	0.296	0.288	0.285
% (compared to control)	100	98	95	94
- Testes	1.066	1.104	1.113	1.100
% (compared to control)	100	103	104	103

Comparison of weight parameters with historical control data

When compared to historical control data of three studies using animals with an age of 83 days at terminal kill, the following observations were made:

- The terminal body weight of both control and test group 3 (6.6 mg/kg bw/d) of this study were slightly lower than historical controls
- Absolute prostate weights were higher in the control of this study than in historical controls, while test group 3 weights were within historical controls.
- Relative prostate weights were within historical controls.
- Absolute and relative seminal vesicle weights were within historical controls.

Historical control data

	Study 1		Study 2		Study 3	
	absolute	relative	absolute	relative	absolute	relative
Number of animals	10	10	10	10	10	10
Terminal body weight	348.01 g		315.95 g		325.74 g	
Prostate	0.606 g	0.174	0.642 g	0.204	0.730	0.226
Seminal vesicles	0.842 g	0.241	0.989 g	0.313	0.906	0.281

Present study

Test group	0	3	0	3
	absolute	absolute	relative	relative
Number of animals	15	15	15	15
Terminal body weight	311 g	307 g		
Prostate	0.757 g	0.676 g	0.243	0.221
Seminal vesicles	0.939 g	0.872 g	0.302	0.285

No relevant weight changes were observed after comparison with historical controls.

2. Gross lesions

All findings occurred either individually or were biologically equally distributed over control and treatment groups. They were considered to be incidental or spontaneous in origin and without any relation to treatment. These findings included discoloration of lymph nodes, foci (liver, glandular stomach, epididymides) or pelvic dilation in the kidney.

3. Histopathology

All findings occurred either individually or were biologically equally distributed over control and treatment groups. They were considered to be incidental or spontaneous in origin and without any relation to treatment.

Specific attention was given to round spermatids and pachytene spermatocytes in stage VII and VIII tubules, as apoptosis in these cells would indicate testosterone depletion (Creasy, 2001). No indication of this finding was found.

4. Immunohistology

Mean values of positive Sertoli cells per tubule perimeter:

		Stage VII	Stage VIII	Stage IX
Test group 0 (0 mg/kg bw/d)	Mean	22.6	20.0	19.7
	SD	2.4	2.6	3.2
	N	15	15	15
Test group 3 (6.6 mg/kg bw/d)	Mean	22.2	21.3	20.7
	SD	2.5	2.9	3.3
	N	15	15	15

Wilcoxon (two-sided): *p ≤ 0.05, **p ≤ 0.01
N = Number of animals per test group
SD = Standard deviation

No significant differences in androgen receptor staining could be detected.

Regarding androgen receptor staining, no differences were observed comparing test groups 0 (control) and 3 (6.6 mg/kg bw/d).

The evaluation of androgen receptor staining was triggered by the following publications:

- Hu JX, Li YF, Li J, Pan C, He Z, Dong HY, Xu LC. (2013) Toxic effects of Cypermethrin on the male reproductive system: with emphasis on the androgen receptor. J Appl Toxicol. 33, 576–585
- Lu Liu, Jin-xia Hu, He Wang, Bao-jun Chen, Zhen He, Li-chun Xu (2010) Environmental Toxicology and Pharmacology 30, 251–256

In publication of Liu et al. (2010), the expression of the androgen receptor was measured using grey pixel scale values on whole slides without counting specific cells. This approach is not appropriate to discover stage-specific differences in androgen receptor staining, as the number of tubules in specific stages was not determined.

Therefore, the stage aware evaluation of Hu et al. (2013) was preferred with the following alterations of their protocol:

- Fixation of testis was in modified Davidson's solution, in contrast to Hu et al. (2013), who fixed their tissue in Bouin's. In the literature, Latendresse et al. (2002) reported that tubular shrinkage can occur with Bouin's fixative and that androgen receptor staining was weaker or absent in central tubules with Bouin's fixative as compared to modified Davidson's solution.
- Hu et al. (2013) evaluated 100 visual fields for each group and five fields per image. All positive cells were counted on those images. In Hu et al. (2013), it was not transparent how many tubules of each stage were evaluated, and how the different sizes of tubules due to the anatomical cut section in histology were taken into account. In this study, we selected approximately 100 tubuli of a given stage for each group. In addition, only the positive Sertoli cells of a given tubule were counted and set in relation to the perimeter of that tubule to normalize the number of positive cells to the size of the tubule. This approach is used in evaluation of cell proliferation of e.g. the nasal cavity where positive cells are given per unit length (Monticello et al., 1990).

- Hu et al. (2013) divided their stage evaluation into a combination of V/VI; VII/VIII, and IX. In this study stages VII, VIII, and IX were evaluated separately as differences in staining between stages VII and VIII could not be excluded at the beginning of the study.
- Slides were counterstained with hematoxylin, as staging is not possible without recognizable nuclear detail. No mention of a counterstained was made in Hu et al. (2013).

Taking into account all considerations mentioned above no differences in androgen receptor staining were observed comparing test group 0 and 3. Results by Hu et al. (2013) reported for cypermethrin could not be confirmed for alpha-cypermethrin.

5. Sperm parameters

Concerning the motility of the sperms and the incidence of abnormal sperms in the cauda epididymidis as well as the sperm head counts in the testis and in the cauda epididymidis no treatment-related effects were observed.

Table 5.8.3-20: Sperm parameters of rats administered alpha-cypermethrin

Sex	**Males**			
Test-item	Control	Alpha-cypermethrin		
Dose level [mg/kg bw]	0	2.0	3.5	6.6
Sperm motility (day 15)				
Mean	83	81	84	82
SD	8	10	6	9
TS/gT (day 15) [Mio/g]				
Mean	120	111	113	109
SD	27	26	32	30
TS/gC (day 15) [Mio/g]				
Mean	541	570	552	511
SD	145	166	131	164
Abnormal (day 15) [%; Cut off 6%]				
Mean	6.1	8.6	6.0	6.0
SD	0.4	9.9	0.0	0.0

TS: total sperm count; T: Testis; C: Cauda epididymis

6. Hormon levels

Since there was no indication for any hormonal change based on reproductive organ weights, sperm analysis and histopathology examinations, measurements of FSH, LH and testosterone in the serum samples were not performed and are not considered necessary to determine the absence of any adverse effects on hormone regulated organs and spermatogenesis.

III. CONCLUSIONS

After administration of alpha-cypermethrin at doses of 2.0, 3.5, and 6.6 mg/kg bw by gavage to adult intact male Wistar rats for 15 days in corn oil no findings with regard to clinical examination,

food consumption, body weight, and organ weights were observed when compared to the control animals. Sperm motility and morphology as well as sperm head counts in testes or in cauda epididymidis were not influenced by the test item. Histopathology and immunohistology androgen receptor staining revealed no substance-related effects.

In conclusion, under the conditions of this study a NOAEL of 6.6 mg/kg bw/day can be derived in adult intact male Wistar rats for effects on hormone regulated male reproductive organs and spermatogenesis.

CA 5.9 Medical Data

Information evaluated in the draft monograph of the Rapporteur Member State Belgium in Sep., 1999: Alpha-cypermethrin belongs to the group of pyrethroids, therefore the reported data were already at the stage of first annex I inclusion broad based on data with cypermethrin and pyrethroids in general. However it was concluded that despite the extensive world-wide use of alpha-cypermethrin, there were relatively few reports of effects seen in humans and the overall summary from the monograph 1999 was as follows:

"The salient clinical effect of alpha-cypermethrin exposure in humans is a transient-sensation experienced usually in the facial skin around the mouth and jaws or cheeks and the forehead following direct skin contact, often with insignificant amounts such that the individual is usually unaware of the exposure at the time. This paresthesia, as the abnormal sensation is called, is somewhat subjective as it is not associated with any visible or objective skin irritation such as erythema, eczematisation or local swelling. The symptom is variously described as being a sensation of coldness or numbness or, alternatively, burning, which can be quite painful, tingling or a sunburn-like smarting. The onset of paresthesia may not occur for several hours after exposure, and is not immediately relieved by washing, but usually wears off within a few hours to 24 hours."

Furthermore, European Chemicals Bureau decided in 2001 (ECBI/64/01, October 2001) to classify alpha-cypermethrin for respiratory irritation based on human observations with cypermethrin and based on animal data for alpha-cypermethrin.

The following conclusion was given in the Review Report (SANCO/4335/2000 final from 13.Feb. 2004) for list of endpoint listing of alpha-cypermethrin:

Medical data	Paresthesiae and peripheral sensory phenomena and irritation of respiratory tract; R37

Information on medical data obtained since then has been collected and evaluated. In order to extend the evaluation basis a search in the literature has been performed.

The relevant document s are summarized in the chapter CA 5.9

Considering all available information, the conclusion for relevant endpoints for the current renewal remains as follows:

Medical data (SANCO/11802 data point 5.9)

.....	Paresthesiae and peripheral sensory phenomena and irritation of respiratory tract; R37

CA 5.9.1 Medical surveillance on manufacturing plant personnel and monitoring studies

All persons handling crop protection products are surveyed by regular medical examinations.
There are no specific parameters available for effect monitoring of alpha-cypermethrin.
Thus, the medical monitoring program is designed as a general health check-up. At the two production sites the following parameters are investigated:

Bayer Vapi Private Limited:

a) Quarterly medical examination of company employees in Form No. 22 as per Insecticide rules 1971. Insecticide Act, 1968 (46 of 1968) Chapter VIIIRule No. 37 – by factory medical officer.

- List of questionaries (symptoms) for all systems including neuromuscular psychological symptoms
- Finger nose test to each employee working in plant to rule out any neurological symptoms

b) Annual medical checkup:

- a general physical examination including anamnesis, physical examination, blood analysis, urine analysis, stool routine examination, Audiometry/Lung Function test, Electrocardiogram, x-ray Chest (PA) view.

Tagros Chemials India Ltd:

The surveillance program includes a general physical examination, blood analysis (Hb, DC, TC, ESR), urine analysis, x-ray Chest (PA) with opinion, Electrocardiogram, lipid profile, borne density test, Vision/Audiometry/Lung Function test,.

Adverse health effects suspected to be related to alpha-cypermethrin exposure have not been observed and no monitoring studies are known to the applicant.

CA 5.9.2 Data collected on humans

The peer review literature search revealed the following new reports:

Report:	CA 5.9.2/1 Costa C. et al., 2013a Cytokine patterns in greenhouse workers occupationally exposed to alpha-Cypermethrin: An observational study 2013/1417820
Guidelines:	none
GLP:	no (certified by none)

Executive Summary of Literature:

An investigation on greenhouse workers occupationally exposed to alpha-cypermethrin showed neither clinical signs of immunosuppression nor alterations in total leukocytes or leukocyte subpopulations This is consistent with findings in experimental studies in rats (see Immunotoxicity study CA 5.8.2/1).

Classification of study: Supplementary information

Report: CA 5.9.2/2
Elfman L. et al., 2005a
Acute health effects on planters of conifer seedlings treated with insecticides
2005/1043540
Guidelines: none
GLP: no
(certified by none)

Executive Summary of Literature:

In male tobacco farmers a lower sensory nerve conduction velocity in the sural nerves was observed as compared to controls. However, this study has several limitations such as office workers as control group, lack of controlling for confounders, coexposure to other pesticides.

Classification of study: Supplementary information

CA 5.9.3 Direct observations

Some cases of slight irritation of the skin and mouth and/or intoxication (headache, nausea, vomiting, ague, fatigue, aching muscles, drowsiness, dizziness, breathing difficulties) have been reported to BASF SE in Ludwigshafen in persons exposed to alpha-cypermethrin containing products in combination with other active ingredients and/or products.

From the open literature, the following clinical cases and poisoning incidents were available:

Report: CA 5.9.3/1
Majumdar B.B. et al., 2012a
Toxic intracerebral demeyelination in a case of suicidal Cypermethrin poisoning
2012/1367262
Guidelines: none
GLP: no
(certified by none)

Executive summary of literature:

A 17-year-old boy was admitted in the General Medicine indoor with complains of vomiting and salivation due to suicidal intake of an unknown poison. Further interrogation revealed an ingestion of 50 ml of "Ostad", having a concentration of 10% of cypermethrin in an emulsifiable concentrate, which is equivalent to 5 g of poison about 8 hrs ago. The calculated exposure assuming a body weight of 70 kg is around 70 mg/kg bw Cypermethrin of unknown isomeric composition.
This case had predominant neurological manifestations; however, tremor and fasciculations were not documented. After gradually recovery on the 3rd day the patient showed neurological deterioration on the 4th day, showing fever, hemodynamic collapse, and intermittently seizures over the next few days. Routine hematological and biochemical investigations were within normal limits. His liver function and renal function tests, hemogram, serum electrolytes and glucose were normal. Over the next 4 days, the patient gradually improved and regained hemodynamic stability, but his neurological symptoms lacked an improvement. An MRI of brain revealed patchy areas of demyelination in subcortical part of the cerebrum.
To the best of knowledge, no reported case of demyelinating plaques has been reported till date following this poison. It also causes an increased salivation, upper gastrointestinal bleeding, and rarely renal failure which might be partly induced by the type of formulation. The patient survived with supportive and symptomatic management and was discharged on the phase of ongoing recovery.

Classification of study: Supplementary information

Report: CA 5.9.3/2
Martinez-Navarrete J. et al., 2008a
Accidental poisoning with Chinese chalk
2013/1417282
Guidelines: none
GLP: no
(certified by none)

Executive summary of literature:

A 1.5-year old child accidentally ingested half of a “Chinese chalk”, which contains deltamethrin and cypermethrin. A day later, the infant showed vomiting, cough, fever, drowsiness, and irritability and, respiratory distress. Eight days after symptomatic treatment the infant was discharged from hospital and no further complications were observed.

Classification of study: Supplementary information

CA 5.9.4 Epidemiological studies

The open literature revealed the following epidemiological study:

Report: CA 5.9.4/1
Elfman L. et al., 2011a
Detection of pesticides used in rice cultivation in streams on the island of Leyte in the Philippines
2009/1131087
Guidelines: none
GLP: no
(certified by none)

Executive summary of literature:

Elfman et al. 2009 examined 19 planters handled untreated und with imidacloprid- and cypermethrin-treated conifer seedlings. No acute health effect could be found regarding to the pesticides.

Classification of study: Supplementary information

Furthermore several investigations are published that try to correlate urinary pyrethroid metabolite excretion with health issues. As surrogate parameter for pyrethroid exposure in general 3-phenoxybenzoicacid (PBA) is used whereas exposure to cypermethrins (zeta-, beta-, alpha-cypermethrin and cypermethrin itself), permethrins, cyfluthrin leads to cis- and trans-dichlorodimethylvinylcyclopropane carboxylic acid in urine. As these are common metabolites, a conclusion on or association to alpha-cypermethrin is not possible but however a summary of exemplary discussed data is given in Table 5.9.4-1.

In summary the following data are presented:
One study (Kimata et al., 2009) reports urinary 3-PBA levels in residents of rural and suburban areas compared to occupational exposure of sprayers and non-sprayers showing higher values in occupational exposed persons.
2 studies (Imai et al; 2014 & Perry et al., 2007) investigating urinary 3-PBA levels in healthy population showing no correlation to any semen parameter. However, semen quality is correlated to several life style factors.
5 studies (Toshima et al., 2012, Ji et al., 2011, Meeker et al., 2008 & 2009 and Han et al., 2008) investigating patients who had infertility consultation at gynecology clinics. These data indicate several correlations between 3-PBA and reduced semen quality or changed hormonal status. But there is no clear and convincing picture and these studies have several limitations such as no standardized sample treatment, lack of control groups, and lack of controlling for confounders. Some of the studies show correlations that lack any reliability. Some of the authors themselves note that associations described could be by chance due to the number of statistical comparisons and analyses that were carried out. Therefore the data are considered only as supplemental information and the following overall conclusion is drawn:

In conclusion, a clear absence of any correlation between urinary pyrethroid metabolites and semen parameters in two studies within the normal (healthy) population group was reported. In the clinical studies among patients with presumed infertility issue the 3-PBA level in urine was repeatedly reported to be correlated with effects on semen parameters, but the spectrum of reported effects was inconsistent. Considering comparable 3-PBA levels in the healthy and the clinical studies, the described correlations between 3-PBA levels in urine and the reported inconsistent changes are clearly not associated but by chance.

Report: CA 5.9.4/2
Kimata A. et al., 2009a
Comparison of urinary concentrations of 3-Phenoxybenzoic acid among general residents in rural and suburban areas and employees of pest control firms
2009/1131086
Guidelines: <none>
GLP: no
(certified by <none>)

Executive Summary of literature:

According to this study 3-PBA levels are lower and comparable for rural and suburban residents which are not occupationally exposed to Pyrethroids compared to higher values found for sprayers the morning after the most recent pesticide spraying, but also for non-sprayers, which are defined as employees who have worked for pest control firms but have not been engaged in active pyrethroid spraying. This study suffers the weakness that it is not age adjusted, however it is discussed that several publications found no correlation between age and urinary excretion of pyrethroid metabolites so that the observed difference in age distribution between the groups is considered to have little impact on the results. The geometric means for 3-PBA in urine for residents are at levels of 0.32 ± 1.1 µg/g creatinine for rural residents and 0.49± 1.2 µg/g creatinine for suburban residents, which is mentioned to be identical to those of US and German populations. The occupational exposure led to a two-order magnitude of increase in geometric means of 3-PBA concentrations between sprayers (23.8 ± 1.4 µg/g creatinine) compared to residents.

Classification of study: Supplementary information

Correlations between urinary pyrethroid metabolite concentration and sperm parameters were investigated several times, some in general population, some among patients who had infertility consultation at a gynecology clinic.
The following two publications were selected to present data for general population:

Report: CA 5.9.4/3
Imai K. et al., 2013a
Pyrethroid insecticide exposure and semen quality of young Japanese men
2014/1242698
Guidelines: none
GLP: no
(certified by none)

Executive Summary of literature:

Imai et al; 2014 (Doc ID2014/1242698) reports a study on 323 Japanese healthy students and showed that environmental exposure to pyrethroid insecticides, measured via the surrogate 3-PBA in urine, did not affect semen quality (sperm motility, sperm count nor volume). 3-PBA was found in 91 % of the urine probes (LOD: 0.08 ng/mL). Geometric mean of unadjusted 3-PBA concentrations in urine were at a 0.68 µg/L (Range: <0.08-13.2µg/L). Fluctuations of 3-PBA concentrations were conditioned by season (higher values in sommer and autumn), whereas sperm effects were described in dependency of age, abstinence, testes size, varicoceles, and frequency of cheese/soy/ non-oily white fish consumption.

Classification of study: Supplementary information

Report: CA 5.9.4/4
Perry M.J. et al., 2006a
Environmental Pyrethroid and organophosphorus insecticide exposures and sperm concentration
2007/1070389
Guidelines: none
GLP: no
(certified by none)

Executive Summary of literature:

Perry et al., 2007 (Doc ID 2007/1070389) screened 18 randomly selected urine samples collected from male participants from China, Anhui in the Winter of 2004, all of reproductive age, for 24 parent compounds and metabolites of pesticides and examined the results in relation to sperm concentration. Results showed high prevalence of exposure to OP and PYR pesticides. 3-PBA was found in 100 % of the urine probes with a median at 1.1 µg/L and maximum values more than 20 times higher. Cis-dichlorodimethylvinylcyclopropane carboxylic acid (CDCCA, cis- DCVA) was found only in samples of the maximum exposure group (at a concentration of 12.1 µg/L). Trans-dichlorodimethylvinylcyclopropane carboxylic acid (TDCCA, trans - DCVA) was found in the median and the maximum exposure group. Although the investigation provide some suggestion that the higher exposure group had lower sperm concentrations, however, no correlation to PBA, cis-DCVA or trans-DCVA was found. A statistically significant difference in sperm concentration was found in correlation with the organophosphate representative diethylthiophosphate (DETP) in urine.

Classification of study: Supplementary information

In summary, among healthy representatives of the population no correlation was found between sperm parameters (sperm motility, sperm count and volume) and PBA, cis-DCVA or trans-DCVA.

Based on the inconsistency of the data presented in the following reports the data were not considered to be convincing. Therefore the reports were not further considered but are here only presented to demonstrate the inconsistency.

Report: CA 5.9.4/5
Toshima H. et al., 2011a
Endocrine disrupting chemicals in urine of Japanese male partners of subfertile couples: A pilot study on exposure and semen quality
2012/1367223
Guidelines: none
GLP: no
(certified by none)

Executive Summary of literature:

Toshima et al., 2012 (Doc ID 2012/1367223) measured urine samples from 42 Japanese male partners from infertility clinic (11 current smokers, only 1 of them was excluded) and found PBA levels in urine at 0.547 ± 2.76 µg/L (geometric mean) with a range of 0.160 - 7.72 µg/L (Corrected for specific gravity, LOD: 0.04µg/L). When the subjects were divided into two groups based on the semen parameter reference values (40% for motility and 15x106 for sperms / ml), Pearson correlation analysis found no correlation of motility or of sperm counts to PBA-concentration. However, in the multiple regressions analysis lower semen motility was associated with urinary 3-PBA concentration. However, the significance of this finding is already limited by the authors which state that some of the associations described could be by chance due to the number of statistical comparisons and analyses that were carried out. One point that strongly reduces the reliability of the study is the significant correlation of smoking and improved semen quality.

Classification of study: Not further considered

Report: CA 5.9.4/6
Ji G. et al., 2010a
Effects of non-occupational environmental exposure to pyrethroids on semen quality and sperm DNA integrity in Chinese men
2011/1296811
Guidelines: none
GLP: no
(certified by none)

Executive Summary of Literature:

240 men were recruited from an infertility clinic through the clinic following strict eligibility screening. Urinary 3-phenoxybenzoic acid (3-PBA) concentration, semen quality (sperm concentration, motility, total sperm count), and sperm DNA integrity were evaluated. After adjustment for potential confounders, a significant inverse correlation was observed between the urinary 3-PBA level and the sperm concentration. Moreover, we also found a significant positive correlation between urinary 3-PBA level and sperm DNA fragmentation (ˇ = 0.27, 95%CI: 0.15–0.39, P < 0.001). Our results suggest that non-occupational environmental pyrethroids exposure may have a negative impact on sperm DNA integrity and semen quality in Chinese males.

Classification of study: Not further considered

Report: CA 5.9.4/7
Meeker J.D. et al., 2008a
Human semen quality and sperm DNA damage in relation to urinary metabolites of Pyrethroid insecticides
2008/1102220
Guidelines: none
GLP: no
(certified by none)

Executive Summary of Literature:

Meeker et al (2008, Doc ID 2008/1102220) investigated the association between urinary pyrethroids metabolites, semen quality, sperm motion parameters and sperm DNA damage (neutral comet assay) in 207 men recruited from an infertility clinic at an age of 18 to 54 years. The metabolites that were determined included 3-phenoxybenzoic acid (3-PBA) and cis- and trans-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylic acid (CDCCA and TDCCA) and others. There was a quite high proportion of samples below the limit of detection (LOD=0.1µg/L) for 3PBA (46%), CDCCA (47%) and TDCCA (49%). Multiple liniear regression as well as multiple logistic regression was performed. Preliminary bivariate analyses demonstrate that a higher metabolite concentration in the urine was associated with greater age whereas semen parameters were positively associated with abstinence period of sexual activity. The regression in semen quality with urinary metabolite groups were therefore adjusted for age and abstinence period. Meeker found that 3-PBA and TDCCA were associated with a reduction in sperm concentration, sperm motility and sperm motion parameters. Conversely, CDCCA and 3PBA were associated with increased sperm DNA damage, measured as percent DNA in the comet tail but not associated with comet extent or tail distribution moment.
However, the significance of this single finding is not convincing, especially due to the divers parameters that are known risk factors for infertility and which were not addressed in the investigation.

Classification of study: Not further considered

Report: CA 5.9.4/8
Meeker J.D. et al., 2008b
Pyrethroid insecticide metabolites are associated with serum hormone levels in adult men
2009/1131084
Guidelines: none
GLP: no
(certified by none)

Executive Summary of Literature:

In a second part Meeker et al., 2009 (Doc ID 2009/1131084) investigated several serum hormone levels within a subset of the former group. The regressions were adjusted for age and body mass index, smoking status, season and time of day but further risk factors were not integrated. The data show no clear picture and some very curious correlations were reported, like current smoking habit is negatively correlated with high cis-DCCA levels. The sum parameter (3-PBA, cis- and trans-DCCA) and 3-PBA alone were reported to be correlated with increased FSH levels.
Any conclusion on causality between urinary pyrethroid metabolite levels and serum hormone levels is not considered reliable based on these data.

Classification of study: Not further considered

This is substantiated by the following investigation which revealed a complete different picture

Report: CA 5.9.4/9
Han Y. et al., 2008a
The relationship of 3-PBA pyrethroids metabolite and male reproductive hormones among non-occupational exposure males
2008/1102218
Guidelines: none
GLP: no
(certified by none)

Executive Summary of Literature:

The authors investigated the association between serum reproductive hormone levels and urinary creatine adjusted concentration of 3-PBA level in Chinese adult men. The study subjects (n = 212) were from the affiliated hospitals of Nanjing Medical University. By using GC–MS, urinary 3-PBA level of each subject was measured and adjusted by urinary creatin. Blood samples were collected for measuring the serum levels of reproductive hormones, including follicle stimulating hormone (FSH), luteinizing hormone (LH), estradiol (E2), testosterone (T) and prolactin (PRL). All the subjects had detectable levels of 3-PBA in their urine samples. The median concentration of 3-PBA was 0.815 µg/g creatin. The results showed that there were positive associations between the levels of serum LH and 3-PBA (p = 0.013) but negative associations between E2 and 3-PBA level (p = 0.022), and the adjusting p-value was 0.044 for LH and E2, which suggested that pyrethroids are capable of disrupting the male endocrine function. A correlation to FSH was not found. The provided scatter plots for individual PBA levels and hormone levels rather show demonstrate a crude distribution of unrelated parameters.

Classification of study: Not further considered

Table 5.9.4-1: Overview of available studies investigating pyrethroid metabolite concentration in urine and possible correlation to health parameters in humans.

Author	Subject	Investigations/questionnaire	Samples	3-PBA concentration (in urine)*	Dependency	Strength	Weakness
Exemplary 3-PBA levels in residents and occupational exposed persons							
Kimata et a., 2009, Doc ID 2009/1131086	143 male from rural area, 66 males from suburban area, pest control workers (sprayers, n= 14; non-sprayers, n= 16) (2005-2007)	Age (years), Current smoking and drinking habits (yes/no) Weight and height were measured	- Urine (3-PBA analysis, creatinine content (CR))	Adjusted (CR): Rural residents: 0.32 ± 1.1 µg/g CR Suburban residents: 0.49± 1.2 µg/g CR Sprayers: 23.8 ± 1.4 µg/g CR Non Sprayers: 5.4 ± 1.3 µg/g CR (LOD=0.02µg/l)	No difference of 3-PBA levels in rural or suburban populations Increased 3-PBA levels in sprayers and also in non-sprayers	Population based	No adjustment for age or seasonal changes
3-PBA levels and sperm parameters in healthy population							
Imai et al., 2014 DocID 2014/1242698	Male student (n=322), 18-24 years, mother/himself born in Japan, healthy (1999-2000, 2002-2003	- Abstinence period - Testis size - Varicoceles - Lifestyle information (smoking, alcohol consumption, frequency of selected food stuff consumption) - Physical examination	- Semen (sperm motility, sperm count, volume) - Urine (3-PBA analysis, specific gravity)	Unadjusted: 0.68 ± 3.26 ng/mL Adjusted (SG): 0.59 ± 3.12 ng/mL	sperm correlates to: 3-PBA: no influence Age, abstinence, testes size, varicoceles: yes Vegetables/fruit consumption: No Cheese/soy/ non-oily white fish: Yes 3-PBA correlates to Season: yes Food composition (beef/vegetables): no	Population based	Students of a limited geographical region

Author	Subject	Investigations/questionnaire	Samples	3-PBA concentration (in urine)*	Dependency	Strength	Weakness
Perry et al., 2007 Doc ID 2007/1070389	Males young (n=202; samples n=18), no pesticide users (but with home villages consisting of agricultural background) (2004)		- Semen (sperm concentration) - Urine (3-PBA, cis/trans-DCCA),	Unadjusted (median; ng/mL): - 3-PBA: 1.1 ng/mL - cis-DCCA: <LOD - trans-DCCA: 0.1 ng/mL	No effects of pyrethroids metabolites but effect of DETP on sperm concentration	Population based, 24 pesticides determined in total	
3-PBA levels and sperm parameters in hospital based popultion							
Toshima et al., 2012 Doc ID 2012/1367223	41 men from infertility clinic, 29-58 years (2010)	- lifestyle information (smoking, dietary habit)	- Semen (sperm motility, sperm count, volume), - Urine (3-PBA, specific gravity), Other metabolites	Adjusted (SG): 0.547±2.76 ng/mL	sperm parameters correlate with: -3-PBA reduce motility; -Isoflavones reduce motility/concentration -Age, Alkohol, Coffee: no effect -Smoking: increased semen quality	Further chemicals investigate (phthalates, isoflavones)	Hospital based, semen sampling at home
Ji et al., 2011 DocID 2011/1296811	Males from infertility clinic (n=240), 28.5±3.6 years, mother/himself born in Japan, healthy, lifestyle information asked (2005-2007)	- abstinence period - physical examination - lifestyle information asked	- Semen (sperm motility, sperm count, volume), - Urine (3-PBA analysis), - DNA fragmentation	0.19-10.37 µg/g of CR Median: 1.12 ng/mL	3-PBA correlates with - Sperm conc.: Yes - Total sperm count: Yes - DNA fragmentation: Yes - Seminal volume: No - Motility: No		Hospital based
Meeker et al., 2008 DocID 2008/1102220	Males from infertility clinic (n=207), 18-54 years (2000-2003)	- Exclusion criteria: varicoceles, orchidopexy, use of exogenous hormones- self-reported - health information asked	- Semen (sperm motility, sperm count, volume, morphology), -urine (3-PBA, cis/trans-DCCA, specific gravity) - DNA fragmentation	Adjusted (SG); 90th percentile; ng/mL): - 3-PBA: 1.31 - cis-DCCA: 0.58 - trans-DCCA: 1.25	3-PBA correlates with: - Sperm conc.: yes - Total sperm count: Yes - DNA fragmentation: Yes - Motility: Yes (Effect when compared >75% percentile with below median; no trend was observed within percentiles)	Higher number of samples	Hospital based

Author	Subject	Investigations/questionnaire	Samples	3-PBA concentration (in urine)*	Dependency	Strength	Weakness
Meeker et al., 2009 DocID 2009/1131084	Males from infertility clinic (n=161), 18-54 years (2000-2003)	- Exclusion criteria: vasectomy, exogenous hormones- self-reported; -nurse administered health questionaire -height and weight measured,	- Urine (3-PBA, cis/trans-DCCA, specific gravity) - Serum (testosterone, SHBG, Inhibin B, LH, FSH, E2, prolactin, T3/4, TSH)	Adjusted (SG); 90th percentile; ng/mL): - 3-PBA: 1.31 - cis-DCCA: 0.6 - trans-DCCA: 1.35	3-PBA correlates with: - ↑ FSH cis-DCCA correlates with: -↓Inhibin ↑FSH ↓LH but not with ↓ T trans-DCCA correlates with: - ↑ FSH, ↓ T, ↓ FAI		Hospital based
Han et al., 2008; Doc ID2008/1102218	Chinese adult men (n=212), 20- 40 years, (2004-2006)	- Detailed physical examination(height, weight) - Questionnaire (lifestyle factors (smoking, drinking, psychologic tension, sleep quality), occupational and environmental exposures, genetic risk factors, sexual and procreate state, medical history, physical activity) - Exclusion criteria: hormonal medications, genetic disease, occupational exposure to pesticides;	Urine (3-PBA, creatinine) - Serum (testosterone, LH, FSH, E2, prolactin)	(n=199) Adjusted (CR): - 3-PBA: 0.925 µg/g Unadjusted: - 3-PBA: 1.398 µg/L	3-PBA correlates with: - ↑ LH and E2		Hospital based

* 3-PBA concentration in urine is given as geometric mean if not mentioned otherwise, correction was done for specific gravity (SG) or creatinine content (CR)

CA 5.9.5 Diagnosis of poisoning (determination of active substance, metabolites), specific signs of poisoning, clinical tests

Expected clinical signs and symptoms of poisoning based on animal studies:
In acute toxicity studies in animals transient signs of neurotoxicty were seen after exposure to moderate doses of alpha-cypermethrin, whereas higher doses higher doses could be lethal. In repeated dose toxicity studies skin irritation effects were repeatedly seen.
Based on the rapid absorption with peak plasma levels reached within 6-9 hours and the nearly complete elimination within 48 hours and assuming a similar absorption kinetic for humans, onset of clinical signs after (sufficiently high oral exposure) may be expected within several hours and potential symptoms of acute poisoning are expected to decrease in parallel to elimination. In acute toxicity studies first symptoms were already seen after 1 hour.

In humans the occurrence of "facial sensations" (Paresthesiae) is an indication of exposure.

Several biomarkers .(e.g. 3-PBA) can be detected in human urine by chromatography.

The passage from the original DAR from 1999 is still valid:
“The facial paresthesiae described following direct contact of the facial skin with alphacypermethrin are highly characteristic, especially in the absence of any visible sign of skin irritation such as erythema, swelling, blistering, exudation or desquamation, etc. The urinary metabolite of alphacypermethrin (the methyl ester of the cis-isomer of 3-(2,2-dichlorovinyl)-2,2-dimethyl-cyclopropane carboxylic acid) can be identified and measured with a limit of detection of 0.003 mg/liter. It would not appear useful, therefore, to perform a urinalysis in most cases of facial paresthesiae, since it is most unlikely that the urinary metabolite would reach the level of detection in the urine following skin contact exposure, sufficient to cause the transient paresthesiae. (Van Sittert, 1982)

The signs of systemic poisoning by alpha-cypermethrin following accidental ingestion, appear to be non-specific. Acute intoxications by synthetic pyrethroids in general have been reported to lead to signs and symptoms such as dizziness, headache, nausea, anorexia, fatigue, gastrointestinal complaints and fever. In severe cases, exposure has resulted in impaired consciousness, muscular fasciculations, convulsions, coma and pulmonary edema. The signs and symptoms of acute intoxication appear to be very similar for all types of pyrethroids, presumably including alpha-cypermethrin.”

From the above, it would appear to be very difficult to differentiate pyrethroid poisoning from either organophosphate or carbamate poisoning. Systemic poisoning by these anticholinergic agents from occupational exposure is much more commonplace, and because specific and effective treatment for the latter is available, a blood cholinesterase test might prove useful to exclude organophosphate or carbamate poisoning.

CA 5.9.6 Proposed treatment: first aid measures, antidotes, medical treatment

In case of skin contact paraesthesia usually resolve in 12 – 24 hours. Specific treatment is not generally required. However, topical application of dl-alpha tocopherol acetate (vitamin E) reduced the serverity of skin reactions to cypermethrin.

In case of ingestion the stomach should be washed out by intubation as soon as possible, since induction of vomiting is not recommended.

For acute systemic intoxication there is no specific antidote. Convulsions should be treated with anti-convulsants such as diazepam. Isolated brief convulsions do not require treatment but intravenous diazepam (5-10mg) should be given if seizures are prolonged. Diazepam is also useful in the treatment of muscle fasciculation. Treatment should otherwise be symptomatic or supportive.
The use of methocarbamol, a skeletal muscle relaxant, to reduce mortality, as reported in the DAR 1999, could not be confirmed in a further antidot study (see CA 5.8.2/3). Also the use of mephenesin did not show any protective effects on the incidence or latency of the toxicity. Halothane protected the animals from the acute toxic signs (salivation and convulsions) although it had no protective effects on the lethality.

The use of atropine, which may be life-saving in the case of organophosphate or carbamate poisoning, needs careful control in case of pyrethroid poisoning as it is contraindicated at high doses. A blood cholinesterase test may, therefore, be of crucial importance in making this discrimination. Intravenous atropine (0.6 – 12 mg in an adult) may be useful in controlling excess salvation, but care should be taken to avoid excess administration. It is unlikely to have an effect on pulmonary edema. (Bradberry et al; (2005); Doc ID 2005/1043400).

In a general pharmacological study (CA 5.8.2/2) effects of alpha-cypermethrin on the autonomic and central nervous system, effects on respiration and circulation, effects on digestive organs, effects on skeletal muscles and renal function were investigated. The test substance produced acute toxicities through its action of nervous and cardiorespiratory systems at sublethal doses close to lethal doses.

CA 5.9.7 Expected effects of poisoning

Acute but normally transient signs of neurotoxicity were seen in animals after exposure to sub-lethal doses of alpha-cypermethrin. Reports on human poisonings indicate similar effects.

After local contact paresthesiae is a well-known symptom.
Typically there may be a delay of one to several hours between facial skin contamination by alpha-cyano-pyrethroids, including alpha-cypermethrin, and the onset of the characteristic paresthesiae. The sensations may then last for several hours up to one day or more before disappearing. It is not clear whether that variation in duration of the symptoms is strictly dependent on the degree of contamination, i.e., the dose delivered to the skin, upon individual variation, or a combination of the aforementioned factors.

BASF SE ▪ 67114 Limburgerhof, Germany

SUPPLEMENT INFORMATION DOCUMENT

Alpha-cypermethrin (BAS 310 I):

Residues studies used for the estimation
of the dietary exposure of alpha-cypermethrin
in plant and animal commodities

and

Detailed dietary exposure risk assessment
of alpha-cypermethrin metabolites

December 2014

BASF Doc ID 2014/1314800

Compiled by:

[redacted]

Table of Contents

List of Tables

Page

List of Figures

Page

1 Introduction

Alpha-cypermethrin (BAS 310 I) belongs to the class of synthetic pyrethroid insecticides. It is a non-systemic insecticide and is registered against insect pests in various crops belonging to the crop groups of cereals, oilseeds, fruits and vegetables. As a consequence of its use, residues might result in plant items destined for human food or animal feed.

This document summarizes supplemental information on the residue situation in crops not belonging to the representative uses for the Annex I renewal. It gives information on residue field and glasshouse studies, processing data and the relevant residue values (MRLs, HRs and STMRs) derived and used for risk assessment purposes in the main dossier. For each crop, a chapter is included summarizing the supervised field trials, processing studies (if applicable) and an assessment of the residue data to derive an MRL, STMR and HR. For the MRL calculations, the current version of the OECD calculator was used. All individual residue data used for the consumer dietary risk assessment of alpha-cypermethrin (BAS 310 I) are summarized in table format in the appendix to this supplement, including document and trial number, PHI, residue value, and the calculated HRs and STMRs (Table 5-1 and Table 5-2).

In the residue trials summarised below, several formulated products were used: BAS 310 03 I (SC), BAS 310 QC I (SC), BAS 310 11 I (EC), BAS 310 40 I (EC), BAS 310 08 I (WG), BAS 310 41 I (SC), Fastac SC (SC), BAS 310 51 I (ME), BAS 310 55 I (ME), alpha-cypermethrin 50 g as./L EC, alpha-cypermethrin 50 g a.s./kg PVP, alpha-cypermethrin 100 g as./L OSK and BAS 310 63 I (CB). Most of the trials were carried out with the formulation BAS 310 40 I. Bridging trials with the formulations BAS 310 40 I and BAS 310 08 I on tomato (see 2.8.1), cauliflower (see 2.12.1) and lettuce (see 2.16.1) show similar residues for both products. The study summaries on grape, olive, tomato and lettuce also include bridging trials, demonstrating that alpha-cypermethrin residues after application of BAS 310 40 I are comparable to residues found after use of BAS 310 51 I or BAS 310 55 I, respectively. According to European Community Guideline 7527/VI/95 of March 2011, EC, WP, WG and SC formulations usually produce comparable residues. Since the formulation BAS 310 55 I is a micro-emulsion (ME) with only minor changes to BAS 310 51 I, and residues after application of all above-mentioned products are demonstrated to be similar, those formulations are considered suitable to describe the residue behaviour of alpha-cypermethrin.

2 Food of plant origin

2.1 Grape

Residue data from supervised trials in grapes were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.1-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Grape	2003	-	-	4	IT, SP	4	2005/1004977
Grape	2003	4	FR	-	-	4	2005/1006474
Grape	2004	-	-	4	FR, GR	4	2005/1007589
Grape	2004	4	DE, FR	-	-	4	2005/1007591
Grape	2005	4	DE, FR	4	FR, GR, IT, SP	8	2006/1026853
Grape	2006	4	DE, FR	4	FR, GR, IT, SP	8	2007/1008492
Grape	2011	2	DE, FR	2	ES, GRC	4	BASF DocID 2013/1066508
Total number of trials per region		**18**	-	**18**	**Total number of trials**	**36**	

Table 2.1-2: Processing studies available for grape

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Grape	2004	4	DE	-	-	4	2005/1014175
Total number of trials per region					Total number of trials	4	

2.1.1 Supervised residue trials in grape

Schulz S (2005)

Full study reference

Schulz S (2005): Study on the Residue Behaviour of Alpha-Cypermethrin in Wine Grapes after Treatment with BAS 310 11 I under Field Conditions in Spain and Italy, 2003; BASF Doc-ID 2005/1004977

Perny A (2005)

Full study reference

Perny A (2005): Study of the residue behaviour of alpha-cypermethrin on grapes after application of BAS 310 11 I under field condition, in France, in 2003; BASF Doc-ID 2005/1006474

Perny A (2005)
Full study reference
Perny A (2005): Study of the behaviour of alphacypermethrin on grapes after application of BAS 310 41 I under field condition, in Southern France and Greece, in 2004; BASF Doc-ID 2005/1007589

Perny A (2005)
Full study reference
Perny A (2005): Study on the behaviour of BAS 310 I in Grapes after Treatment with BAS 310 41 I under Field Conditions, in Germany and in Northern France; BASF Doc-ID 2005/1007591

Jones G (2006)
Full study reference
Jones G (2006): Study on the residue behaviour of BAS 310 I in Grapes after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF Doc-ID 2006/1026853

Diehl M (2007)
Full study reference
Diehl M (2007): Study on the Residue Behaviour of BAS 310 I in Grape after Treatment with BAS 310 40 I under open field Conditions In Southern and Northern Europe, 2006; BASF Doc-ID 2007/1008492

Material and Methods:

In the years 2003, 2004, 2005 and 2006 a field program on grapes was conducted in representative wine growing areas in France (Northern and Southern European region), Germany, Greece, Italy, and Spain to investigate the residue behaviour of alpha-cypermethrin.

During the 2003 growing season, four field trials were conducted in grapes in the Northern part of France. An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 11 I) was foliar applied on grapes on two different plots in each trial. In one variant, one treatment was done at a target rate of 15 g a.s./ha. This was compared to two treatments with the same application rate. The last application took place at growth stages between 85 (softening of berries) and 89 (berries ripe for harvest). Grape specimens were collected directly after the last application as well as 3-4, 7 and 14 days thereafter.
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS with BASF method No. 546/0 which has a limit of quantification of 0.05 mg/kg in all sample materials.

In the same year, four field trials were conducted with wine grapes in Spain and Italy. An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 11 I) was foliar applied to grapes twice at a target rate of 15 g a.s./ha. The last application took place at growth stage 85. Grape specimens were collected directly after the last application as well as 3, 6-7 and 14 days thereafter. The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS with BASF method No. 546/0 which has a limit of quantification of 0.05 mg/kg in all sample materials.

During the 2004 growing season, a total of four field trials were conducted on grapes in Germany and in the Northern part of France. Each trial included separate plots which were treated with alpha-cypermethrin formulated as a soluble concentrate (100 g a.s./L; BAS 310 41 I) either once at a target rate of 15 g a.s./ha, or twice at a target rate of 15 g a.s./ha. The last application took place at growth stages between 85 and 89. Bunches were collected just after the last application, then 3, 7 and 14 days after the last application. The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS using BASF method No. 567/0 which has a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2004 growing season, a total of four field trials were conducted in Southern France and Greece. A soluble concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 41 I) was foliar applied to grapes twice at a target rate of 15 g a.s./ha. The last application took place at growth stages between 79 (majority of berries touching) and 85 (softening of berries). Grape fruit specimens were sampled directly after the last application as well as 2-3, 6-7 and 13-14 days thereafter. The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS using BASF method No. 567/0 which has a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2005 growing season, eight field trials were conducted in representative vineyards in France (Northern and Southern European region), Italy, Spain, Greece and Germany.
An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 40 I), was foliar applied to separate plots of grapes either once or twice at a target rate of 12.5 g a.s./ha. The last application took place at growth stages between 83 (berries developing colour) and 89 (berries ripe for harvest). Specimens of grape bunches were collected immediately after the last application from each plot as well as 2-4, 7-8, and 14 days thereafter.
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS using BASF method No. 567/0 which has a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2006 growing season, eight field trials were conducted in representative vineyards in France (Northern and Southern European region), Germany, Greece, Italy, and Spain.
An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 40 I), was foliar applied to separate plots of grapes either once or twice at a target rate of 12.5 g a.s./ha.
The last application took place at growth stages between 79 (majority of berries touching) and 88. Specimens of grape bunches were collected immediately after the last application (0 DALA) from each plot as well as 3-4, 6-8, and 13-15 days thereafter.
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS using BASF method No. 567/0 which has a limit of quantification of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.1.1-1.

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1006474 Trial No. FAN/19/03 Study to GLP Study carried out in 2003	Wine grapes (variety Chardonnay)	France 67650 Rosheim Alsace (North of EU)	15 g as/ha 03.09.03	89	0 3 7 14	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	BASF analytical method N 546/0 fruit: mean recovery = 85.2%; SD: +/- 17.2; CV: 20.2%; n=4; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1006474 Trial No. FAN/20/03 Study to GLP Study carried out in 2003	Wine grapes (variety Pinot noir)	France 67117 Handschuheim Alsace (North of EU)	15 g as/ha 03.09.03	85	0 3 7 14	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	
BASF Doc ID 2005/1006474 Trial No. FBM/14/03 Study to GLP Study carried out in 2003	Wine grapes (variety Grolleau)	France 49540 Martigné Briand Pays de la Loire (North of EU)	15 g as/ha 11.09.03	85	0 4 7 14	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	
BASF Doc ID 2005/1006474 Trial No. FBM/15/03 Study to GLP Study carried out in 2003	Wine grapes (variety Grolleau)	France 49190 Saint Aubin de Luigné Pays de la Loire (North of EU)	15 g as/ha 11.09.03	85	0 4 7 14	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	
BASF Doc ID 2005/1007591 Trial No. DU2/06/04 Study to GLP Study carried out in 2004	Grape (wine) (variety Spaet-burgunder)	Germany, Stetten	15 g as/ha 28.09.04	85	0 3 7 14	fruit 0.013 fruit 0.011 fruit 0.011 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 82.6%; SD: +/- 5.9; CV: 7.1%; n=4; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007591 Trial No. DU4/06/04 Study to GLP Study carried out in 2004	Grape (wine) (variety Gewuerz-traminer)	France 76831 Eschbach (North of EU)	15 g as/ha 28.09.04	89	0 3 7 14	fruit 0.013 fruit 0.046 fruit <0.01 fruit 0.011	
BASF Doc ID 2005/1007591 Trial No. FAN/12/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Auxerrois)	France 67117 Handschuheim (North of EU)	15 g as/ha 06.09.04	85	0 3 7 14	fruit <0.01 fruit <0.01 fruit 0.010 fruit <0.01	
BASF Doc ID 2005/1007591 Trial No. FBM/06/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Chenin)	France 49540 Martigné Briand (North of EU)	15 g as/ha 28.09.04	85	0 3 7 14	fruit 0.021 fruit 0.012 fruit <0.01 fruit 0.014	

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026853 Trial No. AF/8830/BA/1 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Chardonnay)	France Collongette, Lugny,71260 Saône-et-Loire, (North of EU)	12.5 g as/ha 07.09.05	85	0 2 7 14	bunches 0.049 bunches 0.033 bunches 0.031 bunches 0.033	BASF analytical method N 567/0 bunches: mean recovery = 94.4%; SD: +/- 15.1; CV: 16.0%; n=7; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026853 Trial No. AF/8830/BA/2 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Grolleau)	France, La Ropeliere, St Jean des Mauvrets, 49320, Maine-et-Loire (North of EU)	12.5 g as/ha 13.09.05	85-89	0 3 7 14	bunches 0.019 bunches 0.026 bunches 0.021 bunches 0.015	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/3 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Riesling)	Germany, 69181 Leimen, Baden-Württemberg	12.5 g as/ha 06.09.05	85	0 3 7 14	bunches <0.01 bunches <0.01 bunches <0.01 bunches 0.013	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/4 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Müller-Thurgau)	Germany, 69231 Rauenberg, Baden-Württemberg	12.5 g as/ha 30.08.05	83	0 3 7 14	bunches 0.033 bunches 0.019 bunches 0.030 bunches 0.026	BASF analytical method N 567/0 bunches: mean recovery = 94.4%; SD: +/- 15.1; CV: 16.0%; n=7; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/5 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Gamay)	France, Labastide du Temple, 82100 Les Barthes, Tarn-et-Garonne EU South	12.5- 08.09.05	89	0 4 8 14	bunches 0.035 bunches 0.044 bunches 0.020 bunches 0.027	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/6 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Montuni)	Italy Castel S. Pietro, 40024 Bologna	12.5 g as/ha 29.08.05	88	0 3 7 14	bunches <0.01 bunches <0.01 bunches <0.01 bunches <0.01	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/7 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Macabeo)	Spain, Muela Alta S/N, Malejan 50540	12.5 g as/ha 16.09.05	85	0 3 7 14	bunches 0.021 bunches 0.039 bunches 0.013 bunches 0.019	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/8 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Roditis)	Greece, Anchialos, Thessaloniki, Central Macedonia, GR-57011	12.5 g as/ha 25.08.05	85	0 3 7 14	bunches <0.01 bunches 0.019 bunches 0.012 bunches <0.01	
BASF Doc ID 2007/1008492 Trial No A/NF/I/06/127 Study to GLP/ Study carried out in 2006	Grape (variety Chardonnay)	France Ay Champagne-Ardennes (North of EU)	11 g as/ha 12.09.06	85	0 3 6 15	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.6%; SD: +/- 8.8; CV: 8.9%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008492 Trial No A/NF/I/06/128 Study to GLP/ Study carried out in 2006	Grape (variety Chardonnay)	France Reims Champagne-Ardennes (North of EU)	13 g as/ha 30.08.06	79	0 4 8 14	fruit 0.012 fruit 0.010 fruit 0.011 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.6%; SD: +/- 8.8; CV: 8.9%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008492 Trial No A/GE/I/06/129 Study to GLP/ Study carried out in 2006	Grape (variety Müller-Thurgau)	Germany Geinsheim Rheinland-Pfalz	12 g as/ha 04.09.06	85	0 3 7 14	fruit 0.012 fruit <0.01 fruit <0.01 fruit 0.016	
BASF Doc ID 2007/1008492 Trial No A/GE/I/06/130 Study to GLP/ Study carried out in 2006	Grape (variety Phönix)	Germany Kirchheim Rheinland-Pfalz	14 g as/ha 04.09.06	87	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008492 Trial No A/SF/I/06/131 Study to GLP/ Study carried out in 2006	Grape (variety Grenache)	France Mazan Vaucluse (South of EU)	12 g as/ha 04.09.06	85	0 3 7 14	fruit 0.014 fruit 0.014 fruit <0.01 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.6%; SD: +/- 8.8; CV: 8.9%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008492 Trial No A/GR/I/06/132 Study to GLP/ Study carried out in 2006	Grape (variety Muscat)	Greece Kato Milea Central Macedonia	12 g as/ha 08.09.06	87	0 3 7 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008492 Trial No A/SP/I/06/133 Study to GLP/ Study carried out in 2006	Grape (variety Tempranillo)	Spain Pobla del Duc Valencia	13 g as/ha 25.08.06	85-87	0 3 7 13	fruit 0.043 fruit 0.026 fruit 0.043 fruit 0.023	
BASF Doc ID 2007/1008492 Trial No A/IT/I/06/134 Study to GLP/ Study carried out in 2006	Grape (variety Barbera)	Italy Costigliole D´Asti Piemont	13 g as/ha 04.10.06	88	0 4 6 13	fruit 0.029 fruit 0.016 fruit 0.014 fruit 0.011	
BASF Doc ID 2005/1004977 Trial No. ALO/15/03 Study to GLP Study carried out in 2003	Wine grapes (variety Cardenal, red grape)	Spain 41710 Utrera (Sevilla) Andalucia	15 g as/ha 2 treatm. last date 07.07.03	85 at last treatm.	0 3 7 14	bunches <0.05 bunches <0.05 bunches <u><0.05</u> bunches <0.05	BASF analytical method N 546/0 fruit: mean recovery = 82.8%; SD: +/- 15.6; CV: 18.9%; n=2; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1004977 Trial No. ALO/24/03 Study to GLP Study carried out in 2003	Wine grapes (variety Airen, white grape)	Spain 41720 Los Palacios (Sevilla) Andalucia	15 g as/ha 2 treatm. last date 28.07.03	85 at last treatm.	0 3 7 14	bunches <0.05 bunches <0.05 bunches <u><0.05</u> bunches <0.05	

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1004977 Trial No. ITA/10/03 Study to GLP Study carried out in 2003	Wine grapes (variety Croatina)	Italy 15059 Monleale Piedmont	15 g as/ha 2 treatm. last date 05.09.03	85 at last treatm.	0 3 6 14	bunches <0.05 bunches <0.05 bunches <u>≤0.05</u> bunches <0.05	BASF analytical method N 546/0 fruit: mean recovery = 82.8%; SD: +/- 15.6; CV: 18.9%; n=2; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1004977 Trial No. ITA/11/03 Study to GLP Study carried out in 2003	Wine grapes (variety Barbera)	Italy 15057 Torlona Piedmont	15 g as/ha 2 treatm. last date 05.09.03	85 at last treatm.	0 3 6 14	bunches <0.05 bunches <0.05 bunches <u>≤0.05</u> bunches <0.05	
BASF Doc ID 2005/1006474 Trial No. FAN/19/03 Study to GLP Study carried out in 2003	Wine grapes (variety Chardonnay)	France 67650 Rosheim Alsace (North of EU)	15 g as/ha 2 treatm. last date 03.09.03	89 at last treatm.	0 3 7 14	fruit <0.05 fruit <0.05 fruit <u>≤0.05</u> fruit <0.05	BASF analytical method N 546/0 fruit: mean recovery = 85.2%; SD: +/- 17.2; CV: 20.2%; n=4; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1006474 Trial No. FAN/20/03 Study to GLP Study carried out in 2003	Wine grapes (variety Pinot noir)	France 67117 Handschuheim Alsace (North of EU)	15 g as/ha 2 treatm. last date 03.09.03	85 at last treatm.	0 3 7 14	fruit <0.05 fruit <0.05 fruit <u>≤0.05</u> fruit <0.05	
BASF Doc ID 2005/1006474 Trial No. FBM/14/03 Study to GLP Study carried out in 2003	Wine grapes (variety Grolleau)	France 49540 Martigné Briand Pays de la Loire (North of EU)	15 g as/ha 2 treatm. last date 11.09.03	85 at last treatm.	0 4 7 14	fruit <0.05 fruit <0.05 fruit <u>≤0.05</u> fruit <0.05	BASF analytical method N 546/0 fruit: mean recovery = 85.2%; SD: +/- 17.2; CV: 20.2%; n=4; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1006474 Trial No. FBM/15/03 Study to GLP Study carried out in 2003	Wine grapes (variety Grolleau)	France 49190 Saint Aubin de Luigné Pays de la Loire (North of EU)	15 g as/ha 2 treatm. last date 11.09.03	85 at last treatm.	0 4 7 14	fruit <0.05 fruit <0.05 fruit <u>≤0.05</u> fruit <0.05	
BASF Doc ID 2005/1007591 Trial No. DU2/06/04 Study to GLP Study carried out in 2004	Grape (wine) (variety Spaet-burgunder)	Germany, Stetten	15 g as/ha 2 treatm. last date 28.09.04	85 at last treatm.	0 3 7 14	fruit 0.027 fruit 0.026 fruit <u>0.030</u> fruit 0.024	
BASF Doc ID 2005/1007591 Trial No. DU4/06/04 Study to GLP Study carried out in 2004	Grape (wine) (variety Gewuerz-traminer)	France 76831 Eschbach (North of EU)	15 g as/ha 2 treatm. last date 28.09.04	89 at last treatm.	0 3 7 14	fruit 0.045 fruit 0.010 fruit <u>0.031</u> fruit 0.029	BASF analytical method N 567/0 fruit: mean recovery = 82.6%; SD: +/- 5.9; CV: 7.1%; n=4; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007591 Trial No. FAN/12/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Auxerrois)	France 67117 Handschuheim (North of EU)	15 g as/ha 2 treatm. last date 06.09.04	85 at last treatm.	0 3 7 14	fruit 0.018 fruit 0.015 fruit <u>0.018</u> fruit 0.013	
BASF Doc ID 2005/1007591 Trial No. FBM/06/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Chenin)	France 49540 Martigné Briand (North of EU)	15 g as/ha 2 treatm. last date 28.09.04	85 at last treatm.	0 3 7 14	fruit 0.014 fruit 0.019 fruit 0.019 fruit <u>0.020</u>	BASF analytical method N 567/0 fruit: mean recovery = 82.6%; SD: +/- 5.9; CV: 7.1%; n=4; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1007589 Trial No. FBD/01/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Syrah)	France 26600 Pont d´Isère Rhone-Alpes (South of EU)	15 g as/ha 2 treatm. last date 03.09.04	85 at last treatm.	0 3 6 14	fruit 0.090 fruit 0.084 fruit 0.055 fruit <u>0.069</u>	
BASF Doc ID 2005/1007589 Trial No. FTL/01/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Negrette)	France 31620 Fronton Midi-Pyrenees (South of EU)	15 g as/ha 2 treatm. last date 07.09.04	85 at last treatm.	0 3 7 14	fruit 0.084 fruit 0.034 fruit 0.036 fruit <u>0.061</u>	BASF analytical method N 567/0 fruit: mean recovery = 96.8%; SD: +/- 4.3; CV: 4.5%; n=2; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007589 Trial No. GRE/03/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Xinomavro)	Greece 59200 Naousa Northern Greece-Macedonia	15 g as/ha 2 treatm. last date 16.09.04	79 at last treatm.	0 2 7 13	fruit 0.079 fruit 0.027 fruit <u>0.048</u> fruit 0.045	
BASF Doc ID 2005/1007589 Trial No. GRE/04/04 Study to GLP/ Study carried out in 2004	Grape (wine) (variety Xinomavro)	Greece 59200 Marina Northern Greece-Macedonia	15 g as/ha 2 treatm. last date 16.09.04	81 at last treatm.	0 2 7 13	fruit 0.075 fruit 0.054 fruit <u>0.051</u> fruit 0.043	
BASF Doc ID 2006/1026853 Trial No. AF/8830/BA/1 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Chardonnay)	France Collongette, Lugny,71260 Saône-et-Loire, (North of EU)	12.5 g as/ha 2 treatm. last date 07.09.05	85 at last treatm.	0 2 7 14	bunches 0.082 bunches 0.077 bunches <u>0.064</u> bunches 0.063	BASF analytical method N 567/0 bunches: mean recovery = 94.4%; SD: +/- 15.1; CV: 16.0%; n=7; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026853 Trial No. AF/8830/BA/2 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Grolleau)	France, La Ropeliere, St Jean des Mauvrets, 49320, Maine-et-Loire (North of EU)	12.5 g as/ha 2 treatm. last date 13.09.05	85-89 at last treatm.	0 3 7 14	bunches 0.026 bunches 0.028 bunches <u>0.033</u> bunches 0.013	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/3 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Riesling)	Germany, 69181 Leimen, Baden-Württemberg	12.5 g as/ha 2 treatm. last date 06.09.05	85 at last treatm.	0 3 7 14	bunches 0.016 bunches <0.01 bunches 0.028 bunches <u>0.037</u>	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/4 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Müller-Thurgau)	Germany, 69231 Rauenberg, Baden-Württemberg	12.5 g as/ha 2 treatm. last date 30.08.05	83 at last treatm.	0 3 7 14	bunches 0.049 bunches 0.037 bunches <u>0.043</u> bunches 0.039	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/5 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Gamay)	France, Labastide du Temple, 82100 Les Barthes, Tarn-et-Garonne (South of EU)	12.5 g as/ha 2 treatm. last date 08.09.05	89 at last treatm.	0 4 8 14	bunches 0.071 bunches 0.036 bunches 0.034 bunches <u>0.051</u>	BASF analytical method N 567/0 bunches: mean recovery = 94.4%; SD: +/- 15.1; CV: 16.0%; n=7; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/6 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Montuni)	Italy Castel S. Pietro, 40024 Bologna	12.5 g as/ha 2 treatm. last date 29.08.05	88 at last treatm.	0 3 7 14	bunches 0.020 bunches <0.01 bunches <u><0.01</u> bunches <0.01	BASF analytical method N 567/0 bunches: mean recovery = 94.4%; SD: +/- 15.1; CV: 16.0%; n=7; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/7 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Macabeo)	Spain, Muela Alta S/N, Malejan 50540	12.5 g as/ha 2 treatm. last date 16.09.05	85 at last treatm.	0 3 7 14	bunches 0.035 bunches 0.027 bunches <u>0.045</u> bunches 0.020	
BASF Doc ID 2006/1026853 Trial No AF/8830/BA/8 Study to GLP/ Study carried out in 2005	Grape (wine) (variety Roditis)	Greece, Anchialos, Thessaloniki, Central Macedonia, GR-57011	12.5 g as/ha 2 treatm. last date 25.08.05	85 at last treatm.	0 3 7 14	bunches 0.030 bunches 0.035 bunches <u><0.01</u> bunches <0.01	
BASF Doc ID 2007/1008492 Trial No A/NF/I/06/127 Study to GLP/ Study carried out in 2006	Grape (variety Chardonnay)	France Ay Champagne-Ardennes (North of EU)	14/13 g as/ha 2 treatm. last date 12.09.06	85 at last treatm.	0 3 6 15	fruit 0.018 fruit 0.016 fruit <u>0.012</u> fruit 0.012	BASF analytical method N 567/0 fruit: mean recovery = 98.6%; SD: +/- 8.8; CV: 8.9%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008492 Trial No A/NF/I/06/128 Study to GLP/ Study carried out in 2006	Grape (variety Chardonnay)	France Reims Champagne-Ardennes (North of EU)	11/13 g as/ha 2 treatm. last date 30.08. 06	79 at last treatm.	0 4 8 14	fruit 0.017 fruit 0.025 fruit <u>0.019</u> fruit 0.010	
BASF Doc ID 2007/1008492 Trial No A/GE/I/06/129 Study to GLP/ Study carried out in 2006	Grape (variety Müller-Thurgau)	Germany Geinsheim Rheinland-Pfalz	12 g as/ha 2 treatm. last date 04.09.06	85 at last treatm.	0 3 7 14	fruit 0.016 fruit 0.014 fruit 0.020 fruit <u>0.021</u>	BASF analytical method N 567/0 fruit: mean recovery = 98.6%; SD: +/- 8.8; CV: 8.9%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008492 Trial No A/GE/I/06/130 Study to GLP/ Study carried out in 2006	Grape (variety Phönix)	Germany Kirchheim Rheinland-Pfalz	13/12 g as/ha 2 treatm. last date 04.09.06	87 at last treatm.	0 3 7 14	fruit 0.013 fruit 0.010 fruit 0.014 fruit <u>0.028</u>	
BASF Doc ID 2007/1008492 Trial No A/SF/I/06/131 Study to GLP/ Study carried out in 2006	Grape (variety Grenache)	France Mazan Vaucluse (South of EU)	12/13 g as/ha 2 treatm. last date 04.09.06	85 at last treatm.	0 3 7 14	fruit 0.029 fruit 0.015 fruit <u>0.015</u> fruit 0.011	
BASF Doc ID 2007/1008492 Trial No A/GR/I/06/132 Study to GLP/ Study carried out in 2006	Grape (variety Muscat)	Greece Kato Milea Central Macedonia	13 g as/ha 2 treatm. last date 08.09.06	87 at last treatm.	0 3 7 13	fruit 0.022 fruit 0.011 fruit <u>0.012</u> fruit <0.01	

Table 2.1.1-1: Residues in grapes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days GAP for EU-S is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days							
GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008492 Trial No A/SP/I/06/133 Study to GLP/ Study carried out in 2006	Grape (variety Tempranillo)	Spain Pobla del Duc Valencia	13 g as/ha 2 treatm. last date 25.08.06	85-87 at last treatm.	0 3 7 13	fruit 0.035 fruit 0.029 fruit 0.028 fruit <u>0.039</u>	BASF analytical method N 567/0 fruit: mean recovery = 98.6%; SD: +/- 8.8; CV: 8.9%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008492 Trial No A/IT/I/06/134 Study to GLP/ Study carried out in 2006	Grape (variety Barbera)	Italy Costigliole D´Asti Piemont	13/12 g as/ha 2 treatm. last date 04.10.06	88 at last treatm.	0 4 6 13	fruit 0.043 fruit 0.035 fruit <u>0.030</u> fruit 0.025	

_ underlined values were used for MRL calculation

Findings:

The residue studies in grapes presented in the alpha-cypermethrin dossier were carried out in 5 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 14 days
- for the Southern European region: 2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha – 16 trials including both variants, 8 conducted in the Northern European Region and 8 conducted in the Southern European Region
- one or two treatments at a target rate of 15 g a.s./ha - 8 trials including both variants, all conducted in the Northern European Region
- two treatments at a target rate of 15 g a.s./ha - 8 trials, all conducted in the Southern European Region.

After one application at 12.5 g a.s./ha, initial residues ranged between <0.01 mg/kg and 0.049 mg/kg and declined to <0.01 – 0.044 mg/kg, <0.01 – 0.043 mg/kg and <0.01 – 0.033 mg/kg at 2-4, 6-8 and 13-15 days after application, respectively.

After two applications at 12.5 g a.s./ha, initial residues ranged between 0.013 mg/kg and 0.082 mg/kg and declined to <0.01 – 0.077 mg/kg, <0.01 – 0.064 mg/kg and <0.01 – 0.063 mg/kg at 2-4, 6-8 and 13-15 days after application, respectively.

A total of 16 residue trials was performed to investigate the residue behaviour after application of either one or two treatments at a rate of 15 g a.s./ha. These trials were conducted during the growing seasons 2003 and 2004. Residue analysis of the samples generated in these studies was carried out according to two different methods with different limits of quantification (LOQ): while the samples from the 2003 studies were analysed according to method No 546/0 with a LOQ of 0.05 mg/kg, the samples from the 2004 trials were analysed with method N° 567/0 which has a lower validated LOQ of 0.01 mg/kg.

In the year 2003, 4 trials were conducted in the Northern European Region with either one or two treatments at a rate of 15 g a.s./ha applied to separate plots. In Southern Europe, 4 trials were conducted with two applications at a rate of 15 g a.s./ha. The samples generated in these studies were analysed according to BASF method No. 546/0 with a LOQ of 0.05 mg/kg. No residues above the LOQ were found in any of the treated samples from all of these trials, regardless of number of application or sampling interval (all <0.05).

In the year 2004, 4 trials were conducted in the Northern European Region with either one or two treatments at a rate of 15 g a.s./ha applied to separate plots. In Southern Europe, 4 trials were conducted with two applications at a rate of 15 g a.s./ha. The samples generated in these studies were analysed according to BASF method No. 567/0 with a LOQ of 0.01 mg/kg.
After one application at 15 g a.s./ha, initial residues in fruit ranged between <0.01 and 0.021 mg/kg. Residues between <0.01 – 0.046 mg/kg, <0.01 – 0.011 mg/kg and <0.01 – 0.014 mg/kg were found 3, 7 and 14 days afterwards.

After two applications at 15 g a.s./ha, initial residues between 0.014 and 0.090 mg/kg were found in fruit or bunches, respectively. Residues between 0.010 – 0.084 mg/kg, 0.018 – 0.055 mg/kg and 0.013 – 0.069 mg/kg were found 2-3, 6-7 and 13-14 days afterwards.

Conclusion:

The residue studies presented demonstrate that residues in grapes treated with alpha-cypermethrin according to the proposed GAP are below a level of 0.1 mg/kg at the target PHI.

Report:	**Meyer M., 2013a**
Title:	Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in wine-grapes after treatment with either BAS 310 55 I or BAS 310 40 I under field conditions in Germany, France, Spain and Greece, 2011
Document No:	BASF DocID 2013/1066508
Guidelines:	EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7
GLP	yes

Executive Summary

During the growing season of 2011, a total of four trials with grapes were conducted in Germany, Northern France, Spain and Greece to determine the magnitude of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC). Therefore two solo formulations BAS 310 55 I (50 g/L BAS 310 I, ME) and BAS 310 40 I (100 g/L BAS 310 I, EC) were applied once to the treated plot at a rate equivalent of 0.0125 kg a.s./ha. In all trials the applications were made at crop stages between BBCH 87 and 89.

For the analysis the grape specimens were taken immediately after the last treatment (0 DALA) and at 2-3, 6-8 and 13-15 DALA. The grape specimens were analysed for residues of BAS 310 I according to BASF Method No. 567/0, which has a limit of quantitation (LOQ) of 0.01 mg/kg. Directly after application of BAS 310 55 I and BAS 310 40 I, the residue of alpha-cypermethrin ranged between 0.01-0.06 mg/kg in grape specimens. At 6-8 DALA, residues found were 0.02-0.05 mg/kg, and at 13-15 DALA residues ranged between <0.01-0.04 mg/kg.

The analytical results obtained demonstrate that the treatment with one application of BAS 310 55 I and BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 55 I (ME)
BAS 310 40 I (EC)
Lot/Batch #: 101198 BAS 310 55 I, 50 g/L alpha-cypermethrin
1209 BAS 310 40 I, 100 g/L alpha-cypermethrin
Purity:
CAS#: 67375-30-8 alpha-cypermethrin
Development code:
Spiking levels: 0.01-5.0 mg/kg

2. Test Commodity:

Crop: Grape
Type: Berries and other small fruit
Variety: Vanessa, Topkapi, Asterix, Brillante
Botanical name: *Vitis vinifera*
Crop parts(s) or processed
Commodity: Fruit
Sample size: 1.0 kg (12 bunches)

B. STUDY DESIGN

1. Test procedure

During the growing season of 2011, a total of four trials with grapes were conducted in Germany, Northern France, Spain and Greece to determine the magnitude of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC). Therefore two solo formulations BAS 310 55 I (50 g/L BAS 310 I, ME) and BAS 310 40 I (100 g/L BAS 310 I, EC) were applied once to the treated plot at a rate equivalent of 0.0125 kg a.s./ha.
BAS 310 55 I was spray-applied at a rate of 0.250 L/ha of formulated product, corresponding to 0.0125 kg/ha of BAS 310 I.
BAS 310 40 I was spray-applied at a rate of 0.125 L/ha of formulated product, corresponding to 0.0125 kg/ha of BAS 310 I.
The water volume was 800 L/ha. The applications took place 6 – 8 days before harvest (DBH), at crop stages between BBCH 87 and 89. At the sampling occasion 0 DALA, the untreated specimens were collected immediately before the last application of the treated plots. The treated wine grape specimens were sampled immediately after the application (0 DALA), as well as 2 – 3 DALA, 6 – 8 DALA (PHI) and 13 – 15 DALA.
In all trials the applications were made at crop stages between BBCH 87 and 89.
For the analysis the grape specimens were taken immediately after the last treatment (0 DALA) and at 2-3, 6-8 and 13-15 DALA.

Table 2.1.1-2: Target application rates and timings for grape

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2011	4	1	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	800	1st appl.: 7 ± 1 DBH
		1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	800	1st appl.: 7 ± 1 DBH

DBH: days before harvest

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).
The residues of alpha-cypermethrin in grape specimens were extracted with methanol/water/hydrochloric acid (70:25:5, v/v/v). After centrifugation, an aliquot of the extract was partitioned into cyclohexane. An aliquot of the cyclohexane phase was evaporated and the residue was taken up into methanol/water (80:20, v/v). Final determination was performed by LC/MS/MS using the ammonium adduct of cypermethrin.
The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery in wine-grapes averaged 86±7% (mean ± RSD) for alpha-cypermethrin at fortification levels of 0.01 and 5.0 mg/kg.

Table 2.1.1-3: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in grapes

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
Method No. 567/0		alpha-cypermethrin		
Wine-grapes	0.01	4	84	3
	5.0	4	88	8

II. RESULTS AND DISCUSSION

The residue levels of alpha-cypermethrin (BAS 310 I) in grape specimens taken directly after the last application (0 DALA) of BAS 310 55 I ranged between 0.01-0.06 mg/kg in all trials. In the specimens taken 2-3 DALA 0.02- 0.05 mg/kg were found. The residues remained at this level at 6-8 DALA (0.02- 0.04 mg/kg). At the last sampling (13-15 DALA) < 0.01-0.04 mg/kg were found.

Residues found in grape specimens taken directly after the last application (0 DALA) of BAS 310 40 I contained residues of BAS 310 I in a range of 0.03 mg/kg to 0.06 mg/kg. They decreased slightly to 0.02- 0.03 mg/kg in the specimen taken 2- 3 DALA. At 6-8 DALA 0.02-0.05 mg/kg were found. At the last sampling (13- 15 DALA) the residues remained at this level (0.01- 0.04 mg/kg).

No residues of BAS 310 I (alpha-cypermethrin) above the limit of quantification were found in any of the analysed untreated specimens.
An overall summary of the residues is given in the table below.

Table 2.1.1-4: Summary of residues of BAS 310 I in grape after application of BAS 310 55 I and BAS 310 40 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	alpha-cypermethrin (BAS 310 I)
Wine-grapes	2011	1	BAS 310 55 I (ME)	0	87-89	fruit	0.01 – 0.06
				2-3	87-89		0.02 – 0.05
				6-8	87-89		0.02 – 0.04
				13-15	89		< 0.01 – 0.04
		1	BAS 310 40 I (EC)	0	87-89	fruit	0.03 – 0.06
				2-3	87-89		0.02 – 0.03
				7-8	87-89		0.02 – 0.05
				13-15	89		0.01 – 0.04

DALA = days after last application
BBCH = growth stage at respective sampling

III. CONCLUSION

Directly after application of BAS 310 55 I and BAS 310 40 I, the residue of alpha-cypermethrin ranged between 0.01-0.06 mg/kg in grape specimens. At 6-8 DALA, residues found were 0.02-0.05 mg/kg, and at 13-15 DALA residues ranged between <0.01-0.04 mg/kg. The analytical results obtained demonstrate that the treatment with one application of BAS 310 55 I and BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.

Table 2.1.1-5: Residues of BAS 310 I after one application of the formulation BAS 310 55 I and BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 408606 **Doc ID:** 2013/1066508 **Trial No.:** L110429 **GLP:** yes **Year** 2011	Grape	Germany	BAS 310 55 I BAS 310 I 1 x 0.0125	89	0	fruit	0.06
				89	3	fruit	0.04
				89	7	fruit	0.04
				89	15	fruit	0.03
			BAS 310 40 I BAS 310 I 1 x 0.0125	89	0	fruit	0.03
				89	3	fruit	0.02
				89	7	fruit	0.03
				89	15	fruit	0.02
Study code: 408606 **Doc ID:** 2013/1066508 **Trial No.:** L110430 **GLP:** yes **Year** 2011	Grape	France	BAS 310 55 I BAS 310 I 1 x 0.0125	87	0	fruit	0.05
				87	3	fruit	0.05
				87	7	fruit	0.04
				89	13	fruit	0.04
			BAS 310 40 I BAS 310 I 1 x 0.0125	87	0	fruit	0.06
				87	3	fruit	0.03
				87	7	fruit	0.05
				89	13	fruit	0.04

DALA = days after last application
BBCH = growth stage at respective sampling

Table 2.1.1-6: Residues of BAS 310 I after one application of the formulation BAS 310 55 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 408606 **Doc ID:** 2013/1066508 **Trial No.:** L110431 **GLP:** yes **Year** 2011	Grape	Spain	BAS 310 55 I BAS 310 I 1 x 0.0125	89	0	fruit	0.01
				89	2	fruit	0.02
				89	6	fruit	0.02
				89	13	fruit	<0.01
			BAS 310 40 I BAS 310 I 1 x 0.0125	88	0	fruit	0.04
				88	3	fruit	0.02
				89	7	fruit	0.03
				89	14	fruit	0.02
Study code: 408606 **Doc ID:** 2013/1066508 **Trial No.:** L110432 **GLP:** yes **Year** 2011	Grape	Greece	BAS 310 55 I BAS 310 I 1 x 0.0125	87	0	fruit	0.04
				87	2	fruit	0.04
				89	8	fruit	0.02
				89	15	fruit	0.02
			BAS 310 40 I BAS 310 I 1 x 0.0125	87	0	fruit	0.04
				87	2	fruit	0.03
				89	8	fruit	0.02
				89	15	fruit	0.01

DALA = days after last application
BBCH = growth stage at respective sampling

2.1.2 Processing study in grape

Report:	Raunft E, Hafemann C, Mackenroth C. 2005a Study on the residue behaviour of alpha-cypermethrin in grapes and grape process fractions after application of BAS 310 41 I under field conditions in Germany, 2004 2005/1014175
Guidelines:	EC 91/414 Annex II (Part A Section 6),EEC 1607/VI/97 rev. 2 10.06.1999,EEC 7029/VI/95 rev. 5,EEC 7525/VI/95 rev. 7,EEC 7035/VI/95 rev. 5,EEC 91/414 Annex III (Part A Section 8)
GLP:	yes (certified by Landesamt für Umweltschutz und Gewerbeaufsicht, Mainz, Germany)

Material and methods:

During the 2004 growing season, four field trials were conducted in grapes in different representative growing areas in Germany (Baden-Württemberg, Rheinland-Pfalz) in order to determine the magnitude and distribution of alpha-cypermethrin residues in the various intermediate and end products after processing. Two trials in the wine variety Spätburgunder (DU2/01/04 and DU2/02/04) and two trials the wine variety Portugieser (DU4/01/04 and DU4/02/04) were established.
Two of the trials consisted of two plots (DU2/01/04 and DU4/01/04), one untreated and one treated; the other two consisted of one treated plot each (DU2/02/04 and DU4/02/04).

A soluble concentrate formulation of alpha-cypermethrin was foliar applied 2 times at a target rate of 75 g a.s./ha for each application, resulting in a seasonal target rate of 150 g a.s./ha. Actual rates were within +/10 % of the nominal rate. This exaggerated rate was used in an attempt to generate residue levels sufficiently above the method limit of quantification (LOQ) in the raw agricultural commodity.

The applications were made 20-21 and 14 days before the anticipated harvest date using a spray volume of approximately 1000 I/ha. The growth stage at the last application was BBCH 85 (softening of berries).

Specimens of grapes were collected on the day of the last application for analysis and 14 days later for analysis of the raw agricultural commodity (RAC) and also for processing. RAC specimens were frozen within 24 hours of sampling, and remained frozen at or below -18°C, including during transportation, until analysis. The specimens for processing were maintained at ambient or cool temperatures until processing.
The specimens for processing were sent to the "Dienstleistungszentrum Ländlicher Raum – Rheinpfalz (DLR)" in Neustadt/Weinstraße. All specimens sent to the processing facility arrived within 24 hours of being sampled in good condition.

The following specimens resulted from the processing:
- must (rose and red wine making),
- stalks (red wine making)
- pomace (rose and red wine making)
- wine (rose and red wine making)
- yeast (rose and red wine making)
- raisins from dried grapes

The wine samples were stored at cellar temperatures. The samples of the intermediate products were taken at the relevant stages during the processing procedure and were frozen immediately. The analytical part of the study was performed at the Agricultural Research Center of BASF in Limburgerhof, Germany.
The specimens (RAC) as well as the processed commodity samples were analysed for alpha-cypermethrin by means of HPLC-MS/MS according to BASF method N° 567/0. The LOQ of the method was 0.01 mg/kg for all sample materials analysed.

As part of this study, a validation of method N° 567/0 in the matrix wine was performed. At fortification levels of 0.01 and 0.1 mg/kg, the recovery rates averaged at 95% (Transition 433-191) and at 98% (Transition 435-193).

Method performance was checked by determining the procedural recoveries in grape RAC and processed fractions.
The mean results of the procedural recoveries were 87 % at fortification levels between 0.01 mg/kg and 1.0 mg/kg with a standard deviation of 15.2 and a coefficient of variation of 17.4 %.

Findings:

No residues of alpha-cypermethrin at or above the method LOQ were detected in the untreated RAC specimens or in the processed fractions (must, stalks, pomace, wine, yeast and raisins) obtained from untreated RAC specimens.

Residues of alpha-cypermethrin in treated grape RAC collected at 14 days after the last application ranged from 0.05 to 0.12 mg/kg.

During the process of rose wine making, alpha-cypermethrin residues were found as follows:
in pomace after pressing, residues ranged between 0.12 and 0.22 mg/kg (concentration factors between 1.83 and 3.2). Relatively low residues were found in naturally cloudy must with values between 0.05 and 0.13 mg/kg (concentration factors between 0.83 and 1.6) which were further decreased after the following separation: in the separated must alpha-cypermethrin was found between 0.01 and 0.07 mg/kg (concentration factors between 0.2 and 0.8). Correspondingly, the waste product must deposit contained higher residues between 0.36 and 0.68 mg/kg which leads to concentration factors between 5.67 and 12.8.

In yeast deposit, too, concentrated residues between 0.16 and 0.77 mg/kg were found (concentration factors between 3.2 and 10.0). The final product rosé wine did not contain any alpha-cypermethrin above the limit of quantification (<0.01 mg/kg).

The residue situation in the red wine process was very similar: a slight concentration in the fractions stalks (factors 1.25 to 3.17) and crush (factors 1.42 to 2.2); low residues in the different must portions (naturally cloudy: factors 0.92 to 1.2, separated: factors 0.67 to 1.0).
As with rosé wine processing, the highest concentration was seen in the waste fractions pomace (factors 3.2 to 5.67), must deposit (factors 2.08 to 5.2) and yeast deposit (factors 5.83 to 10.8). The red wine did not show any alpha-cypermethrin above the LOQ.

Raisins are produced by drying which must lead to a concentration of residues. The residues found were between 0.17 and 0.38 mg/kg corresponding to concentration factors between 3.17 and 3.4. The waste fraction stalks which was removed after drying showed the highest residue concentration in this study (concentration factors 9.08 to 46.6).

The residue levels detected in the treated specimens and processed fractions as well as the calculated transfer factors are presented in the following table.

Table 2.1.2-1: Summary of alpha-cypermethrin residues in grapes and transfer factors - DocID 2005/1014175

Process	Sample material	DALA [1)]	Residues of Alpha-cypermethrin (mg/kg)				Transfer factor (Concentration factor) [2) 3)]			
RAC										
	fruit	0	0.16	0.10	0.22	0.11				
	fruit	14	0.06	0.05	0.12	0.05	1.00	1.00	1.00	1.00
Rosé wine making		14								
	pomace		0.17	0.12	0.22	0.16	2.83	2.40	1.83	3.20
	must naturally cloudy		0.05	0.07	0.13	0.08	0.83	1.40	1.08	1.60
	must separated		0.04	0.01	0.07	0.04	0.67	0.20	0.58	0.80
	must deposit		0.36	0.45	0.68	0.64	6.00	9.00	5.67	12.8
	yeast deposit		0.38	0.16	0.77	0.50	6.33	3.20	6.42	10.0
	rosé wine		<0.01	<0.01	<0.01	<0.01	0.17	0.20	0.08	0.20
Red wine making		14								
	Stalks		0.19	0.12	0.15	0.15	3.17	2.40	1.25	3.00
	crush		0.11	0.08	0.17	0.11	1.83	1.60	1.42	2.20
	pomace		0.34	0.23	0.40	0.16	5.67	4.60	3.33	3.20
	must naturally cloudy		0.06	0.06	0.11	0.06	1.00	1.20	0.92	1.20
	must separated		0.04	0.04	0.10	0.05	0.67	0.80	0.83	1.00
	must deposit		0.26	0.26	0.25	0.21	4.33	5.20	2.08	4.20
	yeast deposit		0.35	0.33	0.93	0.54	5.83	6.60	7.75	10.8
	red wine		<0.01	<0.01	<0.01	<0.01	0.17	0.20	0.08	0.20
Raisin production		14								
	Raisin		0.19	0.17	0.38	0.17	3.17	3.40	3.17	3.40
	Stalks (raisin)		1.59	0.85	1.09	2.33	26.5	17.0	9.08	46.6

1. Days after last application
2. Transfer factor =residue in processed fraction *I* residue in RAC (RAC =Raw Agricultural Commodity)
3. for calculation purposes, < 0.01 was set 0.01

Wine processing procedure:
The amount of grapes was divided into two equal portions and then processed in two different ways (rose wine making and red wine making). The final fermentation was the same in both procedures.

Pressing and Separation (Rosé wine)
The grapes were crushed, and pressed on the same day. The must was sulphured with 50 mg SO_2/L, and left overnight for deposit of the must and subsequent separation.

Pressing and Separation (Red wine)
The grapes were stemmed and crushed on the same day. The total amount of the crush was sulphured with 50 mg SO_2/L and heated up to 70°C. After the heating the crush was pressed according to good wine making practice. The must was left to separate overnight.

Fermentation, Separation and Bottling (Rosé and Red Wine)
The separated must was immediately filled into two 25 L glass balloons and mixed with yeast. After fermentation and deposit of the young wine, the yeast was separated, and the young wine was mixed with 100 mg SO_2/L, and approx. 2 g Bentonit/L wine.
The wines of each repetition were mixed and filled into a balloon up to the bung. After another separation and ripening of the wine, a further separation was made, and the wine was filtered using EK filter units. Immediately after the filtration from each plot, 6 x 0.75 L bottles were filled. The bottles were stored in the cellar at cellar temperature until hand over to the Study Director.

Raisin processing procedure:
The grapes were dried in a dry chamber at 45 °C until raisin ripeness. Then the stalks were removed from the raisins.

Wine processing procedure flowcharts:

Figure 2.1.2-1: Rosé wine processing procedure flowchart - DocID 2005/1014175

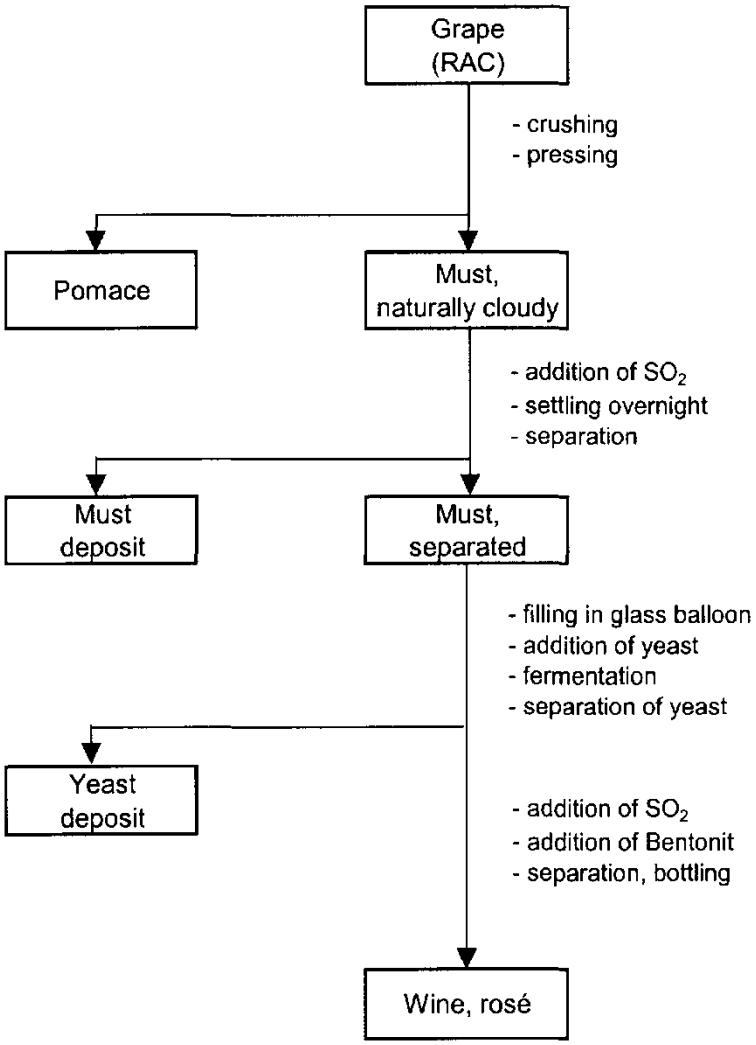


Figure 2.1.2-2: Red wine processing procedure flowchart - DocID 2005/1014175

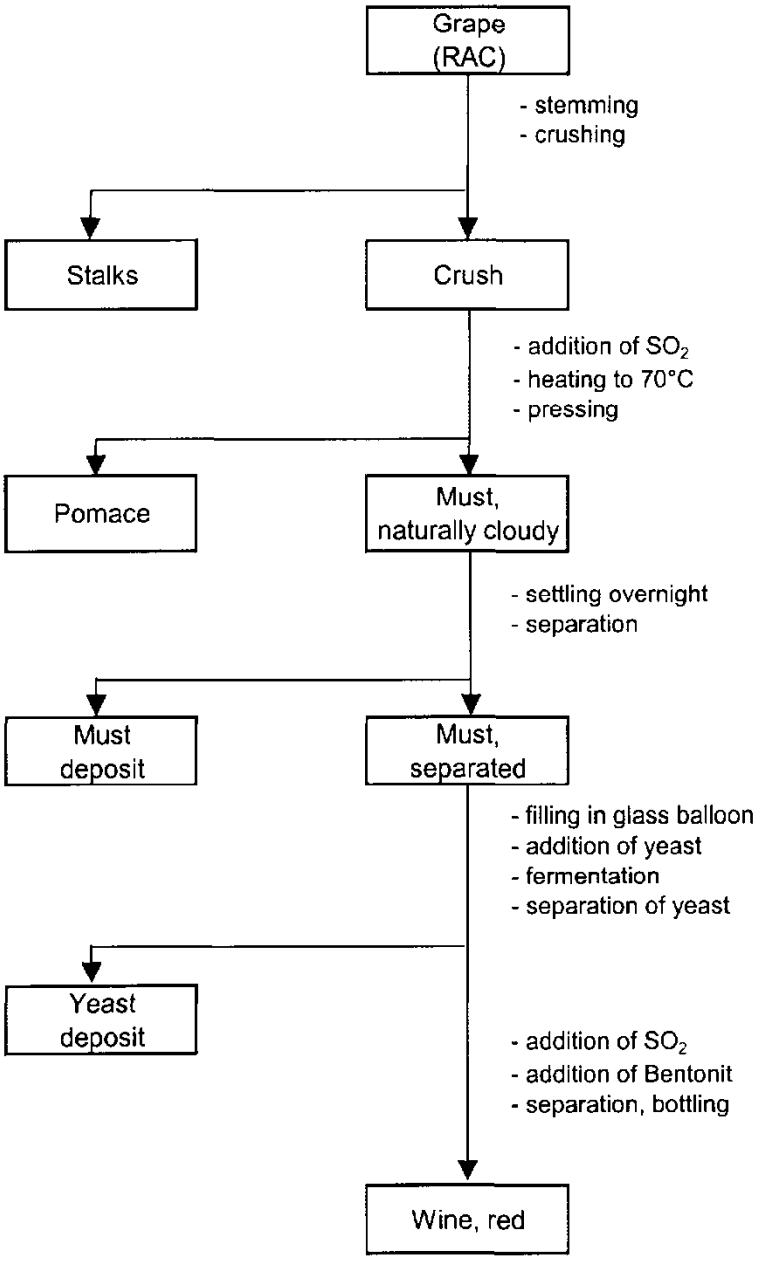


Table 2.1.2-2: Mass balance of grape processing – Rosé wine - DocID 2005/1014175

Process fraction	DU2/01/04	DU2/02/04	DU4/01/04	DU4/02/04
Grape (kg)	100.0	113.0	110.0	100.0
Pomace (kg)	18.1	21.2	20.0	15.5
Must naturally cloudy (kg)	78.0	88.0	85.0	79.0
Sum (kg)	96.1	109.2	105.0	94.5
Yield (%)	96.1	96.6	95.5	94.5
Must naturally cloudy (kg)	78.0	88.0	85.0	79.0
Must deposit (kg)	8.0	9.4	6.1	5.8
Must separated (kg)	70.0	78.0	79.0	73.0
Sum (kg)	78.0	87.4	85.1	78.8
Yield (%)	100.0	99.3	100.1	99.7
Must to fermentation [1] (kg)	40.0	40.0	40.0	40.0
Yeast deposit (kg)	2.9	2.9	2.4	2.5
Wine (kg)	34.3	33.6	33.9	34.0
Sum (kg)	37.2	36.5	36.3	36.5
Yield (%)	93.0	91.3	90.8	91.3

1) the portion "must for fermentation" is an aliquot of "must separated"

Table 2.1.2-3: Mass balance of grape processing – Red wine - DocID 2005/1014175

Process fraction	DU2/01/04	DU2/02/04	DU4/01/04	DU4/02/04
Grape (kg)	100.0	100.0	104.0	100.0
Stalks (kg)	3.5	5.2	5.7	4.0
Crush (kg)	96.0	93.0	97.0	93.0
Sum (kg)	99.5	98.2	102.7	97.0
Yield (%)	99.5	98.2	98.8	97.0
Crush before pressing[1] (kg)	95.0	92.0	96.0	92.0
Pomace (kg)	13.3	13.0	16.1	12.5
Must naturally cloudy (kg)	79.0	80.0	77.0	75.0
Sum (kg)	92.3	93.0	93.1	87.5
Yield (%)	102.9	101.1	97.0	95.1
Must naturally cloudy (kg)	79.0	80.0	77.0	75.0
Must deposit (kg)	11.2	14.2	10.7	9.9
Must separated (kg)	67.0	64.0	65.0	64.0
Sum (kg)	78.2	78.2	75.7	73.9
Yield (%)	99.0	97.8	98.3	98.5
Must to fermentation [2] (kg)	40.0	40.0	40.0	40.0
Yeast deposit (kg)	3.6	3.4	3.1	3.4
Wine (kg)	33.5	33.0	33.1	33.0
Sum (kg)	37.1	36.4	36.2	36.4
Yield (%)	92.8	91.0	90.5	91.0

1) the portion "crush before pressing" is an aliquot of "crush"
2) the portion "must for fermentation" is an aliquot of "must separated

Table 2.1.2-4: Mass balance of raisin processing - DocID 2005/1014175

Process fraction	DU2/01/04	DU2/02/04	DU4/01/04	DU4/02/04
Grape (kg)	5.01	5.08	5.32	5.15
Stalks (kg)	0.02	0.03	0.02	0.03
Raisins (kg)	1.24	1.22	1.03	0.82
Sum (kg)	1.26	1.25	1.05	0.85
Yield (%)	25.1	24.6	19.7	16.5

Conclusion:
The results of this processing study demonstrate clearly that residues in or on the raw agricultural commodity grapes are not transferred into the wine fraction during rosé or red wine making. Residues in the final products red or rosé wine were below the method LOQ (<0.01 mg/kg) in all cases. In raisins, residues are concentrated to a relatively high extent due to the drying process.

2.1.3 Estimation of MRL, HR and STMR for grape

For *grapes*, the following residue studies were considered (BASF DocIDs): 2005/1004977, 2005/1006474, 2005/1007589, 2005/1007591, 2006/1026853 and 2007/1008492.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2005/1004977 (open field)	-	<0.05 (4x)
2005/1006474 (open field)	<0.05 (4x)	-
2005/1007589 (open field)	-	0.048, 0.051, 0.061, 0.069
2005/1007591 (open field)	0.018, 0.02, 0.03, 0.031	-
2006/1026853 (open field)	0.033, 0.037, 0.043, 0.064	<0.01 (2x), 0.045, 0.051
2007/1008492 (open field)	0.012, 0.019, 0.021, 0.028	0.012, 0.015, 0.030, 0.039
OECD-MRL-calculation	**0.1 (n=16, STMR=0.032, HR=0.064)**	**<u>0.15</u> (n=16, STMR=0.049, HR=0.069)**

_ underlined values were used for risk assessment purposes

2.2 Strawberry

Residue data from supervised trials in strawberries were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.2-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Strawberry (field)	2005	4	BE, DE,FR, UK	4	FR, IT, ES	8	2007/1008489
Strawberry (field)	2006	4	DE, FR, NL, UK	4	FR, GR, IT, ES	8	2007/1008493
Strawberry (glasshouse)	2005	4	BE, DE, FR	4	FR, GR, IT, ES	8	2007/1007935
Total number of trials per region		**8 field 4 glasshouse**		**8 field 4 glasshouse**	**Total number of trials**	**16 field 8 glassh.**	

2.2.1 Supervised residue trials in strawberry

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the Residue Behaviour of BAS 310 40 I in Strawberry (field) after Treatment with BAS 310 40 1 under Field Conditions in Southern and Northern Europe, 2005; BASF Doc-ID 2007/1008489

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the Residue Behaviour of BAS 310 40 I in Strawberry after Treatment wit BAS 310 40 I under Greenhouse Conditions in Southern and Northern Europe, 2005; BASF Doc-ID 2007/1007935

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the Residue Behaviour of BAS 310 I in Strawberry after Treatment with BAS 310 40 I under open field Conditions in Southern and Northern Europe, 2006; BASF Doc-ID 2007/1008493

Material and Methods:

In the years 2005 and 2006 a field program on strawberries was conducted in Belgium, France (Northern and Southern Region), Germany, Greece, Italy, the Netherlands, Spain and the United Kingdom under open field and glasshouse conditions to investigate the residue behaviour of alpha-cypermethrin after application of a 100 g a.s./L emulsifiable concentrate formulation (BAS 310 40 I).

During the 2005 growing season, eight trials were conducted in strawberries under open field conditions, four of them in the Northern European region (in Belgium, France, Germany and the United Kingdom) and four in the Southern European region (in France, Italy and Spain).
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I), was foliar applied to strawberry plants in several variants. In four trials located in the Northern European region, the product was applied once at target rate of 12.5 g a.s./ha. This was compared to a second variant in which two applications were made at the same target application rate.
The last application took place at growth stages between 75 (50% of fruit have reached final size) and 89 (second harvest: more fruits coloured).
In four trials located in the Southern European region, a third variant was assayed in which BAS 310 40 I was applied to strawberry plants once at a target rate of 25 g a.s./ha at growth stages between 85 (first fruits have cultivar-specific colour) and 89 (second harvest: more fruits coloured).
Strawberry fruit specimens were collected directly after the last application as well as 1-2, 3 and 6-7 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF method No. 567/0, which has a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2006 growing season, eight trials were conducted in strawberries under open field conditions, four of them in the Northern European region (in France, Germany, the Netherlands and the United Kingdom) and four in the Southern European region (in France, Greece, Italy and Spain).
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I) was foliar applied to strawberry plants in several variants. In four trials located in the Northern European region, the product was applied once at target rate of 12.5 g a.s./ha . This was compared to a second variant in which two applications were made at the same target application rate.
The last application took place at growth stages between 85 (first fruits have cultivar-specific colour) and 87 (main harvest: more fruits coloured).
In four trials located in the Southern European region, a third variant was assayed in which BAS 310 40 I was applied to strawberry plants once at a target rate of 25 g a.s./ha at growth stages between 85 and 87.
Strawberry fruit specimens were collected directly after the last application as well as 1, 2-4 and 6-8 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF method No. 567/0, which has a limit of quantification of 0.01 mg/kg in all sample materials.

Eight glasshouse trials on strawberries were performed during the growing season 2005 in Belgium, France (Northern and Southern Region), Germany, Greece, Italy and Spain.
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I), was foliar applied to strawberry plants at a target rate of 40 g a.s./ha. The growth stages of the plants were between 84 and 89 (second harvest: more fruits coloured).
Strawberry fruit specimens were collected directly after the last application as well as 3 and 7 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF method No. 567/0, which has a limit of quantification of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.2.1-1 and Table 2.2.1-2.

Table 2.2.1-1: Residues in strawberry – open field trials

GAP for EU-N is 2 applications with 12.5 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
GAP for EU-S is 1 applications with 30 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
Indoor GAP is 1 application at 50 g as/ha at infestation as an overall spray, PHI 3

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008489 Trial No. A/BE/I/05/88 Study to GLP Study carried out in 2005	Strawberry (variety Elsanta)	Belgium Nivelles Brabant	13 g as/ha 16.08.05	89	0 1 3 7	fruit <0.01 fruit 0.010 fruit <0.01 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 96.7%; SD: +/- 18.3; CV: 19.0%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008489 Trial No. A/GE/I/05/81 Study to GLP Study carried out in 2005	Strawberry (variety Chandler)	Germany Ladenburg Baden-Württemberg	12 g as/ha 15.06.05	87-89	0 1 3 7	fruit 0.015 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008489 Trial No. A/NF/I/05/80 Study to GLP Study carried out in 2005	Strawberry (variety Cirafine)	France Fontaines-en-Sologne Centre (North of EU)	13 g as/ha 12.07.05	89	0 1 3 6	fruit 0.032 fruit 0.029 fruit 0.022 fruit 0.017	
BASF Doc ID 2007/1008489 Trial No. A/UK/I/05/83 Study to GLP Study carried out in 2005	Strawberry (variety Florence)	United Kingdom Ledbury Herefordshire	12 g as/ha 27.06.05	75-81	0 1 3 7	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/NF/I/06/88 Study to GLP Study carried out in 2006	Strawberry (variety Florence)	France Vraux Marne (North of EU)	12 g as/ha 16.06.06	85	0 1 3 7	fruit 0.011 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.1%; SD: +/- 3.9; CV: 4.0%; n=5; fortification range 0.01 – 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008493 Trial No. A/GE/I/06/89 Study to GLP Study carried out in 2006	Strawberry (variety Florence)	Germany Schmilau Schleswig-Holstein	13 g as/ha 30.06.06	86	0 1 2 8	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/NL/I/06/90 Study to GLP Study carried out in 2006	Strawberry (variety Elsanta)	The Netherlands AK Nymegen Gelderland	13 g as/ha 07.08.06	87	0 1 4 8	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/UK/I/06/91 Study to GLP Study carried out in 2006	Strawberry (variety Pegasus)	United Kingdom Harvington Evesham Worcestershire	14 g as/ha 20.06.06	85	0 1 3 7	fruit 0.012 fruit 0.012 fruit <0.01 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.1%; SD: +/- 3.9; CV: 4.0%; n=5; fortification range 0.01 – 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008489 Trial No. A/BE/I/05/88 Study to GLP Study carried out in 2005	Strawberry (variety Elsanta)	Belgium Nivelles Brabant	13 g as/ha 2 treatm. last date 16.08.05	89 at last treatm.	0 1 3 7	fruit 0.012 fruit 0.012 fruit <0.01 fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 96.7%; SD: +/- 18.3; CV: 19.0%; n=8; fortification range 0.01 – 0.1 mg/kg

Table 2.2.1-1: Residues in strawberry – open field trials

GAP for EU-N is 2 applications with 12.5 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
GAP for EU-S is 1 applications with 30 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
Indoor GAP is 1 application at 50 g as/ha at infestation as an overall spray, PHI 3

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008489 Trial No. A/GE/I/05/81 Study to GLP Study carried out in 2005	Strawberry (variety Chandler)	Germany Ladenburg Baden-Württemberg	12 g as/ha 2 treatm. last date 15.06.05	87-89 at last treatm.	0 1 3 7	fruit 0.018 fruit <0.01 fruit <u><0.01</u> fruit <0.01	Residue analysed as total cypermethrin BASF analytical method N 567/0 fruit: mean recovery = 96.7%; SD: +/- 18.3; CV: 19.0%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008489 Trial No. A/NF/I/05/80 Study to GLP Study carried out in 2005	Strawberry (variety Cirafine)	France Fontaines-en-Sologne Centre (North of EU)	13 g as/ha 2 treatm. last date 12.07.05	89 at last treatm.	0 1 3 6	fruit 0.049 fruit 0.042 fruit <u>0.031</u> fruit 0.020	
BASF Doc ID 2007/1008489 Trial No. A/UK/I/05/83 Study to GLP Study carried out in 2005	Strawberry (variety Florence)	United Kingdom Ledbury Herefordshire	12/13 g as/ha 2 treatm. last date 27.06.05	75-81 at last treatm.	0 1 3 7	fruit <0.01 fruit <0.01 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/NF/I/06/88 Study to GLP Study carried out in 2006	Strawberry (variety Florence)	France Vraux Marne (North of EU)	14/12 g as/ha 2 treatm. last date 16.06.06	85 at last treatm.	0 1 3 7	fruit 0.018 fruit 0.017 fruit <u><0.01</u> fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.1%; SD: +/- 3.9; CV: 4.0%; n=5; fortification range 0.01 – 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008493 Trial No. A/GE/I/06/89 Study to GLP Study carried out in 2006	Strawberry (variety Florence)	Germany Schmilau Schleswig-Holstein	12/ 13 g as/ha 2 treatm. last date 30.06.06	86 at last treatm.	0 1 2 8	fruit <0.01 fruit <0.01 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/NL/I/06/90 Study to GLP Study carried out in 2006	Strawberry (variety Elsanta)	The Netherlands AK Nymegen Gelderland	13 g as/ha 2 treatm. last date 07.08.06	87 at last treatm.	0 1 4 8	fruit <0.01 fruit <0.01 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/UK/I/06/91 Study to GLP Study carried out in 2006	Strawberry (variety Pegasus)	United Kingdom Harvington Evesham Worcestershire	13/ 14 g as/ha 2 treatm. last date 20.06.06	85 at last treatm.	0 1 3 7	fruit 0.020 fruit 0.027 fruit 0.018 fruit <u>0.023</u>	BASF analytical method N 567/0 fruit: mean recovery = 98.1%; SD: +/- 3.9; CV: 4.0%; n=5; fortification range 0.01 – 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008489 Trial No. A/IT/I/05/86 Study to GLP Study carried out in 2005	Strawberry	Italy Gavi Ligure Piemont	24 g as/ha 12.06.05	86-87	0 2 3 7	fruit 0.015 fruit <0.01 fruit <u><0.01</u> fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 96.7%; SD: +/- 18.3; CV: 19.0%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008489 Trial No. A/IT/I/05/87 Study to GLP Study carried out in 2005	Strawberry (variety Peco)	Italy S. Stefano Roero Piemont	28 g as/ha 08.06.05	85-86	0 1 3 7	fruit 0.038 fruit <u>0.028</u> not available fruit 0.022	BASF analytical method N 567/0 fruit: mean recovery = 96.7%; SD: +/- 18.3; CV: 19.0%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total

Table 2.2.1-1: Residues in strawberry – open field trials

GAP for EU-N is 2 applications with 12.5 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
GAP for EU-S is 1 applications with 30 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
Indoor GAP is 1 application at 50 g as/ha at infestation as an overall spray, PHI 3

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008489 Trial No. A/SF/I/05/84 Study to GLP Study carried out in 2005	Strawberry (variety Garigette)	France Feugarolles Lot et Garonne (South of EU)	26 g as/ha 07.06.05	89	0 1 3 7	fruit 0.012 fruit 0.011 fruit <u>≤0.01</u> fruit <0.01	cypermethrin BASF analytical method N 567/0 fruit: mean recovery = 96.7%; SD: +/- 18.3; CV: 19.0%; n=8; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008489 Trial No. A/SP/I/05/85 Study to GLP Study carried out in 2005	Strawberry (variety Plantafrigo)	Spain Quatretonda Valencia	26 g as/ha 03.05.05	87	0 1 3 7	fruit <0.01 fruit <0.01 fruit <u>≤0.01</u> fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/SF/I/06/92 Study to GLP Study carried out in 2006	Strawberry (variety Cleret)	France Lansargue Hérault (South of EU)	26 g as/ha 20.05.06	22	0 1 3 7	fruit 0.015 fruit 0.030 fruit <u>0.018</u> fruit <0.01	BASF analytical method N 567/0 fruit: mean recovery = 98.1%; SD: +/- 3.9; CV: 4.0%; n=5; fortification range 0.01 – 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008493 Trial No. A/SP/I/06/93 Study to GLP Study carried out in 2006	Strawberry (variety camarosa)	Spain Qauatretonda Valencia	25 g as/ha 16.05.06	87	0 1 3 8	fruit 0.026 fruit 0.017 fruit <u>0.017</u> fruit 0.015	
BASF Doc ID 2007/1008493 Trial No. A/GR/I/06/95 Study to GLP Study carried out in 2006	Strawberry (variety Aroma)	Greece Suoronos Central Macedonia	25 g as/ha 02.06.06	85	0 1 3 6	fruit <0.01 fruit <0.01 fruit <u>≤0.01</u> fruit <0.01	
BASF Doc ID 2007/1008493 Trial No. A/IT/I/06/94 Study to GLP Study carried out in 2006	Strawberry (variety Marmohede)	Italy S. Stefano Roero Piemont	27 g as/ha 10.06.06	86	0 1 3 7	fruit 0.016 fruit 0.019 fruit <u>≤0.01</u> fruit <0.01	

_ underlined values were used for MRL calculation

Table 2.2.1-2: Residues in strawberry – glasshouse trials

GAP for EU-N is 2 applications with 12.5 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
GAP for EU-S is 1 applications with 30 g as/ha at infestation as an overall spray, PHI 3 (outdoor)
Indoor GAP is 1 application at 50 g as/ha at infestation as an overall spray, PHI 3

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007935 Trial No. A/BE/I/05/78 Study to GLP Study carried out in 2005	Strawberry (variety Darselect)	Belgium Nivelles Brabant	39.47 g as/ha 06.05.06 Indoor	85-87	0 3 7	fruit 0.047 fruit <u>0.040</u> fruit 0.023	BASF analytical method N 567/0 fruit: mean recovery = 95.6%; SD: +/- 11,7; CV: 12.2%; n=10; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007935 Trial No. A/GE/I/05/72 Study to GLP Study carried out in 2005	Strawberry (variety Avanta)	Germany Ladenburg Baden-Württemberg	37.40 g as/ha 04.05.05 Indoor	85-87	0 3 7	fruit <0.01 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1007935 Trial No. A/GE/I/05/73 Study to GLP Study carried out in 2005	Strawberry (variety Rosella)	Germany Schmilau Schleswig-Holstein	38.70 g as/ha 18.05.05 Indoor	85-89	0 3 7	fruit 0.012 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1007935 Trial No. A/GR/I/05/77 Study to GLP Study carried out in 2005	Strawberry (variety Aroma)	Greece Svoronos Central Macedonia	39.42 g as/ha 19.08.05 Indoor	87	0 3 7	fruit 0.060 fruit <u>0.048</u> fruit 0.035	
BASF Doc ID 2007/1007935 Trial No. A/IT/I/05/76 Study to GLP Study carried out in 2005	Strawberry (variety Alba)	Italy S. Stefano Roero Piemont	45.33 g as/ha 16.05.05 Indoor	84-86	0 3 7	fruit 0.037 fruit <u>0.029</u> fruit 0.015	
BASF Doc ID 2007/1007935 Trial No. A/NF/I/05/70 Study to GLP Study carried out in 2005	Strawberry (variety Cirafine)	France Fontaines-en-Sologne Centre	42.69 g as/ha 03.06.05 Indoor	89	0 3 7	fruit 0.056 fruit <u>0.054</u> fruit 0.041	
BASF Doc ID 2007/1007935 Trial No. A/SF/I/05/71 Study to GLP Study carried out in 2005	Strawberry (variety Darselect)	France Feugarolles Lot et Garonne	39.70 g as/ha 07.06.05 Indoor	87	0 3 7	fruit <0.01 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1007935 Trial No. A/SP/I/05/75 Study to GLP Study carried out in 2005	Strawberry (variety Ventana)	Spain Lloc Nou Del Fenollet Valencia	40.00 g as/ha 06.05.05 Indoor	87	0 3 7	fruit 0.013 fruit <u><0.01</u> fruit <0.01	

_ underlined values were used for MRL calculation

Findings:
The residue studies in strawberries presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 2 applications with 12.5 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 3 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 3 days
- glasshouse GAP is a single application with 50 g a.s./ha with a PHI of 3 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions - 8 trials including both variants, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions - 8 trials, all conducted in the Southern European region
- one treatment at a target rate of 40 g a.s./ha under glasshouse conditions glasshouse - 8 trials, 4 conducted in the Northern European region and 4 conducted in the Southern European region

After one application at a target rate of 12.5 g a.s./ha under open field conditions, initial residues in strawberry fruit ranged between <0.01 mg/kg and 0.032 mg/kg and declined to <0.01 – 0.029 mg/kg and <0.01 – 0.022 mg/kg at 1 and 2-4 days after application, respectively. At the last sampling date, 6-8 days after application, only in one treated sample residues of 0.017 mg/kg were found (trial A/NF/I/05/80), while all other treated samples taken at the same sampling interval did not show residues above the limit of quantification (LOQ) of the analytical method (all <0.01).
After two applications at a target rate of 12.5 g a.s./ha under open field conditions, initial residues in strawberry fruit ranged between <0.01 mg/kg and 0.049 mg/kg and declined to <0.01 – 0.042 mg/kg, <0.01 – 0.031 mg/kg and <0.01 – 0.023 mg/kg at 1, 2-4 and 6-8 days after application, respectively.
After one application at a target rate of 25 g a.s./ha under open field conditions, initial residues in strawberry fruit ranged between <0.01 mg/kg and 0.038 mg/kg and declined to <0.01 – 0.030 mg/kg, <0.01 – 0.018 mg/kg and <0.01 – 0.022 mg/kg at 1-2, 3 and 6-8 days after application, respectively.
Under glasshouse conditions, initial residues between <0.01 mg/kg and 0.060 mg/kg were found in strawberry fruit after a single application of 40 g alpha-cypermethrin/ha. Residues declined to <0.01 – 0.054 mg/kg and <0.01 – 0.041 mg/kg 3 and 7 days after application, respectively.

Conclusion:

After treatment of strawberry plants with alpha-cypermethrin under open field conditions according to the proposed GAP, residues in strawberry fruit harvested at the target PHI were not higher than 0.05 mg/kg.

After treatment of strawberry plants with alpha-cypermethrin according to the GAP for glasshouse use, residues below 0.1 mg/kg can be expected in strawberry fruit.

2.2.2 Estimation of MRL, HR and STMR for strawberry

For *strawberries*, the following residue studies were considered (BASF DocIDs): 2007/1008489, 2007/1007935 and 2007/1008493.

The following residue values (PHI=3±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2007/1008489 (open field)	<0.01 (3x), 0.031	<0.01 (3x), 0.028
2007/1007935 (glasshouse)	<0.01 (4x), 0.029, 0.04, 0.048, 0.054	-
2007/1008493 (open field)	<0.01 (3x), 0.023	<0.01 (2x), 0.017, 0.018
OECD-MRL-calculation (open field)	**0.05 (n=8, STMR=0.01, HR=0.031)**	**0.04 (n=8, STMR=0.01, HR=0.028)**
OECD-MRL-calculation (glasshouse)	**<u>0.1</u> (n=8, STMR=0.02, HR=0.054)**	-

_ underlined values were used for risk assessment purposes

2.3 Olive

Residue data from supervised trials in olives were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.3-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Olive	2003	-	-	4	ES, GR	4	2005/1004975
Olive	2004	-	-	4	ES, GR	4	2005/1007582
Olive	2011	-	-	2	ITA, ESP	2	BASF DocID 2012/1157548
Total number of trials per region		-	-	**6**	**Total number of trials**	**10**	

Table 2.3-2: Processing studies available for olive

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Olive	1998	-	-	1	GR	1	Al-714-001
Olive	1998	-	-	2	GR	2	AL-714-002
Olive	2004	-	-	4	ES	4	2007/1009604
Total number of trials per region					**Total number of trials**	**4**	

2.3.1 Supervised residue trials in olive

Perny A (2005)

Full study reference

Perny A (2005): Study on the residue behaviour of alphacypermethrin (BAS 310 I) on olives after application of BAS 310 11 I under field conditions in Greece and Spain, 2003; BASF Doc-ID 2005/1004975

Schroth E (2005)

Full study reference

Schroth E (2005): Study on the residue behavior of Alpha-cypermethrin on olives after application of BAS 31041 I under field conditions in Spain and Greece, 2004; BASF Doc-ID 2005/1007582

Material and Methods:

In the years 2003 and 2004 a field program on olives was conducted in Greece and Spain.

During the growing season 2003, four trials were performed. An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L, BAS 310 11 I) was foliar applied to olive trees in several variants. In one variant, the product was applied once at target rate of 15 g a.s./ha . This was compared to a second variant in which two applications were made at the same target application rate.
The last application took place at growth stages between 78 (80% of fruits have reached final size) and 80 (fruit deep green colour becomes light green, yellowish.).
Olive fruit specimens were collected directly after the last application as well as 3-4, 6-8 and 14-15 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF method No. 546/0, which has a limit of quantification of 0.05 mg/kg in olives.

During the growing season 2004, four trials were performed. A soluble concentrate formulation of alpha-cypermethrin (100 g a.s./L, BAS 310 41 I) was foliar applied to olive trees in several variants. In one variant, the product was applied once at target rate of 15 g a.s./ha . This was compared to a second variant in which two applications were made at the same target application rate.
The last application took place at growth stages between 78 and 79 (fruit size about 90 % of final size; fruit suitable for picking green olives).
Olive fruit specimens were collected directly after the last application as well as 3, 7 and 13-14 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF method No. 567/0, which has a limit of quantification of 0.01 mg/kg in olives.

The trial data and residue results are summarized in Table 2.3.1-1.

Table 2.3.1-1: Residues in olives

GAP for EU-S is 2 applications at 15 g a.s./ha, PHI = 7 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1004975 Trial No. ALO/16/03 Study to GLP Study carried out in 2003	Olive (variety Manzanilla)	Spain 41500 Alcala Sevilla Andalucia	15 g a.s./ha 29.08.03	78	0 3 6 14	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	BASF analytical method No. 546/0: fruit: mean recovery = 84.3%; SD: +/- 7.4; CV: 8.7%; n=4; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1004975 Trial No. ALO/17/03 Study to GLP Study carried out in 2003	Olive (variety Manzanilla)	Spain 41600 Arahal Sevilla Andalucia	15 g a.s./ha 29.08.03	78	0 3 6 14	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	
BASF Doc ID 2005/1004975 Trial No. GRE/11/03 Study to GLP Study carried out in 2003	Olive (variety Koroneiki)	Greece 60063 Leptokaria Northern Greece - Macedonia	15 g a.s./ha 17.10.03	80	0 3 7 14	fruit <0.05 fruit <0.05 fruit 0.066 fruit <0.05	
BASF Doc ID 2005/1004975 Trial No. GRE/12/03 Study to GLP Study carried out in 2003	Olive (variety Amfisis)	Greece 59100 Veria Northern Greece - Macedonia	15 g a.s./ha 10.10.03	80	0 4 8 15	fruit <0.05 fruit <0.05 fruit <0.05 fruit <0.05	
BASF Doc ID 2005/1007582 Trial No. ALO/23/04 Study to GLP Study carried out in 2004	Olive (variety Manzanilla)	Spain E-41500 Alcalá de Guadaira Andalucia	15 g a.s./ha 27.08.04	78	0 3 7 13	fruit 0.01 fruit 0.02 fruit 0.02 fruit 0.02	BASF analytical method No. 567/0: fruit: mean recovery = 78.1%; SD: +/- 10.9; CV: 14.0%; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007582 Trial No. ALO/24/04 Study to GLP Study carried out in 2004	Olive (variety Manzanilla)	Spain E-41600 Arahal, Andalucia	15 g a.s./ha 27.08.04	78	0 3 7 13	fruit 0.09 fruit 0.10 fruit 0.09 fruit 0.06	
BASF Doc ID 2005/1007582 Trial No. GRE/14/04 Study to GLP Study carried out in 2004	Olive (variety Halkidikis)	Greece G-59100 Imathia Veria Macedonia	15 g a.s./ha 20.09.04	79	0 3 7 14	fruit 0.02 fruit 0.02 fruit 0.05 fruit 0.01	
BASF Doc ID 2005/1007582 Trial No. GRE/15/04 Study to GLP Study carried out in 2004	Olive (variety Halkidikis)	Greece G-591 00 Veroia, Veroia Macedonia	15 g a.s./ha 29.09.04	79	0 3 7 14	fruit 0.02 fruit <0.01 fruit <0.01 fruit <0.01	

Table 2.3.1-1: Residues in olives

GAP for EU-S is 2 applications at 15 g a.s./ha, PHI = 7 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1004975 Trial No. ALO/16/03 Study to GLP Study carried out in 2003	Olive (variety Manzanilla)	Spain 41500 Alcaia Andalucia	15 g a.s./ha 2 treatm. last date 29.08.03	78 at last treatm.	0 3 6 14	fruit <0.05 fruit <0.05 fruit <u><0.05</u> fruit <0.05	BASF analytical method No. 546/0: fruit: mean recovery = 84.3%; SD: +/- 7.4; CV: 8.7%; n=4; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1004975 Trial No. ALO/17/03 Study to GLP Study carried out in 2003	Olive (variety Manzanilla)	Spain 41600 Arahal Sevilla Andalucia	15 g a.s./ha 2 treatm. last date 29.08.03	78 at last treatm.	0 3 6 14	fruit <0.05 fruit <0.05 fruit <u><0.05</u> fruit <0.05	
BASF Doc ID 2005/1004975 Trial No. GRE/11/03 Study to GLP Study carried out in 2003	Olive (variety Koroneiki)	Greece 60063 Leptokaria Northern Greece - Macedonia	15 g a.s./ha 2 treatm. last date 17.10.03	80 at last treatm.	0 3 7 14	fruit <0.05 fruit <0.05 fruit <u><0.05</u> fruit <0.05	BASF analytical method No. 546/0: fruit: mean recovery = 84.3%; SD: +/- 7.4; CV: 8.7%; n=4; fortification range 0.05 – 0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1004975 Trial No. GRE/12/03 Study to GLP Study carried out in 2003	Olive (variety Amfisis)	Greece 59100 Veria Northern Greece - Macedonia	15 g a.s./ha 2 treatm. last date 10.10.03	80 at last treatm.	0 4 8 15	fruit <0.05 fruit <0.05 fruit <u><0.05</u> fruit <0.05	
BASF Doc ID 2005/1007582 Trial No. ALO/23/04 Study to GLP Study carried out in 2004	Olive (variety Manzanilla)	Spain E-41500 Alcalá de Guadaira Andalucia	15 g a.s./ha 2 treatm. last date 27.08.04	78 at last treatment	0 3 7 13	fruit 0.04 fruit 0.04 fruit <u>0.04</u> fruit 0.04	BASF analytical method No. 567/0: fruit: mean recovery = 78.1%; SD: +/- 10.9; CV: 14.0%; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007582 Trial No. ALO/24/04 Study to GLP Study carried out in 2004	Olive (variety Manzanilla)	Spain E-41600 Arahal, Andalucia	15 g a.s./ha 2 treatm. last date 27.08.04	78 at last treatment	0 3 7 13	fruit 0.16 fruit 0.08 fruit <u>0.09</u> fruit 0.07	
BASF Doc ID 2005/1007582 Trial No. GRE/14/04 Study to GLP Study carried out in 2004	Olive (variety Halkidikis)	Greece G-59100 Imathia Veria Macedonia	15 g a.s./ha 2 treatm. last date 20.09.04	79 at last treatment	0 3 7 14	fruit 0.02 fruit 0.04 fruit <u>0.04</u> fruit 0.02	
BASF Doc ID 2005/1007582 Trial No. GRE/15/04 Study to GLP Study carried out in 2004	Olive (variety Halkidikis)	Greece G-591 00 Veroia, Veroia Macedonia	15 g a.s./ha 2 treatm. last date 29.09.04	79 at last treatment	0 3 7 14	fruit 0.03 fruit <0.01 fruit <u><0.01</u> fruit <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in olives presented in the alpha-cypermethrin dossier were carried out in 2 different EU countries and provide data relevant to conditions in the Southern European region.

The following residue studies were presented:

- one or two treatments at a target rate of 15 g a.s./ha under open field conditions - 8 trials including both variants, all conducted in the Southern European region

The trials conducted during the growing season 2003 were analysed with a LOQ of 0.05 mg/kg, while the analytical method applied in the 2004 study had a LOQ of 0.01 mg/kg.

After one application at a target rate of 15 g a.s./ha, initial residues in olive fruit ranged between 0.01 mg/kg and 0.09 mg/kg and were <0.01 – 0.10 mg/kg, <0.01 – 0.09 mg/kg and <0.01 – 0.06 mg/kg at 3-4, 6-8 and 14-15 days after application, respectively.

After two applications at a target rate of 15 g a.s./ha, initial residues in olive fruit ranged between 0.02 mg/kg and 0.16 mg/kg and declined to <0.01 – 0.08 mg/kg, <0.01 – 0.09 mg/kg and <0.01 – 0.07 mg/kg at 3, 7 and 13-14 days after application, respectively.

Conclusion:

After two treatments with BAS 310 I at a target rate of 15 g a.s./ha, residues ranged between <0.01 – 0.09 mg/kg at 7 days after the last application.

Report:	**Moreno S., 2013a**
Title:	Study on the residue behaviour of Alpha-Cypermethrin in olive after treatment with either BAS 310 40 I or BAS 310 55 I under field conditions in South Europe, season 2011
Document No:	BASF DocID 2012/1157548
Guidelines:	EEC 87/18 (18 December 1986), International guidelines for distribution and pesticides application AEPLA FAO 1985, EC 1107/2009 of the European Parliament and of the Council of 21 Oct 2009, EEC 79/117, EEC 91/414, EEC 7029/VI/95 rev. 5 Appendix B, EEC 7525/VI/95 rev. 9 (March 2011)
GLP	yes

Executive Summary

During the 2011 growing season a total of two trials (L110427 and L110428) were conducted in representative olive growing areas in Spain and Italy, in order to determine the residue level of alpha-cypermethrin after application of either BAS 310 40 I or BAS 310 55 I.
The test item BAS 310 40 I (100 g a.s./L BAS 310 I, EC) was foliar applied twice on plot 3, and the test item BAS 310 55 I (50 g a.s./L BAS 310 I, ME) was foliar applied twice on plot 2. Both of them were applied at 14±1 and 7±1 days before harvest and at a nominal rate of 15 g alpha-cypermethrin/ha in a nominal spray volume of 1000 l/ha using backpack spraying equipment.
Plot 4 was treated with a mixture of the test item BAS 310 55 I and a commercial bait applied at a concentration of 2%. The spray mixture was foliar applied twice at 14±1 and 7±1 days before harvest at a nominal rate of 9 g alpha-cypermethrin/ha in a low spray volume of 30 l/ha using backpack spraying equipment. Olive fruits were taken directly after the last application as well as 3, 7 and 14 days thereafter.
Specimens were analysed for alpha-cypermethrin using BASF method No. 567/0. The method has a limit of quantitation of 0.01 mg/kg.
Directly after the last of two applications of either BAS 310 55 I or BAS 310 40 I in a spray volume of 1000 L/ha, the residue of alpha-cypermethrin ranged between 0.03-0.16 mg/kg in olive fruit specimens. At 7±1 days, residues found were 0.08-0.19 mg/kg.
The analytical results obtained demonstrate that the treatment with two applications of BAS 310 55 I and BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.
Directly after the last of two applications of BAS 310 55 I with 2% bait in a spray volume of 30 L/ha the residue of alpha-cypermethrin ranged between 0.08-0.47 mg/kg in olive fruit specimens. At 7±1 days, residues found were 0.07-0.26 mg/kg.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 55 I (ME)
BAS 310 40 I (EC)
Lot/Batch #: 101198 BAS 310 55 I, 50 g/L alpha-cypermethrin
1209 BAS 310 40 I, 100 g/L alpha-cypermethrin
Purity:
CAS#: 67375-30-8 alpha-cypermethrin
Development code:
Spiking levels: 0.01-5.0 mg/kg

2. Test Commodity:

Crop: Olive
Type: Miscellaneous fruit
Variety: Picual, Cima di Melfi
Botanical name: *Olea europaea*
Crop parts(s) or processed
Commodity: Fruit
Sample size: 1.050-2.6 kg

B. STUDY DESIGN

1. Test procedure

Two residue trials in olive (L110427 and L110428) were conducted during the growing season 2011 in a representative olive growing area in Spain and Italy, respectively, to determine the residue level of alpha-cypermethrin after application of either BAS 310 40 I or BAS 310 55 I.
Both trials had a single untreated plot (Plot 1) and three treated plots (Plot 2, Plot 3, and Plot 4).
The test item BAS 310 40 I (100 g/L BAS 310 I, EC) was foliar applied twice on plot 3, and the test item BAS 310 55 I (50 g/L BAS 310 I, ME) was foliar applied twice on plot 2. Both of them were applied at 14±1 and 7±1 days before harvest and at a nominal rate of 15 g alpha-cypermethrin/ha in a nominal spray volume of 1000 l/ha using backpack spraying equipment.
Plot 4 was treated with a very low water volume of 30 l/ha and together with commercially available bait (mixing the product with water and the bait (2%)). Commercial bait for trial L110427 was named Hydrolyzed proteins 30% and was added to the mixture with BAS 310 55 I at a rate of 2%. Commercial bait for trial L110428 was named NUBAIT and was added to the mixture with BAS 310 55 I at a rate of 2%. The spray mixture was foliar applied twice at 14±1 and 7±1 days before harvest at a nominal rate of 9 g alpha-cypermethrin/ha in a low spray volume of 30 l/ha using backpack spraying equipment.

Olive fruits were taken directly after the last application as well as 3, 7 and 14 days thereafter. Olives collected 7 and 14 DALA were separated into flesh and stones and individual weights were recorded.

Table 2.3.1-2: Target application rates and timings for olive

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2011	2	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.015	1000	1st appl.: 14±1 DBH 2nd appl.: 7±1 DBH
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.015	1000	1st appl.: 14±1 DBH 2nd appl.: 7±1 DBH
		2	F	BAS 310 55 I (ME) + 2% bait	alpha-cypermethrin	0.009	30	1st appl.: 14±1 DBH 2nd appl.: 7±1 DBH

DBH: days before harvest

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).
The residues of alpha-cypermethrin in olive specimens were extracted with acetonitrile/n-hexane. After centrifugation and a further partitioning step with n-hexane, the acetonitrile phase was cleaned up by solid-phase extraction on silica gel phase. Alpha-cypermethrin was eluted with dichloromethane. The solvent was evaporated, the residue was taken up into acetonitrile and diluted with acetonitrile/water (80:20, v/v) for LC/MS/MS determination. Final determination was performed by LC/MS/MS using the ammonium adduct of alpha-cypermethrin.
The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery in olive fruit averaged 77±12% (mean ± RSD) for alpha-cypermethrin at fortification levels of 0.01 and 5.0 mg/kg.

Table 2.3.1-3: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in olive fruit

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
Method No. 567/0		alpha-cypermethrin		
Olive fruit	0.01	6	76	12
	5.0	8	78	13

II. RESULTS AND DISCUSSION

The residue levels of alpha-cypermethrin (BAS 310 I) in olive specimens taken directly after the last application (0 DALA) of BAS 310 55 I at 0.015 kg a.s./ha ranged between 0.03-0.15 mg/kg. In the specimens taken 3 DALA 0.08-0.10 mg/kg were found. The residues remained at this level in the PHI specimens (7 DALA) (0.08-0.13 mg/kg). At the last sampling (14 DALA) 0.06-0.09 mg/kg were found.

Residues found in olive specimens taken directly after the last application (0 DALA) of BAS 310 40 I at 0.015 kg a.s./ha contained residues of BAS 310 I in a range of 0.12 mg/kg to 0.16 mg/kg. After 3 days (3 DALA) residues ranged between 0.10-0.17 mg/kg. At 7 DALA 0.14-0.19 mg/kg were found. At the last sampling (14 DALA) the residue level lay at 0.12-0.13 mg/kg).

After the last treatment with BAS 310 55 I with 2% bait at a rate of 0.009 kg a.s.//ha, initial residues (0 DALA) ranged between 0.08-0.47 mg/kg which declined to 0.05-0.07 mg/kg after 3 days (3 DALA). At 7 days (7 DALA), residues between 0.07-0.26 mg/kg were detected. 14 days after the last treatment (14 DALA), residues ranged between 0.04-0.21 mg/kg.

In untreated samples residues of alpha-cypermethrin were not detectable.

An overall summary of the residues is given in the table below.

Table 2.3.1-4: Summary of residues of BAS 310 I in olive after application of BAS 310 55 I and BAS 310 40 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	alpha-cypermethrin (BAS 310 I)
Olive / fruit	2011	2	BAS 310 55 I (ME) at 0.015 kg a.s./ha	0	85-88	fruit	0.03 – 0.15
				3	88-89		0.08 – 0.10
				7	88-89		0.08 – 0.13
				14	89		0.06 – 0.09
		2	BAS 310 40 I (EC) at 0.015 kg a.s./ha	0	85-88	fruit	0.12 – 0.16
				3	88-89		0.10 – 0.17
				7	88-89		0.14 – 0.19
				14	89		0.12 – 0.13
		2	BAS 310 55 I at 0.009 kg a.s./ha + commercial bait (2%)	0	85-88	fruit	0.08 – 0.47
				3	88-89		0.05 – 0.07
				7	88-89		0.07 – 0.26
				14	89		0.04 – 0.21

DALA = days after last application
BBCH = growth stage at respective sampling

III. CONCLUSION

Directly after the last of two applications of either BAS 310 55 I or BAS 310 40 I at a rate of 0.015 kg a.s./ha, the residues of alpha-cypermethrin ranged between 0.03-0.16 mg/kg in olive fruit specimens. At 7±1 days, residues found were 0.08-0.19 mg/kg.
The analytical results obtained demonstrate that the treatment with two applications of BAS 310 55 I and BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.
Directly after the last of two applications of BAS 310 55 I at a rate of 0.009 kg a.s./ha with 2% bait the residue of alpha-cypermethrin ranged between 0.08-0.47 mg/kg in olive fruit specimens. At 7±1 days, residues found were 0.07-0.26 mg/kg.

Table 2.3.1-5: Residues of BAS 310 I after two applications of the formulations BAS 310 55 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408605 **Doc ID:**2012/1157548 **Trial No.:**L110427 **GLP:**yes **Year**2011	Olive	Spain	BAS 310 55 I BAS 310 I 2 x 0.015	85-88	0	fruit	0.15
				88-89	3	fruit	0.10
				88-89	7	fruit	0.08
				89	14	fruit	0.06
			BAS 310 40 I BAS 310 I 2 x 0.015	85-88	0	fruit	0.16
				88-89	3	fruit	0.17
				88-89	7	fruit	0.14
				89	14	fruit	0.12
			BAS 310 55 I BAS 310 I 2 x 0.009 +2% bait	85-88	0	fruit	0.08
				88-89	3	fruit	0.07
				88-89	7	fruit	0.07
				89	14	fruit	0.04
Study code:408605 **Doc ID:**2012/1157548 **Trial No.:**L110428 **GLP:**yes **Year**2011	Olive	Italy	BAS 310 55 I BAS 310 I 2 x 0.015	88	0	fruit	0.03
				88	3	fruit	0.08
				89	7	fruit	0.13
				89	14	fruit	0.09
			BAS 310 40 I BAS 310 I 2 x 0.015	88	0	fruit	0.12
				88	3	fruit	0.10
				89	7	fruit	0.19
				89	14	fruit	0.13
			BAS 310 55 I BAS 310 I 2 x 0.009 +2% bait	88	0	fruit	0.47
				88	3	fruit	0.05
				89	7	fruit	0.26
				89	14	fruit	0.21

DALA = days after last application
BBCH = growth stage at respective sampling

2.3.2 Processing studies in olive

Report:
Schulz H. 2007d
Study on the Residue Behaviour of Alpha-Cypermethrin in Olives and Processed Products after Treatment with BAS 310 41 I under Field Conditions in Spain, 2004
2007/1009604

Guidelines: EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8),EEC 1607/VI/97 rev. 2 10.06.1999,EEC 7029/VI/95 rev. 5,EEC 7525/VI/95 rev. 7,EEC 7035/VI/95 rev. 5

GLP: yes
(certified by Hessisches Ministerium für Umwelt, Laendlichen Raum und Verbraucherschutz)

Material and methods:

During the 2004 growing season, four field trials were conducted in olives in Spain in order to determine the magnitude and distribution of alpha-cypermethrin residues in the various intermediate and end products after processing.
Two of the trials (ALO/19/04 and ALO/21/04) consisted of two plots, one untreated and one treated; the other two consisted of one treated plot each (ALO/20/04 and ALO/22/04).

A soluble concentrate formulation of alpha-cypermethrin (BAS 310 41 I) was foliar applied 2 times at a target rate of 75 g a.s./ha for each application, resulting in a seasonal target rate of 150 g a.s./ha. Actual rates were within +/10 % of the nominal rate. This exaggerated rate was used in an attempt to generate residue levels sufficiently above the method limit of quantification (LOQ) in the raw agricultural commodity.

The applications were made 21 (± 1) and 7 (± 1) days before the anticipated harvest date using a spray volume of approximately 1000 I/ha. The growth stage at the last application was BBCH 88, i.e. fruit ripening.

Specimens of olives were collected on the day of the last application for analysis and 6-7 days later for analysis of the raw agricultural commodity (RAC) and also for processing. From every trial, 30 kg of olives were harvested to be processed to olive oil and 4 kg to fermented olives (7 days = PHI).

The olives were processed according to the BBA guideline Part IV 3 - 4 dated June 1990. In order to simulate common industrial procedures, the processing was performed in the pilot plant of Pilot Pflanzenöltechnologie Magdeburg e.V. (PPM) in D-39114 Magdeburg.

The following specimens resulted from the processing phases:

- wash water (olives), press cake, press water, oil meal, crude oil, n-hexane phase, slime/water mixture, soap, wash water (refining), filter cake, fatty acids and refined olive oil

- wash water (fermented olives), fermented olives, broth

The specimens were analysed for residues of alpha-cypermethrin using BASF method No. 567/0 for the determination of alpha-cypermethrin. The limit of quantification (LOQ) of the method was 0.01 mg/kg for all specimen materials analysed.

In order to validate the analytical method, control specimens were fortified with alpha-cypermethrin. In total, 33 fortified specimens were analysed. At fortification levels of 0.01, 0.1 and 1.0 mg/kg, the recovery rates averaged at 78.8 %. The standard deviation (SD) was 8.4% and the coefficient of variation (CV) was 10.6%.
The fortified specimens comprised both the olives and the processed products.

Findings:

Residues of alpha-cypermethrin in olives treated with BAS 310 41 I were determined. Specimens were collected immediately after application at growth stage 88 and 6 - 7 days later (7 days = PHI) at BBCH 89 (harvest maturity).

Directly after application, the alpha-cypermethrin residues in the olive specimens ranged from 0.154 to 0.478 mg/kg. They decreased substantially to 0.119 to 0.555 mg/kg in the specimens, sampled at 6 – 7 DAT.

In the olive oil processed specimens, the alpha-cypermethrin residues in wash water were between 0.048 and 0.124 mg/kg. After pressing of the olives, the alpha-cypermethrin residues were only found only in press cake, and no residues above the LOQ were found in press water. After extraction and distillation, the alpha-cypermethrin residues were concentrated in crude oil, whereas only small amounts were observed in oil meal and no residues above the LOQ were found in the extraction solvent n-hexane. In the processed products of the subsequent refining step, high alpha-cypermethrin residues were found in filter cake (0.473 – 1.78 mg/kg) and in the final product olive oil (1.11 – 3.42 mg/kg). In the other refining products, the residues were between < 0.01 mg/kg and 1.03 mg/kg.

In the fermentation processed products, alpha-cypermethrin residues were found in wash water (0.013 – 0.204 mg/kg) and in the consumer product fermented olives (0.190 – 0.607 mg/kg). No residues were observed in broth.

No residues at or above the LOQ were detected in the analysed untreated RAC specimens or in the processed fractions.

The residue levels detected in the treated specimens and processed fractions as well as the calculated transfer factors are presented in the following table. The transfer factors in the olive fruits (RAC) were set as 1.

Table 2.3.2-1: Summary of alpha-cypermethrin residues in olives and transfer factors - DocID 2007/1009604

	Residue Concentration of Alpha-Cypermethrin in mg/kg				Transfer Factors				
	ALO/ 19/04	ALO/ 20/04	ALO/ 21/04	ALO/ 22/04	ALO/ 19/04	ALO/ 20/04	ALO/ 21/04	ALO/ 22/04	mean
Olives (day 0)	0.478	0.369	0.252	0.154	N/A	N/A	N/A	N/A	N/A
Olives (RAC, day 6 - 7)	0.555	0.422	0.119	0.119	1	1	1	1	1
Wash water (olive oil proc.)	0.124	0.060	0.016	0.048	0.22	0.14	0.13	0.40	0.22
Press cake	0.754	0.827	0.403	0.303	1.36	1.96	3.39	2.55	2.32
Press water	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	< 0.02	< 0.08	< 0.08	< 0.05
Oil meal	0.054	0.051	0.028	0.012	0.10	0.12	0.24	0.10	0.14
Crude oil	3.72	3.55	2.09	1.67	6.70	8.41	17.56	14.03	11.68
n-hexane phase	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	< 0.02	< 0.08	< 0.08	< 0.05
Slime/water mixture	0.083	1.03	0.467	0.489	0.15	2.44	3.92	4.11	2.66
Soap	0.022	0.032	0.018	0.030	0.04	0.08	0.15	0.25	0.13
Wash water (refining proc.)	< 0.01	0.067	< 0.01	< 0.01	< 0.02	0.16	< 0.08	< 0.08	< 0.09
Filter cake	1.78	1.31	0.675	0.473	3.21	3.10	5.67	3.97	3.99
Fatty acids	0.080	0.087	0.165	< 0.01	0.14	0.21	1.39	< 0.08	0.46
Refined oil	3.42	3.04	1.52	1.11	6.16	7.20	12.77	9.33	8.87
Wash water (ferment. proc.)	0.204	0.124	0.013	0.082	0.37	0.29	0.11	0.69	0.37
Fermented olives	0.607	0.457	0.237	0.190	1.09	1.08	1.99	1.60	1.44
Broth	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	< 0.02	< 0.08	< 0.08	< 0.05

Procedure for processing of olives to olive oil

Preparation of olives (RAC)

Cleaning

Dirt was removed from the field olives manually and discarded. Only the cleaned olives were used for processing.

Washing

The cleaned olives were washed manually with tap water. An aliquot of the wash water (olives) was taken as a specimen.

Crushing

The washed olives were crushed mechanically using a compression roll equipped with two coarse rollers. During the crushing process, the olive stones were crashed.

Preparation of crude oil

Pressing

The crushed olives were pressed using a platen press up to 280 bar to obtain press cake and oil/water mixture. An aliquot of the press cake was taken as specimen. The oil/water mixture was drained and allowed to separate in a container for 16 to 66 hours to obtain press oil and press water. The press oil was vacuum filtrated via Filtrasit, in order to remove solid particles (exception press oil of the olives of ALO/19/04, T, which was centrifuged). The filter residue and an aliquot of the press water were taken for residue analysis, the rest was discarded. The press oil was taken aside and combined with the extracted oil (see below) to obtain crude oil.

Drying of Press Cake

The press cake was dried in a vacuum drum dryer between room temperature up to 62 °C and between 61 and 140 mbar. The vacuum drying lasted between 1 hour and 1:30 hours.

Extraction and Distillation

The dried press cake was transferred into an extraction apparatus. After filling the solvent tank with 65 L of n-hexane, the heating supply was switched on, the heating supply was switched on (starting point of the extraction) while n-hexane was pumped in circle. After 2 hours, the extraction was stopped and the heating supply switched off.

The miscella was pumped into a 50 L distillation flask. The main part of the n-hexane was distilled off at 41 °C to 49 °C and between 410 mbar and 420 mbar. The oil remained in the distillation flask. The loss of n-hexane was compensated by adding fresh n-hexane into the tank. Thereafter, the second extraction run of the press cake was started. Then, the miscella was pumped into the distillation flask where the main part of n-hexane was distilled off. Finally, the oil phase was drained off, and the remaining n-hexane was removed in several batches using a rotary evaporator. The n-hexane phases were combined and an aliquot was taken as a specimen. The extracted oil and the press oil were combined to obtain crude oil. A specimen was taken for residue analysis. The rest of the crude oil was used for refining.

The extraction residue (oil meal) was transferred into plastic vats and allowed to stand at ambient temperature. The evaporation of n-hexane was supported by an exhaustive device. The weight of the oil meal was determined and a specimen taken for residue analysis. The remaining oil meal was discarded.

Refining

Hydration

About 1500 g of crude oil were heated-up to 60 °C using two infrared lamps under stirring, after which about 150 g of water (60 - 70 °C) were added. The mixture was heated up to about 85 °C and stirred for a further contact time of 45 min. and then allowed to rest until phase separation was achieved. The aqueous phase 1 containing voluminous precipitates was drained off and combined with the aqueous phase 2 obtained from the desliming step.

Desliming

The mixture obtained from hydration was heated up to 85 °C. When 60 °C were reached, between about 6 g of conc. phosphoric acid / water (1/1, v/v) were added drop by drop. At 85 °C, the mixture was stirred for a total time of 45 min. at 350 rpm. Then, about 150 g of water (100 °C) were added and the mixture was allowed to rest until slime separation was achieved. Because of the high phosphatite content, the deslimig step of the treated olives from trial ALO/21/04 was carried out twice. 2 x 151 g of water (100 °C) were added. The aqueous phase 2 was drained off by means of a discharge cock and combined with the aqueous phase 1 of the hydration step to obtain the slime/water specimen.

Neutralization

In order to determine the amount of sodium hydroxide necessary for the neutralization of the oil, the acid value was determined (see de-sliming). The oil was stirred and heated up to 90 °C. Then, a calculated volume of sodium hydroxide solution (7 %, T = ~ 90 °C) were added and the mixture stirred for 20 minutes while the temperature was kept at 90 °C. Subsequently, about 150 g of hot water were added and the stirring continued for a further 5 min. after which the mixture was allowed to cool down and rest for about 18 hours. The aqueous phase “soap” was taken as a specimen.
The acid value was between 0.06 and 0.11.

Washing

The oil phase obtained during neutralization was heated up to about 90 °C while stirred at 250 rpm and about 150 g of water at 100 °C were added. Subsequently, the mixture was stirred continuously for 20 min. and then allowed to rest until phase separation was achieved. Then, the aqueous phase was drained off. The pH value of the aqueous phase was determined and the washing procedure was repeated two to three times until pH 7 was achieved. The aqueous phases were taken as the specimen “wash water (refining)”.

Drying

The weight of the oil was determined after which 60 mg/kg of citric acid dissolved in water, were added while the oil was stirred at about 400 rpm and heated up to about 95 °C. Then, the remaining water was removed by applying a vacuum of 8 mm under stirring, while the temperature was kept at about 95 °C. This drying step lasted for 10 to 22 minutes.

Bleaching
The temperature of the dried oil was about 95 °C and 1 % of bleaching earth were added while the oil was stirred at 400 to 500 rpm. This mixture was stirred for 5 min. (contact time) at 95 °C. Then, vacuum of about 30 mm was applied and the mixture stirred for a further 30 min. after which time it was filtered through a cellulose acetate filter at about 80 °C by means of a nitrogen pressure of 3 bar. The filter cake was taken as a specimen.

Deodorization
Volatile substances in the bleached oil were removed by deodorization. During the deodorization process, condensed reaction products were trapped in a vacuum trap with dry-ice and taken as a specimen.
About 600 g of the bleached oil were deodorized at 1 mm. The oil was heated up. When 160 °C were reached, the water stream was allowed to pass through the oil at a rate of about 0.2 mL/minutes. When 240 °C were reached, this temperature was kept for a contact time of 20 minutes. Thereafter, the heating supply was switched off and the oil allowed to cool down. When 160 °C were reached in the cooling down phase, the water stream was stopped. The total water stream treatment lasted for 29 to 33 minutes. Subsequently, the oil was allowed to cool down to 80 °C (or lower). The deodorized oil was taken as a specimen.

This deodorization procedure was repeated once whereby the refined oils were combined.

Characterization of the refined olive oil specimens
In order to characterize the produced, refined olive oil specimens, fatty acid spectra were recorded at the processing facility using GC/FID. These data were not recorded according to GLP guidelines.

Procedure for processing of olives to fermented olives

Sorting
Blemished and rotten olives were sorted out from the field olives manually and discarded. Only the healthy olives were used for processing.

Washing
4 kg of olives were transferred into a stainless steel basin, after which about 2 kg of tap water was added. The olives were washed by hand for a few seconds and then allowed to stand for 24 hours. Thereafter, the olives were separated from the wash water using a sieve and subsequently rinsed with about 0.5 kg of fresh water, which was added to the water portion previously used. A specimen of the washing water was taken for residue analysis.

Fermentation
The washed olives were transferred into a glass bottle which was then filled up with sodium chloride solution (7 %). The fermentation process was started using Vege-Start (8 mg/kg olives). Thereafter, the glass bottles were closed using fermentation tubes with water as sealing liquid and stored protected from light for 3 months at room temperature.

After 3 months, the olives were separated from the broth using a sieve. Both the fermented olives and the broth were taken as specimens.

Figure 2.3.2-1: Olive oil processing procedure flowchart – DocID 2007/1009604

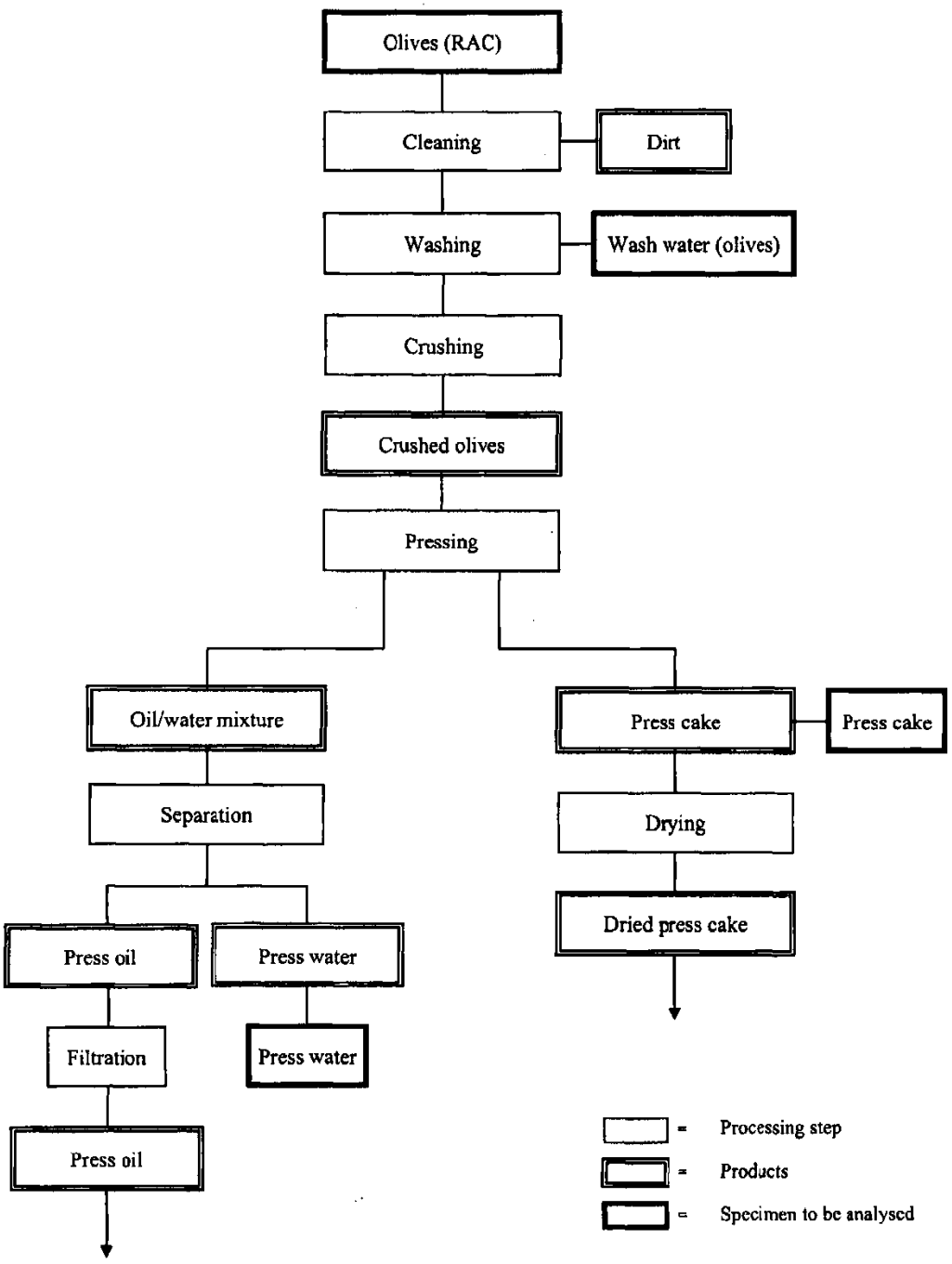


Figure 2.3.2-2: Olive oil processing procedure flowchart, continued – DocID 2007/1009604

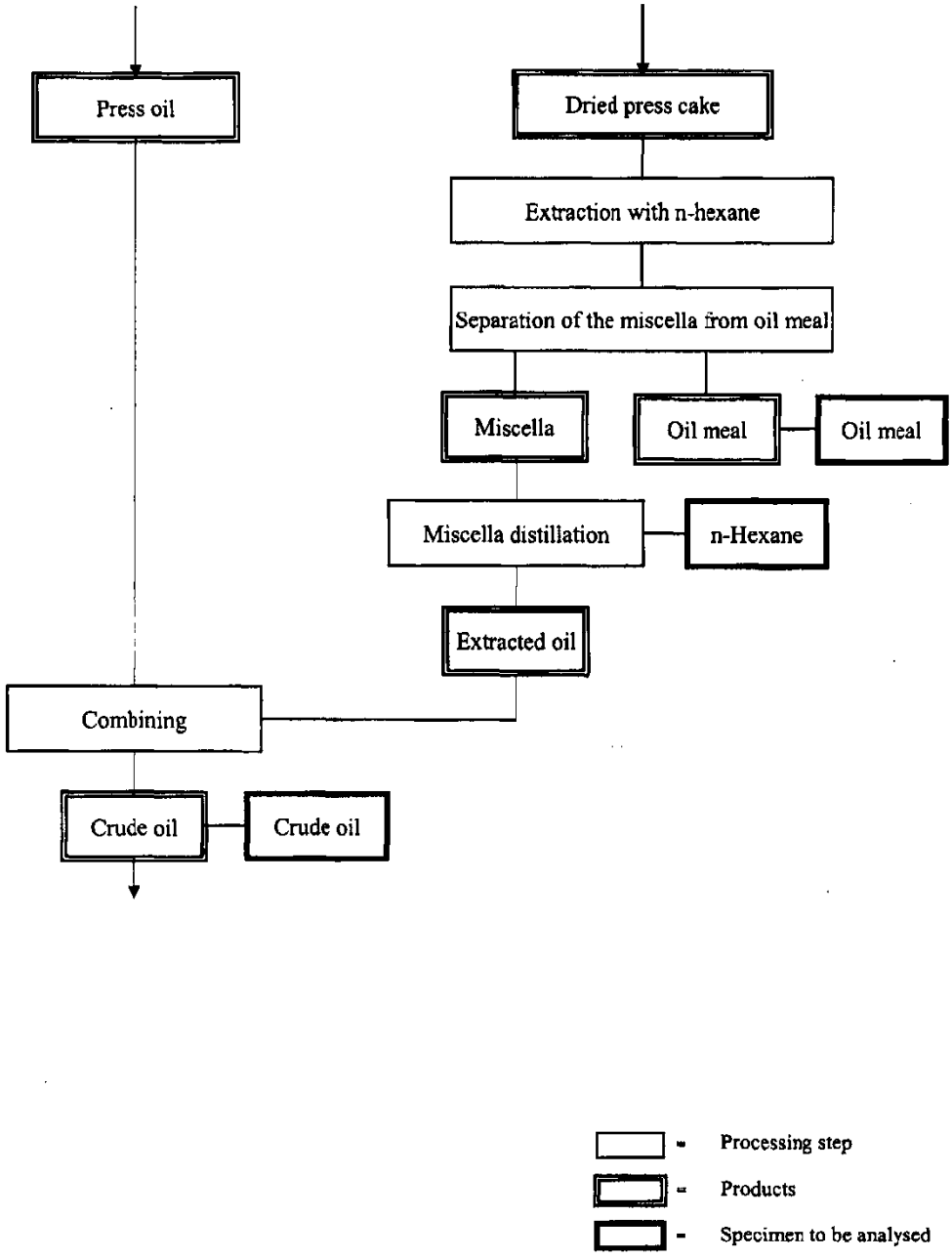
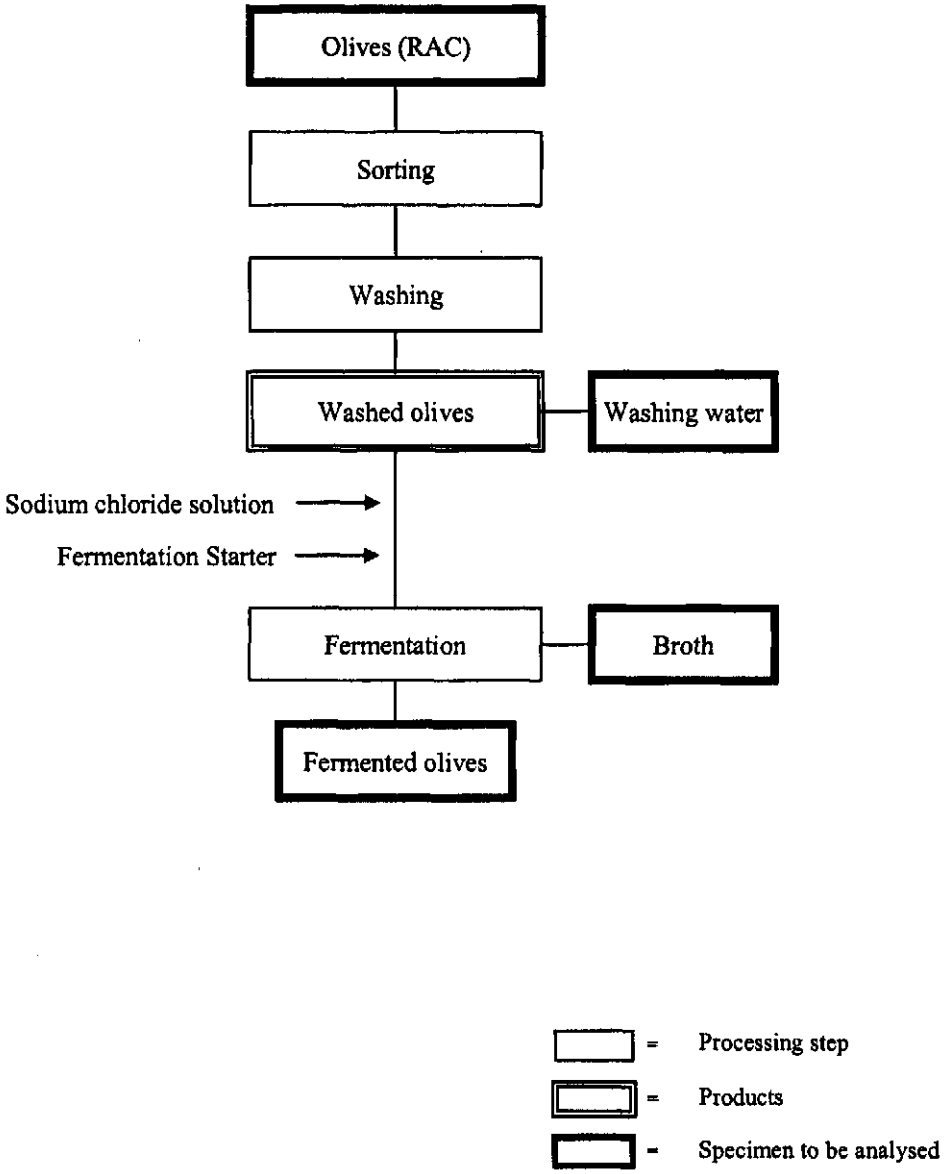


Figure 2.3.2-3: Fermented olives processing procedure flowchart – DocID 2007/1009604



Based on the weight of the starting material and the final products, the following mass balances were calculated:

Table 2.3.2-2: Mass balance of the production of crude olive oil - DocID 2007/1009604

Trial No. / Treatment		ALO/ 19/04 U	ALO/ 19/04 T	ALO/ 20/04 T	ALO/ 21/04 U	ALO/ 21/04 T	ALO/ 22/04 T
Olives (RAC), field specimen	g	30350.0	31000.0	30400.0	29500.0	30050.0	28650.0
Retain specimen	g	none	none	none	1051.3	1008.1	1027.1
Olives (RAC) starting material	g	30350.0	31000.0	30400.0	28448.7	29041.9	27622.9
Cleaning							
Olives (RAC)	g	30350.0	31000.0	30400.0	28448.7	29041.9	27622.9
Dirt	g	34.3	51.1	35.0	64.8	184.2	168.1
Weight of cleaned olives	g	30315.7	30948.9	30365.0	28383.9	28857.7	27454.8
Washing							
Wash water	g	30050.0	32860.0	30360.0	24250.0	22034.0	27000.0
Crushing							
Weight of crushed olives	g	30350.0	29300.0	30200.0	28383.9	28857.7	27454.8
Pressing							
Crushed olives used for pressing	g	30268.3	29214.5	30065.1	28303.0	28728.7	27305.0
Press cake	g	16500.0	19250.0	16800.0	13800.0	14150.0	13750.0
Press oil	g	1845.0	870.4	2268.0	994.7	951.5	707.8
Press water	g	10590.0	5610.0	9630.0	11240.0	10790.0	10740.0
Filter residue	g	263.2	1529.1	376.5	297.2	256.4	239.0
Total	g	29198.2	27259.5	29074.5	26331.9	26147.9	25436.8
Yield related to crushed olives	%	96.5	93.3	96.7	93.0	91.0	93.2
Drying of Press Cake							
Press cake used for drying	g	15403.5	18126.8	15807.8	12811.6	13115.4	12557.2
Dried press cake	g	10100.0	12750.0	11100.0	8200.0	8005.0	7950.0
Yield	%	65.6	70.3	70.2	64.0	61.0	63.3
Extraction and Distillation							
Dried press cake	g	10100.0	12750.0	11100.0	8200.0	8005.0	7950.0
Oil meal	g	7350.0	10310.0	7950.0	5800.0	5450.0	5550.0
Yield related to dried press cake	%	72.8	80.9	71.6	70.7	68.1	69.8
Extracted oil post n-hexane removal	g	3007.9	2936.0	2905.1	2584.4	2896.1	2620.7
Yield related to dried press cake	%	29.8	23.0	26.2	31.5	36.2	33.0
Total weight	g	10357.9	13246.0	10855.1	8384.4	8346.1	8170.7
Total yield of the extraction step	%	102.6	103.9	97.8	102.2	104.3	102.8
Total weight of n-hexane [1)]	g	39828.0	39828.0	39828.0	39828.0	39828.0	39828.0

[1)]: Approximate value since a non defined part was evaporated during distillation and vacuum evaporation whereas about 5 L of fresh n-hexane were added to compensate this loss.

Table 2.3.2-3: Mass balance of the production of refined olive oil - DocID 2007/1009604

Trial No. / Treatment		ALO/ 19/04 U	ALO/ 19/04 T	ALO/ 20/04 T	ALO/ 21/04 U	ALO/ 21/04 T	ALO/ 22/04 T
Total yield of crude oil production [1]							
Crushed olives used for pressing	g	30350.0	31000.0	30400.0	28448.7	29041.9	27622.9
Press oil	g	1850.0	873.0	2278.2	997.5	955.8	711.7
Extracted oil	g	3230.7	3127.1	3101.3	2791.7	3138.6	2885.4
Total of crude oil		5080.7	4000.0	5379.5	3789.2	4094.4	3597.1
Yield of crude olive oil	%	16.7	12.9	17.7	13.3	14.1	13.0
Refining Process							
Hydratation							
Crude oil	g	1504.8	1513.0	1510.6	1502.2	1510.5	1492.1
Addition of water	g	150.0	151.0	151.0	150.0	151.0	150.0
Desliming							
Addition of H_3PO_4/H_2O 1:1	g	6.0	6.1	6.0	6.1	12.1	6.0
Addition of water (approx.)	g	150.0	151.0	151.0	150.0	302.0	150.0
Weight of specimen: Slime/water mixture	g	463.2	308.2	415.0	443.5	631.0	473.0
Neutralisation							
Addition of NaOH (7 %) [2]	g	23.8	25.3	25.5	32.1	39.3	48.6
Addition of water	g	150.0	151.0	151.0	150.0	151.0	150.0
Weight of specimen: Soap	g	181.0	182.6	185.2	213.6	222.3	220.9
Washing							
Addition of water	g	450.0	453.0	453.0	604.0	600.0	450.0
Weight of specimen: Wash water	g	454.3	442.0	451.0	593.1	595.9	441.7
Drying and Bleaching							
Addition of citric acid solution [2]	g	5.0	5.0	5.0	5.0	5.0	5.0
Addition of bleaching earth	g	12.3	13.9	12.7	12.0	12.0	12.2
Weight of specimen: Filter cake	g	20.4	23.3	19.5	19.6	19.4	18.9
Bleached oil (no sample taken)	g	1158.6	1289.7	1149.1	1121.1	1085.8	1150.3
Deodorization							
Weight of bleached oil	g	632.4	633.0	633.9	614.0	629.2	608.6
Weight of refined oil	g	618.9	627.4	624.5	606.6	617.7	601.3
Weight of fatty acids	g	7.8	6.9	4.9	7.5	4.5	7.5
Yield of the deodorization step	%	99.1	100.2	99.3	100.0	98.9	100.0
Total yield of refined oil production from crude oil							
Initial weight of starting materials	g	2451.9	2469.3	2465.8	2611.4	2782.9	2463.9
Final weight of products	g.	2267.1	2248.5	2211.7	2391.1	2542.0	2305.2
Yield	%	92.5	91.1	89.7	91.6	91.3	93.6

[1] extrapolated to the mass of the starting material of olives (RAC)

[2] added in mL, but calculated as g to facilitate the calculation of the mass balance

Table 2.3.2-4: Mass balance of the production of fermented olives - DocID 2007/1009604

Trial No. **Treatment**		**ALO/ 19/04 U**	**ALO/ 19/04 T**	**ALO/ 20/04 T**	**ALO/ 21/04 U**	**ALO/ 21/04 T**	**ALO/ 22/04 T**
Olives (RAC) starting material	g	4140	4470	4260	4535	4343	4300
Olives sorted out	g	135	70	160	335	295	370
Olives, used for fermentation	g	4005	4400	4100	4200	44048	3930
Washing							
Wash water	g	2580	2520	2715	2805	2800	2805
Fermentation							
Starting material:							
Weight of washed olives	g	4265	4730	4230	4235	4095	3990
Weight of 7 % NaCl solution	g	5000	5000	5000	5000	5000	5000
Total	g	9265	9730	9230	9235	9095	8990
Products:							
Fermented olives	g	3970	4475	4220	4180	3995	3795
Broth	g	5205	5145	4905	4975	5035	5125
Total	g	9175	9620	9125	9155	9030	8920
Yield	%	99.0	98.9	98.9	99.1	99.3	99.2

Conclusion:

The distribution of alpha-cypermethrin residues during processing of olives to olive oil and during the production of fermented olives was investigated.
In the olive oil fractions, the alpha-cypermethrin residues were concentrated in press cake (mean transfer factor = 2.32) and in crude oil (mean transfer factor = 11.68). In the subsequent refining steps, the alpha-cypermethrin residues were concentrated in slime/water mixture (mean transfer factor = 2.66), in filter cake (mean transfer factor = 3.99) and in the edible portion refined oil (mean transfer factor = 8.87). In the other processing products, the mean transfer factors were below 1.

In the fermentation process the alpha-cypermethrin residues were slightly concentrated in the fermented olives (mean transfer factor 1.44).

Report:
Klitsinaris A. 1998a
Alphacypermethrin (CL 900049) 100 g a.i./l OESC: Decline Curve Residue Study on Alphacypermethrin in olives and olive oil - Hellas 1998
BASF DocID AL-714-002
BASF DocID 2000/7000997

Guidelines: EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8),EEC 96/68,EEC 7029/VI/95 rev. 5

GLP: yes
(Agrolab: Helenic Republic Ministry of Finance, General Chemical State Laboratory, Division of Environment)

Material and methods:

Two trials were laid down in Greece in 1998. Olives trees were treated with alpha-cypermethrin 100 g a.s./l OESC and received six applications, 21 days apart at a target dose of 30 g a.s./hL. Spray volumes in the first trial (98-01-01) ranged between 125 - 133 L/ha and in the second trial (98-01-03) between 67 - 72 L/ha. The application method was foliar with 300 cc spray solution applied on 10% of the foliage of each tree of the plot. The last application was made in the first trial (98-01-01) at crop growth stage BBCH 81 (beginning of fruit colouring) and in the second trial (98-01-03) at BBCH 87 (fruit ripe for picking). Olives were sampled 0- (before last application), 0+ (immediately after last application), 14-15, 26-28, 40-42 and 56 days after the last application.
Specimens were frozen within 24 hours and remained frozen until residue analysis.

The processing phase of the olive oil production was conducted at Agrolab-Sindos Laboratories, VI.PE.TH Thessaloniki, Greece according to an internal AGROLAB method.

The analysis of the olive oil and olive specimens was conducted at Agrolab-Sindos Analytical Laboratories, VI.PE.TH Thessaloniki Greece. The analysis of the olive oil specimens was performed according to method AGROLABSOP-RAD.M.004.This method is applicable for the determination of alpha-cypermethrin residues in olive oil with a limit of quantification of 0.020 mg/kg.
The analysis of the olive fruit specimens was performed according to Sittingbourne Analytical
Method “Method for determination of residues of Alpha-cypermethrin in Crops-Gas Chromatographic Method”, SAMS 351-2.This method is intended for the determination of Alpha-cypermethrin residues in dry, oily and aqueous crops, with a limit of quantification of 0.01 mg/kg.
Method performance was checked by determining the procedural recoveries in olive and olive oil matrices. In olive fruit, the recovery rates averaged 102.4% with a standard deviation of 24.8 and a coefficient of variation of 24.2% at fortification levels of 0.01 mg/kg and 0.10 mg/kg. In olive oil, the recovery rates averaged 97.4% with a standard deviation of 15.9 and a coefficient of variation of 16.3% at fortification levels of 0.02 mg/kg and 1.00 mg/kg.

Findings:
No residues of alpha-cypermethrin were detected in the untreated specimens at or above the
limit of quantification of the analytical methods used.

Residues of alpha-cypermethrin were determined in the treated olive fruit (RAC) specimens and in the corresponding oil fractions 0- (before last application), 0+ (immediately after last application), 14-15, 26-28, 40-42 and 56 days after the last application.
The results are summarized below:

Table 2.3.2-5: Summary of alpha-cypermethrin residues in olives and transfer factors – DocID AL-714-002

	Residue Concentration of Alpha-cypermethrin in mg/kg		Transfer Factors		
	Trial 98-01-01	Trial 98-01-03	Trial 98-01-01	Trial 98-01-03	mean
Olive fruit (day 0-, before last application)	0.126	0.035	N/A	N/A	N/A
Olive fruit (day 0+, after last application)	0.215	0.079	N/A	N/A	N/A
Olive fruit (day 14-15)	0.097	0.063	N/A	N/A	N/A
Olive fruit (day 26-28)	0.099	0.045	N/A	N/A	N/A
Olive fruit (day 40-42)	0.040	0.043	N/A	N/A	N/A
Olive fruit (day 56)	0.117	0.063	N/A	N/A	N/A
Olive oil (day 0-, before last application)	0.363	0.194	2.88	5.54	4.21
Olive oil (day 0+, after last application)	0.668	0.220	3.11	2.78	2.95
Olive oil (day 14-15)	0.850	0.171	8.76	2.71	5.74
Olive oil (day 26-28)	0.413	0.182	4.17	4.04	4.11
Olive oil (day 40-42)	0.318	0.107	7.95	2.49	5.22
Olive oil (day 56)	0.075	0.158	0.64	2.51	1.58

Procedure for processing of olives to olive oil

The processing was conducted separately for each individual specimen. The final oil sub-specimens were pooled at the end of the processing phase.
Before processing, the initial net weight of each specimen was recorded.

Homogenization
For the homogenization step, a Tecator homogenizer model 1094 was used. The olive fruits were homogenized for approximately 1-2 minutes. The pulp produced was weighed and transferred to the pressing device or stored in cool and dark conditions for later use. Pulp compression was conducted within 24 hours from pulp production. The homogenizer was thoroughly cleaned after each use.

Pulp pressing
The press device container was filled with pulp and was pressed for about 1 hour in 100 bar pressure in a hydraulic press device Karakasis Bros.

Juice extraction
The juice that was extracted, was weighed and collected into special plastic containers for centrifugation, while the cake was weighed and then discarded.

Centrifugation and olive – oil separation
The containers containing the extracted juice are centrifuged for 10 minutes at a 9000 rounds/min. speed in a Sigma 6-10 centrifugal device.
After centrifugation, the oily phase was collected in a plastic container and then was weighed and labelled. The water phase and the solid deposit were discarded.
At the end of the processing phase for each individual specimen, the processed oil sub-specimens were collected in the same vessel and labelled with the trial number, the specimen code number, the type of product after processing and the date of processing.
The samples were deep-frozen at <-18° C. At the end of the processing, the processed specimens were delivered to the analytical laboratory for residue analysis.

Figure 2.3.2-4: Olive oil processing procedure flowchart - DocID AL-714-002

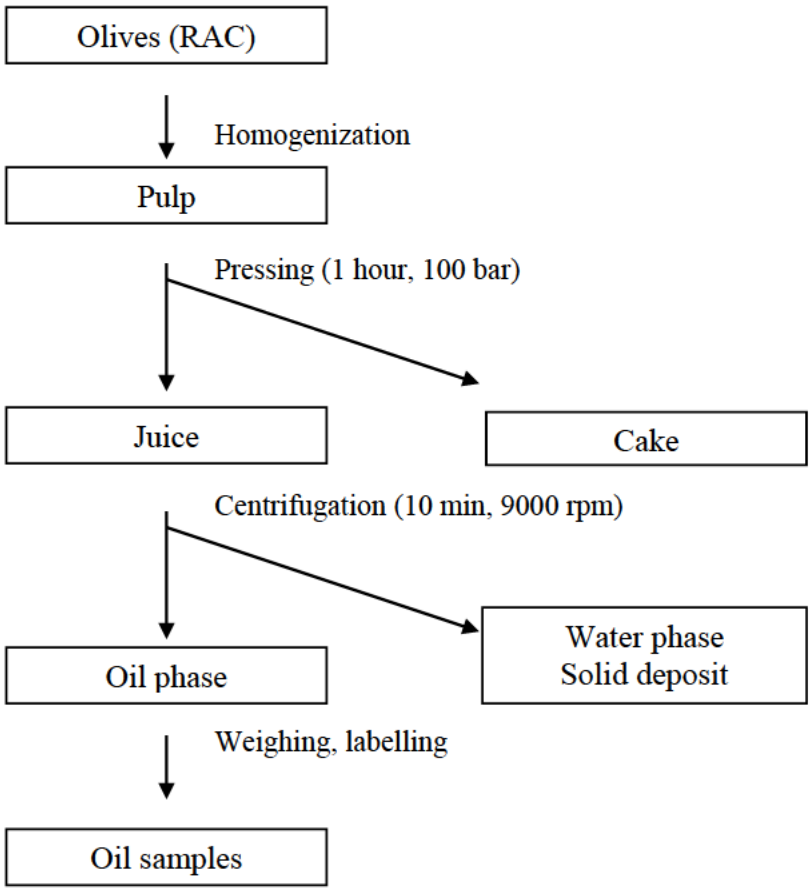


Table 2.3.2-6: Weights of processed olive oil fractions - DocID AL-714-002

Trial No.	Sample code	Treatment	Initial weight of specimen (g)	Olive oil produced (g)
98-01-01	98-01-01-01	UTR	8050	626
	98-01-01-03	TR	8110	973
	98-01-01-05	TR	7820	1039
	98-01-01-07	TR	1980	247
	98-01-01-09	UTR	2065	248
	98-01-01-011	TR	2040	242
	98-01-01-013	TR	3950	541
	98-01-01-015	UTR	3000	338
	98-01-01-017	TR	3300	410
98-01-03	98-01-03-01	UTR	4285	372
	98-01-03-03	TR	4090	500
	98-01-03-05	TR	3970	510
	98-01-03-07	TR	3000	243
	98-01-03-09	UTR	2900	322
	98-01-03-011	TR	2620	377
	98-01-03-013	TR	3000	483
	98-01-03-015	UTR	2900	618
	98-01-03-017	TR	2935	667

Conclusion:

After treatment of olives trees with alpha-cypermethrin, residues are concentrated in the oil fraction. The mean transfer factors found in this study ranged between 1.58 – 5.74.

Report:
Trewhitt J. A. 1999a
Alphacypermethrin (CL 900049) 100 g a.s./L OESC (CF 07493): Decline Curve Residue Study on Alphacypermethrin in Olives and Olive Oil Hellas 1999
BASF DocID AL-714-001
BASF DocID 2000/7000996

Guidelines: EEC 96/68,EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8),EEC 7029/VI/95 rev. 5

GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Material and methods:

One trial was laid down in Greece in 1999 in which olive trees were treated with alpha-cypermethrin 100 g a.s./L OESC. The olive trees received 6 applications, 21-26 days apart at a dose rate of 3 g a.s./hL with spray volumes ranging between 998 and 1001 L/ha. The crop growth stage at the final application was BBCH 85 (advanced ripening). Olive specimens were sampled at 0- and 0+ days after treatment no. 6 (just before and just after the final application), and then at 14, 28, 42 and 56 days after the final treatment. At each sampling, separate specimens were taken for olive oil and olive fruit analysis.

The specimens for processing into olive oil were sent to AGROLAB in Thessaloniki (Greece) at ambient temperature. There, the olive oil was extracted from the olives and was then frozen prior to despatch to the Product Research and Development Laboratory (PRDL), Cyanamid Agriculture Ltd., Gosport, Hampshire, UK, for analysis.

The specimens taken for olive fruit analysis were frozen within 24 hours of being taken and remained frozen, including during transportation, until analysis.

The analysis of the specimens was conducted at PRDL according to method RLA 12594.01.
Procedural recoveries performed during the analysis of the specimens from this study, AL-HE-99-530, confirmed the validity of the method.
In olive fruit, a recovery of 70% was found at a fortification level of 0.05 mg/kg. In olive oil, the recovery was 83% at 0.5 mg/kg.

Findings:

No residues, at or above the limit of quantification, of alpha-cypermethrin were detected in the untreated olive fruit and olive oil specimens.

Residue levels in the treated olive oil specimens ranged between 0.24 and 0.79 mg/kg. Residue levels in the treated olive fruit specimens ranged between <0.05 and 0.15 mg/kg.

The results are summarized in the following table:

Table 2.3.2-7: Summary of alpha-cypermethrin residues in olives and transfer factors - DocID AL-714-001

	Trial 99-530-01		
	Residue concentration of alpha-cypermethrin in mg/kg	Transfer Factors	
Olive fruit flesh (day 0-, before last application)	<0.05	N/A	
Olive fruit flesh (day 0+, after last application)	0.06	N/A	
Olive fruit flesh (day 14)	<0.05	N/A	
Olive fruit flesh (day 28)	<0.05	N/A	
Olive fruit flesh (day 42)	<0.05	N/A	
Olive fruit flesh (day 56)	0.15	N/A	
Whole olive fruit (day 0-, before last application) [1)]	<0.05	N/A	
Whole olive fruit (day 0+, after last application) [1)]	<0.05	N/A	
Whole olive fruit (day 14) [1)]	<0.05	N/A	
Whole olive fruit (day 28) [1)]	<0.05	N/A	
Whole olive fruit (day 42) [1)]	<0.05	N/A	
Whole olive fruit (day 56) [1)]	0.11	N/A	
		calc. for flesh	calc. for whole fruit
Olive oil (day 0-, before last application)	0.07	>1.40	>1.40
Olive oil (day 0+, after last application)	0.42	7.00	>8.40
Olive oil (day 14)	0.58	>11.60	>11.60
Olive oil (day 28)	0.79	>15.80	>15.80
Olive oil (day 42)	0.34	>6.80	>6.80
Olive oil (day 56)	0.24	1.60	2.18

1) The residue in the whole fruit can be calculated as follows:
Weight of flesh = 75% of whole fruit 75% of 0.15 mg/kg = 0.11 mg/kg = total residues in the whole fruit.
(The actual residue in flesh of <0.05 mg/kg is taken as 0.05 mg/kg for the purposes of the calculation.)

Procedure for processing of olives to olive oil

The processing was conducted separately for each individual specimen. The final oil sub-specimens were pooled at the end of the processing phase.
Before processing, the initial net weight of each specimen was recorded.

Homogenization
The olive fruits were homogenized in a Tecator 1094 homogenizer. The stones were then removed from the pulp produced.

Pulp pressing
The pulp was pressed in a laboratory hydraulic press at 100 bar pressure and the juice that was extracted was collected and equally distributed in six centrifugation vessels of 300 ml capacity each.

Centrifugation and olive – oil separation
The juice was then centrifuged at 9000 rpm for ten minutes and the overlaying fraction of oil was removed from each vessel using a pipette. The oil produced in each centrifugation was weighed and collected in a plastic container.
Because specimens were too large to be processed in one individual processing step, they were processed in smaller quantities (sub-specimens) and then mixed in one individual container.
The processed specimens were packed in plastic bags and labelled.
The samples were stored deep-frozen at <-18° C. At the end of the processing, the processed specimens were delivered to the analytical laboratory for residue analysis.

Figure 2.3.2-5: Olive oil processing procedure flowchart - DocID AL-714-001

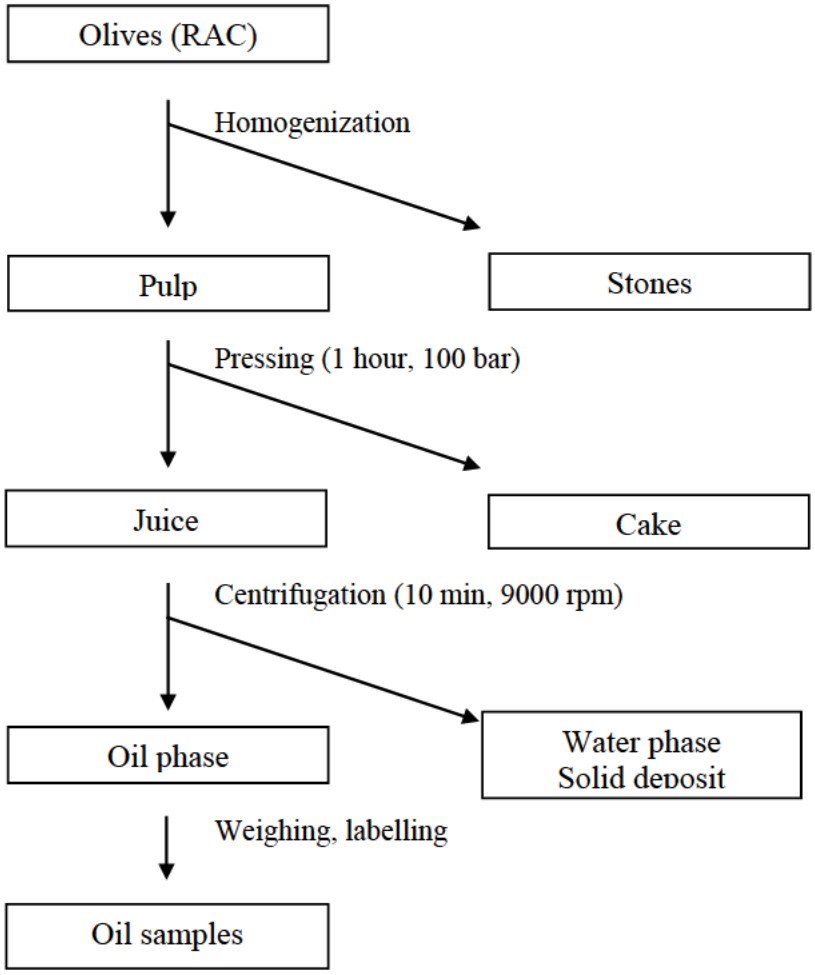


Table 2.3.2-8: Weights of processed olive oil fractions - DocID AL-714-001

Trial No.	Sample code	Treatment	Weight per processing stage			
			Fruit (kg)	Pulp (kg)	Juice (g)	Oil (g)
99-530-01	99-530-01-01	UTR	5.8	5.4	1615	522
	99-530-01-03	TR	5.6	5.3	1597	353
	99-530-01-05	TR	5.0	4.8	1438	206
	99-530-01-07	TR	5.4	5.1	1443	333
	99-530-01-09	UTR	5.4	5.1	1442	514
	99-530-01-11	TR	5.4	4.8	1232	516
	99-530-01-13	TR	5.5	5.3	1324	436
	99-530-01-15	UTR	5.0	4.8	1202	788
	99-530-01-17	TR	5.0	4.8	1264	632

Conclusion:

After treatment of olive trees with alpha-cypermethrin, a residue concentration was observed in the oil fraction. The transfer factors found in this study ranged between >1.40 – >15.80.

2.3.3 Estimation of MRL, HR and STMR for olive

For *olives*, the following residue studies were considered (BASF DocIDs): 2005/1004975, 2005/1007582 and 2012/1157548.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Southern Europe (S-EU) [mg/kg]
2005/1004975 (open field)	<0.05 (4x)
2005/1007582 (open field)	<0,01, 0.04 (2x), 0.09
2012/1157548 (open field)	0,14, 0,26
OECD-MRL-calculation (open field)	**0.4 (n=10, STMR=0.05, HR=0.26)**

_ underlined values were used for risk assessment purposes

2.4 Potato

Residue data from supervised trials in potatoes were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.4-1: Number of residue trials conducted in potato per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Potato	2001	4	DE, FR, NL, UK	4	ES, FR, GR, IT	8	AL-721-049
Potato	2004	-	-	2	GR, IT	2	2005/1007592
Potato	2005	4	DE, FR, NL, UK	4	ES, FR, GR, IT	8	2006/1026846
Potato	2006	4	DE, DK, FR, UK	4	ES, FR, GR, IT	8	2007/1007945
Potato	2007	2	FR, NL	2	ES, FR	4	BASF DocID 2008/1002704
Potato	2011	2	DE, UK	2	IT, ES	4	BASF DocID 2012/1157550
Total number of trials per region		**16**	**-**	**18**	**Total number of trials**	**34**	

2.4.1 Supervised residue trials in potato

Jones S (2002)

Full study reference

Jones S (2002): Study on the residue behaviour of BAS 310 I in potato after application of BAS 310 08 I under field conditions in Great Britain, Germany, Spain, France (N &S) Italy, Greece, Netherlands, 2001; BASF RDI No. AL-721-049

Perny A (2005)

Full study reference

Perny A (2005): Study on the behaviour of BAS 310 I in Potatoes after Treatment with BAS 310 41 I under Field Conditions, in Italy and in Greece, in 2004; BASF DocID 2005/1007592

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of alpha-cypermethrin in potato after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026846

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alpha-cypermethrin in potato after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007945

Material and Methods:

In the years 2001, 2004, 2005 and 2006 a residue program in potatoes was conducted in representative potato growing areas in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.

Eight residue decline trials were conducted in potatoes in the United Kingdom, Germany, Spain, France (North and South), Italy, Greece and the Netherlands during 2001. Potato plants were treated with a 150 g/kg wettable granule formulation (WG; BAS 310 08 I) formulation of alpha-cypermethrin. Separate plots of potatoes were treated with either a single application at a target rate of 15 g a.s./ha or two applications at a target rate of 15 g a.s./ha. The single applications were intended to be made 28 days before harvest. The growth stage of the potato plants at application was between 32 (20% of plants meet between rows) and 91(beginning of leaf yellowing). Specimens of potatoes were taken immediately after the last application, 6-8, 20-22, 28-29 and 34-36 days after the final application.
The specimens were analysed for residues of BAS 310 I using BASF Agro Research method RLA 12513.03V (GC-ECD) which has a limit of quantitation of 0.05 mg/kg for potatoes.

During the 2004 growing season, two field trials were conducted in Italy and Greece in order to determine the residues of alpha-cypermethrin in potatoes after application of a 100 g a.s./L soluble concentrate formulation (BAS 310 41 I).
The product was foliar applied to potatoes in different amounts: In the first variant, one application was done at a target rate of 15 g a.s./ha. This was compared to a second variant, consisting of two treatments at a target rate of 15 g a.s./ha performed at the same application rate. The growth stage of the potato plants was 49 (skin set complete: (skin at apical end of tuber not removable with thumb) 95% of tubers in this stage) at the last application. Potato tuber specimens were sampled directly after the last application as well as 3, 7-8 and 14 days thereafter.
The samples were analysed for alpha-cypermethrin by means of HPLC-MS/MS according to BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg.

During the 2005 growing season, alpha-cypermethrin was tested in potato at eight locations. Different application schemes were compared: In the Northern European trials performed in the United Kingdom, France, Germany and the Netherlands an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I) was foliar applied once or twice at a target rate of 12.5 g a.s./ha to separate plots of potato plants. The growth stage of the potato plants was between 46 (60% of total final tuber mass reached) and 69 (end of flowering in the first inflorescence) at the last application.
In the four Southern European trials performed in France, Italy, Spain and Greece one single treatment at a target rate of 25 g a.s./ha was foliar applied to potato plants. The growth stage of the potato plants was between 42 (20% of total final tuber mass reached) and 47 (70% of total final tuber mass reached) at application. Potato tuber specimens were collected directly after the last application as well as 7, 14 and 20-22 days thereafter. They were analysed for alpha-cypermethrin by means of HPLC MS/MS with BASF analytical method No. 567/0 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, alpha-cypermethrin was tested in potato at eight locations. Different application schemes were compared: In the Northern European trials performed in France, Denmark, Germany and the United Kingdom an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I) was foliar applied once or twice at a target rate of 12.5 g a.s./ha to separate plots of potato plants. The growth stage of the potato plants was between 45 (50% of total final tuber mass reached) and 71 (10% of berries in the first fructification have reached full size (main stem)) at the last application.
In the four Southern European trials performed in France, Italy, Spain and Greece one single treatment at a target rate of 25 g a.s./ha was foliar applied to potato plants. The growth stage of the potato plants was between 46 (60% of total final tuber mass reached) and 83 at application. Potato tuber specimens were collected directly after the last application as well as 7, 14 and 21 days thereafter. They were analysed for alpha-cypermethrin by means of HPLC MS/MS with BASF method No. 567/0 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.4.1-1.
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Table 2.4.1-1: Residues in potatoes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID AL-721-049 Trial No. AGR/27/01 Study to GLP Study carried out in 2001	Potato (variety Cilena)	The Netherlands 6595 Ottersum Zandsteeg 18 Limburg	15 g a.s./ha 07.08.01	43	0 7 20 28 34	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	BASF analytical method No. RLA 12513.03.V potato, tuber: mean recovery = 89.0 %; SD: +/- 10.7; CV: 12.1; n=8; fortification range 0.05 mg/kg Method check: potato, tuber: mean recovery = 96.3 %; SD: +/- 8.0; CV: 8.3; n=8; fortification range 0.05-0.5 mg/kg
BASF Doc ID AL-721-049 Trial No. ALO/39/01 Study to GLP Study carried out in 2001	Potato (variety Spunta)	Spain E-41710 Virgen del Castillo 31, Sevilla, Andalucia	15 g a.s./ha 03.05.01	48	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. DU4/10/01 Study to GLP Study carried out in 2001	Potato (variety Solana)	Germany 67376 Herthausen, Rheinland-Pfalz	15 g a.s./ha 13.08.01	41	0 8 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. FBD/05/01 Study to GLP Study carried out in 2001	Potato (variety Mana)	France 26380 Perins, Le plan Rhone-Alpes (South of EU)	16 g a.s./ha 24.07.04	32	0 8 22 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. FBM/06/01 Study to GLP Study carried out in 2001	Potato (variety Nicolas)	France 72800 Thoree les Pins, le Point du Jour Pays de la Loire (North of EU)	17 g a.s./ha 28.08.01	91	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. HEL/01/01 Study to GLP Study carried out in 2001	Potato (variety Spunta)	Greece Edessa, Nisi Northern Greece - Macedonia	15 g a.s./ha 28.09.01	89	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. ITA/07/01 Study to GLP Study carried out in 2001	Potato (variety Monalisa)	Italy 27050 Casei Gerola Strada Voghera Pavia	15 g a.s./ha 13.07.01	85	0 8 22 29 36	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. OAT/08/01 Study to GLP Study carried out in 2001	Potato (variety Wilja)	United Kingdom Akeman Street Farm, Combe OX8 8EW Oxfordshire	14 g a.s./ha 17.08.01	79	0 6 20 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	

Table 2.4.1-1: Residues in potatoes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1007592 Trial No. GRE/02/04 Study to GLP Study carried out in 2004	Potato (variety Agria)	Greece 60100 Kozani Northern Greece - Macedonia	15 g a.s./ha 28.07.04	49	0 3 8 14	tuber <0.01 tuber <0.01 tuber <0.01 tuber <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 88.5 %; SD: +/- 5.5; CV: 6.2; n=3; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007592 Trial No. ITA/01/04 Study to GLP Study carried out in 2004	Potato (variety Agata)	Italy 27050 Casel Gerola Pavia	15 g a.s./ha 27.07.04	49	0 3 7 14	tuber <0.01 tuber <0.01 tuber <0.01 tuber <0.01	
BASF Doc ID 2005/1007592 Trial No. GRE/02/04 Study to GLP Study carried out in 2004	Potato (variety Agria)	Greece 60100 Kozani Northern Greece - Macedonia	15 g a.s./ha 2 treatm. last date 28.07.04	49 at last treatm.	0 3 8 14	tuber <0.01 tuber <0.01 tuber <0.01 tuber <u><0.01</u>	
BASF Doc ID 2005/1007592 Trial No. ITA/01/0**4** Study to GLP Study carried out in 2004	Potato (variety Agata)	Italy 27050 Casel Gerola Pavia	15 g a.s./ha 2 treatm. last date 27.07.04	49 at last treatm.	0 3 7 14	tuber <0.01 tuber <0.01 tuber <0.01 tuber <u><0.01</u>	
BASF Doc ID AL-721-049 Trial No. AGR/27/01 Study to GLP Study carried out in 2001	Potato (variety Cilena)	The Netherlands 6595 Oltersum Zandsteeg 18 Limburg	15 g a.s./ha 2 treatm. last date 07.08.01	43 at last treatm.	0 7 20 28 34	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	BASF analytical method No. RLA 12513.03.V potato, tuber: mean recovery = 89.0 %; SD: +/- 10.7; CV: 12.1; n=8; fortification range 0.05 mg/kg Method check: potato, tuber: mean recovery = 96.3 %; SD: +/- 8.0; CV: 8.3; n=8; fortification range 0.05-0.5 mg/kg
BASF Doc ID AL-721-049 Trial No. ALO/39/01 Study to GLP Study carried out in 2001	Potato (variety Spunta)	Spain E-41710 Virgen del Castillo 31, Sevilla, Andalucia	15 g a.s./ha 2 treatm. last date 03.05.01	48 at last treatm.	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. DU4/10/01 Study to GLP Study carried out in 2001	Potato (variety Solana)	Germany 67376 Herthausen, Rheinland-Pfalz	15 g a.s./ha 2 treatm. last date 27.08.01	33 at last treatm.	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. FBD/05/01 Study to GLP Study carried out in 2001	Potato (variety Mana)	France 26380 Perins, Le plan Rhone-Alpes (South of EU)	15/16 g a.s./ha 2 treatm. last date 24.07.04	32 at last treatm.	0 8 22 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. FBM/06/01 Study to GLP Study carried out in 2001	Potato (variety Nicolas)	France 72800 Thoree les Pins, le Point du Jour Pays de la Loire (North of EU)	17/15 g a.s./ha 2 treatm. last date 28.08.01	91 at last treatm.	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	

Table 2.4.1-1: Residues in potatoes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID AL-721-049 Trial No. HEL/01/01 Study to GLP Study carried out in 2001	Potato (variety Spunta)	Greece Edessa, Nisi Northern Greece - Macedonia	16/15 g a.s./ha 2 treatm. last date 28.09.01	89 at last treatm.	0 7 21 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	BASF analytical method No. RLA 12513.03.V potato, tuber: mean recovery = 89.0 %; SD: +/- 10.7; CV: 12.1; n=8; fortification range 0.05 mg/kg Method check: potato, tuber: mean recovery = 96.3 %; SD: +/- 8.0; CV: 8.3; n=8; fortification range 0.05-0.5 mg/kg
BASF Doc ID AL-721-049 Trial No. ITA/07/01 Study to GLP Study carried out in 2001	Potato (variety Monalisa)	Italy 27050 Casei Gerola Strada Voghera Pavia	15/17 g a.s./ha 2 treatm. last date 13.07.01	85 at last treatm.	0 8 22 29 36	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID AL-721-049 Trial No. OAT/08/01 Study to GLP Study carried out in 2001	Potato (variety Wilja)	United Kingdom Akeman Street Farm, Combe OX8 8EW Oxfordshire	15 g a.s./ha 2 treatm. last date 17.08.01	79 at last treatm.	0 6 20 28 35	tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05 tubers <0.05	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/1 Study to GLP Study carried out in 2005	Potato (variety Wilja)	United Kingdom Barn farm Cranebrook Lane; Hilton, Lichfield Staffordshire	12.5 g a.s./ha 02.09.05	48	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 90.2 %; SD: +/- 7.9; CV: 8.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/2 Study to GLP Study carried out in 2005	Potato (variety Synthomas)	France Le Quart Mallet Uchizy 71700 North of EU	12.5 g a.s./ha 03.08.05	46	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/3 Study to GLP Study carried out in 2005	Potato (variety Agria)	Germany Waldstraße 5 69256 Mauer	12.5 g a.s./ha 23.08.05	47	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/4 Study to GLP Study carried out in 2005	Potato (variety Agria)	The Netherlands Reethsestraat 6662 PK Elst Gelderland	12.5 g a.s./ha 24.08.05	69	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	

Table 2.4.1-1: Residues in potatoes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/1 Study to GLP Study carried out in 2006	Potato (variety Mona Lisa)	France St Hilaire St Mesmin Loiret, 45160 (North of EU)	12.5 g a.s./ha 19.07.06	47	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 86.4 %; SD: +/- 6.8; CV: 7.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/2 Study to GLP Study carried out in 2006	Potato (variety Hamlet)	Denmark Agrolab A/S Røjleskovvej 18 Middelfart, Fyn	12.5 g a.s./ha 26.07.06	45	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/3 Study to GLP Study carried out in 2006	Potato (variety Bernadette)	Germany Peiner Weg 60 31303 Burgdorf Lower saxony	12.5 g a.s./ha 07.07.06	65-69	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 86.4 %; SD: +/- 6.8; CV: 7.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/9 Study to GLP Study carried out in 2006	Potato (variety King Edward)	United Kingdom Hemmington, Derby, Derbyshire	12.5 g a.s./ha 19.09.06	71	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/1 Study to GLP Study carried out in 2005	Potato (variety Wilja)	United Kingdom Barn farm Cranebrook Lane Hilton, Lichfield Staffordshire	12.5 g a.s./ha 2 treatm. last date 02.09.05	48 at last treatm	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 90.2 %; SD: +/- 7.9; CV: 8.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/2 Study to GLP Study carried out in 2005	Potato (variety Synthomas)	France Le Quart Mallet Uchizy 71700 North of EU	12.5 g a.s./ha 2 treatm. last date 03.08.05	46 at last treatm	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/3 Study to GLP Study carried out in 2005	Potato (variety Agria)	Germany Waldstraße 5 69256 Mauer	12.5 g a.s./ha 2 treatm. last date 23.08.05	47 at last treatm	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/4 Study to GLP Study carried out in 2005	Potato (variety Agria)	The Netherlands Reethsestraat 6662 PK Elst Gelderland	12.5 g a.s./ha 2 treatm. last date 24.08.05	69 at last treatm	0 7 14 21	tubers <0.01 tubers <0.01 tubers <0.01 tubers <0.01	

Table 2.4.1-1: Residues in potatoes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/1 Study to GLP Study carried out in 2006	Potato (variety Mona Lisa)	France St Hilaire St Mesmin, Loiret, 45160 (North of EU)	12.5 g a.s./ha 2 treatm. last date 19.07.06	47 at last treatm.	0 7 14 21	tubers <0.01 tubers <u><0.01</u> tubers <0.01 tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 86.4 %; SD: +/- 6.8; CV: 7.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/2 Study to GLP Study carried out in 2006	Potato (variety Hamlet)	Denmark Agrolab A/S Røjleskovvej 18 Middelfart, Fyn	12.5 g a.s./ha 2 treatm. last date 26.07.06	45 at last treatm	0 7 14 21	tubers <0.01 tubers <u><0.01</u> tubers <0.01 tubers <0.01	
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/3 Study to GLP Study carried out in 2006	Potato (variety Bernadette)	Germany Peiner Weg 60 31303 Burgdorf Lower saxony	12.5 g a.s./ha 2 treatm. last date 07.07.06	65-69 at last treatm	0 7 14 21	tubers <0.01 tubers <u><0.01</u> tubers <0.01 tubers <0.01	
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/9 Study to GLP Study carried out in 2006	Potato (variety King Edward)	United Kingdom Hemmington, Derby, Derbyshire	12.5 g a.s./ha 2 treatm. last date 19.09.06	71 at last treatm	0 7 14 21	tubers <0.01 tubers <u><0.01</u> tubers <0.01 tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 86.4 %; SD: +/- 6.8; CV: 7.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/5 Study to GLP Study carried out in 2006	Potato (variety Mona Lisa)	France St Jory Haute-Garonne (South of EU)	25 g a.s./ha 04.07.06	46	0 7 14 21	tubers <0.01 tubers <0.01 tubers <u><0.01</u> tubers <0.01	
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/6 Study to GLP Study carried out in 2006	Potato (variety Agria)	Spain Alcala del Moncayo, Zaragoza, Aragon	25 g a.s./ha 08.08.06	81-83	0 7 14 21	tubers <0.01 tubers <0.01 tubers <u><0.01</u> tubers <0.01	
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/7 Study to GLP Study carried out in 2006	Potato (variety Almera)	Italy Budrio-BO, Emilia-Romagna	25 g a.s./ha 14.07.06	46-48	0 7 14 21	tubers <0.01 tubers <0.01 tubers <u><0.01</u> tubers <0.01	
BASF Doc ID 2007/1007945 Trial No. AF/10497/BA/8 Study to GLP Study carried out in 2006	Potato (variety Agria)	Greece Nea Magnisia, Thessaloniki, Central Macedonia	25 g a.s./ha 16.06.06		0 7 14 21	tubers <0.01 tubers <0.01 tubers <u><0.01</u> tubers <0.01	

Table 2.4.1-1: Residues in potatoes

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/5 Study to GLP Study carried out in 2005	Potato (variety Agatha)	France Route de Monclar de Quercy, Montauban 82000 South of EU	25 g a.s./ha 07.10.05	47	0 7 14 21	tubers <0.01 tubers <0.01 tubers <u>≤0.01</u> tubers <0.01	BASF analytical method No. 567/0 potato, tuber: mean recovery = 90.2 %; SD: +/- 7.9; CV: 8.8; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/6 Study to GLP Study carried out in 2005	Potato (variety Agata)	Italy Via Olmo 60-Budrio, Bologna 40054	25 g a.s./ha 29.06.05	45-46	0 7 14 20	tubers <0.01 tubers <0.01 tubers <u>≤0.01</u> tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/7 Study to GLP Study carried out in 2005	Potato (variety Agria)	Spain Replazeto S/N, Villarreal 50490	25 g a.s./ha 29.08.05	46	0 7 14 22	tubers <0.01 tubers <0.01 tubers <u>≤0.01</u> tubers <0.01	
BASF Doc ID 2006/1026846 Trial No. AF/8831/BA/8 Study to GLP Study carried out in 2005	Potato (variety Spunta)	Greece Zoodochus Pygi Central Macedonia GR-50100	25 g a.s./ha 01.08.05	42	0 7 14 21	tubers <0.01 tubers <0.01 tubers <u>≤0.01</u> tubers <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in potatoes presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 14 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha–8 trials including both variants, all conducted in the Northern European region
- one or two treatments at a target rate of 15 g a.s./ha–10 trials including both variants, 4 conducted in the Northern European region and 6 conducted in the Southern European region
- one treatment at a target rate of 25 g a.s./ha conditions–8 trials conducted in the Southern European region

In all trials presented above no residues above the method LOQ (0.05 mg/kg for the 2001 study-8 trials with either one or two applications at 15 g a.s./ha; 0.01 mg/kg for all other trials) were found in any of the treated potato tuber samples, regardless of rate or sampling interval.

Conclusion:

In the years 2001, 2004, 2005 and 2006 a residue program in potatoes was conducted in representative potato growing areas in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.
A total of 26 trials were performed that support the proposed GAPs for the Northern and Southern European region. In all trials no residues above the method LOQ (0.05 mg/kg for the 2001 study-8 trials with either one or two applications at 15 g a.s./ha; 0.01 mg/kg for all other trials) were found in any of the treated potato tuber samples, regardless of rate or sampling interval.

Report:	**Kreke N.,Gehl J., 2008c**
Title:	Determination of residues of Alpha-Cypermethrin and Acetamiprid in potato (RAC tubers) following two treatments with BAS 370 00 I, BAS 310 40 I or BAS 9111 0 I from four open field trials in Northern and Southern Europe in 2007
Document No:	BASF DocID 2008/1002704
Guidelines:	EEC 96/68, EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7
GLP	yes

Executive Summary

During the 2007 growing season, a total of four trials were conducted on potato in order to determine the magnitude of residues of active ingredient(s) in or on Raw Agricultural Commodities (RAC).
Plot 101: Untreated (control)
Plot 102: BAS 370 00 I, a SL formulation of alpha-cypermethrin (25 g/L) and acetamiprid (100 g/L), was applied twice at the rate of 500 mL/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).
Plot 103: BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g/L), was applied twice at the rate of 125 mL/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).
Plot 104: BAS 9111 0 I, a WP formulation of acetamiprid (200 g/kg), was applied twice at the rate of 250 g/ha, in order to determine the magnitude of the residues of active ingredient in or on
Raw Agricultural Commodities (RAC).
In context of this summary, only the results of alpha-cypermethrin (plot 102 & plot 103) are reported.
Potato specimens were collected immediately after the last application and 7 ± 1, 14 ± 1 and 21 ± 1 day after the last application.
The specimens were analyzed according to BASF analytical method number 567/0 for alpha-cypermethrin and to Nippon Soda CO. LTD method RD-9991 N2 for acetamiprid.
Both methods have a limit of quantification of 0.01 mg/kg in all sample materials. Procedural recoveries averaged 92% for alpha-cypermethrin and 84% for acetamiprid, respectively, at fortification levels between 0.01 and 1 mg/kg.
For alpha-cypermethrin, at all samplings from plot 102 and 103, in potato tubers no residues above the LOQ (0.01 mg/kg) were determined.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description:	BAS 370 00 I BAS 310 40 I BAS 9111 0 I
Lot/Batch #:	1463 (BAS 370 00 I, 25 g/L alpha-cypermethrin, 100 g/L acetamiprid, SL) 1209 (BAS 310 40 I, 100 g/L alpha-cypermethrin, EC) FRE-000429 (BAS 9111 0 I, 200 g/kg acetamiprid)
Purity:	not relevant
CAS#:	alpha-cypermethrin: 67375-30-8 Acetamiprid: 160430-64-8
Development code:	not applicable
Spiking levels:	0.01-1.0 mg/kg

2. Test Commodity:

Crop:	Potatoes
Type:	Root and tuber vegetables
Variety:	Agria, Condor, Mona Lisa
Botanical name:	*Solanum tuberosum* L.
Crop parts(s) or processed commodity:	Potato tubers
Sample size:	min. 2.0 kg

B. STUDY DESIGN

1. Test procedure

During the 2007 growing season, a total of four trials were conducted on potato in order to determine the magnitude of residues of active ingredient(s) in or on Raw Agricultural Commodities (RAC).

Plot 101: Untreated (control)

Plot 102: BAS 370 00 I, a SL formulation of alpha-cypermethrin (25 g/L) and acetamiprid (100 g/L), was applied twice at the rate of 500 mL/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).

Plot 103: BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g/L), was applied twice at the rate of 125 mL/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).

Plot 104: BAS 9111 0 I, a WP formulation of acetamiprid (200 g/kg), was applied twice at the rate of 250 g/ha, in order to determine the magnitude of the residues of active ingredient in or on

Raw Agricultural Commodities (RAC).

In context of this summary, only the results of alpha-cypermethrin (plot 102 & plot 103) are reported.
Potato specimens were collected immediately after the last application and 7 ± 1, 14 ± 1 and 21 ± 1 day after the last application.

Table 2.4.1-2: Target application rates and timings for potatoes

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2007	4	2	F	BAS 370 00 I (SL)	alpha-cypermethrin acetamiprid	0.0125 0.05	400	21±1 DBH 14±1 DBH
			F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125		
			F	BAS 9111 0 I (WP)	acetamiprid	0.05		

DBH = Days before harvest

2. Description of analytical procedures

The specimens were analyzed according to BASF analytical method number 567/0 for alpha-cypermethrin and to Nippon Soda CO. LTD method RD-9991 N2 for acetamiprid.
Both methods have a limit of quantification of 0.01 mg/kg in all sample materials. Procedural recoveries averaged 92% for alpha-cypermethrin and 84% for acetamiprid, respectively, at fortification levels between 0.01 and 1 mg/kg.
Alpha-cypermethrin: the residues of alpha-cypermethrin are extracted from plant matrices using a mixture of methanol, water and HCl 2 mol/L. For clean-up a liquid/liquid partition against cyclohexane is used. The final determination of alpha-cypermethrin is performed by LC/MS/MS. The limit of quantification (LOQ) ofthe method is 0.01 mg/kg.
Acetamiprid: The residues of acetamiprid are extracted from plant matrices using a mixture of methanol and water. For clean-up a liquid/liquid partition against dichloromethane is used. The final determination of acetamiprid is performed by LC/MS/MS. The limit of quantification (LOQ) ofthe method is 0.01 mg/kg.

Table 2.4.1-3: Summary of recoveries of alpha-cypermethrin and acetamiprid in potato tubers

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
potato tubers	0.01 / 1.0	6	92	7	6
RD-9991 N2		acetamiprid			
potato tubers	0.01 / 1.0	5	84	4	3

II. RESULTS AND DISCUSSION

For alpha-cypermethrin, at all samplings from plot 102 and 103, in potato tubers no residues above the LOQ (0.01 mg/kg) were determined.

An overall summary of the residues is given in the table below.

Table 2.4.1-4: Summary of residues of BAS 310 I in potato tubers from trials according to critical GAP after application of BAS 310 40 I and BAS 370 00 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin (BAS 310 I)
Potato	2007	2	BAS 370 00 I (SL)	0	44-68	tubers	<0.01
				7±1	45-75		<0.01
				14±1*	47-85		<0.01
				21±1	48-95		<0.01
			BAS 310 40 I (EC)	0	44-68	tubers	<0.01
				7±1	45-75		<0.01
				14±1*	47-85		<0.01
				21±1	48-95		<0.01

DALA = days after last application
BBCH = growth stage at respective sampling
* harvest

III. CONCLUSION

At all sampling events (between 0 and 21 days after the last application of either formulation BAS 370 00 I or BAS 310 40 I), in potato tuber specimens no residues of alpha-cypermethrin above the LOQ (0.01 mg/kg) were determined.

Table 2.4.1-5: Residues of BAS 310 I after two applications of the formulations BAS 370 00 I and BAS 310 40 I in Northern and Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 264991 **Doc ID:** 2008/1002704 **Trial No.:** L070446 **GLP:** yes **Year** 2007	Potato	France (N)	BAS 370 00 I 2 x 0.0125	48	0	tubers	<0.01
					6	tubers	<0.01
					14	tubers	<0.01
					21	tubers	<0.01
			BAS 310 40 I 2 x 0.0125	48	0	tubers	<0.01
					6	tubers	<u><0.01</u>
					14	tubers	<0.01
					21	tubers	<0.01
Study code: 264991 **Doc ID:** 2008/1002704 **Trial No.:** L070447 **GLP:** yes **Year** 2007	Potato	The Netherlands (N)	BAS 370 00 I 2 x 0.0125	44	0	tubers	<0.01
					8	tubers	<0.01
					14	tubers	<0.01
					22	tubers	<0.01
			BAS 310 40 I 2 x 0.0125	44	0	tubers	<0.01
					8	tubers	<u><0.01</u>
					14	tubers	<0.01
					22	tubers	<0.01
Study code: 264991 **Doc ID:** 2008/1002704 **Trial No.:** L070448 **GLP:** yes **Year** 2007	Potato	Spain (S)	BAS 370 00 I 2 x 0.0125	65	0	tubers	<0.01
					8	tubers	<0.01
					15	tubers	<0.01
					21	tubers	<0.01
			BAS 310 40 I 2 x 0.0125	65	0	tubers	<0.01
					8	tubers	<0.01
					15	tubers	<u><0.01</u>
					21	tubers	<0.01
Study code: 264991 **Doc ID:** 2008/1002704 **Trial No.:** L070449 **GLP:** yes **Year** 2007	Potato	France (S)	BAS 370 00 I 2 x 0.0125	68	0	tubers	<0.01
					7	tubers	<0.01
					14	tubers	<0.01
					21	tubers	<0.01
			BAS 310 40 I 2 x 0.0125	68	0	tubers	<0.01
					7	tubers	<0.01
					14	tubers	<u><0.01</u>
					21	tubers	<0.01

DALA = days after last application
BBCH = growth stage at respective sampling
_ underlined values were used for MRL calculation

Report:	**Moreno S., 2013b**
Title:	Study on the residue behaviour of Alpha-Cypermethrin in potato after treatment with either BAS 310 40 I or BAS 310 55 I under field conditions in North and South Europe, season 2011
Document No:	BASF DocID 2012/1157550
Guidelines:	EEC 87/18 (18 December 1986), International guidelines for distribution and pesticides application AEPLA FAO 1985, EC 1107/2009 of the European Parliament and of the Council of 21 Oct 2009, EEC 79/117, EEC 91/414, EEC 7029/VI/95 rev. 5 Appendix B
GLP	yes

Executive Summary

During the growing season of 2011, a total of four trials with potato was conducted in Germany, The United Kingdom, Italy and Spain, to determine the magnitude and decline of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC). Therefore two solo formulations BAS 310 55 I (50 g/L BAS 310 I, ME) and BAS 310 40 I (100 g/L BAS 310 I, EC) were applied twice to the treated plots at a rate equivalent of 0.015 kg a.s./ha in a nominal spray volume of 200 l/ha at 21±1 and 14±1 days before harvest. The applications were made at crop stages between BBCH 43 and 48.

Potato tuber specimen were taken for analysis directly after the last application as well as 7-8, 13-15 and 20-21 days thereafter. Specimens were analysed for alpha-cypermethrin using BASF method No. 567/0. The method has a limit of quantitation of 0.01 mg/kg.

At all the field trials, residues of alpha-cypermethrin in all treated potato samples were below the LOQ (< 0.01 mg/kg) after treatment with both formulations at all sampling times.

The analytical results obtained demonstrate that the treatment with two applications of BAS 310 55 I and BAS 310 40 I did not lead to different residue results in the raw agricultural commodity at harvest.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 55 I (ME)
BAS 310 40 I (EC)
Lot/Batch #: 101198 BAS 310 55 I, 50 g/L alpha-cypermethrin
1209 BAS 310 40 I, 100 g/L alpha-cypermethrin
Purity:
CAS#: 67375-30-8 alpha-cypermethrin
Development code:
Spiking levels: 0.01-5.0 mg/kg

2. Test Commodity:

Crop: Potato
Type: Root and tuber vegetables
Variety: Bintje, Cara, Spunta, Liseta
Botanical name: *Solanum tuberosum*
Crop parts(s) or processed
Commodity: Fruit
Sample size: min. 24 units/1.0-5.70 kg

B. STUDY DESIGN

1. Test procedure

During the growing season of 2011, a total of four trials with potato was conducted in Germany, The United Kingdom, Italy and Spain, to determine the magnitude and decline of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC). Therefore two solo formulations BAS 310 55 I (50 g/L BAS 310 I, ME) and BAS 310 40 I (100 g/L BAS 310 I, EC) were applied twice to the treated plots at a rate equivalent of 0.015 kg a.s./ha in a nominal spray volume of 200 l/ha at 21±1 and 14±1 days before harvest. The applications were made at crop stages between BBCH 43 and 48.
Potato tuber specimen were taken for analysis directly after the last application as well as 7-8, 13-15 and 20-21 days thereafter.

Table 2.4.1-6: Target application rates and timings for potato

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2008	4	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.015	200	1st appl.: 21±1 DBH 2nd appl.: 14±1 DBH
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.015	200	1st appl.: 21±1 DBH 2nd appl.: 14±1 DBH

1) 2 litres of water plus 1 litre of product to make up a spray volume of 3 litres
2) DBH: days before harvest

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).

The residues of alpha-cypermethrin in potato specimens were extracted using a mixture of methanol/water/hydrochloric acid (70:25:5, v/v/v). After centrifugation, an aliquot of the extract was partitioned into cyclohexane. An aliquot of the cyclohexane phase was evaporated and the residue was taken up into methanol/water (80:20, v/v). The final determination of alpha-cypermethrin was performed by LC-MS/MS using the ammonium adduct of alpha-cypermethrin. The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery averaged 103±7% (mean±RSD) for alpha-cypermethrin at fortification levels of 0.01 mg/kg -5.0 mg/kg.

Table 2.4.1-7: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in potato

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
Method No. 567/0		alpha-cypermethrin		
Potatoes	0.01	5	99	9
	5.0	5	107	3

II. RESULTS AND DISCUSSION

The residue levels of alpha-cypermethrin (BAS 310 I) in potato specimens taken directly after the last application of BAS 310 55 I (0 DALA) and at 7-8, 13-15 and 20-21 DALA were all below the limit of quantitation of the analytical method (LOQ, <0.01 mg/kg).
Residues found in potato specimens after the application of BAS 310 40 I lead to similar residue results of alpha-cypermethrin, with all residue results at 0, 7-8, 13-15 and 20-21 DALA being below the LOQ. The analytical results obtained demonstrate that the treatment with two applications of BAS 310 55 I and BAS 310 40 I did not lead to different residue results in the raw agricultural commodity at harvest.
In all untreated samples from trials L110423 through L110426, residues of alpha-cypermethrin were below the LOQ (<0.01 mg/kg).
An overall summary of the residues is given in the table below.

Table 2.4.1-8: Summary of residues of BAS 310 I in potato after application of BAS 310 55 I and BAS 310 40 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	alpha-cypermethrin (BAS 310 I)
Potato	2011	2	BAS 310 55 I (ME)	0	47-48	tuber	< 0.01
				7-8	47-48		< 0.01
				13-15	48-49		< 0.01
				20-21	48-49		< 0.01
		2	BAS 310 40 I (EC)	0	47-48	tuber	< 0.01
				7-8	47-48		< 0.01
				13-15	48-49		< 0.01
				20-21	48-49		< 0.01

DALA = days after last application
BBCH = growth stage at respective sampling

III. CONCLUSION

At all the field trials, residues of alpha-cypermethrin in all treated potato samples were below the LOQ (< 0.01 mg/kg) after treatment with both formulations at all sampling times.
The analytical results obtained demonstrate that the treatment with two applications of BAS 310 55 I and BAS 310 40 I did not lead to different residue results in the raw agricultural commodity at harvest.

Table 2.4.1-9: Residues of BAS 310 I after two applications of the formulation BAS 310 55 I and BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408608 **Doc ID:**2012/1157550 **Trial No.:**L110423 **GLP:**yes **Year**2011	Potato	Germany	BAS 310 55 I BAS 310 I 2 x 0.015	47	0	tuber	<0.01
				48	7	tuber	<u><0.01</u>
				49	13	tuber	<0.01
				49	20	tuber	<0.01
			BAS 310 40 I BAS 310 I 2 x 0.015	47	0	tuber	<0.01
				48	7	tuber	<0.01
				49	13	tuber	<0.01
				49	20	tuber	<0.01
Study code:408608 **Doc ID:**2012/1157550 **Trial No.:**L110424 **GLP:**yes **Year**2011	Potato	UK	BAS 310 55 I BAS 310 I 2 x 0.015	47-48	0	tuber	<0.01
				47-48	7	tuber	<u><0.01</u>
				48-49	15	tuber	<0.01
				49	20	tuber	<0.01
			BAS 310 40 I BAS 310 I 2 x 0.015	47-48	0	tuber	<0.01
				47-48	7	tuber	<0.01
				48-49	15	tuber	<0.01
				49	20	tuber	<0.01

DALA = days after last application
BBCH = growth stage at respective sampling
_ underlined values were used for MRL calculation

Table 2.4.1-10: Residues of BAS 310 I after two applications of the formulation BAS 310 55 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408608 **Doc ID:**2012/1157550 **Trial No.:**L110425 **GLP:**yes **Year**2011	Potato	Italy	BAS 310 55 I BAS 310 I 2 x 0.015	47	0	tuber	<0.01
				48	8	tuber	<0.01
				49	14	tuber	<u><0.01</u>
				49	21	tuber	<0.01
			BAS 310 40 I BAS 310 I 2 x 0.015	47	0	tuber	<0.01
				48	8	tuber	<0.01
				49	14	tuber	<0.01
				49	21	tuber	<0.01
Study code:408608 **Doc ID:**2012/1157550 **Trial No.:**L110426 **GLP:**yes **Year**2011	Potato	Spain	BAS 310 55 I BAS 310 I 2 x 0.015	47	0	tuber	<0.01
				48	7	tuber	<0.01
				48-49	14	tuber	<u><0.01</u>
				48-49	21	tuber	<0.01
			BAS 310 40 I BAS 310 I 2 x 0.015	47	0	tuber	<0.01
				48	7	tuber	<0.01
				48-49	14	tuber	<0.01
				48-49	21	tuber	<0.01

DALA = days after last application
BBCH = growth stage at respective sampling
_ underlined values were used for MRL calculation

2.4.2 Estimation of MRL, HR and STMR for potato

For *potato*, the following residue studies were considered (BASF DocIDs): 2006/1026846, 2007/1007945, 2008/1002704, and 2012/1157550.

The following residue values (PHI=7±1 days for EU-N or PHI=14±1 days for EU-S) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026846	<0.01 (4 x)	<0.01 (4 x)
2007/1007945	<0.01 (4 x)	<0.01 (4 x)
2008/1002704	<0.01 (2 x)	<0.01 (2 x)
2012/1157550	<0.01 (2 x)	<0.01 (2 x)
OECD-MRL-calculation	**0.01 (n=12, STMR=0.01, HR=0.01)**	**0.01 (n=12, STMR=0.01, HR=0.01)**

_ underlined values were used for risk assessment purposes

2.5 Carrot

Residue data from supervised trials in carrots were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.5-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Carrot	2004	3	DE	-	-	3	2005/1035294
Carrot	2005	6	DE	-	-	6	2006/1029533
Total number of trials per region		**9**			**Total number of trials**	**9**	

2.5.1 Supervised residue trials in carrot

Residue trials in carrot were performed in 2004 the results of which were submitted to Bundesamt für Verbraucherschutz und Lebensmittelsicherheit (BVL) in 2006 (Submission No. **024018-00).**

Anonymous (2006)

Full study reference

Anonymous (2006): Residue behaviour of Alpha-Cypermethrin in/on cucumbers, carrots, celeriac and salad onions (onions including leaves) after outdoor application of Fastac SC (SC 100) in Germany, 2004; BASF DocID 2005/1035294

Anonymous (2006)

Full study reference

Anonymous (2006): Residue behaviour of Alpha-Cypermethrin in/on fennel, curly cale, cucumbers, carrots, radies, celery, celeriac and bunshing or green onions after outdoor application of Fastac SC (SC 100) in Germany, 2005; BASF DocID 2006/1029533

Material and Methods:

In the years 2004 and 2005 a residue program in carrots was conducted in representative carrot growing areas in Germany.

Three residue decline trials were conducted in carrots in Germany during 2004. Carrots plants were treated with a 100 g a.s./L soluble concentrate formulation (BAS 310 41 I). Plots of carrots were treated with two applications at a rate of 12.5 g a.s./ha. The last application was intended to be made 14 days before harvest. Specimens of carrots were taken immediately after the last application, 3, 7 and 14 days after the final application.
The specimens were analysed for residues of BAS 310 I using DFG method S19 which has a limit of quantitation of 0.01 mg/kg for carrots.

During the 2005 growing season, six field trials were conducted in Germany in order to determine the residues of alpha-cypermethrin in carrots after application of a 100 g a.s./L soluble concentrate formulation (BAS 310 41 I). Plots of carrots were treated with two applications at a rate of 12.5 g a.s./ha. The growth stage of the carrot plants was between 6-7 leaves and BBCH 77. Carrot specimens were sampled directly after the last application as well as 3, 7 and 14-15 days thereafter.

The samples were analysed for alpha-cypermethrin by means of HPLC-MS/MS according to BASF analytical method No. 567/0, which has a limit of quantitation of 0.005 mg/kg.

The trial data and residue results are summarised in Table 2.5.1-1.

Table 2.5.1-1: Residues in carrots

GAP for Germany is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3-7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF DocID 2005/1035294 Trial No. RU-I-13 04 RP NW 2/1 Study to GLP (analytical part) Study carried out in 2004	Carrot (variety Nandera)	Germany 67105 Schifferstadt	12.5 g a.s./ha 2 treatm. last date 21.07.04	14 days before harvest	0 3 7 14	beet with leaf <0.01 beet <u>≤0.01</u> beet <0.01 beet <0.01	DFG method S19 carrot, beets: mean recovery =80.6 %; SD: +/- 10.3; CV: 12.8; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as alpha-cypermethrin
BASF DocID 2005/1035294 Trial No. RU-I-13 04 RP NW 2/2 Study to GLP (analytical part) Study carried out in 2004	Carrot (variety Nandera)	Germany 67105 Schifferstadt	12.8 g a.s./ha 2 treatm. last date 03.08.04	14 days before harvest	0 3 7 14	beet with leaf <0.01 beet <u>≤0.01</u> beet <0.01 beet <0.01	
BASF DocID 2005/1035294 Trial No. RU-I-13 04 RP NW 2/3 Study to GLP (analytical part) Study carried out in 2004	Carrot (variety Napoli)	Germany 67105 Schifferstadt	12.5 g a.s./ha 2 treatm. last date 30.08.04	BBCH 405/407	0 3 7 14	beet with leaf <0.01 beet <u>≤0.01</u> beet <0.01 beet <0.01	
BASF DocID 2006/1029533 Trial No. RU-I-06 05 NW BN 2/1 = GLP 05/029 Study to GLP (analytical part) Study carried out in 2005	Carrot (variety Dordogne)	Germany 53299 Bonn	12.5 g a.s./ha 2 treatm. last date 27.09.05	6-7 leaves	0 3 7 14	beet <0.01 beet <u>≤0.01</u> beet <0.01 beet <0.01	BASF analytical method No. 567/0 carrot, beets: mean recovery = 92-97 %; CV: 2.7-5.2 %; fortification range 0.005-0.1 mg/kg Residue analysed as alpha-cypermethrin
BASF DocID 2006/1029533 Trial No. RU-I-06 05 NW BN 221 = GLP 05/030 Study to GLP (analytical part) Study carried out in 2005	Carrot (variety Yukon)	Germany 53299 Bonn	12.5 g a.s./ha 2 treatm. last date 04.10.05	6-7 leaves	7 14	beet <u>≤0.01</u> beet <0.01	
BASF DocID 2006/1029533 Trial No. RU-I-06 05 SH KI 2/1 Study to GLP (analytical part) Study carried out in 2005	Carrot (variety Napoli)	Germany 24229 Birkenmoor	12.5 g a.s./ha 2 treatm. last date 29.07.05	EC 47-48	7	beet <u>≤0.01</u>	
BASF DocID 2006/1029533 Trial No. RU-I-06 05 SH KI 2/2 Study to GLP (analytical part) Study carried out in 2005	Carrot (variety Bolero)	Germany 24229 Birkenmoor	12.5 g a.s./ha 2 treatm. last date 29.07.05	EC 44-45	7	beet <u>≤0.01</u>	
BASF DocID 2006/1029533 Trial No. RU-I-06 05 BY FS 2/1 Study to GLP (analytical part) Study carried out in 2005	Carrot (variety Tino)	Germany 85452 Eichenried	12.5 g a.s./ha 2 treatm. last date 14.09.05	BBCH 76	7 14	beet <u>≤0.01</u> beet <0.01	

Table 2.5.1-1: Residues in carrots

GAP for Germany is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3-7 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF DocID 2006/1029533 Trial No. RU-I-06 05 BY FS 2/2 Study to GLP (analytical part) Study carried out in 2005	Carrot (variety Tino)	Germany 85452 Eichenried	12.5 g a.s./ha 2 treatm. last date 19.09.05	BBCH 77	7 15	beet <u>≤0.01</u> beet <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in carrots presented in the alpha-cypermethrin dossier were carried out in 1 EU countries and do not provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for Germany: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 3-7 days

The following residue studies were presented:

- two treatments at a target rate of 12.5 g a.s./ha–9 trials, all conducted in Germany

In all trials presented above no residues above the method LOQ (0.01 mg/kg) were found in any of the treated carrot samples.

Conclusion:

In the years 2004 and 2005 a two residue studies in carrots were conducted in Germany. A total of 9 trials were performed that support the proposed GAP for Germany. In all trials no residues above the method LOQ (0.01 mg/kg) were found in any of the treated carrot samples.

2.5.2 Estimation of MRL, HR and STMR for carrot

For *carrot*, the following residue studies were considered (BASF DocIDs): 2005/1035294 and 2006/1029533.

The following residue values (PHI=3-7 days) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2005/1035294	<0.01 (3x)	-
2006/1029533	<0.01 (6x)	-
OECD-MRL-calculation	**0.01 (n=9, STMR=0.01, HR=0.01)**	**-**

_ underlined values were used for risk assessment purposes

Turnip, swedes, rutabaga, radish, horseradish, salsify, parsley roots and celeriac

As the critical GAPs for carrots, turnip, swedes, rutabaga, radish, horseradish, salsify, parsley roots and celeriac are identical, according to European Community Guideline 7525/VI/95 rev. 9 dated March 2011 extrapolation from carrots to turnip, swedes, rutabaga, radish, horseradish, salsify, parsley roots and celeriac is adequate.

2.6 Onions

Residue data from supervised trials in onions were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.6-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Onion	2005	4	FR, NL, UK	-	-	4	2006/1026854
Onion	2006	4	DE, FR, NL	-	-	4	2007/1008499
Total number of trials per region		**8**		**-**	**Total number of trials**	**8**	

2.6.1 Supervised residue trials in onions

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of BAS 310 I in bulb onion after treatment with BAS 310 40 I under field conditions in Northern Europe during 2005; BASF DocID 2006/1026854

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the residue behaviour of BAS 310 I in onion after treatment with BAS 310 40 I under open field conditions in Northern Europe, 2006, BASF DocID 2007/1008499

Material and Methods:

A residue program in bulb onions was conducted in the years 2005 and 2006 in representative onion growing areas in France (Northern European region), Germany, the Netherlands and the United Kingdom under open field conditions.

During the 2005 growing season, four trials were conducted in bulb onions in France (Northern European region), the Netherlands and the United Kingdom. Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I) was foliar applied to onion plants in two variants. In one variant, the product was applied once at target rate of 12.5 g a.s./ha. This was compared to another variant in which two applications were made at the same target rate. The last application took place at growth stages between 47 (bolting begins; in 10% of the plants leaves bent over) and 48 (leaves bent over in 50% of plants). Onion bulb specimens were collected directly after the last application as well as 3-4, 7 and 14 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, four trials were conducted in bulb onions in France (Northern European region), Germany and the Netherlands. Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I) was foliar applied to onion plants in two variants. In one variant, the product was applied once at target rate of 12.5 g a.s./ha. This was compared to another variant in which two applications were made at the same target rate. The last application took place at growth stages between 44 (bulb enlarges) and 47 (bolting begins; in 10% of the plants leaves bent over). Onion bulb specimens were collected directly after the last application as well as 3-4, 7-8 and 14-15 days thereafter. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.6.1-1.

Table 2.6.1-1: Residues in onions

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/1 Study to GLP Study carried out in 2005	Bulb onion (variety Hyskin)	United Kingdom Southery, Downham Market Norfolk PE38 0NL	12.5 g a.s./ha 08.08.05	47	0 3 7 14	bulb <0.01 bulb 0.012 bulb <0.01 bulb <0.01	BASF analytical method No. 567/0 onion, bulb: mean recovery = 87.2%; SD: +/- 13.9; CV: 15.9%; n=3; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/2 Study to GLP Study carried out in 2005	Bulb onion (variety Summit)	France 45300 Rouvres St Jean (North of EU)	12.5 g a.s./ha 05.08.05	47-48	0 3 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/3 Study to GLP Study carried out in 2005	Bulb onion (variety Sumit)	France 45 Aulnay La Rivière, Loiret, 45390 (North of EU)	12.5 g a.s./ha 05.08.05	47-48	0 3 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/4 Study to GLP Study carried out in 2005	Bulb onion (variety Hyfort)	The Netherlands 6678 PB Oosterhout Gelderland	12.5 g a.s./ha 18.08.05	47	0 4 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2007/1008499 Trial No. A/NF/I/06/150 Study to GLP Study carried out in 2006	Onion (variety Barito)	France Bignicourt Champgne-Ardennes (North of EU)	13 g a.s./ha 22.08.06	47	0 3 8 15	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	BASF analytical method No. 567/0 onion, bulb: mean recovery = 85.6%; SD: +/- 5.5; CV: 6.5%; n=5; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008499 Trial No. A/GE/I/06/151 Study to GLP Study carried out in 2006	Onion (variety Stuttgarter Riesen)	Germany Schmilau Schleswig-Holstein	13 g a.s./ha 13.08.06	44	0 3 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2007/1008499 Trial No. A/GE/I/06/152 Study to GLP Study carried out in 2006	Onion (variety Stüron)	Germany Crossen (Sachsen)	12 g a.s./ha 24.08.06	44-45	0 4 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2007/1008499 Trial No. A/NL/I/06/153 Study to GLP Study carried out in 2006	Onion (variety Donna)	The Netherlands NB Elst Gelderland	12 g a.s./ha 09.08.06	46-47	0 3 7 15	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/1 Study to GLP Study carried out in 2005	Bulb onion (variety Hyskin)	United Kingdom Southery, Downham Market Norfolk PE38 0NL	12.5 g a.s./ha 2 treatm. last date 08.08.05	47 at last treatm.	0 3 7 14	bulb 0.017 bulb 0.014 bulb <u>0.010</u> bulb <0.01	BASF analytical method No. 567/0 onion, bulb: mean recovery = 87.2%; SD: +/- 13.9; CV: 15.9%; n=3; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin

Table 2.6.1-1: Residues in onions

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/2 Study to GLP Study carried out in 2005	Bulb onion (variety Summit)	France 45300 Rouvres St Jean (North of EU)	12.5 g a.s./ha 2 treatm. last date 05.08.05	47-48 at last treatm.	0 3 7 14	bulb 0.013 bulb <0.01 bulb 0.010 bulb <0.01	
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/3 Study to GLP Study carried out in 2005	Bulb onion (variety Sumit)	France 45 Aulnay La Rivière, Loiret, 45390 (North of EU)	12.5 g a.s./ha 2 treatm. last date 05.08.05	47-48 at last treatm.	0 3 7 14	bulb 0.016 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2006/1026854 Trial No. AF/8827/BA/4 Study to GLP Study carried out in 2005	Bulb onion (variety Hyfort)	The Netherlands 6678 PB Oosterhout Gelderland	12.5 g a.s./ha 2 treatm. last date 18.08.05	47 at last treatm.	0 4 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2007/1008499 Trial No. A/NF/I/06/150 Study to GLP Study carried out in 2006	Onion (variety Barito)	France Bignicourt Champgne-Ardennes (North of EU)	13 g a.s./ha 2 treatm. last date 22.08.06	47 at last treatm.	0 3 8 15	bulb 0.014 bulb <0.01 bulb <0.01 bulb <0.01	BASF analytical method No. 567/0 onion, bulb: mean recovery = 85.6%; SD: +/- 5.5; CV: 6.5%; n=5; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008499 Trial No. A/GE/I/06/151 Study to GLP Study carried out in 2006	Onion (variety Stuttgarter Riesen)	Germany Schmilau Schleswig-Holstein	13 g a.s./ha 2 treatm. last date 13.08.06	44 at last treatm	0 3 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2007/1008499 Trial No. A/GE/I/06/152 Study to GLP Study carried out in 2006	Onion (variety Stüron)	Germany Crossen (Sachsen)	13 g a.s./ha 2 treatm. last date 24.08.06	44-45 at last treatm.	0 4 7 14	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	
BASF Doc ID 2007/1008499 Trial No. A/NL/I/06/153 Study to GLP Study carried out in 2006	Onion (variety Donna)	The Hetherlands NB Elst Gelderland	13 g a.s./ha 2 treatm. last date 09.08.06	46-47 at last treatm.	0 3 7 15	bulb <0.01 bulb <0.01 bulb <0.01 bulb <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue data presented in the alpha-cypermethrin dossier were carried out in 4 different EU countries and provide data relevant to conditions in the Northern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha–8 trials including both variants, all conducted in the Northern European region

After one application at a target rate of 12.5 g a.s./ha, no residues above the limit of quantitation of the analytical method (LOQ, 0.01 mg/kg) were found in treated onion bulbs sampled 0–15 days after application. The only exception was one isolated finding at the LOQ (0.012 mg/kg) 3 days after application in trial AF/8827/BA/1. Residues in all other treated bulb samples taken from the same plot 0, 7 and 14 days after application were <0.01 mg/kg.

After two applications at a target rate of 12.5 g a.s./ha, initial residues in onion bulbs ranged between <0.01–0.017 mg/kg. Residues declined to <0.01-0.014 mg/kg and <0.01-0.010 mg/kg after 3-4 and 7-8 days and were below the LOQ (<0.01 mg/kg) in all trials 14-15 days after the last application.

Conclusion:

A residue program in bulb onions was conducted in the years 2005 and 2006 in representative onion growing areas in France (Northern European region), Germany, the Netherlands and the United Kingdom under open field conditions. In 8 trials supporting the proposed GAP for the Northern European region, residues in onion bulbs ranged between <0.01-0.010 mg/kg at the target PHI of 7±1 days.

2.6.2 Estimation of MRL, HR and STMR for onions

For *onions*, the following residue studies were considered (BASF DocIDs): 2006/1026854 and 2007/1008499.

The following residue values (PHI=7±1 days) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026854	<0.01 (2x), 0.01 (2x)	-
2007/1008499	<0.01 (4x)	-
OECD-MRL-calculation	**0.01 (n=8, STMR=0.01, HR=0.01)**	**-**

_ underlined values were used for risk assessment purposes

2.7 Pepper, sweet

Residue data from supervised trials in sweet pepper were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.7-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Pepper, sweet (field)	2005	-	-	4	ES, FR, GR, IT	4	2006/1026860
Pepper, sweet (field)	2006	-	-	4	ES, FR, GR, IT	4	2007/1008497
Pepper, sweet (glasshouse)	2005	1	BE	5	ES, FR, GR, IT	5	2006/1036933
Total number of trials per region		**1 (glasshouse)**			**Total number of trials**	**8 (field) 5 (glasshouse)**	

2.7.1 Supervised residue trials in pepper, sweet

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of alpha-cypermethrin in Peppers after treatment with BAS 310 40 I under field conditions in Southern Europe during 2005; BASF Doc-ID 2006/1026860

Schulz H (2007)

Full study reference

Schulz H (2005): Study on the Residue Behaviour of Alpha-cypermethrin in Peppers after Treatment with BAS 310 40 I under Greenhouse Conditions in Southern France, Belgium, Italy, Spain and Greece, 2005; BASF Doc-ID 2006/1036933

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the residue behaviour of BAS 310 I in sweet pepper after treatment with BAS 310 40 I under open field conditions in Southern Europe, 2006; BASF Doc-ID 2007/1008497

Material and Methods:

During the growing seasons 2005 and 2006, a residue program in sweet pepper was conducted in different representative pepper growing areas in Belgium, France (Southern European region), Greece, Italy, and Spain under open field and glasshouse conditions.

In the year 2005, four field trials were conducted in France (Southern European region), Greece, Italy and Spain to determine the residue level of alpha-cypermethrin after application as a emulsifiable concentrate (EC 100 g a.s./L; BAS 310 40 I). The product was foliar applied once at a target rate of 25 g a.s./ha 7 days before expected harvest. The growth stages of the plants at application were between 68 (8^{th} inflorescence: first flower open) and 87 (70% of fruits show typical fully ripe colour).
Specimens of fruit were collected at the day of application from each plot as well as 2-3, 6-7 and 13-14 days thereafter.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS using BASF method no. 567/0 with a limit of quantification of 0.01 mg/kg in all sample materials.

In the year 2006, four field trials were conducted in France (Southern European region), Greece, Italy and Spain to determine the residue level of alpha-cypermethrin after application as a emulsifiable concentrate (EC 100 g a.s./L; BAS 310 40 I). A single treatment was applied at a target rate of 25 g a.s./ha 7 days before expected harvest. The growth stages of the plants at application were between 72 (2nd fruit has reached typical size and form) and 87 (70% of fruits show typical fully ripe colour).
Specimens of fruit were collected at the day of application from each plot as well as 3-4, 7-8 and 13-14 days thereafter.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS using BASF method no. 567/0 with a limit of quantification of 0.01 mg/kg in all sample materials.

Six glasshouse trials in sweet pepper were performed during the growing season 2005 in Belgium, France (Southern European region), Greece, Italy and Spain.
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L; BAS 310 40 I) was foliar applied to sweet pepper plants once at a target rate of 40 g a.s./ha. The growth stages of the plants were between 72 (2nd fruit has reached typical size and form) and 89 (Fully ripe: fruits have typical fully ripe colour) at application. Samples of sweet pepper fruit were collected immediately after application as well as 3, 7 and 14 days afterwards.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS using BASF method no. 567/0 with a limit of quantification of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.7.1-1 and Table 2.7.1-2.

Table 2.7.1-1: Residues in sweet pepper – open field trials

GAP for EU-S is 1 applications with 30 g as/ha at infestation as an overall spray, PHI 7 (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026860 Trial No. AF/8820/BA/1 Study to GLP Study carried out in 2005	Pepper (variety Albi)	France Labarthe; Tarn-et-Garonne 82220 (South of EU)	25 g a.s./ha 28.06.05	68	0 3 7 14	fruit <0.01 fruit <0.01 fruit <u><0.01</u> fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 105.2%(104.3-106.1%); SD: +/- n/a; CV: n/a; n=2; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026860 Trial No. AF/8820/BA/2 Study to GLP Study carried out in 2005	Pepper (variety Negrillo)	Spain El Viso Del Alcor Sevilla, Andalucia 41520	25 g a.s./ha 20.06.05	71	0 3 7 14	fruit 0.010 fruit <0.01 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2006/1026860 Trial No. AF/8820/BA/3 Study to GLP Study carried out in 2005	Pepper (variety Senior)	Italy Castenaso, Bologna Emilia Romana, 40055	25 g a.s./ha 13.09.05	81-82	0 2 6 13	fruit 0.018 fruit 0.024 fruit <u>0.028</u> fruit <0.01	
BASF Doc ID 2006/1026860 Trial No. AF/8820/BA/4 Study to GLP Study carried out in 2005	Pepper (variety Florinis)	Greece Thessalonikis Central Macedonia GR-57200	25 g a.s./ha 30.07.05	87	0 3 7 14	fruit 0.032 fruit 0.026 fruit 0.014 fruit <u>0.015</u>	
BASF Doc ID 2007/1008497 Trial No. A/SF/I/06/162 Study to GLP Study carried out in 2006	Sweet pepper (variety Galilo)	France Chateaurenard Bouches-de-Rhône (South of EU)	25.1 g a.s./ha 03.07.06	81	0 4 8 14	fruit 0.065 fruit 0.011 fruit <u><0.01</u> fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 89.4%; SD: +/- 3.4; CV: 3.8%; n=3; fortification range 0.01 – 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008497 Trial No. A/IT/I/06/163 Study to GLP Study carried out in 2006	Sweet pepper (variety Quadrato)	Italy Costigliole D´Asti Piemont	25 g a.s./ha 29.08.06	8-802	0 3 8 13	fruit 0.032 fruit 0.015 fruit <u><0.01</u> fruit <0.01	
BASF Doc ID 2007/1008497 Trial No. A/SP/I/06/164 Study to GLP Study carried out in 2006	Sweet pepper (variety Stilo)	Spain Almussafes Valencia	24.7 g a.s./ha 20.06.06	72	0 3 8 13	fruit 0.033 fruit 0.025 fruit <u>0.024</u> fruit 0.012	
BASF Doc ID 2007/1008497 Trial No. A/GR/I/06/165 Study to GLP Study carried out in 2006	Sweet pepper (variety Laser F1)	Greece Profidis Thessaloniki Central Macedonia	24.8 g a.s./ha 11.07.06	87	0 3 7 14	fruit 0.019 fruit 0.030 fruit <u>0.017</u> fruit <0.01	

_ underlined values were used for MRL calculation

Table 2.7.1-2: Residues in sweet pepper – glasshouse trials

no current glasshouse GAP available							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1036933 Trial No. 05I CL FR P35 Study to GLP Study carried out in 2005	Sweet pepper (variety Hannibal)	France Chemin Madeleine 84800 Isle sur la Sorgue Provence (South of EU)	40 g a.s./ha 26.07. 05 glasshouse	73	0 3 7 14	fruit 0.016 fruit <u>0.016</u> fruit 0.015 fruit 0.011	BASF analytical method No. 567/0 fruit: mean recovery = 94.1%; SD: +/- 4.8; CV: 5.1%; n=4; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1036933 Trial No. 05I CL FR P39 Study to GLP Study carried out in 2005	Sweet pepper (variety Galileo)	France Chemin d´Arles 13870 Rognonas Provence (South of EU)	40 g a.s./ha 08.08. 05 glasshouse	72	0 3 7 14	fruit 0.016 fruit <u>0.017</u> fruit <0.01 fruit 0.013	
BASF Doc ID 2006/1036933 Trial No. 05ES086R Study to GLP Study carried out in 2005	Sweet pepper (variety Italico)	Spain Finca la Dehesilla 41710 Utrera Andalucia Sevilla	40 g a.s./ha 01.07. 05 glasshouse	75	0 3 7 14	fruit 0.031 fruit 0.026 fruit <u>0.029</u> fruit 0.023	
BASF Doc ID 2006/1036933 Trial No. 05RF047 Study to GLP Study carried out in 2005	Sweet pepper (variety Staboli)	Greece Profitis Thessaloniki Central Macedonia GR 57200	40 g a.s./ha 08.07. 05 glasshouse	81	0 3 7 14	fruit 0.049 fruit <u>0.016</u> fruit 0.016 fruit 0.016	
BASF Doc ID 2006/1036933 Trial No. G023-05 I Study to GLP Study carried out in 2005	Sweet pepper (variety Dazzle)	Belgium Rue Dominique Seret, 34 6210 Villers-Perwin Wagnelée (Hainaut)	40 g a.s./ha 20.09. 05 glasshouse	87-89	0 3 7 14	fruit 0.019 fruit 0.028 fruit 0.029 fruit <u>0.033</u>	
BASF Doc ID 2006/1036933 Trial No. IR05BASL51PL01 Study to GLP Study carried out in 2005	Sweet pepper (variety Quadrato d´Asti)	Italy Via Foggia, 110 70056 Molfetta (BA) Bari	40 g a.s./ha 27.07. 05 glasshouse	74	0 3 7 14	fruit 0.044 fruit 0.011 fruit <u>0.018</u> fruit 0.013	

_ underlined values were used for MRL calculation

Findings:

The residue trials in sweet pepper presented in the alpha-cypermethrin dossier were carried out in 5 different EU countries and provide data relevant to conditions in the Southern European region and to glasshouse conditions.
The proposed GAP is

- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days under open field conditions
- glasshouse GAP is a single application with 50 g a.s./ha applied at infestation as an overall spray with a PHI of 3 days

The following residue studies were presented:

- one treatment at a target rate of 25 g a.s./ha under open field conditions; 8 trials, all conducted in the Southern European region
- one treatment at a target rate of 40 g a.s./ha under glasshouse conditions – 6 trials, 5 of them conducted in the Southern European region and one conducted in the Northern European region

After one application at 25 g a.s./ha in the field, initial residues in fruit ranged between <0.01 mg/kg and 0.065 mg/kg and declined to <0.01 – 0.030 mg/kg, <0.01 – 0.028 mg/kg and <0.01 – 0.015 mg/kg 2-4, 6-8 and 13-14 days after application, respectively.

Under glasshouse conditions, initial residues between 0.016 mg/kg and 0.049 mg/kg were found after application of 40 g alpha-cypermethrin/ha. Residues declined to 0.011 – 0.028 mg/kg, <0.01 – 0.029 mg/kg and 0.011 – 0.033 mg/kg after 3, 7 and 14 days, respectively.

Conclusion:

After application of alpha-cypermethrin to sweet pepper plants according to the proposed GAP, determinable residues within a range below 0.1 mg/kg have to be expected in pepper fruit.

Residues found in sweet pepper fruit after treatment with alpha-cypermethrin according to the GAP for the glasshouse use show a tendency to higher values as compared to the open field situation. The residue levels determined range below 0.1 mg/kg.

2.7.2 Estimation of MRL, HR and STMR for pepper, sweet

For *sweet pepper*, the following residue studies were considered (BASF DocIDs): 2006/1026860, 2006/1036933 and 2007/1008497.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Southern Europe (S-EU) [mg/kg]
2006/1026860 (open field)	<0.01 (2x), 0.015, 0.028
2007/1008497 (open field)	<0.01 (2x), 0.0170, 0.024
OECD-MRL-calculation (open field)	**<u>0.05</u> (n=8, STMR=0.013, HR=0.028)**
2006/1036933 (glasshouse)	0.016 (2x), 0.017, 0.018, 0.029, 0.033
OECD-MRL-calculation (glasshouse)	**<u>0.07</u> (n=6, STMR=0.018, HR=0.033)**

_ underlined values were used for risk assessment purposes

2.8 Tomato

Residue data from supervised trials in tomatoes were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.8-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Tomato (field)	2005	4	BE, DE, FR	4	ES, FR, IT	8	2007/1008488
Tomato (field)	2006	4	DE, FR	4	ES, FR, GR, IT	8	2007/1008494
Tomato (field)	2006	2	DE; FR	2	ES, FR	4	2007/1007937
Tomato (field)	2008	2	DE, FR	2	IT, ES	4	2009/1090704
Tomato (glasshouse)	2004	4	DK, DE, FR, NL,	4	ES, FR, GR, IT	8	2004/5000719
Tomato (glasshouse)	2005	4	BE, DE, FR	4	ES, FR, GR, IT	8	2007/1007934
Total number of trials per region		**12 (field) 8 (glasshouse)**	-	**12 (field) 8 (glasshouse)**	**Total number of trials**	**24 (field) 16 (glasshouse)**	

Table 2.8-2: Processing studies available for tomato

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Tomato (glasshouse)	2004	4	DE, NL	-	-	4	2006/1021295
Total number of trials per region		**4**			**Total number of trials**	**4**	

2.8.1 Supervised residue trials in tomato

Report:	**Klaas P.,Ziske J., 2009c**
Title:	Study on the residue behaviour of Alpha-Cypermethrin in tomato after treatment with BAS 310 51 I and BAS 310 40 I under field conditions in Germany, Northern France, Italy and Spain, 2008
Document No:	BASF DocID 2009/1090704
Guidelines:	EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 1607/VI/97 rev. 2 10.06.1999
GLP	Yes

Executive Summary

During the growing season of 2008, a total of four trials with tomato was conducted in Germany, Northern France, Italy and Spain, to determine the magnitude and decline of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC). Therefore two solo formulations BAS 310 51 I (50 g/L BAS 310 I, ME) and BAS 310 40 I (100 g/L BAS 310 I, EC) were applied twice or once to the treated plot at a rate equivalent of 0.0150 kg a.s./ha or 0.0300 kg a.s./ha. In all trials the applications were made at crop stages BBCH 71 and 89.
For the analysis the tomato specimens were taken immediately after the last treatment (0 DALA) and at 2±1, 7±1 and 14 DALA. The tomato specimens were analysed for residues of BAS 310 I according to BASF Method No. 567/0, which has a limit of quantitation (LOQ) of 0.01 mg/kg. The analytical results obtained demonstrate that the treatment with one or two applications of BAS 310 51 I and BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 51 I (ME)
BAS 310 40 I (EC)
Lot/Batch #: 101156 BAS 310 51 I, 50 g/L alpha-cypermethrin
1209 BAS 310 40 I, 100 g/L alpha-cypermethrin
Purity:
CAS#: 67375-30-8 alpha-cypermethrin
Development code:
Spiking levels: 0.01-1.0 mg/kg

2. Test Commodity:

Crop: Tomato
Type: Fruiting vegetables
Variety: Vanessa, Topkapi, Asterix, Brillante
Botanical name: *Lycopersicon esculentum*
Crop parts(s) or processed
Commodity: Fruit
Sample size: 1.0-2.0 kg

B. STUDY DESIGN

1. Test procedure

During the growing season of 2008, a total of four trials with tomato were conducted in Germany, Northern France, Italy and Spain to determine the magnitude and decline of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC).
Each field trial consisted of three plots, one control plot (plot 1), one plot treated with BAS 310 51 I (plot 2) and another plot treated with BAS 310 40 I (plot 3).
In two of four trials (L080 173 and L080 174) both formulations (BAS 310 51 I, BAS 310 40 I) were applied twice at a rate equivalent to 0.015 kg alpha-cypermethrin/ha and for the others trials (L080175 and L080176) the application was made once at a rate equivalent to 0.0300 kg alpha-cypermethrin/ha. Both formulations were applied with the same GAP. The applications took place at BBCH 71-89 with an application rate of 400 L/ha. For the analysis the tomato specimens were taken immediately after the last treatment (0 DALA) and at 2±1, 7±1 and 14 DALA.

Table 2.8.1-1: Target application rates and timings for tomato

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2008	4	2	F	BAS 310 51 I (ME)	alpha-cypermethrin	0.0150	400	1st appl.: 72-76 BBCH 2nd appl.: 82-89 BBCH
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0150	400	1st appl.: 72-76 BBCH 2nd appl.: 82-89 BBCH
		1	F	BAS 310 51 I (ME)	alpha-cypermethrin	0.0300	400	1st appl.: 71-85 BBCH
		1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0300	400	1st appl.: 71-85 BBCH

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin). The residues of alpha-cypermethrin in the tomato specimens were extracted from plant matrices using a mixture of methanol/water/hydrochloric acid. A liquid/liquid partition against cyclohexane was used for clean up. The final determination of alpha-cypermethrin was performed by LC-MS/MS. The limit of quantitation (LOQ) of the method is 0.01 mg/kg. Procedural recoveries averaged 99±9% for alpha-cypermethrin at fortification levels of 0.01 mg/kg and 1.0 mg/kg for each matrix in all specimens.

Table 2.8.1-2: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in tomato

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Fruit	0.01-1.0	9	99	9	9

II. RESULTS AND DISCUSSION

The residue levels of alpha-cypermethrin (BAS 310 I) in tomato specimens taken directly after the last application (0 DALA) and at 2±1 DALA of BAS 310 51 I ranged between <0.01-0.03 mg/kg in all trials. At 7±1 DALA and at 14 DALA, residues were <0.01-0.02 mg/kg.

Residues found in tomato specimens after the application of BAS 310 40 I lead to similar residue results of alpha-cypermethrin.

None of the analysed untreated specimens showed alpha-cypermethrin residues exceeding the respective method LOQ.

An overall summary of the residues is given in the table below.

Table 2.8.1-3: Summary of residues of BAS 310 I in tomato from trials according to critical GAP after application of BAS 310 51 I and BAS 310 40 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	alpha-cypermethrin (BAS 310 I)
Tomato	2008	1	BAS 310 51 I (ME)	0	71-85	fruit	0.02
				3	73-87		<0.01
				7	88-89		<0.01
				14	88-89		<0.01
		2		0	82-89	fruit	0.01-0.03
				2-3	85-89		<0.01-0.03
				7-8	86-89		0.01-0.02
				14	88-89		0.01-0.02
		1	BAS 310 40 I (EC)	0	71-85	fruit	0.01-0.03
				3	73-87		<0.01
				7	88-89		<0.01
				14	88-89		<0.01
		2		0	82-89	fruit	0.02
				2-3	85-89		<0.01-0.03
				7-8	86-89		0.01-0.02
				14	88-89		<0.01-0.02

DALA = days after last application
BBCH = growth stage at respective sampling

III. CONCLUSION

Directly after the last application of BAS 310 51 I and BAS 310 40 I, the residue of alpha-cypermethrin ranged between 0.01-0.03 mg/kg in tomato specimens. At 2±1 days, residues ranged between <0.01-0.03 mg/kg. At 7±1 days and later, residues found were <0.01-0.02 mg/kg.
The analytical results obtained demonstrate that the treatment with one or two applications of BAS 310 51 I and BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.

Table 2.8.1-4: Residues of BAS 310 I after two applications of the formulations BAS 310 51 I and BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 319699 **Doc ID:** 2009/1090704 **Trial No.:** L080173 **GLP:** yes **Year** 2008	Tomato	Germany	BAS 310 51 I BAS 310 I 2 x 0.0150	82	0	fruit	0.03
					2		0.03
					8		0.02
					14		0.02
			BAS 310 40 I BAS 310 I 2 x 0.0150	82	0	fruit	0.02
					2		0.03
					8		0.02
					14		0.02
Study code: 319699 **Doc ID:** 2009/1090704 **Trial No.:** L080174 **GLP:** yes **Year** 2008	Tomato	France	BAS 310 51 I BAS 310 I 2 x 0.0150	89	0	fruit	0.01
					3		<0.01
					8		0.01
					14		0.01
			BAS 310 40 I BAS 310 I 2 x 0.0150	89	0	fruit	0.02
					3		<0.01
					8		0.01
					14		<0.01

DALA = days after last application
BBCH = growth stage at respective sampling

Table 2.8.1-5: Residues of BAS 310 I after one application of the formulations BAS 310 51 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 319699 **Doc ID:** 2009/1090704 **Trial No.:** L080175 **GLP:** yes **Year** 2008	Tomato	Italy	BAS 310 51 I BAS 310 I 1 x 0.0300	85	0	fruit	0.02
					3		<0.01
					7		<0.01
					14		<0.01
			BAS 310 40 I BAS 310 I 1 x 0.0300	85	0	fruit	0.03
					3		<0.01
					7		<0.01
					14		<0.01
Study code: 319699 **Doc ID:** 2009/1090704 **Trial No.:** L080176 **GLP:** yes **Year** 2008	Tomato	Spain	BAS 310 51 I BAS 310 I 1 x 0.0300	71	0	fruit	0.02
					3		<0.01
					7		<0.01
					14		<0.01
			BAS 310 40 I BAS 310 I 1 x 0.0300	71	0	fruit	0.01
					3		<0.01
					7		<0.01
					14		<0.01

DALA = days after last application
BBCH = growth stage at respective sampling

Leonard R C, Saha M (2005)

Full study reference

Leonard R C, Saha M (2005): Study on the residue behavior of BAS 310 I in tomatoes (glasshouse) after application of BAS 310 41 I in Germany, France (N), Denmark, Netherlands, France (S), Spain, Italy and Greece, 2004; BASF DocID 2004/5000719

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the Residue Behaviour of BAS 310 40 I in Tomato (field) after Treatment with BAS 310 40 I under Field Conditions in Southern and Northem Europe, 2005; BASF DocID 2007/1008488

Diehl M (2007)

Full study reference

Diehl M 2007: Study on the Residue Behaviour of BAS 310 40 I in Tomato after Treatment with BAS 310 40 I under Greenhouse Conditions in Southern and Northern Europe, 2005; BASF DocID 2007/1007934

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alphacypermethrin in field tomato after treatment with BAS 310 40 I or BAS 310 08 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007937

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the residue behaviour of BAS 310 I in tomato after treatment with BAS 310 40 I under open field conditions in Southern and Northern Europe, 2006; BASF DocID 2007/1008494

Material and Methods:

A residue program in tomatoes was conducted in the years 2004, 2005 and 2006 in representative tomato growing areas in Belgium, Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain and the Netherlands under open field and glasshouse conditions.

During the 2005 growing season, eight trials were conducted in tomatoes under open field conditions, four of them in the Northern European region (in Belgium, France and Germany) and four in the Southern European region (in France, Italy and Spain).
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I) was foliar applied to tomato plants in several variants. In four trials located in the Northern European region, the product was applied once at target rate of 12.5 g a.s./ha. This was compared to another variant in which two applications were made at the same target application rate. The last application took place at growth stages between 69 (9 or more inflorescences with open flowers) and 85 (50% of fruits show typical fully ripe colour).

At four other trials located in the Southern European region, a third variant was assayed in which BAS 310 40 I was applied once at a target rate of 25 g a.s./ha at growth stages between 81 (10% of fruits show typical fully ripe colour) and 89 (fully ripe). Tomato specimens were collected directly after the last application as well as 3, 7 and 14 days thereafter.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

A total of 8 residue trials were conducted during the 2006 growing season under open field conditions in Germany, France (Northern and Southern European region), Greece, Italy and Spain.
All trials were performed with an emusifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L, BAS 310 40 I).
In the trials performed in the Northern European region (4 sites) the product was sprayed on separate plots of tomato plants at two different rates: variant 1 received one treatment at a target rate of 12.5 g a.s./ha, while in variant 2, two treatments at 12. 5 g a.s./ha were applied. The last application took place at growth stages between 79 (9th fruit cluster: first fruit has reached typical size) and 84 (40% of fruits show typical fully ripe colour). Samples of tomatoes were collected 0, 3, 7 and 14 days after the last application.
In the trials performed in the Southern European region (4 sites) the product was applied once at a target rate of 25 g a.s./ha. The application took place at growth stages between 73 (3rd fruit cluster: first fruit has reached typical size) and 88 (80% of fruits show typical fully ripe colour). Samples of tomatoes were collected 0, 2-4, 6-8 and 14 days after application.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, four bridging trials were conducted in France (Northern and Southern European region), Germany and Spain under open field conditions to compare the residue behaviour after application of two different formulations of alpha-cypermethrin.
An emulsifiable concentrate formulation (EC 100 g a.s./L, BAS 310 40 I) and a wettable granule formulation (WG 150 g/kg, BAS 310 08 I) were foliar applied to separate plots of tomato plants, each one at the following rates: One treatment at 12.5 g a.s./ha (variant 1), two treatments at 12.5 g a.s./ha (variant 2), or one treatment at 25 g a.s./ha (variant 3). Variants 1 and 2 were included in the trials located in the Northern European region and variant 3 was included in the trials located in the Southern European region. Samples of tomato fruit were collected immediately after the last application as well as 2-4, 6-8, and 14 days afterwards.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2004 growing season, eight residue trials were conducted in tomatoes grown in glasshouses.
Alpha-cypermethrin, formulated as a soluble concentrate (SC 100 g a.s./L, BAS 310 41 I) was foliar applied to tomatoes on two different plots. In one variant, one application was made at a target rate of 15 g a.s./ha. This was compared to another variant in which two applications were made at the same target application rate. The last application took place at growth stages between 76 (6th fruit cluster: first fruit has reached typical size) and 85 (50% of fruits show typical fully ripe colour). Tomato fruit specimens were collected directly after the last application from each plot at all locations, and at 2-4, 6-7 and 13-14 days after the last application. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

Eight glasshouse trials were conducted during the 2005 growing season in Belgium, France (Northern and Southern European region), Germany, Greece, Italy and Spain.
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L, BAS 310 40 I) was foliar applied to tomato once at a target rate of 40 g a.s./ha at growth stages between 71 (first fruit cluster: first fruit has reached typical size) and 88 (80% of fruits show typical fully ripe colour).
Tomato samples were collected 0, 2-3, 7 and 14-15 days after application.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.8.1-6 and Table 2.8.1-7.

Table 2.8.1-6: Residues in tomatoes – open field trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008488 Trial No. A/NF/I/05/60 Study to GLP Study carried out in 2005	Tomato (variety Joker)	France Dame Marie les Bois Centre (North of EU)	13 g a.s./ha 29.08.05	82	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/NF/I/05/61 Study to GLP Study carried out in 2005	Tomato (variety Hector)	France Saint Martin des Bois Centre (North of EU)	13 g a.s./ha 09.09.05	84	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 87.1%; SD: +/- 8.8; CV: 10.1%; n=5; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008488 Trial No. A/GE/I/05/62 Study to GLP Study carried out in 2005	Tomato (variety Vanessa)	Germany Lambsheim Rheinland-Pfalz	13 g a.s./ha 24.08.05	81-85	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/BE/I/05/63 Study to GLP Study carried out in 2005	Tomato (variety Felice)	Belgium Villers-Perwin (Hainaut)	13 g a.s./ha 06.09.05	69-83	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008494 Trial No. A/NF/I/06/97 Study carried out in 2006	Tomato (variety Medina)	France Warmeriville Marne (North of EU)	12 g a.s./ha 23.08.06	83	0 4 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008494 Trial No. A/GE/I/06/98 Study carried out in 2006	Tomato (variety Harzfeuer)	Germany Schleswig Schleswig-Holstein	14 g a.s./ha 29.07.06	84	0 3 8 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.9%; SD: +/- 11.9; CV: 13.1%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008494 Trial No. A/GE/I/06/99 Study carried out in 2006	Tomato (variety Vanessa)	Germany Lambsheim Rheinland-Pfalz	13 g a.s./ha 18.07.06	79	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008494 Trial No. A/GE/I/06/100 Study carried out in 2006	Tomato (variety Tombolino St Pierre Fiaschetto Mix)	Germany Kirchheim Rheinland-Pfalz	12 g a.s./ha 03.08.06	80	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/1 Study carried out in 2006	Field tomato (variety Topkapi)	France Allonnes Maine-et-Loire 49650 (North of EU)	12.5 g a.s./ha (EC) 25.08.06	81	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.5%; SD: +/- 10.2; CV: 11.3%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/1 Study carried out in 2006	Field tomato (variety Topkapi)	France Allones Maine-et-Loire 49650 (North of EU)	12.5 g a.s./ha (WG)- 25.08.06	81	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	

Table 2.8.1-6: Residues in tomatoes – open field trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/2 Study carried out in 2006	Field tomato (variety Vanessa)	Germany Ringstraße 46 67245 Lambsheim Baden-Württemberg	12.5 g a.s./ha (EC) 26.07.06	73	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/2 Study carried out in 2006	Field tomato (variety vanessa)	Germany Ringstr. 46 67245 Lambsheim Baden-Wurttemberg	12.5 g a.s./ha (WG) 26.07.06	73	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/NF/I/05/60 Study to GLP Study carried out in 2005	Tomato (variety Joker)	France Dame Marie des Bois Centre (North of EU)	13 g a.s./ha 2 treatm. last date 29.08.05	82 at last treatm.	0 3 7 14	fruit 0.013 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/NF/I/05/61 Study to GLP Study carried out in 2005	Tomato (variety Hector)	France Saint Martin de Bois Centre (North of EU)	13 g a.s./ha 2 treatm. last date 09.09.05	84 at last treatm.	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 87.1%; SD: +/- 8.8; CV: 10.1%; n=5; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008488 Trial No. A/GE/I/05/62 Study to GLP Study carried out in 2005	Tomato (variety Vanessa)	Germany Lambsheim Rheinland-Pfalz	13 g a.s./ha 2 treatm. last date 24.08.05	81-85 at last treatm.	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/BE/I/05/63 Study to GLP Study carried out in 2005	Tomato (variety Felice)	Belgium Villers-Perwin Hainaut	13 g a.s./ha 2 treatm. last date 06.09.05	69-83	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008494 Trial No. A/NF/I/06/97 Study carried out in 2006	Tomato (variety Medina)	France Warmeriville Marne (North of EU)	12/13 g a.s./ha 2 treatm. last date 23.08.06	83 at last treatm.	0 4 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.9%; SD: +/- 11.9; CV: 13.1%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008494 Trial No. A/GE/I/06/98 Study carried out in 2006	Tomato (variety Harzfeuer)	Germany Schleswig Schleswig-Holstein	14/12 g a.s./ha 2 treatm. last date 29.07.06	84 at last treatm.	0 3 8 14	fruit <0.01 fruit <u>0.010</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.9%; SD: +/- 11.9; CV: 13.1%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008494 Trial No. A/GE/I/06/99 Study carried out in 2006	Tomato (variety Vanessa)	Germany Lambsheim Rheinland-Pfalz	13/12 g a.s./ha 2 treatm. last date 18.07.06	79 at last treatm.	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	

Table 2.8.1-6: Residues in tomatoes – open field trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008494 Trial No. A/GE/I/06/100 Study carried out in 2006	Tomato (variety Tombolino St Pierre Fiaschetto Mix)	Germany Kirchheim Rheinland-Pfalz	13/13 g a.s./ha 2 treatm. last date 03.08.06	80 at last treatm.	0 3 7 14	fruit 0.012 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/1 Study carried out in 2006	Field tomato (variety Topkapi)	France Allonnes Maine-et-Loire 49650 (North of EU)	12.5 g a.s./ha (EC) 2 treatm. last date 25.08.06	81 at last treatm.	0 3 7 14	fruit <0.01 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.5%; SD: +/- 10.2; CV: 11.3%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/1 Study carried out in 2006	Field tomato (variety Topkapi)	France Allones Maine-et-Loire 49650 (North of EU)	12.5 g a.s./ha (WG) 2 treatm. last date 25.08.06	81 at last treatm.	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/2 Study carried out in 2006	Field tomato (variety Vanessa)	Germany Ringstraße 46 67245 Lambsheim Baden-Württemberg	12.5 g a.s./ha (EC) 2 treatm. last date 26.07.06	73 at last treatm.	0 3 7 14	fruit 0.011 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/2 Study carried out in 2006	Field tomato (variety vanessa)	Germany Ringstr. 46 67245 Lambsheim Baden-Wurttemberg	12.5 g a.s./ha (WG) 2 treatm. last date 26.07.06	73 at last treatm.	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/SF/I/05/64 Study to GLP Study carried out in 2005	Tomato (variety All R 50)	France Boisseron Languedoc Roussillon (South of EU)	28 g a.s./ha 16.09.05	81	0 3 7 14	fruit <0.01 fruit <0.01 fruit <u>0.013</u> fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 87.1%; SD: +/- 8.8; CV: 10.1%; n=5; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008488 Trial No. A/SF/I/05/65 Study to GLP Study carried out in 2005 181987	Tomato (variety Coudoulet)	France Caumont Midi-Pyrenées (South of EU)	25 g a.s./ha 05.08.05	87	0 3 7 14	fruit 0.015 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008488 Trial No. A/SP/I/05/66 (TRC05-8R1) Study to GLP Study carried out in 2005	Tomato (variety Rio Grande)	Spain Turis Valencia	25 g a.s./ha 29.08.05	81	0 3 7 14	fruit <0.01 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 87.1%; SD: +/- 8.8; CV: 10.1%; n=5; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008488 Trial No. A/IT/I/05/67 Study to GLP Study carried out in 2005	Tomato (variety Rio Grande)	Italy Costigliole d´Asti Piemont	27 g a.s./ha 17.08.05	89	0 3 7 15	fruit <0.01 fruit <u>0.021</u> fruit <0.01 fruit <0.01	

Table 2.8.1-6: Residues in tomatoes – open field trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008494 Trial No. A/SF/I/06/101 Study carried out in 2006	Tomato (variety Perfectil)	France Grillon Vaucluse (South of EU)	25 g a.s./ha 11.08.06	81	0 3 7 14	fruit 0.012 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.9%; SD: +/- 11.9; CV: 13.1%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008494 Trial No. A/GR/I/06/102 Study carried out in 2006	Tomato (variety Alma)	Greece Profidis Thessaloniki Central Macedonia	24 g a.s./ha 18.07.06	88	0 4 8 14	fruit <0.01 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008494 Trial No. A/SP/I/06/103 Study carried out in 2006	Tomato (variety Bodar)	Spain Benicarlo Castellon	25 g a.s./ha 14.07.06	73	0 3 7 14	fruit 0.021 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1008494 Trial No. A/IT/I/06/104 Study carried out in 2006	Tomato (variety Rio Grande)	Italy Costigliole D´Asti Piemont	25 g a.s./ha 12.08.06	81	0 2 6 14	fruit 0.013 fruit <u>0.019</u> fruit 0.013 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/3 Study carried out in 2006	Field tomato (variety Tina)	Spain, Enrique Olmos Pellicena Santa Engracia	25 g a.s./ha (EC) 11.09.06	85	0 3 7 14	fruit <0.01 fruit <u>0.013</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 90.5%; SD: +/- 10.2; CV: 11.3%; n=7; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/3 Study carried out in 2006	Field tomato (variety Tina)	Spain Enrique Olmos Pellicena Santa Engracia	25 g a.s./ha (WG) 11.09.06	85	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/4 Study carried out in 2006	Field tomato (variety Leader)	France Lagarrigue Castelmayran Tarn-et-Garonne	25 g a.s./ha (EC) 26.07.06	81-85	0 3 7 14	fruit 0.015 fruit <u>≤0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007937 Trial No. AF/10504/BA/4 Study carried out in 2006	Field tomato (variety Leader)	France, Lagarrigue Castelmayran Tarn-et-Garonne (South of EU)	25 g a.s./ha (WG) 27.06.06	81-85	0 3 7 14	fruit 0.014 fruit <0.01 fruit <0.01 fruit <0.01	

_ underlined values were used for MRL calculation

Table 2.8.1-7: Residues in tomatoes - glasshouse trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/5000719 Trial No. ACK/07/04 Study to GLP Study carried out in 2004	Tomato (variety Swift)	Germany Brandenburg (glasshouse)	15 g a.s./ha 25.05.04	85	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 79.9%; SD: +/- 14.1; CV: 17.6%; n=10; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2004/5000719 Trial No. AGR/09/04 Study to GLP Study carried out in 2004	Tomato (variety Cedrico)	Netherlands Limburg (glasshouse)	15 g a.s./ha 12.10.04	85	0 3 7 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. ALB/06/04 Study to GLP Study carried out in 2004	Tomato (variety Aromata)	Denmark South Jutland (glasshouse)	15 g a.s./ha 10.06.06	83	0 4 7 14	fruit <0.01 fruit <0.01 fruit 0.019 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. ALO/16/04 Study to GLP Study carried out in 2004	Tomato (variety Antilla)	Spain Andalucia (glasshouse)	15 g a.s./ha 3.11.04	81	0 3 7 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. FBM/05/04 Study to GLP Study carried out in 2004	Tomato (variety Petula)	France Pays de la Loire (glasshouse) (North of EU)	15 g a.s./ha 16.07.04	83	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. FTL/12/04 Study to GLP Study carried out in 2004	Tomato (variety Brenda)	France Midi-Pyreneés (glasshouse) (South of EU)	15 g a.s./ha 16.09.04	85	0 2 6 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. GRE/11/04 Study to GLP Study carried out in 2004	Tomato (variety Alma)	Greece Northern Greece-Macedonia (glasshouse)	15 g a.s./ha 17.09.04	76	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. ITA/08/04 Study to GLP Study carried out in 2004	Tomato (variety Seni)	Italy Torino (glasshouse)	15 g a.s./ha 13.09.04	85	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 79.9%; SD: +/- 14.1; CV: 17.6%; n=10; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2004/5000719 Trial No. ACK/07/04 Study to GLP Study carried out in 2004	Tomato (variety Swift)	Germany Brandenburg (glasshouse)	15 g a.s./ha 2 treatm. last date 25.05.04	85 at last treatm.	0 3 7 14	fruit 0.012 fruit 0.012 fruit 0.010 fruit <0.01	

Table 2.8.1-7: Residues in tomatoes - glasshouse trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/5000719 Trial No. AGR/09/04 Study to GLP Study carried out in 2004	Tomato (variety Cedrico)	Netherlands Limburg (glasshouse)	15 g a.s./ha 2 treatm. last date 12.10.04	85 at last treatm.	0 3 7 13	fruit 0.017 fruit 0.018 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. ALB/06/04 Study to GLP Study carried out in 2004	Tomato (variety Aromata)	Denmark South Jutland (glasshouse)	15 g a.s./ha 2 treatm. last date 10.06.06	83 at last treatm.	0 4 7 14	fruit 0.019 fruit <0.01 fruit 0.013 fruit 0.011	
BASF Doc ID 2004/5000719 Trial No. ALO/16/04 Study to GLP Study carried out in 2004	Tomato (variety Antilla)	Spain Andalucia (glasshouse)	15 g a.s./ha 2 treatm. last date 3.11.04	81 at last treatm.	0 3 7 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. FBM/05/04 Study to GLP Study carried out in 2004	Tomato (variety Petula)	France Pays de la Loire (glasshouse) (North of EU)	15 g a.s./ha 2 treatm. last date 16.07.04	83 at last treatm.	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. FTL/12/04 Study to GLP Study carried out in 2004	Tomato (variety Brenda)	France Midi-Pyreneés (glasshouse) (South of EU)	15 g a.s./ha 2 treatm. last date 16.09.04	85 at last treatm.	0 2 6 13	fruit 0.011 fruit <0.01 fruit <0.01 fruit 0.015	
BASF Doc ID 2004/5000719 Trial No. GRE/11/04 Study to GLP Study carried out in 2004	Tomato (variety Alma)	Greece Northern Greece-Macedonia (glasshouse)	15 g a.s./ha 2 treatm. last date 17.09.04	76 at last treatm.	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000719 Trial No. ITA/08/04 Study to GLP Study carried out in 2004	Tomato (variety Seni)	Italy Torino (glasshouse)	15 g a.s./ha 2 treatm. last date 13.09.04	85 at last treatm.	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007934 Trial No. A/NF/I/05/50 Study to GLP Study carried out in 2005	Tomato (variety Sympathic)	France St Genouph Tourraine	39 g a.s./ha 20.06.05 Indoor	81	0 3 7 14	fruit <0.01 fruit 0.010 fruit 0.011 fruit <u>0.016</u>	BASF analytical method No. 567/0 fruit: mean recovery = 94.8%; SD: +/- 13.9; CV: 14.6%; n=6; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007934 Trial No. A/SF/I/05/51 Study to GLP Study carried out in 2005	Tomato (variety Brenda)	Frances St Remy de Provence Bouches du Rhone (South of EU)	42 g a.s./ha 19.07.05 Indoor	74-81	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	

Table 2.8.1-7: Residues in tomatoes - glasshouse trials

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 3 days (outdoor) GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor) or Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days , or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007934 Trial No. A/BE/I/05/54 Study to GLP Study carried out in 2005	Tomato (variety Piole)	Belgium Villers-Perwin Hainaut	42 g a.s./ha 26.07.05 Indoor	71-85	0 3 7 14	fruit <0.01 fruit 0.015 fruit 0.01 fruit <0.01	
BASF Doc ID 2007/1007934 Trial No. A/SP/I/05/55 Study to GLP Study carried out in 2005	Tomato (variety Marmande RAF)	Spain Benifayo Valencia	40 g a.s./ha 07.06.05 Indoor	85	0 3 7 14	fruit 0.030 fruit 0.024 fruit 0.015 fruit 0.013	
BASF Doc ID 2007/1007934 Trial No. A/GR/I/05/57 Study to GLP Study carried out in 2005	Tomato (variety Alma)	Greece Profitis Thessaloniki Central Macedonia	39 g a.s./ha 29.07.05 Indoor	84	0 3 7 14	fruit <0.01 fruit <0.01 fruit 0.014 fruit <0.01	
BASF Doc ID 2007/1007934 Trial No. A/GE/I/05/52 Study to GLP Study carried out in 2005	Tomato (variety Pipo)	Germany Hassloch-Meckenheim Rheinland-Pfalz	37 g a.s./ha 13.07.05 Indoor	85-88	0 2 7 14	fruit 0.019 fruit 0.025 fruit <0.01 fruit 0.013	
BASF Doc ID 2007/1007934 Trial No. A/GE/I/05/53 Study to GLP Study carried out in 2005	Tomato (variety Alma)	Germany Engelbrechtsche Wildnis Schleswig-Holstein	42 g a.s./ha 01.08.05 Indoor	85-87	0 3 7 15	fruit 0.012 fruit 0.016 fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007934 Trial No. A/IT/I/05/56 Study to GLP Study carried out in 2005	Tomato (variety Cuor Olibue (hybrid))	Italy Motta di Costigliole, Loc. Remoneirini Piemont	41 g a.s./ha 11.07.05 Indoor	85	0 3 7 14	fruit 0.029 fruit 0.037 fruit 0.024 fruit 0.032	

_ underlined values were used for MRL calculation

Findings:

The residue data in tomatoes presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region and to glasshouse conditions.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 3 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days
- glasshouse GAP is a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 2 days or a single application with 50 g a.s./ha with a PHI of 7 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-10 trials including both variants on different plots, 2 of them bridging trials with two different formulations, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions-10 trials, 2 of them bridging trials with two different formulations, all conducted in the Southern European region
- one or two treatments at a target rate of 15 g a.s./ha under glasshouse conditions-8 trials including both variants on different plots, 4 conducted in the Northern European region and 4 conducted in the Southern European region
- one treatment at target rate of 40 g a.s./ha under glasshouse conditions-8 trials, 4 conducted in the Northern European region and 4 conducted in the Southern European region

In the open field trials, consistently very low residues were found.

After one application at a target rate of 12.5 g a.s./ha, no residues above the limit of quantitation of the analytical method were found in any treated tomato fruit sample, regardless of sampling interval (all <0.01 mg/kg).

After two applications at a target rate of 12.5 g a.s./ha, initial residues in tomato fruit samples from three out of twelve plots ranged between 0.011-0.013 mg/kg. In all other samples taken immediately after the last application residues were below the LOQ (<0.01 mg/kg). A residue at the LOQ (0.01 mg/kg) was found in one trial three days after the last application. Besides this finding, residues in all other treated samples were below the LOQ (<0.01 mg/kg).

After one application at a target rate of 25 g a.s./ha, initial residues tomato fruit ranged between <0.01-0.021 mg/kg. Residues between 0.013-0.021 mg/kg were found in three trials three days after application, while in all other trials the residues were below the LOQ (<0.01 mg/kg) at this sampling point. Seven days after application, a residue of 0.013 mg/kg was determined in two trials, while all other treated samples taken at the same time as well as all treated samples taken 14 days after application did not show residues above the LOQ (all <0.01 mg/kg).

The residue situation in the glasshouse trials was as follows:

After one application at a target rate of 15 g a.s./ha under glasshouse conditions, residues in tomato fruit were below the method LOQ (<0.01 mg/kg) in all trials at all sampling times with only one exception (trial ALB/06/04): 7 days after application, a residue of 0.019 mg/kg was found in fruit, while all other samples from the same plot taken earlier or later did not show residues above the LOQ (all <0.01 mg/kg).

After two applications at a target rate of 15 g a.s./ha under glasshouse conditions, in four out of eight trials no determinable residues were found at any sampling interval (all <0.01 mg/kg). In the other four trials, initial residues in tomato fruit ranged between 0.011-0.019 mg/kg and declined to <0.01-0.018 mg/kg, <0.01-0.013 mg/kg and <0.01-0.015 mg/kg after 2-4, 6-7 and 13-14 days, respectively.

After one application at a target rate of 40 g a.s./ha under glasshouse conditions, initial residues in tomato fruit ranged between <0.01-0.030 mg/kg.
Residue levels found at later sampling points were <0.01-0.037 mg/kg, <0.01-0.024 mg/kg and <0.01-0.032 mg/kg after 3, 7 and 14 days, respectively.

Conclusion:

A residue program in tomatoes was conducted in the years 2004, 2005 and 2006 in representative tomato growing areas in Belgium, Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain and the Netherlands under open field and glasshouse conditions.

Treatment of tomato plants with alpha-cypermethrin under open field conditions according to the proposed GAP leads to a favourable residue situation. Residue levels in tomato fruit are very low with the majority of values within a range below the limit of quantitation of the analytical method.
In 10 trials (including 12 treated plots) supporting the proposed field GAP for the Northern European region, a residue at the LOQ (0.01 mg/kg) was found in one trial three days after the last application. Besides this finding, residues in all other treated samples were below the LOQ (<0.01 mg/kg) at the target PHI of 3±1 days.
In 10 trials (including 12 treated plots) supporting the proposed field GAP for the Southern European region, residues between 0.013-0.021 mg/kg were found in three trials at the target PHI of 3±1 days, while in all other trials the residues were below the LOQ (<0.01 mg/kg) at this sampling point.

Results showed that residues after BAS 10 40 I and BAS 310 08 I treatment were comparable.

Treatment of tomato plants with alpha-cypermethrin under glasshouse conditions represents a worst case situation with regard to residues with a tendency to higher residue levels at application rates that are comparable to the open field GAP.
In 8 trials supporting the proposed glasshouse GAP, residues ranged between <0.01-0.037 mg/kg at the target PHI of 3 days and between <0.01-0.024 mg/kg at the target PHI of 7 days.

2.8.2 Processing study in tomato

Report:

Schulz H. 2006a
Study on the Residue Behaviour of Alpha-Cypermethrin in Tomato and Processed Products after Treatment of BAS 310 41 I under Greenhouse Conditions in Germany and The Netherlands, 2004
2006/1021295

Guidelines: EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8),EEC 1607/VI/97 rev. 2 10.06.1999,EEC 7029/VI/95 rev. 5,EEC 7525/VI/95 rev. 7,EEC 7035/VI/95 rev. 5

GLP: yes
(certified by Hessisches Ministerium für Umwelt, Laendlichen Raum und Verbraucherschutz)

Material and methods:

During the 2004 growing season, four greenhouse trials in tomato were conducted in Germany (Brandenburg; ACK/09/04 and ACK/10/04) and the Netherlands (Limburg; AGR/12/04 and AGR/13/04) in order to determine the magnitude and distribution of alpha-cypermethrin residues in a number of intermediate and end products after processing.

Two of the trials consisted of two plots, one untreated and one treated (ACK/09/04 and AGR/12/04); the other two (ACK/10/04 and AGR/13/04) consisted of one treated plot each.

Tomato plants (varieties: Clothilde in the German trials and Cedrico in the Dutch trials) were foliar sprayed twice with a soluble concentrate formulation of alpha-cypermethrin (SC 100 g/L, code: BAS 310 41 I) at a rate of 75 g a.s./ha. Actual rates were within +/10 % of the nominal rate. This exaggerated treatment rate was used in an effort to generate finite alpha-cypermethrin residues in tomato so that the distribution of the residue in the processed fractions could be better quantified.

The applications were made 14(±1) and 7(±1) days before harvest. The growth stage at the last application was BBCH 85 in all trials (50% of fruits show typical fully ripe colour). The spray volume used was 400 I/ha. Tomato fruit samples were collected immediately after the second application and 7 days thererafter.
Field samples for residue analysis from all trials were frozen within 24 hours of sampling and remained frozen (including during transportation) during analysis.

18 - 23 kg of tomatoes were picked for processing at 7 days after application from every trial.
The processing phase was conducted at NIG GmbH Nahrungs-Ingenieurtechnik GmbH, Wasserkunststr. 26, O-39124 Magdeburg, Germany.

The following specimens resulted from the processing:
- washed tomatoes
- wash water
- peeled tomatoes
- peel
- dipping water
- canned tomatoes
- vegetable stock
- tomato juice
- remainder of the straining process
- tomato puree
- tomato paste
- condensed water

These processing products fulfil the requirements for a balance study.

The analytical part of the study was performed at the Agricultural Research Center of BASF in Limburgerhof, Germany.
The specimens were analysed for residues of alpha-cypermethrin by means of HPLC-MS/MS according to BASF method N° 567/0. This method quantifies the residues in tomatoes and processed products with a limit of quantification (LOQ) of 0.01 mg/kg.
Method performance was checked by determining the procedural recoveries in tomato RAC and processed fractions.
At fortification levels between 0.01 and 1.0 mg/kg, the recovery rates averaged at 95% with a standard deviation of 8.4 and a coefficient of variation of 8.9%.

Findings:

No residues of alpha-cypermethrin at or above the validated method LOQ (0.01 mg/kg) were detected in the untreated RAC species.

Directly after the last application, alpha-cypermethrin was found between 0.07 and 0.10 mg/kg in tomato fruit. After 7 days, the residues had declined to levels between 0.04 and 0.09 mg/kg.

No detectable residues were found above the validated method LOQ (0.01 mg/kg) in any untreated tomato processed fractions (washed tomatoes, wash water, peeled tomato, peel, dipping water, canned tomato, vegetable stock, tomato juice, remainder of straining, tomato puree, tomato paste and condensed water).
After washing, the treated tomatoes still showed residues between 0.04 and 0.07 mg/kg, in the wash water were no residues above the LOQ. After peeling; residues of alpha-cypermethrin were only found on the peel (0.39 to 1.58 mg/kg), the peeled fruits as well as the canned tomatoes made thereof were free of alpha-cypermethrin (< 0.01).
Tomato juice contained alpha-cypermethrin residues between 0.01 and 0.02 mg/kg. In the concentrated fractions puree and paste higher levels (0.02-0.04 mg/kg and 0.04-0.11 mg/kg) were found according to the degree of concentration.

The total alpha-cypermethrin residues in tomato and processed products as well as the corresponding transfer factors were as follows, whereby the transfer factors in the tomato fruit (RAC) were set as 1:

Table 2.8.2-1: Summary of alpha-cypermethrin residues in tomatoes and transfer factors - DocID 2006/1021295

Specimen	DALA [1)]	Total Alpha-cypermethrin Residues mg/kg	Transfer Factors Trial No.			
			ACK/09/04	ACK/10/04	AGR/12/04	AGR/13/04
Tomato fruit (RAC)	7	0.04 -0.09	1	1	1	1
Washed tomato	-	0.04 -0.07	1.5	1.0	1.2	0.4
Wash water	-	< 0.01	< 0.25	< 0.2	< 0.2	< 0.1
Peeled tomato	-	< 0.01	< 0.25	< 0.2	< 0.2	< 0.1
Peel	-	0.39 -1.58	9.8	18.0	15.5	17.6
Dipping water	-	< 0.01	< 0.25	< 0.2	< 0.2	< 0.1
Canned tomato	-	< 0.01	< 0.25	< 0.2	< 0.2	< 0.1
Vegetable stock	-	< 0.01	< 0.25	< 0.2	< 0.2	< 0.1
Tomato juice	-	0.01 -0.02	0.25	0.3	0.3	0.2
Remainder of straining	-	0.21 -0.50	5.3	8.3	7.3	3.1
Tomato puree	-	0.02 -0.04	0.5	0.5	0.7	0.3
Tomato paste	-	0.04 -0,.11	1.0	1.8	1	1.1
Condensed water	-	< 0.01	< 0.25	< 0.2	< 0.2	< 0.1

1) days after last application

Compared to the alpha-cypermethrin residues in tomato fruit taken at 7 days after last application, no remarkable residue changes occurred in the washed tomatoes. In the peeled tomatoes, no residues above the LOQ were found, whereas the highest residue concentration was obtained in the peel, indicating that alpha-cypermethrin was concentrated on the tomato surface. Transfer factors between 3.1 and 8.3 were obtained in the remainder of the straining process, which contained a high content of peel. In tomato juice and in tomato puree, the alpha-cypermethrin residues were lower than those of the RAC specimens. A slight enrichment was found in tomato paste where the transfer factors were between 1.0 and 1.8. No residues above the limit of quantification were obtained in the aqueous specimens wash water, dipping water, vegetable stock and condensed water.

Tomato processing summary – preparation of canned tomatoes, sterilized juice, tomato puree and tomato paste:

Tomatoes suitable for processing were sorted out by hand. The tomatoes were washed in water (18 °C, 9 washing batches, 3 min duration each). Peeling was achieved by dipping in hot (90 °C) and cold water. The tomatoes were then canned and sterilized at 120 °C for 18 min.
For preparation of juice, the tomatoes were strained (3 runs, heating up to 85 °C, duration of straining for each run approx. 1 min). The juice was sterilized at 120 °C for 18 min.
Paste and puree were prepared by concentration of juice through evaporation under reduced pressure.

Figure 2.8.2-1: Canned tomatoes and tomato juice processing procedure flowchart - DocID 2006/1021295

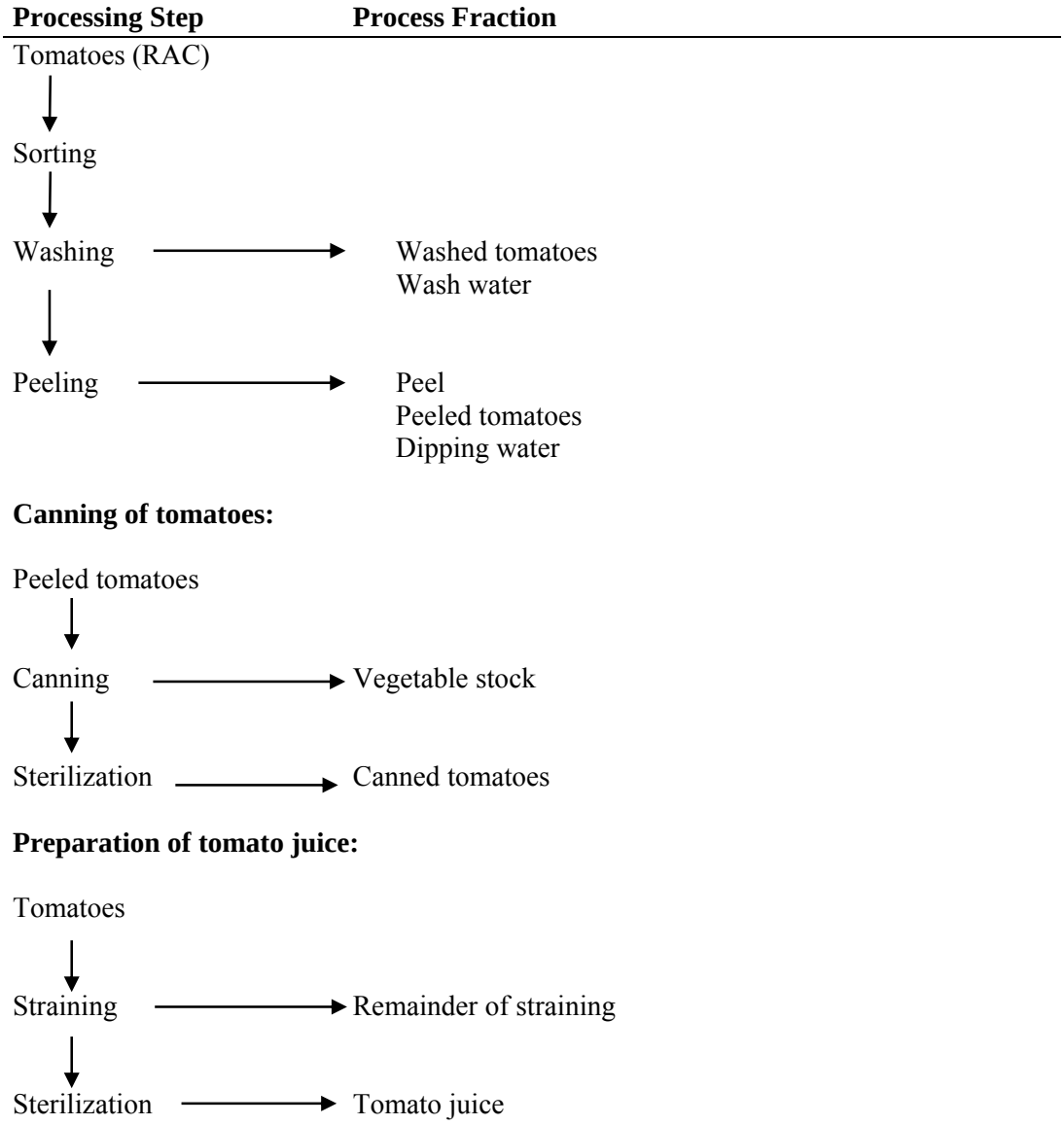

Figure 2.8.2-2: Tomato puree and tomato paste processing procedure flowchart - DocID 2006/1021295

Processing Step	Process Fractions
Preparation of tomato puree:	
Juice	
Concentration →	Tomato puree Condensed water
Preparation of tomato paste:	
Juice ↓	
Concentration →	Tomato paste Condensed water

Table 2.8.2-2: Mass balance of tomato processing - DocID 2006/1021295

Trial No. Treatment	ACK 09/04 U	ACK 09/04 T	ACK 10/04 T	AGR 12/04 U	AGR 12/04 T	AGR 13/04 T
Weight of tomatoes used for processing	20523	22792	21923	18156	18660	18625
Sorting						
Sorted out tomatoes (g)	1087	565	0	400	1437	2328
Tomatoes appropriate for processing (g)	19436	22227	21923	17756	17223	16297
Washing						
Tomatoes used for washing (g)	19436	18038	18248	17756	17223	16297
Wash water (g)	24839	23736	23472	23240	22697	21462
Weight of washed tomatoes (g)	19527	18102	18319	17871	17290	16349
Peeling						
Weight of washed tomatoes (g)	5001	4992	4996	5105	4950	5027
Weight of water used for dipping (g)	24122	24746	22741	25255	24417	24331
Total (g)	29123	29738	27737	30360	29367	29358
Weight of peeled tomatoes (g)	4597	4512	4707	4651	4512	4657
Weight of peels (g)	391	451	357	344	328	298
Weiqht of dipping water (g)	24122	24746	22741	25255	24417	24331
Total (g)	29110		27805			29286
Yield of the peelinq process (%)	100.0	29709 99.9	100.2	30250 99.6	29257 99.6	99.8
Canning Weight of peeled tomatoes (g) Sodium chloride solution (1%) g	2325 1335	2512 1248	2564 1162	2584 1197	2577 1203	2689 1198
Total (g)						
Weight of canned tomatoes (g)	3660 1157	3760 1094	3726 1420	3781 1275	3780 1382	3887 867
Weight of vegetable stock (g)	2282	1778	2077	2212	2014	815
Total (g)	3439	2872	3497	3487	3396	1682
Yield of the canning process (%)	94.0	76.4[1)]	93.9	92.2	89.8	nc [2)]
Preparation of tomato juice						
Weight of washed tomatoes (g)	13409	10851	11088	11727	11194	10263
Remainder of the straining process (g)	1965	2241	1467	1203	1076	1075
Juice (g)	10289	7517	8642	9583	9223	8417
Total (g)	12254	9758	10109	10786	10299	9492
Yield (%)	91.4	89.9	91.2	92.0	92.0	92.5
Preparation of tomato puree						
Weight of tomato juice (g)	2617	1375	1500	2512	2529	1924
Weight of tomato puree (g)	1403	738	791	1252	1081	901
Weight of condensed water (g)	1184	608	688	1225	1430	1004
Total (g)	2587	1346	1479	2477	2511	1905
Yield (%)	98.9	97.9	98.6	98.6	99.3	99
Preparation of tomato paste						
Weight of tomato juice (g)	7011	4931	5327	4159	4635	4510
Weight of tomato paste (g)	1420	876	942	586	557	668
Weight of condensed water (g)	4747	3411	4297	3558	3546	3824
Total (g)	6167	4287	5239	4144	4103	4492
Yield (%)	88.0	86.9	98.3	99.6	88.5	99.6

1) Low yield, because one out of three glasses was broken 2): Because of an error of the autoclav, the second specimen batch must be discarded. Yield was incalculable

Conclusion:

The residue data obtained in this processing study clearly demonstrate that alpha-cypermethrin residues are reduced compared to the levels in the raw agricultural commodity during processing to peeled and canned tomatoes, juice and pureé. This is due to the fact that alpha-cypermethrin residues are located on the peel and are retained in the peel fraction during processing.
A slight enrichment of alpha-cypermethrin residues was only found in tomato purée which can be attributed to the high concentration of the juice used as starting material.

2.8.3 Estimation of MRL, HR and STMR for tomato

For *tomato*, the following residue studies were considered (BASF DocIDs): 2007/1008488, 2007/1008494, 2007/1007937, 2009/1090704 and 2007/1007937.

The following residue values (PHI=3±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (S-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2007/1008488 (open field)	<0.01 (4x)	<0.01 (2x), 0.013, 0.021
2007/1008494 (open field)	<0.01 (3x), 0.01	<0.01 (3x), 0.019
2007/1007937 (open field)	<0.01 (2x)	<0.01, 0.013
2009/1090704 (open field)	0.01, 0,03	<0.01 (2x)
OECD-MRL-calculation (open field)	**<u>0.04</u> (n=12, STMR=0.01, HR=0.03)**	**<u>0.03</u> (n=12, STMR=0.01, HR=0.021)**
2007/1007934 (glasshouse)	<0.01, 0.014, 0.015, 0.016 (2x), 0.024, 0.025, 0.037	
OECD-MRL-calculation (glasshouse)	**<u>0.06</u> (n=8, STMR=0.016, HR=0.037)**	

_ underlined values were used for risk assessment purposes

2.9 Cucumber / courgette

Besidues the new studies presented in Section M-CA 6.3, no additional residue data were considered for cucumber / courgette.

2.10 Melon

Residue data from supervised trials in melons were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.10-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Melon (field)	2005	-	-	4	ES, FR, IT	4	2006/1024607 2007/1011007
Melon (field)	2006	-	-	4	ES, FR, GR, IT	4	2007/1007940
Melon (glasshouse)	2004	2	DK, NL	4	ES, FR, IT	6	2004/5000721
Melon (glasshouse)	2005	3	BE, DE, DK	5	ES, FR, GR, IT	8	2006/1037507
Total number of trials per region		**5 (glasshouse)**		**8 (field) 9 (glasshouse)**	**Total number of trials**	**8 (field) 14 (glasshouse)**	

Table 2.10-2: Processing studies available for melon

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Melon (glasshouse)	2004	-	-	4	ES	4	2006/8038660
Total number of trials per region		**-**			**Total number of trials**	**4**	

2.10.1 Supervised residue trials in melon

Leonard R C (2005)

Full study reference

Leonard R C (2005): Study on the residue behavior of BAS 310 I in melons (glasshouse) after application of BAS 310 41 I in Denmark, Netherlands, France (S), Spain, and Italy, 2004; BASF DocID 2004/5000721

Evans L (2007)

Full study reference

Evans L (2007): Study on the residue behaviour of alpha-cypermethrin in melon after treatment with BAS 310 40 I under field conditions in Southern Europe during 2005; BASF DocID 2006/1024607

North L (2007)
Full study reference
North L (2007): Final Report Amendment No. 1, Study on the residue behaviour of alpha-cypermethrin in melon after treatment with BAS 310 40 I under field conditions in Southern Europe during 2005; BASF DocID 2007/1011007

North L (2007)
Full study reference
North L (2007): Study on the residue behaviour of alpha-cypermethrin in Melon after treatment with BAS 310 40 I under field conditions in Southern Europe during 2006; BASF DocID 2007/1007940

Schulz H (2007)
Full study reference
Schulz H (2007): Study on the Residue Behaviour of Alpha-cypermethrin in melon after Treatment with BAS 310 40 I under Greenhouse Conditions in Southern France, Germany, Belgium, Denmark, Italy, Spain and Greece, 2005; BASF DocID 2006/1037507

Material and Methods:

In the years 2004-2006 a residue program in melons was conducted in Belgium, Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain and the Netherlands under open field and glasshouse conditions.

During the 2005 growing season, four trials were conducted in melon under open field conditions in France (Southern European region), Italy and Spain. Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to melon plants once at a target rate of 25 g a.s./ha. The growth stage of the plants at application was between 72 (2nd fruit on main stem has reached typical size and form) and 85 (50% of fruits show typical fully ripe colour). Melon fruit specimens were collected directly after the last application as well as approximately 3, 7 and 14 days thereafter.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, four trials were conducted in melon under open field conditions in France (Southern European region), Greece, Italy and Spain. Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to melon plants once at target rate of 25 g a.s./ha. The growth stage of the plants at application was between 71 (first fruit on main stem has reached typical size and form) and 88 (80% of fruits show typical fully ripe colour). Melon fruit specimens were collected directly after the last application as well as 2-3, 7 and 14 days thereafter.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2004 growing season, six trials were conducted in Denmark, France (Southern European region), Italy, Spain and the Netherlands in melons grown in glasshouses. A soluble concentrate formulation of alpha-cypermethrin (SC 100 g a.s./L; BAS 310 41 I) was foliar applied to melon plants on two different plots. In one variant, one application was made at a rate of 15 g a.s./ha. This was compared to another variant in which two applications were made at the same application rate. The growth stage of the plants at application was between 77 (7th fruit on main stem has reached typical size and form) and 87 (70% of fruits show typical fully ripe colour). Melon specimens were collected directly after the last application as well as about 2-4, 7 and 13-15 days thereafter.
The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

Eight glasshouse trials were performed in the year 2005 in Belgium, Denmark, France (Southern European region), Germany, Greece, Italy and Spain to determine the magnitude of the residue after application of a 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC; BAS 310 40 I). The product was foliar applied once at a rate of 40 g a.s./ha. The growth stage of the plants at application was between 71 (first fruit on main stem has reached typical size and form) and 87 (70% of fruits show typical fully ripe colour). Samples of melon fruit were collected 0, 2-3, 7-8 and 13-14 days after application. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.10.1-1 and Table 2.10.1-2

Table 2.10.1-1: Residues in melons – open field trials

GAP for EU-N is 1 application at 30 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)
Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days, or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1024607 and 2007/1011007 Trial No. AF/8816/BA/1 Study to GLP Study carried out in 2005	Melon (variety Anasta)	SCA Rouge Gorge du Thouet Charteau, Taize, Noize, 79100 (South of EU)	25 g a.s./ha 19.08.05	74	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 96.9%; SD: +/- 10.7; CV: 11.0; n=3; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1024607 and 2007/1011007 Trial No. AF/8816/BA/2 Study to GLP Study carried out in 2005	Melon (variety Anasta)	SCA Rouge Gorge du Thouet Charteau, Taize, Noize, 79100 (South of EU)	25 g a.s./ha 22.08.05	73	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2006/1024607 and 2007/1011007 Trial No. AF/8816/BA/3 Study to GLP Study carried out in 2005	Melon (variety Lusitano)	Spain C/Alferez Carlos No. 18 Villamanrique de la Condesa, 21450 Spain	25 g a.s./ha 03.06.06	72	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2006/1024607 and 2007/1011007 Trial No. AF/8816/BA/4 Study to GLP Study carried out in 2005	Melon (variety C5)	Italy Via Nuove 44, Funo, Bologna, 40050	25 g a.s./ha 20.07.05	85	0 2 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007940 Trial No. AF/10490/BA/1 Study to GLP Study carried out in 2006	Melon (variety Cezanne)	France Meauzac 82290 South of EU	25 g a.s./ha 11.07.06	81	0 2 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 84.2% (79.4-89.0%); SD: +/- n/a; CV: n/a%; n=2; fortification range 0.01-0 1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007940 Trial No. AF/10490/BA/2 Study to GLP Study carried out in 2006	Melon (variety Massada F1)	Greece Platanos GR-50032 Imathia Central Macedonia	25 g a.s./ha 30.08.06	88	0 3 7 14	fruit 0.014 fruit <u>0.014</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007940 Trial No. AF/10490/BA/3 Study to GLP Study carried out in 2006	Melon (variety Piel de Sapo)	Spain Los Palacios y Villafranca DNI-34070932-C	25 g a.s./ha 09.06.06	71-83	0 3 7 14	fruit <0.01 fruit <u><0.01</u> fruit <0.01 fruit <0.01	
BASF Doc ID 2007/1007940 Trial No. AF/10490/BA/4 Study to GLP Study carried out in 2006	Melon (variety Tamaris)	Italy Budrio, 40054	25 g a.s./ha 27.07.06	87-88	0 3 7 14	fruit 0.014 fruit <u><0.01</u> fruit <0.01 fruit <0.01	

_ underlined values were used for MRL calculation

Table 2.10.1-2: Residues in melons – glasshouse trials

GAP for EU-N is 1 application at 30 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)
Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days, or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/5000721 Trial No. AGR/11/04 Study to GLP Study carried out in 2004	Melon (variety Lunabel)	Netherlands Limburg (glasshouse)	15 g a.s./ha 01.09.04	87	0 2 7 14	fruit 0.013 fruit <0.01 fruit 0.010 fruit 0.011	BASF analytical method No. 567/0 fruit: mean recovery = 87.5%; SD: +/- 4.6; CV: 5.2; n=6; fortification range 0.01-1.0 mg/kg. Residue analysed as total cypermethrin
BASF Doc ID 2004/5000721 Trial No. ALB/08/04 Study to GLP Study carried out in 2004	Melon (variety Aroma)	Denmark South Jutland (glasshouse)	15 g a.s./ha 27.07.04	79	0 3 7 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. ALO/18/04 Study to GLP Study carried out in 2004	Melon (variety Maxdimon)	Spain Andalucia (glasshouse)	15 g a.s./ha 26.10.04	79	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. FBD/12/04 Study to GLP Study carried out in 2004	Melon (variety Nagaro)	France Rhone-Alpes (glasshouse) (South of EU)	15 g a.s./ha 19.08.04	83	0 4 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. FTL/13/04 Study to GLP Study carried out in 2004	Melon (variety Panchito)	France Languedoc-Roussillon (glasshouse) (South of EU)	15 g a.s./ha 08.06.04	77	0 3 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. ITA/10/04 Study to GLP Study carried out in 2004	Melon (variety Macigno)	Italy Piemonte (glasshouse)	15 g a.s./ha 07.06.04	85	0 3 7 15	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. AGR/11/04 Study to GLP Study carried out in 2004	Melon (variety Lunabel)	Netherlands Limburg (glasshouse)	15 g a.s./ha 2 treatm. last date 01.09.04	87 at last treatm.	0 2 7 14	fruit 0.014 fruit 0.010 fruit 0.019 fruit 0.012	
BASF Doc ID 2004/5000721 Trial No. ALB/08/04 Study to GLP Study carried out in 2004	Melon (variety Aroma)	Denmark South Jutland (glasshouse)	15 g a.s./ha 2 treatm. last date 27.07.04	79 at last treatm.	0 3 7 13	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. ALO/18/04 Study to GLP Study carried out in 2004	Melon (variety Maxdimon)	Spain Andalucia (glasshouse)	15 g a.s./ha 2 treatm. last date 26.10.04	79 at last treatm.	0 3 7 14	fruit 0.011 fruit 0.011 fruit <0.01 fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 87.5%; SD: +/- 4.6; CV: 5.2; n=6; fortification range 0.01-1.0 mg/kg. Residue analysed as total cypermethrin

Table 2.10.1-2: Residues in melons – glasshouse trials

GAP for EU-N is 1 application at 30 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)
Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days, or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/5000721 Trial No. FBD/12/04 Study to GLP Study carried out in 2004	Melon (variety Nagaro)	France Rhone-Alpes (glasshouse) (South of EU)	15 g a.s./ha 2 treatm. last date 19.08.04	83 at last treatm.	0 4 7 14	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2004/5000721 Trial No. FTL/13/04 Study to GLP Study carried out in 2004	Melon (variety Panchito)	France Languedoc-Roussillon (glasshouse) (South of EU)	15 g a.s./ha 2 treatm. last date 08.06.04	77 at last treatm.	0 3 7 14	fruit 0.011 fruit 0.011 fruit 0.010 fruit 0.013	
BASF Doc ID 2004/5000721 Trial No. ITA/10/04 Study to GLP Study carried out in 2004	Melon (variety Macigno)	Italy Piemonte (glasshouse)	15 g a.s./ha 2 treatm. last date 07.06.04	85 at last treatm.	0 3 7 15	fruit <0.01 fruit <0.01 fruit <0.01 fruit <0.01	
BASF Doc ID 2006/1037507 Trial No. 05I CL FR P34 Study to GLP Study carried out in 2005	Melon (variety Anasta)	France 13560 Senas Provence (South of EU)	40 g a.s./ha 14.06.05 glasshouse	71	0 3 7 13	fruit <0.01 fruit <0.01 fruit <u>≤0.01</u> fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 96.3%; SD: +/- 10.5; CV: 10.9%; n=4; fortification range 0.01-1.0 mg/kg. Residue analysed as total cypermethrin
BASF Doc ID 2006/1037507 Trial No. AGR/54/05 Study to GLP Study carried out in 2005	Melon (variety Delta F1)	Belgium 3454 Rummen Limburg	40 g a.s./ha 30.08.05 glasshouse	87	0 3 7 13	fruit 0.058 fruit 0.057 fruit <u>0.022</u> fruit 0.030	
BASF Doc ID 2006/1037507 Trial No. AGR/53/05 Study to GLP Study carried out in 2005	Melon (variety Delta F1)	Germany 47574 Goch-Hülm North Rhine-Westphalia	40 g a.s./ha 29.08.05 glasshouse	87	0 3 8 14	fruit 0.047 fruit 0.045 fruit <u>0.029</u> fruit 0.022	
BASF Doc ID 2006/1037507 Trial No. ALB/190508-01 Study to GLP Study carried out in 2005	Melon (variety Aroma)	Denmark 7080 Børkop	40 g a.s./ha 13.07.05 glasshouse	75	0 2 7 14	fruit 0.012 fruit 0.010 fruit <u>≤0.01</u> fruit <0.01	
BASF Doc ID 2006/1037507 Trial No. 05 I CL FR P38 Study to GLP Study carried out in 2005	Melon (variety Luna Star)	France 13560 Senas Provence (South of EU)	40 g a.s./ha 08.06.05 glasshouse	71	0 3 7 13	fruit <0.01 fruit 0.012 fruit <u>≤0.01</u> fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 96.3%; SD: +/- 10.5; CV: 10.9%; n=4; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1037507 Trial No. IR05BASG61LG01 Study to GLP Study carried out in 2005	Melon (variety Creso)	Italy 40014 Palata Pepoli Bologna	40 g a.s./ha 10.06.05 glasshouse	87	0 3 7 14	fruit <0.01 fruit <0.01 fruit <u>≤0.01</u> fruit <0.01	BASF analytical method No. 567/0 fruit: mean recovery = 96.3%; SD: +/- 10.5; CV: 10.9%; n=4; fortification range 0.01-1.0 mg/kg Residue analysed as total

Table 2.10.1-2: Residues in melons – glasshouse trials

GAP for EU-N is 1 application at 30 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)
Glasshouse GAP is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days, or 1 application with 50 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1037507 Trial No. 05ES/085R Study to GLP Study carried out in 2005	Melon (variety Maxdimon)	Spain 41710 Utrera Andalucia Sevilla	40 g a.s./ha 18.11.05 glasshouse	79	0 3 7 13	fruit 0.074 fruit 0.070 fruit <u>0.048</u> fruit 0.045	cypermethrin
BASF Doc ID 2006/1037507 Trial No. 05RF046 Study to GLP Study carried out in 2005	Melon (variety Gali F1)	Greece Profitis Thessaloniki Central Macedonia	40 g a.s./ha 30.07.05 glasshouse	84	0 3 7 14	fruit <0.01 fruit <0.01 fruit <u><0.01</u> fruit <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in melons presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to open field conditions in the Southern European region and to glasshouse conditions.
The proposed GAP is

- for the Northern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days under open field conditions
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 2 days under open field conditions
- glasshouse GAP is a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 2 days or a single application with 50 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days

The following residue studies were presented:

- one treatment at a target rate of 25 g a.s./ha under open field conditions-8 trials conducted in the Southern European region
- one or two treatments at a target rate of 15 g a.s./ha under glasshouse conditions-6 trials including both variants, 2 of them conducted in the Northern European region and 4 of them conducted in the Southern European region
- one treatment at a target rate of 40 g a.s./ha under glasshouse conditions-8 trials, 3 of them conducted in the Northern European region and 5 of them conducted in the Southern European region

After one application at 25 g a.s./ha under field conditions, initial residues of 0.014 mg/kg were found in two out of eight trials. Residues of 0.014 mg/kg were found in one treated sample 3 days after application. No residues above the limit of quantitation of the analytical method (LOQ; 0.01 mg/kg) were found in any other treated sample from these eight trials, regardless of sampling interval (all <0.01 mg/kg).

After one application of alpha-cypermethrin at a rate of 15 g a.s./ha under glasshouse conditions, residues were below the LOQ (<0.01 mg/kg) in all treated melon fruit samples in five out of six trials. Determinable residues were found only in three samples from trial AGR/11/04; these samples were collected at 0-, 7- and 14-days after application, and contained residues of 0.013, 0.010, and 0.011 mg/kg, respectively.
Immediately after the last of two applications at a rate of 15 g a.s./ha under glasshouse conditions, residues in melon fruit ranged from below the LOQ (<0.01 mg/kg) to 0.014 mg/kg, and were generally below or near the LOQ for the remainder of the study. Residues were between <0.01 mg/kg (LOQ) and 0.013 mg/kg in melon samples collected at the end of the study, 13-15 days after the last application.

After one treatment with 40 g alpha-cypermethrin/ha under glasshouse conditions, initial residues ranged between <0.01 mg/kg and 0.074 mg/kg. Residues declined to <0.01-0.070 mg/kg, <0.01-0.048 mg/kg and <0.01-0.045 mg/kg at 2-3, 7-8 and 13-14 days after application, respectively.

Conclusion:

In the years 2004-2006 a residue program in melons was conducted in Belgium, Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain and the Netherlands under open field and glasshouse conditions.

The trial data demonstrate that residues in melon fruit arising from treatment with alpha-cypermethrin according to GAP are very low under field conditions.
In 8 trials supporting the proposed field GAP for the Southern European region, residues of 0.014 mg/kg were found in one treated sample 3 days after application. No residues above the limit of quantitation of the analytical method (LOQ; 0.01 mg/kg) were found in any other treated sample at the target PHI of 3±1 days.

As expected, there is a tendency to higher residue values under glasshouse conditions as compared to the open field situation.

In 8 trials supporting the proposed glasshouse GAP, residues ranged between <0.01-0.07 mg/kg at the target PHI of 3±1 days and between <0.01-0.048 mg/kg at the target PHI of 7±1 days.

2.10.2 Processing study in melon

Report:	Schroth E. 2006a Study on the residue behavior of Alpha-cypermethrin on melons after application of BAS 310 41 I under greenhouse conditions in Spain, 2004 2006/1038660
Guidelines:	91/414/EEC, 1607VI/97 rev. 2, 10.6.1999 EC 7029/VI/95 rev. 5, 22.7.1997 EC 7525/VI/95 rev. 7, 12.6.2001
GLP:	yes (certified by: Testing facility: not reported, Analytical lab.: Landesamt für Umweltschutz und Gewerbeaufsicht, Main, Germany)

Material and methods:

During the 2004 growing season, 4 greenhouse trials were conducted in different representative melon growing areas in Spain to determine the distribution of residue levels of alpha-cypermethrin (BAS 310 I) between peel and pulp of melon fruit. Two trials (ALO/01/04 and ALO/02/04) were conducted in climbing melons (variety Ciclo), and two (ALO/03/04 and ALO/04/04) were conducted in ground melons (varieties Maxdimon and Piñonet).

A soluble concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 41 I) was foliar applied twice with a target application rate of 75 g a.s./ha. Actual rates were within +/10 % of the nominal rate. This exaggerated treatment rate was used in an effort to generate finite alpha-cypermethrin residues in melons so that the distribution of the residue between peel and pulp could be better quantified.

The applications were made 14(±1) and 7(±1) days before harvest. The growth stage at the last application was BBCH 81 (10% of fruits show typical fully ripe colour) for trials ALO/01/04 and ALO/02/04; and BBCH 79 (9 or more fruits on main stem have reached typical size and form) for trials ALO/03/04 and ALO/04/04. The spray volume used was 400 I/ha.

Specimens of fruits were collected at the day of the last application as well as 6 (ALO/01/04 and ALO/02/04) or 7 days (ALO/03/04 and ALO/04/04) thereafter. Specimens of peel and pulp were collected 6-7 days after the last application. The samples from the four trials were frozen within 24 hours of sampling remained frozen at or below -18°C, including during transportation, until analysis.
The analytical part of the study was performed at the Agricultural Research Center of BASF in Limburgerhof, Germany.

The specimens (RAC) as well as the processed commodity samples were analysed for alpha-cypermethrin by means of HPLC-MS/MS according to the BASF method no. 567/0. The limit of quantification (LOQ) of the method was 0.01 mg/kg for all sample materials analysed.
Method performance was checked by determining the procedural recoveries in melon matrices.
At fortification levels between 0.01 and 1.0 mg/kg, the recovery rates averaged at 76% with a standard deviation of 3.9 and a coefficient of variation of 5.1%.

Findings:

No detectable alpha-cypermethrin residues above the validated method LOQ (0.01 mg/kg) were found in any of the untreated melon fruit specimens or processed fractions (peel and pulp) obtained from untreated melon fruit specimens.

No residues above the LOQ (0.01 mg/kg) were found in any of the treated samples from trial ALO/03/04 or in the processed fractions (peel and pulp) obtained from treated melon fruit.

In the remaining three trials, initial residues ranged between 0.02 – 0.03 mg/kg. On day 6-7 after the last application, residues in melon fruit ranged between 0.01 – 0.02 mg/kg. No detectable residues were found in pulp specimens obtained from treated melon fruit (all <0.01 mg/kg), while the peel fractions showed residues between 0.01-0.07 mg/kg.

The residue levels in the individual fractions are summarized below:

Table 2.10.2-1: Residue distribution between peel and pulp of melons – DocID 2006/8038660

Trial No:	Alpha-cypermethrin residue (mg/kg)			
	Melon fruit	Melon fruit	Peel	Pulp
	0 DALA [1)]	6-7 DALA	6-7 DALA	6-7 DALA
ALO/0104	0.03	0.02	0.07	<0.01
ALO/0204	0.02	0.02	0.05	<0.01
ALO/03/04	<0.01	<0.01	<0.01	<0.01
ALO/0404	0.02	0.01	0.01	<0.01

1) days after last application

Conclusion:

The data from this processing study clearly demonstrate that there are no detectable residues of alpha-cypermethrin in the pulp (edible part) of melons. The residue is located exclusively on the peel.

2.10.3 Estimation of MRL, HR and STMR for melon

For *melon*, the following residue studies were considered (BASF DocIDs): 2006/1024607, 2007/1011007, 2007/1007940 and 2006/1037507.

The following residue values (PHI=3±1 days for the open field trials and PHI=7±1for the glassnouse trials) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1024607 2007/1011007 (field)	-	<0.01 (4x)
2007/1007940 (field)	-	<0.01 (3x), 0.014
OECD-MRL-calculation	-	**<u>0.02</u> (n=8, STMR=0.01, HR=0.014)**
2006/1037507 (glasshouse)		<0.01 (5x), 0.022, 0.029, 0.048
OECD-MRL-calculation		**<u>0.08</u> (n=8, STMR=0.01, HR=0.048)**

_ underlined values were used for risk assessment purposes

<u>Pumpkin</u>

As the critical GAPs for melons and pumpkins are identical, according to European Community Guideline 7525/VI/95 rev. 9 dated March 2011 extrapolation from melons to pumpkins is adequate.

2.11 Broccoli

Residue data from supervised trials in broccoli were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.11-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Broccoli (field)	2005	4	DE, FR, NL, UK	2	FR, IT	6	2006/1026863
Broccoli (field)	2006	4	DE, DK, FR, UK	2	ES, GR	6	2007/1013274
Total number of trials per region		**8**		**4**	**Total number of trials**	**12**	

2.11.1 Supervised residue trials in broccoli

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of BAS 310 I in broccoli after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026863

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the Residue Behaviour of BAS 310 I in Broccoli after Treatment with BAS 310 40 I under open field Conditions in Southern and Northern Europe, 2006; BASF DocID 2007/1013274

Material and Methods:

In the years 2005 and 2006 a residue program on broccoli was conducted in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, The Netherlands and the United Kingdom under field conditions.

During the 2005 growing season, six field trials were conducted in different representative broccoli growing areas in the United Kingdom, Germany, the Netherlands, France and Italy.

In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to broccoli plants either once or twice to separate plots at a target rate of 12.5 g a.s./ha.
The growth stage of the plants at application was between 45 (50% of the expected head diameter reached) and 55 (first individual flowers visible (still closed)). Specimens of broccoli inflorescences were collected immediately after the last application from each plot, as well as 3, 7 and 14 days thereafter.

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to broccoli plants once at a target rate of 25 g a.s./ha.
The growth stage of the plants at application was between 46 and 47 (60 - 70% of the expected head diameter reached). Specimens of broccoli inflorescence were collected immediately after the last application, as well as 3, 7 and 14 days thereafter.
Specimens were analysed using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2006 growing season, six field trials were conducted in different representative broccoli growing areas in Denmark, France (Northern European region), Germany, Greece, Spain and the United Kingdom.

In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to broccoli plants either once or twice to separate plots at a target rate of 12.5 g a.s./ha.
The growth stage of the plants at application was between 43 (30% of the expected head diameter reached) and 49 (typical size and form reached; head tightly closed).
Specimens of broccoli inflorescence were collected immediately after the last application from each plot, as well as 3, 7 and 14 days thereafter.

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to broccoli plants once at a target rate of 25 g a.s./ha.
The growth stage of the plants at application was between 47 (70% of the expected head diameter reached) and 49 (typical size and form reached; head tightly closed). Specimens of broccoli inflorescence were collected immediately after the last application, as well as 2-4, 6-7 and 14 days thereafter.
Specimens were analysed using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantification of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.11.1-1.

Table 2.11.1-1: Residues in broccoli

GAP for EU-N is 1-2 applications at 15 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/1 Study to GLP Study carried out in 2005	Broccoli (variety Ovation)	Germany 67227 Frankenthal	12.5 g as/ha 27.06.05	55	0 3 7 14	inflorescence 0.163 inflorescence 0.048 inflorescence <0.01 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 94.2 %; SD: +/- 17.8; CV: 18.8; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/2 Study to GLP Study carried out in 2005	Broccoli (variety Volta)	The Netherlands 4756 SB Kruisland, Brabant	12.5 g as/ha 20.09.05	44	0 3 7 14	inflorescence 0.044 inflorescence 0.029 inflorescence 0.015 inflorescence <0.01	
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/3 Study to GLP Study carried out in 2005	Broccoli (variety Chevalier)	United Kingdom Westgate Farm, Guide Road, Hesketh Bank, Lancashire PR4 6XS	12.5 g as/ha 10.10.05	44	0 3 7 14	inflorescence 0.056 inflorescence 0.033 inflorescence 0.016 inflorescence <0.01	
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/4 Study to GLP Study carried out in 2005	Broccoli (variety Marathon)	France Varennes-sur-Loire, 49730 Maine-et-Loire (North of EU)	12.5 g as/ha 07.10.05	45	0 3 7 14	inflorescence 0.035 inflorescence 0.018 inflorescence 0.013 inflorescence <0.01	
BASF Doc ID 2007/1013274 Trial No. A/NF/I/06/120 Study to GLP Study carried out in 2006	Broccoli (variety Monterey)	France Sedan Champagne-Ardennes (North of EU)	13 g as/ha 13.09.06	45	0 3 7 13	inflorescence 0.036 inflorescence 0.016 inflorescence 0.010 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 89.7%; SD: +/- 13.7; CV: 15.3; n=6; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013274 Trial No. A/GE/I/06/121 Study to GLP Study carried out in 2006	Broccoli (variety Ironman)	Germany Erfurt Thüringen	12.5 g as/ha 25.10.06	48	0 3 6 14	inflorescence 0.032 inflorescence 0.030 inflorescence 0.017 inflorescence 0.017	BASF method N 567/0 inflorescences: mean recovery = 89.7%; SD: +/- 13.7; CV: 15.3; n=6; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013274 Trial No. A/DK/I/06/122 Study to GLP Study carried out in 2006	Broccoli (variety Marathon)	Denmark Middelfart Fyn	13 g as/ha 12.07.06	43	0 2 7 14	inflorescence 0.053 inflorescence 0.030 inflorescence 0.014 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 89.7%; SD: +/- 13.7; CV: 15.3; n=6; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013274 Trial No. A/UK/I/06/123 Study to GLP Study carried out in 2006	Broccoli (variety Marathon)	United Kingdom Chipping Campden Gloucestershire	13 g as/ha 04.08.06	48	0 3 7 15	inflorescence 0.044 inflorescence 0.032 inflorescence 0.025 inflorescence <0.01	

Table 2.11.1-1: Residues in broccoli

GAP for EU-N is 1-2 applications at 15 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/1 Study to GLP Study carried out in 2005	Broccoli (variety Ovation)	Germany 67227 Frankenthal	12.5 g as/ha 2 treatm. last date 27.06.05	55 at last treatm.	0 3 7 14	inflorescence 0.259 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 94.2 %; SD: +/- 17.8; CV: 18.8; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/2 Study to GLP Study carried out in 2005	Broccoli (variety Volta)	The Netherlands 4756 SB Kruisland, Brabant	12.5 g as/ha 2 treatm. last date 20.09.05	44 at last treatm.	0 3 7 14	inflorescence 0.074 inflorescence 0.039 inflorescence 0.015 inflorescence <0.01	
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/3 Study to GLP Study carried out in 2005	Broccoli (variety Chevalier)	United Kingdom Westgate Farm, Guide Road, Hesketh Bank, Lancashire PR4 6XS	12.5 g as/ha 2 treatm. last date 10.10.05	44 at last treatm.	0 3 7 14	inflorescence 0.048 inflorescence 0.015 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/4 Study to GLP Study carried out in 2005	Broccoli (variety Marathon)	France Varennes-sur-Loire, 49730 Maine-et-Loire (North of EU)	12.5 g as/ha 2 treatm. last date 07.10.05	45 at last treatm.	0 3 7 14	inflorescence 0.069 inflorescence 0.034 inflorescence 0.018 inflorescence <0.01	
BASF Doc ID 2007/1013274 Trial No. A/NF/I/06/120 Study to GLP Study carried out in 2006	Broccoli (variety Monterey)	France Sedan Champagne-Ardennes (North of EU)	14/12 g as/ha 2 treatm. last date 13.09.06	45 at last treatm.	0 3 7 13	inflorescence 0.050 inflorescence 0.017 inflorescence 0.011 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 89.7%; SD: +/- 13.7; CV: 15.3; n=6; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013274 Trial No. A/GE/I/06/121 Study to GLP Study carried out in 2006	Broccoli (variety Ironman)	Germany Erfurt Thüringen	12.5 g as/ha 2 treatm. last date 25.10.06	49 at last treatm.	0 3 6 14	inflorescence 0.052 inflorescence 0.035 inflorescence 0.029 inflorescence 0.018	
BASF Doc ID 2007/1013274 Trial No. A/DK/I/06/122 Study to GLP Study carried out in 2006	Broccoli (variety Marathon)	Denmark Middelfart Fyn	12/13 g as/ha 2 treatm. last date 12.07.06	46 at last treatm.	0 2 7 14	inflorescence 0.052 inflorescence 0.032 inflorescence 0.017 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 89.7%; SD: +/- 13.7; CV: 15.3; n=6; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013274 Trial No. A/UK/I/06/123 Study to GLP Study carried out in 2006	Broccoli (variety Marathon)	United Kingdom Chipping Campden Gloucestershire	12/13 g as/ha 2 treatm. last date 04.08.06	48 at last treatm.	0 3 7 15	inflorescence 0.050 inflorescence 0.036 inflorescence 0.018 inflorescence <0.01	

Table 2.11.1-1: Residues in broccoli

GAP for EU-N is 1-2 applications at 15 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/5 Study to GLP Study carried out in 2005	Broccoli (variety Chevalier)	France Canals, 82170 Tarn-et-Garonne (South of EU)	25 g as/ha 06.06.05	47	0 3 7 14	inflorescence 0.070 inflorescence <u>0.028</u> inflorescence 0.017 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 94.2 %; SD: +/- 17.8; CV: 18.8; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026863 Trial No. AF/8814/BA/6 Study to GLP Study carried out in 2005	Broccoli (variety Eyron)	Italy 40057 Granarolo (Bo), Emilia Romagne	25 g as/ha 17.10.05	46-47	0 3 7 14	inflorescence 0.065 inflorescence <u>0.047</u> inflorescence 0.027 inflorescence 0.011	
BASF Doc ID 2007/1013274 Trial No. A/GR/I/06/124 Study to GLP Study carried out in 2006	Broccoli (variety Marathon)	Greece Chalkidona Thessaloniki Central Macedonia	25 g as/ha 03.10.06	47-48	0 3 7 14	inflorescence 0.056 inflorescence <u>0.032</u> inflorescence 0.014 inflorescence <0.01	BASF method N 567/0 inflorescences: mean recovery = 89.7%; SD: +/- 13.7; CV: 15.3; n=6; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013274 Trial No. A/SP/I/06/125 Study to GLP Study carried out in 2006	Broccoli (variety Monaco)	Spain Nava Campana (Hellin) Abacete Castilla la Mancha	27 g as/ha 24.10.06	49	0 4 7 13	inflorescence 0.050 inflorescence <u>0.026</u> inflorescence 0.013 inflorescence <0.01	

_ underlined values were used for MRL calculation

Findings:
The residue studies in broccoli presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions – 8 trials including both variants; all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions – 4 trials conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues in broccoli inflorescences ranged between 0.032 mg/kg and 0.163 mg/kg. Residues declined to levels between <0.01 – 0.048 mg/kg, and <0.01 – 0.025 mg/kg at 2-3 and 6-7 days after application. At the last sampling, 13-15 days after application, only in one single trial a residue of 0.017 mg/kg was found. All other treated samples did not show determinable residues (all <0.01 mg/kg).

After two applications at 12.5 g a.s./ha, initial residues in broccoli inflorescences ranged between 0.259 mg/kg and 0.048 mg/kg. Residues declined to levels between <0.01 – 0.039 mg/kg and <0.01 – 0.029 mg/kg at 2-3 and 6-7 days after the last application. At the last sampling, 13-15 days after application, only in one single trial a residue of 0.018 mg/kg was found. All other treated samples did not show determinable residues (all <0.01 mg/kg).

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in broccoli ranged between 0.050 mg/kg and 0.070 mg/kg. Residues declined to levels between 0.026 – 0.047 mg/kg, 0.013 – 0.027 mg/kg and <0.01 – 0.011 mg/kg at 3-4, 7 and 13-14 days after application.

Conclusion:

In 8 trials supporting the GAP for the Northern European region, residues in broccoli inflorescences ranged between <0.01 – 0.029 mg/kg at the target PHI of 7 (± 1) days.
In 4 trials supporting the GAP for the Southern European region, residues in broccoli inflorescences ranged between 0.026 – 0.047 mg/kg at the target PHI of 3 (± 1) days.

2.11.2 Estimation of MRL, HR and STMR for broccoli

For *broccoli*, the following residue studies were considered (BASF DocIDs): 2006/1026863 and 2007/1013274.

The following residue values (EU-N: PHI=7±1 days / EU-S: 3±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026863 (open field)	<0.01 (2x), 0.015, 0.018	0.028, 0.047
2007/1013274 (open field)	0.011, 0.017, 0.018, 0.029	0.026, 0.032
OECD-MRL-calculation (open field)	**0.05 (n=8, STMR=0.016, HR=0.029)**	**<u>0.1</u> (n=4, STMR=0.030, HR=0.047)**

_ underlined values were used for risk assessment purposes

2.12 Cauliflower

Residue data from supervised trials in cauliflower were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.12-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Cauliflower (field)	2005	4	DE, FR, NL, UK	2	FR, IT	6	2006/1026864
Cauliflower (field)	2006	2	DE, UK	2	FR, IT	4	2007/1007936
Cauliflower (field)	2006	4	DK, FR, NL, UK	2	ES, GR	6	2007/1008495
Total number of trials per region		**10**		**6**	**Total number of trials**	**16**	

2.12.1 Supervised residue trials in cauliflower

<u>Evans L (2007)</u>

Full study reference

Evans L (2007): Study on the residue behaviour of BAS 310 I in cauliflower after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026864

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alphacypermethrin in Cauliflower after treatment with BAS 310 40 I or BAS 310 08 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007936

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the residue behaviour of BAS 310 I in cauliflower after treatment with BAS 310 40 I under open field conditions in Southern and Northern Europe, 2006; BASF DocID 2007/1008495

Material and Methods:

In the years 2005 and 2006 a residue program on cauliflower was conducted in the field in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.

A total of six residue trials was conducted during the growing season 2005 in France (Northern and Southern European region), Germany, Italy, the Netherlands and the United Kingdom.
In four of these trials located in the Northern European region, separate plots of cauliflower plants received either one or two treatments with a 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC; BAS 310 40 I) at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last treatment was between 41 (cauliflower heads begin to form; width of growing tip > 1 cm) and 47 (70% of the expected head diameter reached). Samples of cauliflower heads were collected immediately after application as well as approximately 3, 7 and 14 days after the last application.
In two trials located in the Southern European region, cauliflower plants received a single treatment with a 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC; BAS 310 40 I) at a target rate of 25 g a.s./ha. The growth stage of the plants at the last treatment was between 43 (30% of the expected head diameter reached) and 47 (70% of the expected head diameter reached). Samples of cauliflower heads were collected immediately after application as well as 3, 7 and 14 days after the last application.
Residue analysis was performed according to BASF analytical method No. 567/0, which determines total cypermethrin residues by means of HPLC-MS/MS with a LOQ of 0.01 mg/kg.

A total of six residue trials was conducted during the growing season 2006 in Denmark, France (Northern European region), Greece, Spain, the Netherlands and the United Kingdom.
In four of these trials located in the Northern European region, separate plots of cauliflower plants received either one or two treatments with a 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC; BAS 310 40 I) at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last treatment was between 42 and 47 (70% of the expected head diameter reached). Samples of cauliflower heads were collected immediately after application as well as 3, 6-8 and 13-14 days after the last application.

In two trials located in the Southern European region, cauliflower plants received a single treatment with a 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC; BAS 310 40 I) at a target rate of 25 g a.s./ha. The growth stage of the plants at the last treatment was between 42 and 48 (80% of the expected head diameter reached). Samples of cauliflower heads were collected immediately after application as well as 3-4, 7 and 14 days after the last application.
Residue analysis was performed according to BASF analytical method No. 567/0, which determines total cypermethrin residues by means of HPLC-MS/MS with a LOQ of 0.01 mg/kg.

During the 2006 growing season, four bridging trials were conducted in France (Southern European region), Germany, Italy and the United Kingdom under open field conditions to compare the residue behaviour after application of two different formulations of alpha-cypermethrin.
An emulsifiable concentrate formulation (EC 100 g a.s./L, BAS 310 40 I) and a wettable granule formulation (WG 150 g/kg, BAS 310 08 I) were foliar applied to separate plots of cauliflower plants, each one at the following rates: One treatment at 12.5 g a.s./ha (variant 1), two treatments at 12.5 g a.s./ha (variant 2), or one treatment at 25 g a.s./ha (variant 3). Variants 1 and 2 were included in the trials located in the Northern European region and variant 3 was included in the trials located in the Southern European region.
At the last application, the growth stages of the plants were between 43 (30% of the expected head diameter reached) and 45 (50% of the expected head diameter reached) in the Northern European trials and at stage 45 in the Southern European trials. Samples of cauliflower heads were collected immediately after the last application as well as 2-4, 6-8, and 13-14 days afterwards. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.12.1-1.

Table 2.12.1-1: Residues in cauliflower

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/1 Study carried out in 2005	Cauliflower (variety Optimist)	France Rue des Bouches d´Or Coutures, 49320 North of EU	12.5 g a.s./ha 11.10.05	41-43	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 88.4%; SD: +/- 15.1; CV: 17.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/2 Study carried out in 2005	Cauliflower (variety Unifor)	United Kingdom Marston, Cudworth Warwickshire	12.5 g a.s./ha 16.09.05	43	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/3 Study carried out in 2005	Cauliflower (variety Freedom)	Germany Ippenstedter Straße 16 30982 Pattensen/Jeinsen	12.5 g a.s./ha 30.09.05	43-47	0 3 7 14	inflorescence 0.123 inflorescence 0.042 inflorescence 0.053 inflorescence 0.011	
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/4 Study carried out in 2005	Cauliflower (variety Fremont)	The Netherlands Rijksstraatweg 92, 328 L W Numansdorp, Zuid-Holland	12.5 g a.s./ha 16.08.05	43-45	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1008495 Trial No. A/NF/I/06/136 Study carried out in 2006	Cauliflower (variety Cartier)	France Sedan Champagne-Ardennes (North of EU)	12.7 g a.s./ha 08.10.06	47	0 3 6 13	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 96.0%; SD: +/- 8.8; CV: 9.2; n=6; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008495 Trial No. A/UK/I/06/137 Study carried out in 2006	Cauliflower (variety Freemont)	United Kingdom Chipping Campden Gloucestershire	12.6 g a.s./ha 17.09.06	42	0 3 8 13	inflorescence 0.022 inflorescence 0.025 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1008495 Trial No. A/NL/I/06/138 Study carried out in 2006	Cauliflower (variety Fremont)	The Netherlands LB Angeren Gelderland	12.5 g a.s./ha 26.09.06	45-46	0 3 7 14	inflorescence 0.028 inflorescence 0.017 inflorescence 0.013 inflorescence <0.01	
BASF Doc ID 2007/1008495 Trial No. A/DK/I/06/139 Study carried out in 2006	Cauliflower (variety Fremont)	Denmark Middelfart Fyn	12.1 g a.s./ha 01.08.06	45	0 3 8 14	inflorescence 0.028 inflorescence 0.032 inflorescence 0.012 inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 96.0%; SD: +/- 8.8; CV: 9.2; n=6; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin

Table 2.12.1-1: Residues in cauliflower

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/1 Study carried out in 2006	Cauliflower (variety Gregor)	Germany Oggersheimer Straße 125 a/b 67071 Ludwigshafen Ruchheim Rheinland-Pfalz	12.5 g a.s./ha (EC) 13.09.06	43-45	0 2 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescence: mean recovery = 77.5%; SD: +/- 15.7; CV: 20.2%; n=8; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/1 Study carried out in 2006	Cauliflower (variety Gregor)	Germany Oggersheimer Straße 125 a/b 67071 Ludwigshafen Ruchheim Rheinland-Pfalz	12.5 g a.s./ha (WG) 13.09.06	43-45	0 2 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/2 Study carried out in 2006	Cauliflower (variety Correll)	United Kingdom Marshalls Butterwick, Boston Lincolnshire PE20 3SS	12.5 g a.s./ha (EC) 23.09.06	43-45	0 4 6 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/2 Study carried out in 2006	Cauliflower (variety Correll)	United Kingdom Marshalls Butterwick, Boston Lincolnshire PE20 3SS	12.5 g a.s./ha (WG) 23.09.06	43-45	0 4 6 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/1 Study carried out in 2005	Cauliflower (variety Optimist)	France Rue des Bouches d´Or Coutures, 49320 North of EU	12.5 g a.s./ha 2 treatm. last date 11.10.05	41-43 at last treatm.	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 88.4%; SD: +/- 15.1; CV: 17.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/2 Study carried out in 2005	Cauliflower (variety Unifor)	United Kingdom Marston, Cudworth Warwickshire	12.5 g a.s./ha 2 treatm. last date 16.09.05	43 at last treatm.	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/3 Study carried out in 2005	Cauliflower (variety Freedom)	Germany Ippenstedter Straße 16 30982 Pattensen/Jeinsen	12.5 g a.s./ha 2 treatm. last date 30.09.05	43-47 at last treatm.	0 3 7 14	inflorescence 0.201 inflorescence 0.075 inflorescence 0.085 inflorescence 0.027	BASF analytical method No. 567/0 inflorescences: mean recovery = 88.4%; SD: +/- 15.1; CV: 17.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/4 Study carried out in 2005	Cauliflower (variety Fremont)	The Netherlands Rijksstraatweg 92, 328 L W Numansdorp, Zuid-Holland	12.5 g a.s./ha 2 treatm. last date 16.08.05	43-45 at last treatm.	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	

Table 2.12.1-1: Residues in cauliflower

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008495 Trial No. A/NF/I/06/136 Study carried out in 2006	Cauliflower (variety Cartier)	France Sedan Champagne-Ardennes (North of EU)	12.0/ 12.3 g a.s./ha 2 treatm. last date 08.10.06	47 at last treatm.	0 3 6 13	inflorescence <0.01 inflorescence <0.01 inflorescence <u><0.01</u> inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 96.0%; SD: +/- 8.8; CV: 9.2; n=6; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008495 Trial No. A/UK/I/06/137 Study carried out in 2006	Cauliflower (variety Freemont)	United Kingdom Chipping Campden Gloucestershire	12.0/12.3 g a.s./ha 2 treatm. last date 17.09.06	42 at last treatm.	0 3 7 14	inflorescence 0.056 inflorescence 0.017 inflorescence <u><0.01</u> inflorescence <0.01	
BASF Doc ID 2007/1008495 Trial No. A/NL/I/06/138 Study carried out in 2006	Cauliflower (variety Fremont)	The Netherlands LB Angeren Gelderland	12.4/12.7 g a.s./ha 2 treatm. last date 26.09.06	45-46 at last treatm.	0 3 7 14	inflorescence 0.036 inflorescence 0.017 inflorescence <u>0.010</u> inflorescence <0.01	
BASF Doc ID 2007/1008495 Trial No. A/DK/I/06/139 Study carried out in 2006	Cauliflower (variety Fremont)	Denmark Middelfart Fyn	12.6/12.4 g a.s./ha 2 treatm. last date 01.08.06	45 at last treatm.	0 3 8 14	inflorescence 0.048 inflorescence 0.024 inflorescence <u>0.011</u> inflorescence <0.01	
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/1 Study carried out in 2006	Cauliflower (variety Gregor)	Germany Oggersheimer Straße 125 a/b 67071 Ludwigshafen Ruchheim Rheinland-Pfalz	12.5 g a.s./ha (EC) 2 treatm. last date 13.09.06	43-45 at last treatm.	0 2 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <u><0.01</u> inflorescence <0.01	BASF analytical method No. 567/0 inflorescence: mean recovery = 77.5%; SD: +/- 15.7; CV: 20.2%; n=8; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/1 Study carried out in 2006	Cauliflower (variety Gregor)	Germany Oggersheimer Straße 125 a/b 67071 Ludwigshafen Ruchheim Rheinland-Pfalz	12.5 g a.s./ha (WG) 2 treatm. last date 13.09.06	43-45 at last treatm.	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescence: mean recovery = 77.5%; SD: +/- 15.7; CV: 20.2%; n=8; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/2 Study carried out in 2006	Cauliflower (variety Correll)	United Kingdom Marshalls Butterwick, Boston Lincolnshire PE20 3SS	12.5 g a.s./ha (EC) 2 treatm. last date 23.09.06	43-45 at last treatm.	0 4 6 14	inflorescence <0.01 inflorescence <0.01 inflorescence <u><0.01</u> inflorescence <0.01	
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/2 Study carried out in 2006	Cauliflower (variety Correll)	United Kingdom Marshalls Butterwick, Boston Lincolnshire PE20 3SS	12.5 g a.s./ha (WG) 2 treatm. last date 23.09.06	43-45 at last treatm.	0 3 7 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	

Table 2.12.1-1: Residues in cauliflower

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/5 Study carried out in 2005	Cauliflower (variety Fremont)	France 2274 Ir de Phyppodrome Mereville, 31330 South of EU	25 g a.s./ha 30.09.05	43-47	0 3 7 14	inflorescence <0.01 inflorescence <u>≤0.01</u> inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 88.4%; SD: +/- 15.1; CV: 17.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026864 Trial No. AF/8813/BA/6 Study carried out in 2005	Cauliflower (variety Emeraude)	Italy Az Agr Gamberini, Vie Cadriono 60/2 Viadagola, 40057	25 g a.s./ha 08.11.05	43-46	0 3 7 14	inflorescence 0.014 inflorescence <u>≤0.01</u> inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1008495 Trial No. A/SP/I/06/140 Study carried out in 2006	Cauliflower (variety Flamenco)	Spain Benicarlo-Castello Castellon	27.0 g a.s./ha 19.10.06	46	0 4 7 14	inflorescence 0.016 inflorescence <u>≤0.01</u> inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescences: mean recovery = 96.0%; SD: +/- 8.8; CV: 9.2; n=6; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008495 Trial No. A/GR/I/06/141 Study carried out in 2006	Cauliflower (variety Siria)	Greece Chalkidona Thessaloniki Central Macedonia	26.2 g a.s./ha 06.10.06	42-48	0 3 7 14	inflorescence 0.014 inflorescence <u>≤0.01</u> inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/3 Study carried out in 2006	Cauliflower (variety Dunia)	Italy Agr. Gamberini Via Cadriano 60 Granarolo 40054 Bologna	25 g a.s./ha (EC)- 20.09.06	45	0 2 8 13	inflorescence 0.16 inflorescence 0.05 inflorescence <0.01 inflorescence <0.01	BASF analytical method No. 567/0 inflorescence: mean recovery = 77.5%; SD: +/- 15.7; CV: 20.2%; n=8; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/3 Study carried out in 2006	Cauliflower (variety Dunia)	Italy Agr. Gamberini Via Cadriano 60 Granarolo 40054 Bologna	25 g a.s./ha (WG)- 20.09.06	45	0 2 8 13	inflorescence 0.13 inflorescence <u>0.083</u> inflorescence 0.014 inflorescence <0.01	BASF analytical method No. 567/0 inflorescence: mean recovery = 77.5%; SD: +/- 15.7; CV: 20.2%; n=8; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/4 Study carried out in 2006	Cauliflower (variety Kintore)	France, 12 Chemin des Garosses 31330 France (South of EU)	25 g a.s./ha (EC)- 18.09.06	45	0 3 8 14	inflorescence <0.01 inflorescence <u>≤0.01</u> inflorescence <0.01 inflorescence <0.01	
BASF Doc ID 2007/1007936 Trial No. AF/10502/BA/4 Study carried out in 2006	Cauliflower (variety Kintore)	France, 12 Chemin des Garosses 31330 France (South of EU)	25 g a.s./ha (WG)- 18.09.06	45	0 3 8 14	inflorescence <0.01 inflorescence <0.01 inflorescence <0.01 inflorescence <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in cauliflower presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-10 trials including both variants in different plots, 2 of them bridging trials with two different formulations, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions-6 trials, 2 of them bridging trials with two different formulations, all conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues of alpha-cypermethrin in cauliflower inflorescences ranged between <0.01 mg/kg and 0.123 mg/kg. Residues declined to levels between <0.01-0.042 mg/kg and <0.01-0.053 mg/kg at 3 and 7 days after application. At the last sampling date, 13-14 days after application, determinable residues at the limit of quantitation of the analytical method (LOQ: 0.01 mg/kg) were found only in one sample out of ten treated plots (0.011 mg/kg), while samples from the other nine plots showed residues below the LOQ (all <0.01 mg/kg).

After two applications at 12.5 g a.s./ha, initial residues in cauliflower inflorescences ranged between <0.01 mg/kg and 0.201 mg/kg. Residues declined to levels between <0.01-0.075 mg/kg and <0.01-0.085 mg/kg and at 2-4 and 6-8 days after the last application. At the last sampling date, 13-14 days after the last application, determinable residues were found only in one sample out of ten treated plots (0.027 mg/kg), while samples from the other nine plots showed residues below the limit of quantitation of the analytical method (LOQ; <0.01 mg/kg).
The residue levels found in one of the trials performed in the Northern European region exceeded by far those of the other trials conducted under comparable conditions (trial AF/8813/BA/3). This situation was found in both application variants.

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in cauliflower inflorescences ranged between <0.01 mg/kg and 0.16 mg/kg. Residues declined to levels between <0.01-0.083 mg/kg at 2-4 days after application. After 7-8 days, determinable residues of alpha-cypermethrin were found only in one treated sample (trial AF/10502/BA/3; 0.014 mg/kg), while all other treated samples showed residues below the LOQ at this sampling point (all <0.01 mg/kg). At the last sampling, 13-14 days after application, residues were below the LOQ in all treated samples (all <0.01 mg/kg).

Conclusion:

In the years 2005 and 2006 a residue program on cauliflower was conducted in the field in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.
In 10 trials (12 plots) supporting the GAP for the Northern European region, residues ranged between <0.01-0.085 mg/kg at the target PHI of 7±1 days.
In 6 trials (8 plots) supporting the GAP for the Southern European region, residues ranged between <0.01-0.083 mg/kg at the target PHI of 3±1 days.

2.12.2 Estimation of MRL, HR and STMR for cauliflower

For *cauliflower*, the following residue studies were considered (BASF DocIDs): 2006/1026864, 2007/1008495 and 2007/1007936.

The following residue values (PHI=7±1 days for EU-N and PHI = 7±1 days for EU-S) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026864 (field)	<0.01 (3x), 0.085	<0.01 (2x)
2007/1008495 (field)	<0.01 (2x), 0.010, 0,011	<0.01 (2x)
2007/1007936 (field)	<0.01 (2x)	<0.01, 0.083
OECD-MRL-calculation	**0.15 (n=10, STMR=0.01, HR=0.085)**	**0.15 (n=6, STMR=0.01, HR=0.083)**

_ underlined values were used for risk assessment purposes

2.13 Brussels sprouts

Residue data from supervised trials in brussels sprouts were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.13-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Brussels sprouts (field)	2005	4	DE, FR, NL, UK	2	ES, GR		2006/1026859
Brussels sprouts (field)	2006	4	BE, DE, FR, UK	2	FR, IT		2007/1007943
Total number of trials per region		**8**			**Total number of trials**	**4**	

2.13.1 Supervised residue trials in brussels sprouts

Evans L (2007)
Full study reference
Evans L (2007): Study on the residue behaviour of alpha-cypermethrin in Brussels Sprouts after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026859

North L (2007)
Full study reference
North L (2007): Study on the residue behaviour of alpha-cypermethrin in Brussels Sprout after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007943

Material and Methods:

In the years 2005 and 2006 a residue program on brussels sprouts was conducted in Belgium, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom under field conditions.

Eight trials were conducted in the Northern European region in order to determine the magnitude of the residue after application of a 100 g a.s./L alpha-cypermethrin emulsifiable concentrate formulation (EC 100 g/L; BAS 310 40 I). Separate plots of brussels sprouts were foliar treated with either a single application at a target rate of 12.5 g a.s./ha or two applications at a target rate of 12.5 g a.s./ha. Samples of sprouts were collected approximately 0, 3, 7 and 14 days after last the application. Residue analysis was performed according to BASF analytical method 567/0, which determines total cypermethrin residues by means of HPLC/MS-MS with a LOQ of 0.01 mg/kg.

Four trials were performed with the same emulsifiable concentrate formulation in the Southern European region. The product was applied at a rate of 25 g a.s./ha. Samples of brussels sprouts were collected approximately 0, 3, 7 and 14 days after last the application. Residue analysis was performed according to BASF analytical method 567/0, which determines total cypermethrin residues by means of HPLC/MS-MS with a LOQ of 0.01 mg/kg.

The trial data and residue results are summarized in Table 2.13.1-1.

Table 2.13.1-1: Residues in brussels sprouts

GAP for EU-N is 1-2 applications at 15 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/1 Study to GLP Study carried out in 2005	Brussels Sprouts	United Kingdom Marston; Curdworth	12.5 g as/ha 09.11.05	43-45	0 3 7 14	sprouts <0.01 sprouts <0.01 sprouts <0.01 sprouts <0.01	BASF method N 567/0 sprouts: mean recovery = 83.4%; SD: +/- 11.4; CV: 13.6; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/2 Study to GLP Study carried out in 2005	Brussels Sprouts	France Le Pot Dore 49650 Allonnes (North of EU)	12.5 g as/ha 26.09.05	43-45	0 3 7 14	sprouts 0.011 sprouts <0.01 sprouts <0.01 sprouts <0.01	
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/3 Study to GLP Study carried out in 2005	Brussels Sprouts	Germany 67365 Schwegenheim	12.5 g as/ha 06.12.05	76	0 3 7 14	sprouts 0.018 sprouts 0.016 sprouts 0.016 sprouts 0.014	
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/4 Study to GLP Study carried out in 2005	Brussels Sprouts	The Netherlands 5809 EJ Leunen, Limburg	12.5 g as/ha 04.11.05	45-47	0 3 7 14	sprouts <0.01 sprouts <0.01 sprouts <0.01 sprouts <0.01	
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/1 Study to GLP Study carried out in 2006	Brussels Sprouts (variety Diablo)	France Liguay-Liger GAEC 19 Chemin St Remy Dampierre en Burly 45570 (North of EU)	12.5 g as/ha 05.12.06	47-49	0 3 7 14	sprouts <0.01 sprouts <0.01 sprouts 0.012 sprouts 0.010	BASF method N 567/0 sprouts: mean recovery = 102.0%; SD: +/- 8.8; CV: 8.6; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/2 Study to GLP Study carried out in 2006	Brussels Sprouts (variety Millenium)	Belgium Alain Boudrez Rue de Thuin, 50 6534 Gozee	12.5 g as/ha 23.10.06	47	0 4 7 14	sprouts 0.010 sprouts 0.010 sprouts <0.01 sprouts <0.01	
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/3 Study to GLP Study carried out in 2006	Brussels Sprouts	Germany	12.5 g as/ha 22.11.06		0 3 7 14	sprouts 0.019 sprouts 0.024 sprouts 0.011 sprouts <0.01	BASF method N 567/0 sprouts: mean recovery = 102.0%; SD: +/- 8.8; CV: 8.6; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/4 Study to GLP Study carried out in 2006	Brussels Sprouts	United Kingdom Taylors Farm Marsh Road Banks Lancashire PR9 8DX	12.5 g as/ha 24.11.06	48-49	0 3 7 14	sprouts <0.01 sprouts <0.01 sprouts <0.01 sprouts <0.01	
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/1 Study to GLP Study carried out in 2005	Brussels Sprouts	United Kingdom Marston; Curdworth	12.5 g as/ha 2 treatm. last date 09.11.05	43-45 at last treatm.	0 3 7 14	sprouts <0.01 sprouts <0.01 sprouts 0.014 sprouts <0.01	BASF method N 567/0 sprouts: mean recovery = 83.4%; SD: +/- 11.4; CV: 13.6; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin

Table 2.13.1-1: Residues in brussels sprouts

GAP for EU-N is 1-2 applications at 15 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/2 Study to GLP Study carried out in 2005	Brussels Sprouts	France Le Pot Dore 49650 Allonnes (North of EU)	12.5 g as/ha 2 treatm. last date 26.09.05	43-45 at last treatm.	0 3 7 14	sprouts 0.017 sprouts 0.013 sprouts <u>0.017</u> sprouts 0.014	BASF method N 567/0 sprouts: mean recovery = 83.4%; SD: +/- 11.4; CV: 13.6; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/3 Study to GLP Study carried out in 2005	Brussels Sprouts	Germany 67365 Schwegenheim	12.5 g as/ha 2 treatm. last date 06.12.05	76 at last treatm.	0 3 7 14	sprouts 0.051 sprouts 0.055 sprouts <u>0.046</u> sprouts 0.033	
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/4 Study to GLP Study carried out in 2005	Brussels Sprouts	The Netherlands 5809 EJ Leunen, Limburg	12.5 g as/ha 2 treatm. last date 04.11.05	45-47 at last treatm.	0 3 7 14	sprouts <0.01 sprouts <0.01 sprouts <u><0.01</u> sprouts <0.01	
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/1 Study to GLP Study carried out in 2006	Brussels Sprouts (variety Diablo)	France Liguay-Liger GAEC 19 Chemin St Remy Dampierre en Burly 45570 (North of EU)	12.5 g as/ha 2 treatm. last date 05.12.06	47-49 at last treatm.	0 3 7 14	sprouts 0.026 sprouts 0.017 sprouts <u>0.025</u> sprouts 0.018	BASF method N 567/0 sprouts: mean recovery = 102.0%; SD: +/- 8.8; CV: 8.6; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/2 Study to GLP Study carried out in 2006	Brussels Sprouts (variety Millenium)	Belgium Alain Boudrez Rue de Thuin, 50 6534 Gozee	12.5 g as/ha 2 treatm. last date 23.10.06	47 at last treatm	0 4 7 14	sprouts 0.022 sprouts 0.018 sprouts <u>0.018</u> sprouts 0.014	
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/3 Study to GLP Study carried out in 2006	Brussels Sprouts	Germany	12.5 g as/ha 2 treatm. last date 22.11.06		0 3 7 14	sprouts 0.037 sprouts 0.022 sprouts <u>0.020</u> sprouts 0.016	
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/4 Study to GLP Study carried out in 2006	Brussels Sprouts	United Kingdom Taylors Farm Marsh Road Banks Lancashire PR9 8DX	12.5 g as/ha 2 treatm. last date 24.11.06	48-49	0 3 7 14	sprouts <0.01 sprouts 0.015 sprouts <u>0.010</u> sprouts <0.01	BASF method N 567/0 sprouts: mean recovery = 102.0%; SD: +/- 8.8; CV: 8.6; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/5 Study to GLP Study carried out in 2006	Brussels Sprouts	Spain 50641 Boquineni	25 g as/ha 27.01.06	46	0 3 7 14	sprouts <0.01 sprouts <u><0.01</u> sprouts <0.01 sprouts <0.01	BASF method N 567/0 sprouts: mean recovery = 83.4%; SD: +/- 11.4; CV: 13.6; n=5; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026859 Trial No. AF/8818/BA/6 Study to GLP Study carried out in 2006	Brussels Sprouts	Greece Chalkidona Thessaloniki Central Macedonia GR-57007	25 g as/ha 01.11.05	47	0 3 7 14	sprouts 0.016 sprouts <u>0.013</u> sprouts 0.011 sprouts <0.01	

Table 2.13.1-1: Residues in brussels sprouts

GAP for EU-N is 1-2 applications at 15 g a.s./ha at infestation as an overall spray, PHI = 7 (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/5 Study to GLP Study carried out in 2006	Brussels Sprouts (variety Dominator)	France Nicolas Henrat 13 Au Gascogne Mondonville 31700 (South of EU)	25 g as/ha 05.12.06	47	0 4 7 14	sprouts 0.021 sprouts <u>0.029</u> sprouts 0.023 sprouts 0.019	BASF method N 567/0 sprouts: mean recovery = 102.0%; SD: +/- 8.8; CV: 8.6; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007943 Trial No. AF/10499/BA/6 Study to GLP Study carried out in 2006	Brussels Sprouts (variety Grosso di Cassano)	Italy Via Cadriano 60/2 40057 Cassette di Cadriano	25 g as/ha 21.11.06	47	0 3 7 14	sprouts <0.01 sprouts <u><0.01</u> sprouts <0.01 sprouts <0.01	

_ underlined values were used for MRL calculation

Findings:
The residue studies in brussels sprouts presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions – 8 trials conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions – 4 trials conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues of alpha-cypermethrin in brussels sprouts ranged between <0.01 mg/kg and 0.019 mg/kg. Residues ranged at levels between <0.01 – 0.024 mg/kg, <0.01 – 0.016 mg/kg and <0.01 – 0.014 mg/kg at 3, 7 and 14 days after application.

After two applications at 12.5 g a.s./ha, initial residues ranged between <0.01 mg/kg and 0.051 mg/kg. Residues ranged at levels between <0.01 – 0.055 mg/kg, <0.01 – 0.046 mg/kg and <0.01 – 0.033 mg/kg at 3, 7 and 14 days after application.

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in brussels sprouts ranged between <0.01 mg/kg and 0.021 mg/kg. Residues declined to levels between <0.01 – 0.029 mg/kg, <0.01 – 0.023 mg/kg and <0.01 – 0.019 mg/kg at 3, 7 and 14 days after application.

Conclusion:

In 8 trials supporting the GAP for the Northern European region, residues ranged between <0.01 – 0.046 mg/kg at the target PHI of 7±1 days.
In 4 trials supporting the GAP for the Southern European region, residues ranged between <0.01 – 0.029 mg/kg at the target PHI of 3±1 days.

2.13.2 Estimation of MRL, HR and STMR for brussels sprouts

For *brussels sprouts*, the following residue studies were considered (BASF DocIDs): 2006/1026859 and 2007/1007943.

The following residue values (EU-N: PHI=7±1 days / EU-S: 3±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026859 (open field)	<0.01, 0.014, 0.017, 0.046	<0.01, 0.013
2007/1007943 (open field)	0.01, 0.018, 0.02, 0.025	<0.01, 0.029
OECD-MRL-calculation (open field)	**0.07 (n=8, STMR=0.018, HR=0.046)**	**0.06 (n=4, STMR=0.012, HR=0.029)**

_ underlined values were used for risk assessment purposes

2.14 Head cabbage

Residue data from supervised trials in head cabbage were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.14-1: Number of residue trials conducted in head cabbage per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Cabbage, head (field)	2003	4	DE, DK, FR, UK	-	-	4	2004/1006470
Cabbage, head (field)	2005	4	DE, FR, UK	2	FR, IT	6	2006/1026852
Cabbage, head (field)	2006	4	BE, DE; FR, UK	2	ES, FR	6	2007/1013148
Total number of trials per region		**12**		**4**	**Total number of trials**	**16**	

Table 2.14-2: Processing studies available for head cabbage

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Head cabbage	2000	-	-	1	IT	1	AL-721-044
Head cabbage	2001	3	DE	-	-	3	2002/1004078
Total number of trials per region		**3**		**1**	**Total number of trials**	**4**	

2.14.1 Supervised residue trials in head cabbage

Raunft E, Rabe U, Mackenroth C (2004)

Full study reference

Raunft E, Rabe U, Mackenroth C (2004): Study on the residue behaviour of alpha-cypermethrin in headed cabbage after application of BAS 310 11 I under field conditions in France (N), Germany, United Kingdom and Denmark, 2003; BASF DocID 2004/1006470

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of aIpha-cypermethrin in head cabbage after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026852

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the residue behaviour of BAS 310 I in head cabbage after treatment with BAS 310 40 I under open field conditions in Southern and Northern Europe, 2006; BASF DocID 2007/1013148

Material and Methods:

In the years 2003, 2005 and 2006 a residue program on head cabbage was conducted in Belgium, Denmark, France (Northern and Southern European region), Germany, Italy, Spain and the United Kingdom under field conditions.

During the 2003 growing season, a total of 4 trials was conducted in headed cabbage. An emulsifiable concentrate formulation of alpha-cypermethrin (SC 100 g a.s./L; BAS 310 11 I) was foliar sprayed on cabbage on two different plots. In one variant, one treatment was done with a rate of 15 g a.s./ha. This was compared to two treatments with the same application rate. The growth stage of the plants at the last application was between BBCH 47-48 (70-80% of the expected head size reached). Cabbage head specimens were collected directly after the last application as well as 2-3, 6-7 and 13-14 days thereafter.
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS with BASF analytical method No. 546/0 which has a limit of quantitation of 0.05 mg/kg in all sample materials.

During the 2005 growing season, six field trials were conducted in different representative head cabbage growing areas in the United Kingdom, France (Northern and Southern European region), Italy and Germany.

In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to cabbage plants either once or twice to separate plots at a target rate of 12.5 g a.s./ha. The growth stage of the plants at application was between 43 (30% of the expected head diameter reached) and 47 (30% of the expected head diameter reached).

Specimens of cabbage heads were collected immediately after the last application from each plot, as well as 3, 7 and 14 days thereafter.

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to cabbage plants once at a target rate of 25 g a.s./ha.

The growth stage of the plants at application was between 47 and 48 (70-80% of the expected head diameter reached).

Specimens of cabbage heads were collected immediately after the last application, as well as 3, 7 and 14 days thereafter.

Specimens were analysed using BASF analytical method No. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, six field trials were conducted in different representative head cabbage growing areas in Belgium, the United Kingdom, France (Northern and Southern European region), Spain and Germany.

In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to cabbage plants either once or twice to separate plots at a target rate of 12.5 g a.s./ha. The growth stage of the plants at application was between 44 (40% of the expected head diameter reached) and 48 (80% of the expected head diameter reached).

Specimens of cabbage heads were collected immediately after the last application from each plot, as well as 3-4, 7-8 and 14 days thereafter.

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to cabbage plants once at a target rate of 25 g a.s./ha at a growth stage up to 48 (80% of the expected head diameter reached).

Specimens of cabbage heads were collected immediately after the last application, as well as 3, 6-7 and 13-14 days thereafter.

Specimens were analysed using BASF analytical method No. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.14.1-1.

Table 2.14.1-1: Residues in head cabbage

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/1006470 Trial No. ACK/14/03 Study to GLP Study carried out in 2003	Cabbage (variety Rodon F1; red cabbage)	Germany 16833 Lentzke Brandenburg	15 g a.s./ha 08.10.03	48	0 2 7 14	cabbage head <0.05 cabbage head <0.05 cabbage head <0.05 cabbage head <0.05	BASF analytical method No. 546/0 cabbage, head : mean recovery = 93.1 %; SD: +/- 16.8; CV: 18.1; n=6; fortification range 0.05-0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2004/1006470 Trial No. ALB/12/03 Study to GLP Study carried out in 2003	Cabbage (variety Mila, savoy cabbage)	Denmark 5500 Middelfart Fuenen	15 g a.s./ha 20.08.03	48	0 2 7 14	cabbage head <0.05 cabbage head <0.05 cabbage head <0.05 cabbage head <0.05	
BASF Doc ID 2004/1006470 Trial No. FAN/21/03 Study to GLP Study carried out in 2003	Cabbage (variety Zerlina, white cabbage)	France North 67203 Oberschaeffolsheim Alsace	15 g a.s./ha 09.09.03	47	0 3 7 14	cabbage head <0.05 cabbage head <0.05 cabbage head <0.05 cabbage head <0.05	
BASF Doc ID 2004/1006470 Trial No. OAT/18/03 Study to GLP Study carried out in 2003	Cabbage (variety Rona, red cabbage)	United Kingdom Oxfordshire	15 g a.s./ha 02.09.03	48	0 3 6 13	cabbage head <0.05 cabbage head <0.05 cabbage head <0.05 cabbage head <0.05	
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/1 Study to GLP Study carried out in 2005	Head cabbage (variety Summer greens)	United Kingdom Banks Lancashire PR9 8DX	12.5 g a.s./ha 11.10.05	43	0 3 7 14	heads 0.069 heads 0.035 heads 0.012 heads <0.01	BASF analytical method No. 567/0 heads: mean recovery = 85.7%; SD: +/- 12.0; CV: 14.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/2 Study to GLP Study carried out in 2005	Head cabbage (variety Spitfire)	France 49650 Allones (North of EU)	12.5 g a.s./ha 29.11.05	47	0 3 7 14	heads <0.01 heads 0.012 heads <0.01 heads <0.01	
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/4 Study to GLP Study carried out in 2005	Head cabbage (variety Nula)	Germany 30982 Pattensen Jeinsen	12.5 g a.s./ha 04.11.05	48	0 3 7 14	heads 0.084 heads 0.082 heads 0.062 heads 0.072	
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/7 Study to GLP Study carried out in 2005	Head cabbage (variety Wirosa F1)	United Kingdom Kings Newton Derbyshire DE73 1DD	12.5 g a.s./ha 29.11.05	47	0 3 7 14	heads 0.015 heads 0.030 heads <0.01 heads <0.01	BASF analytical method No. 567/0 heads: mean recovery = 85.7%; SD: +/- 12.0; CV: 14.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin

Table 2.14.1-1: Residues in head cabbage

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1013148 Trial No. A/NF/I/06/143 Study to GLP Study carried out in 2006	Head cabbage (variety Guisor)	France Sedan Champagne-Ardennes (North of EU)	12.5 g a.s./ha 18.09.06	47	0 4 8 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	BASF analytical method No. 567/0 cabbage, head: mean recovery = 90.0 %; SD: +/- 2.4; CV: 2.6; n=6; fortification range 0.01-0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013148 Trial No. A/BE/I/06/144 Study to GLP Study carried out in 2006	Head cabbage (variety Galaxy)	Belgium Lille Antwerpen	12.5 g a.s./ha 25.09.06	44-45	0 3 7 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	
BASF Doc ID 2007/1013148 Trial No. A/UK/I/06/145 Study to GLP Study carried out in 2006	Head cabbage (variety Stonehead)	United Kingdom Chipping Campden Gloucestershire	12.6 g a.s./ha 26.06.06	45	0 3 7 14	heads 0.067 heads 0.077 heads 0.051 heads 0.030	
BASF Doc ID 2007/1013148 Trial No. A/GE/I/06/146 Study to GLP Study carried out in 2006	Head cabbage (variety Kilatou)	Germany Pegau Sachsen	12.9 g a.s./ha 18.09.06	48	0 3 7 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	
BASF Doc ID 2004/1006470 Trial No. ACK/14/03 Study to GLP Study carried out in 2003	Cabbage (variety Rodon F1; red cabbage)	Germany 16833 Lentzke Brandenburg	15 g a.s./ha 2 treatm. last date 08.10.03	48 at last treatment	0 2 7 14	cabbage head <0.05 cabbage head <0.05 cabbage head <u>≤0.05</u> cabbage head <0.05	BASF analytical method No. 546/0 cabbage, head: mean recovery = 93.1 %; SD: +/- 16.8; CV: 18.1; n=6; fortification range 0.05-0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2004/1006470 Trial No. ALB/12/03 Study to GLP Study carried out in 2003	Cabbage (variety Mila, savoy cabbage)	Denmark 5500 Middelfart Fuenen	15 g a.s./ha 2 treatm. last date 20.08.03	48 at last treatment	0 2 7 14	cabbage head <0.05 cabbage head <0.05 cabbage head <u>≤0.05</u> cabbage head <0.05	
BASF Doc ID 2004/1006470 Trial No. FAN/21/03 Study to GLP Study carried out in 2003	Cabbage (variety Zerlina, white cabbage)	France North 67203 Oberschaeffolsheim Alsace	15 g a.s./ha 2 treatm. last date 09.09.03	47 at last treatment	0 3 7 14	cabbage head <0.05 cabbage head <0.05 cabbage head <u>≤0.05</u> cabbage head <0.05	
BASF Doc ID 2004/1006470 Trial No. OAT/18/03 Study to GLP Study carried out in 2003	Cabbage (variety Rona, red cabbage)	United Kingdom Oxfordshire	15 g a.s./ha 2 treatm. last date 02.09.03	48 at last treatment	0 3 6 13	cabbage head <0.05 cabbage head <0.05 cabbage head <u>≤0.05</u> cabbage head <0.05	BASF analytical method No. 546/0 cabbage, head: mean recovery = 93.1 %; SD: +/- 16.8; CV: 18.1; n=6; fortification range 0.05-0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/1 Study to GLP Study carried out in 2005	Head cabbage (variety Summer greens)	United Kingdom Banks Lancashire PR9 8DX	12.5 g a.s./ha 2 treatm. last date 11.10.05	43 at last treatm.	0 3 7 14	heads 0.088 heads 0.055 heads <u>0.018</u> heads <0.01	BASF analytical method No. 567/0 heads: mean recovery = 85.7%; SD: +/- 12.0; CV: 14.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total

Table 2.14.1-1: Residues in head cabbage

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/2 Study to GLP Study carried out in 2005	Head cabbage (variety Spitfire)	France 49650 Allones (North of EU)	12.5 g a.s./ha 2 treatm. last date 29.11.05	47 at last treatm.	0 3 7 14	heads 0.017 heads <0.01 heads <0.01 heads <0.01	cypermethrin
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/4 Study to GLP Study carried out in 2005	Head cabbage (variety Nula)	Germany 30982 Pattensen Jeinsen	12.5 g a.s./ha 2 treatm. last date 04.11.05	48 at last treatm.	0 3 7 14	heads 0.157 heads 0.152 heads 0.104 heads 0.097	
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/7 Study to GLP Study carried out in 2005	Head cabbage (variety Wirosa F1)	United Kingdom Kings Newton Derbyshire DE73 1DD	12.5 g a.s./ha 2 treatm. last date 29.11.05	47 at last treatm.	0 3 7 14	heads 0.025 heads 0.042 heads 0.014 heads <0.01	
BASF Doc ID 2007/1013148 Trial No. A/NF/I/06/143 Study to GLP Study carried out in 2006	Head cabbage (variety Guisor)	France Sedan Champagne-Ardennes (North of EU)	12.0/11.7 g a.s./ha 2 treatm. last date 18.09.06	47	0 4 8 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	BASF analytical method No. 567/0 cabbage, head: mean recovery = 90.0 %; SD: +/- 2.4; CV: 2.6; n=6; fortification range 0.01-0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013148 Trial No. A/BE/I/06/144 Study to GLP Study carried out in 2006	Head cabbage (variety Galaxy)	Belgium Lille Antwerpen	12.7/12.4 g a.s./ha 2 treatm. last date 25.09.06	44-45	0 3 7 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	
BASF Doc ID 2007/1013148 Trial No. A/UK/I/06/145 Study to GLP Study carried out in 2006	Head cabbage (variety Stonehead)	United Kingdom Chipping Campden Gloucestershire	13.2 /13.2 g a.s./ha 2 treatm. last date 26.06.06	45	0 3 7 14	heads 0.164 heads 0.182 heads 0.105 heads 0.032	
BASF Doc ID 2007/1013148 Trial No. A/GE/I/06/146 Study to GLP Study carried out in 2006	Head cabbage (variety Kilatou)	Germany Pegau Sachsen	13.4 /12.9 g a.s./ha 2 treatm. last date 18.09.06	48	0 3 7 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/5 Study to GLP Study carried out in 2005	Head cabbage (variety Castello)	France 82170 Canals (South of EU)	25 g a.s./ha 16.08.05	47	0 3 7 14	heads <0.01 heads <0.01 heads <0.01 heads <0.01	BASF analytical method No. 567/0 heads: mean recovery = 85.7%; SD: +/- 12.0; CV: 14.0%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026852 Trial No. AF/8829/BA/6 Study to GLP Study carried out in 2005	Head cabbage (variety Primero)	Italy 40057 Viadagola	25 g a.s./ha 17.10.05	47-48	0 3 7 14	heads 0.015 heads 0.013 heads <0.01 heads <0.01	

Table 2.14.1-1: Residues in head cabbage

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1013148 Trial No. A/SF/I/06/147 Study to GLP Study carried out in 2006	Head cabbage (variety Clarissa HF 1)	France ChateaurenardBouches-de-Rhône (South of EU)	31.3 g a.s./ha 13.09.06	48	0 3 7 14	heads 0.045 heads <u>0.010</u> heads <0.01 heads <0.01	BASF analytical method No. 567/0 cabbage, head: mean recovery = 90.0 %; SD: +/- 2.4; CV: 2.6; n=6; fortification range 0.01-0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1013148 Trial No. A/SP/I/06/148 Study to GLP Study carried out in 2006	Head cabbage (variety n.a.)	Spain Almussafes Valencia	41.5 g a.s./ha 26.07.06	26	0 3 6 13	heads 0.010 heads 0.010 heads <u>0.013</u> heads <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in head cabbage presented in the alpha-cypermethrin dossier were carried out in 7 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-8 trials including both variants, all conducted in the Northern European region
- one or two treatments at a target rate of 15 g a.s./ha under open field conditions-4 trials including both variants, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions-4 trials conducted in the Southern European region

After one or two treatments with alpha-cypermethrin at a rate of 15 g a.s./ha, no residues of alpha-cypermethrin above the limit of quantitation of the analytical method (LOQ; 0.05 mg/kg) were detected in any of the cabbage specimens analysed regardless the cabbage species or number of treatment.

After one application at 12.5 g a.s./ha, initial residues of alpha-cypermethrin in cabbage heads ranged between <0.01 mg/kg and 0.084 mg/kg. Residues declined to levels between <0.01-0.082 mg/kg, <0.01-0.062 mg/kg and <0.01-0.072 mg/kg 3, 7 and 14 days after application.

After two applications at 12.5 g a.s./ha, initial residues ranged between <0.01 mg/kg and 0.164 mg/kg. Residues declined to levels between <0.01-0.182 mg/kg, <0.01-0.105 mg/kg and <0.01-0.097 mg/kg at 3-4, 7-8 and 14 days after application.

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in cabbage heads ranged between <0.01 mg/kg and 0.045 mg/kg. Residues declined to levels between <0.01-0.013 mg/kg on days 3 and 6-7, respectively. No residues above the limit of quantitation of the analytical method (LOQ; 0.01 mg/kg) were found in any of the treated cabbage head samples 13-14 days after the last application.

Conclusion:

In the years 2003, 2005 and 2006 a residue program on head cabbage was conducted in Belgium, Denmark, France (Northern and Southern European region), Germany, Italy, Spain and the United Kingdom under field conditions.
In 8 trials supporting the GAP for the Northern European region, residues ranged between <0.01-0.105 mg/kg at the target PHI of 7 days.
In 4 trials supporting the GAP for the Southern European region, residues ranged between <0.01-0.013 mg/kg at the target PHI of 3 days.

2.14.2 Processing studies in head cabbage

Within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 this processing study in cabbage (AL-721-044) was submitted. The study was assessed as not being required (3rd Addendum to the Monograph: Addendum Chapter B-7 (Residue data), January 2003). Therefore, a review was not performed at this time. The study was not included in the application as it had been submitted already with the alpha-cypermethrin addendum dossier. Below it is summarized for the reviewer´s convenience.

Report:	Grolleau G. 2001a Alphacypermethrin (AC 900049) 150 g a.s./ WG (RLM 11203): At harvest residue study on alphacypermethrin in head cabbage and processed fractions (cooked and sauerkraut), Italy, 2000. BASF RDI No: AL-721-044 DocNo. 2001/7001643
Guidelines:	EEC 7029/VI/95 rev. 5,Decret No. 98-1312 (31-12-1998),EEC 1999/11/EC
GLP:	yes (certified by Groupe Interministeriel des Produits Chimiques, France)

Materials and Methods:

In 2000, a single residue trial was established in Italy. Savoy cabbage (variety: Famosa) was treated with the 150 g a.s./kg WG (RLM 11203) alpha-cypermethrin formulation.

In this trial, cabbage plants received either a single 100 g a.s./ha foliar application 7 days prior to typical commercial harvest or three 100 g a.s./ha foliar applications 21, 14, and 7 days prior to typical commercial harvest.

These exaggerated treatment rates were used in an effort to generate finite alpha-cypermethrin residues in cabbage so that any effects of processing on alpha-cypermethrin residues could be better quantitated.

Actual applications were within 10% of nominal and were applied at 960 L/ha. The single application occurred at the BBCH 47 (70% of the expected head size reached) growth stage; the last of the three applications also occurred at BBCH 47. Cabbage specimens were sampled at typical commercial harvest, 7 days following the single 100 g a.s./ha application (7 DAT1) and 7 days following the last of the three 100 g a.s./ha applications (7 DAT3).

Field samples collected for residue analysis were frozen within 24 hours of sampling and remained frozen (including during transportation) until analysis. These specimens were shipped frozen to CEM Analytical Services Ltd. (CEMAS), Glendale Park, Fernbank Road, North Ascot, Berkshire, UK.

Bulk cabbage samples, sufficient for processing, were shipped unfrozen to the processor [Viticulture Recherche et Developpement (VITI R & D).

The processing phase was conducted at VITI R&D, 101 impasse des Capitelles, VILLETELLE, France, according to VITI R&D Processing Phase Plan GRA 0006 EUR.

Specimen analysis was conducted at CEMAS, Glendale Park, Fernbank Road, North Ascot, Berkshire, UK. All specimens were homogenized with dry ice. Cabbage and cabbage process fraction specimens were analyzed using BASF Agro Research analytical method RLA 12513.03V entitled:

> "Analysis of Alphacypermethrin (AC 900049) in Vining Peas, Field Beans, Strawberries, Cherries and Green Beans."

This methodology has been validated with a 0.05 mg/kg alpha-cypermethrin LOQ in:

- BASF Agro Research report 4452 (study 98452V) for green beans;
- BASF Agro Research report 4454 (study 98332V) for strawberries;
- BASF Agro Research report 4453 (study 98330V) for cherries;

A method performance check was included with study AL-UK-00-972 (CEMR-1421) and validated the 0.05 mg/kg alpha-cypermethrin LOQ in cabbage substrates. A method performance check for sauerkraut was included with study AL-IT-00-605 (CEMR-1462) and validated the 0.05 mg/kg alpha-cypermethrin LOQ in cabbage process fractions.

Concurrent procedural recoveries at the LOQ [0.05 mg/kg] and 10 times the LOQ in cabbage and sauerkraut were within the acceptable 70% to 110% range.

Findings:

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any untreated cabbage specimens or processed fractions (cooked cabbage and sauerkraut) obtained from untreated cabbage specimens.

Alpha-cypermethrin residues, ranging from 0.57 mg/kg to 1.0 mg/kg were found in cabbage sampled 7 days after the single 100 g a.s./ha application; alpha-cypermethrin residues ranged from 0.90 mg/kg to 1.3 mg/kg in cabbage sampled 7 days after the third 100 g a.s./ha application.

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any process fractions (cooked cabbage and sauerkraut) obtained from cabbage taken 7 days after a single 100 g a.s./ha application or 7 days after the third 100 g a.s./ha application.

Cabbage processing procedure summary-cooked cabbage:

Outer leaves and cores were removed from approximately 3 kg savoy cabbage. The dressed cabbage was then shred into thin strips and blanched in boiling water (approximately 4 kg water/kg cabbage) for one minute.

Figure 2.14.2-1: Cooked cabbage processing procedure flowchart - DocID AL-721-044

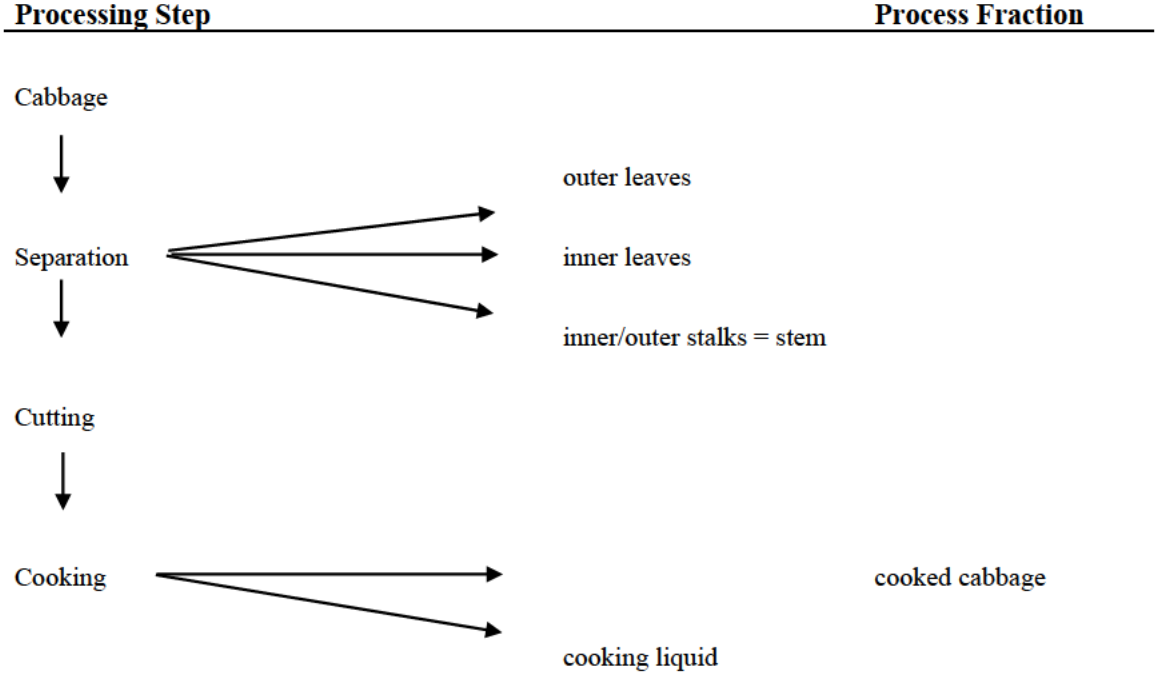

Table 2.14.2-1: Mass balance of the production of cooked cabbage - DocID AL-721-044

Processing Step	U1	T2	T3
weight of cabbage used for cooking process (kg)	3.05	3.10	3.25
dressed cabbage weight (kg)	1.70	0.80	2.00
waste weight (external leaves + core) (kg)	1.35	2.30	1.25
cut cabbage weight (kg)	1.00	0.60	0.75
blanched cabbage weight (kg)	1.10	0.70	0.80
U1 = untreated cabbage.			
T2 = cabbage treated at 1 x 100 g a.s./ha.			
T3 = cabbage treated at 3 x 100 g a.s./ha.			

Cabbage processing procedure summary - sauerkraut:

Outer leaves and cores were removed from approximately 15 kg savoy cabbage. The dressed cabbage was then shred into thin strips. Next cooking salt (25 g/kg cabbage) and lactic bacteria (0.03 g/kg cabbage) were added. Fermentation to sauerkraut followed and once fermentation was complete, the sauerkraut was pasteurized at 85°C for one minute.

Figure 2.14.2-2: Sauerkraut processing procedure flowchart - DocID AL-721-044

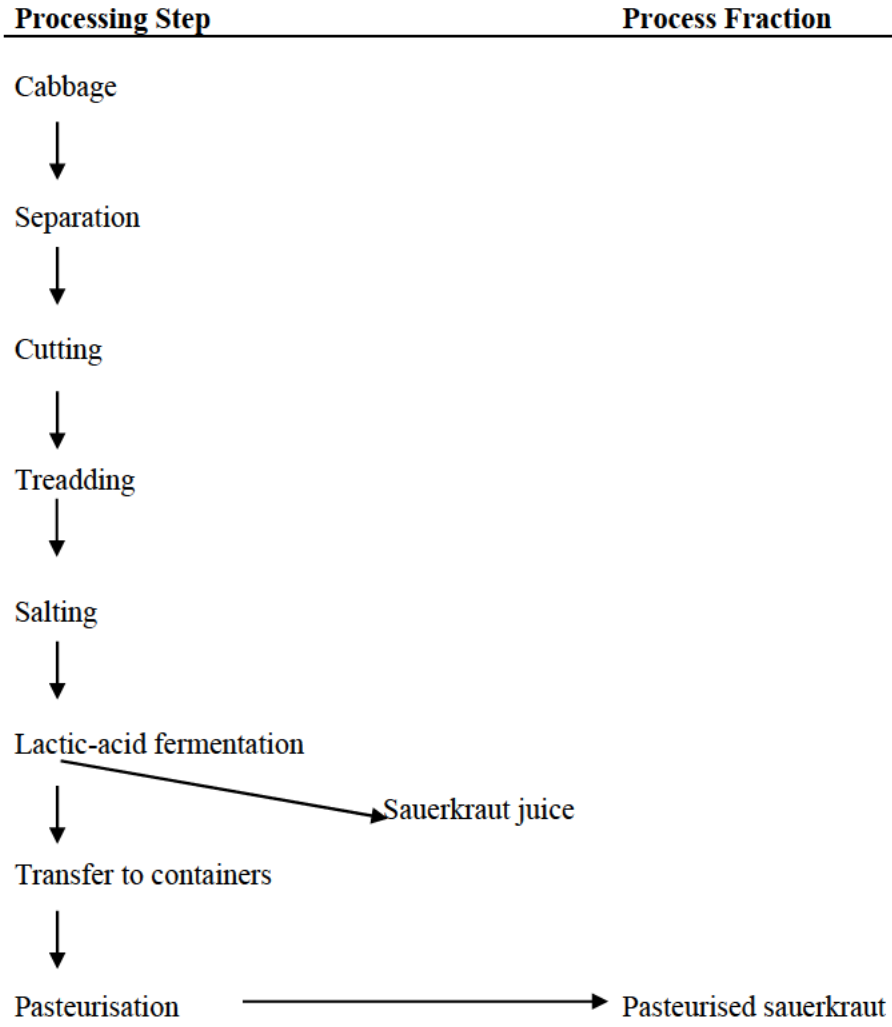

Table 2.14.2-2: Mass balance of the production of sauerkraut - DocID AL-721-044

Processing Step	U1	T2	T3
weight of cabbage used for sauerkraut process (kg)	15.20	15.15	15.30
dressed cabbage weight (kg)	4.30	3.35	4.40
waste weight (external leaves + core) (kg)	10.75	11.80	10.80
cut cabbage weight (kg)	2.05	2.40	2.95
sauerkraut weight (kg)	1.95	2.25	2.60
U1 = untreated cabbage.			
T2 = cabbage treated at 1 x 100 g a.s./ha.			
T3 = cabbage treated at 3 x 100 g a.s./ha.			

Table 2.14.2-3: Transfer factors for cooked cabbage and sauerkraut - DocID AL-721-044

	alpha-cypermethrin residue found (mg/kg)		Transfer Factor	
	T2	T3	T2	T3
cabbage (RAC)*	0.57	0.90	1	1
cabbage (RAC)**	1.00	1.33	1	1
cooked cabbage	<0.05	<0.05	<1	<1
sauerkraut	<0.05	<0.05	<1	<1
* cabbage taken from the field.				
**unprocessed cabbage sampled at the processor.				
U1 = untreated cabbage.				
T2 = cabbage treated at 1 x 100 g a.s./ha.				
T3 = cabbage treated at 3x 100 g a.s./ha.				

Conclusion:

The data from this processing study demonstrate clearly that there is little or no likelyhood of detectable alpha-cypermethrin residues resulting in the edible process fractions from cabbage treated at the GAP proposed rate.

Within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 this processing study in cabbage (DocID 2002/1004078) was submitted. The study was assessed as not being required (3rd Addendum to the Monograph: Addendum Chapter B-7 (Residue data), January 2003). Therefore, a review was not performed at this time. The study was not included in the application as it had been submitted already with the alpha-cypermethrin addendum dossier. Below it is summarized for the reviewer´s convenience.

Report:	Pollmann B. 2002a Determination of residues of alphacypermethrin in field samples and in processed goods after application of BAS 310 08 I in head cabbage at 3 sites in Germany in 2001. 2002/1004078
Guidelines:	IVA Guidelines for Residue Studies Sections IA and IB 2nd edition 1992,BBA IV 3-3,EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8),EEC 91/414 (1607/IV/97 Rev. 1)
GLP:	yes (certified by Ministerium für Umwelt und Verkehr Baden-Wuerttemberg, Stuttgart)

Materials and Methods:

In 2001, three residue trials were established in Germany; two trials in Northern Germany [variety: Krautmann (G01N063R) and variety: Megatonn (G01N064R)] and one trial in Southern Germany [variety: Custello (G01N065R)]. Each trial was treated with the 150 g a.s./kg WG (BAS 310 08 I) alpha-cypermethrin formulation. In all three residue trials, cabbage received three 100 g a.s./ha foliar applications 21, 13 (G01N065R) or 14 (G01N063R and G01N064R), and 7 (G01N065R) or 6 (G01N063R and G01N064R) days prior to typical commercial harvest. This exaggerated treatment rate was used in an effort to generate finite alpha-cypermethrin residues in cabbage so that any effects of processing on alpha-cypermethrin residues could be better quantitated.

Actual applications were within 10% of nominal, ranging from 97 g a.s./ha to 111 g a.s./ha. The last of three applications occurred at BBCH 46 for G01N063R (50% of the expected head size reached), BBCH 47 for G01N064R (70% of the expected head size reached), and BBCH 48 for G01N065R (>70% of the expected head size reached). Application volumes ranged from 567 L/ha to 647 L/ha. Cabbage specimens were sampled immediately after (0+ DAT3) the last of the three applications and 7 days after the third application (7 DAT3), at typical commercial harvest.

Field samples for residue analysis from the three trials were frozen within 24 hours of sampling and remained frozen (including during transportation) until analysis. These specimens were shipped frozen to GAB (Niefern-Öschelbronn), where they were stored until shipment to the analytical laboratory.

Bulk cabbage from all three trials (G01N063R, G01N064R, and G01N065R), sufficient for processing, were shipped unfrozen to the processor, GAB (Niefern-Öschelbronn).

The processing phase for cooked cabbage was conducted by GAB (Niefern-Öschelbronn), while the sauerkraut processing was conducted by Forschungsanstalt Geisenheim, Fachgebiet Weinanalytik und Getränkeforschung, Geisenheim, Germany.

Specimen analysis was conducted at the BASF Agricultural Research Center, Limburgerhof, Crop Protection Division, Ecology and Environmental Analytics, APD/MC. The cabbage and cabbage process fraction specimens were analyzed using BASF method 989/1 (BASF Agro Research analytical method RLA 12513.03V with minor modifications) entitled:

> "Analysis of Alphacypermethrin (AC 900049) in Vining Peas, Field Beans, Strawberries, Cherries and Green Beans."

This methodology has been validated with a 0.05 mg/kg alpha-cypermethrin LOQ in:

- BASF Agro Research report 4452 (study 98452V) for green beans;
- BASF Agro Research report 4454 (study 98332V) for strawberries;
- BASF Agro Research report 4453 (study 98330V) for cherries;

A method performance check was included with study AL-UK-00-972 (CEMR-1421) and validated the 0.05 mg/kg alpha-cypermethrin LOQ in cabbage. A method performance check for sauerkraut was included in this study and validated the 0.05 mg/kg alpha-cypermethrin LOQ in cabbage process fractions.

Concurrent procedural recoveries at the LOQ [0.05 mg/kg] and 10 times the LOQ [in cabbage, inner and outer cabbage leaves, cabbage stems, cooking liquid, sauerkraut, and sauerkraut juice] were acceptable however, not all individual recoveries were within the 70% to 110% range. None-the-less, the overall alpha-cypermethrin procedural recovery at both levels in all substrates was 100% ± 22%, considering the methodology was optimized for all substrates.

Findings:

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any untreated cabbage specimens or processed fractions (outer leaves, inner leaves, stem, cooked cabbage, cooking liquid, sauerkraut, and sauerkraut juice) obtained from untreated cabbage specimens.

Trace alpha-cypermethrin residues, ranging from <0.05 mg/kg (method LOQ) to 0.07 mg/kg were found in cabbage sampled immediately after the third 100 g a.s./ha application.

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in cabbage sampled 6 days after the third 100 g a.s/kg application (G01N063R and G01N064R) or 7 days after the third 100 g a.s/kg (G01N065R) .

With the exception of the outer cabbage leaves, no detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any process fractions (inner leaves, stem, cooked cabbage, cooking liquid, sauerkraut, and sauerkraut juice) obtained from cabbage sampled 6 days after the third 100 g a.s/kg application (G01N063R and G01N064R) or 7 days after the third 100 g a.s/kg (G01N065R) .

Trace alpha-cypermethrin residues, ranging from 0.15 mg/kg to 0.27 mg/kg were found in the outer leaves obtained from cabbage sampled 6 days after the third 100 g a.s/kg application (G01N063R and G01N064R) or 7 days after the third 100 g a.s/kg (G01N065R) .

Figure 2.14.2-3: Cooked cabbage processing procedure flowchart - DocID 2002/1004078

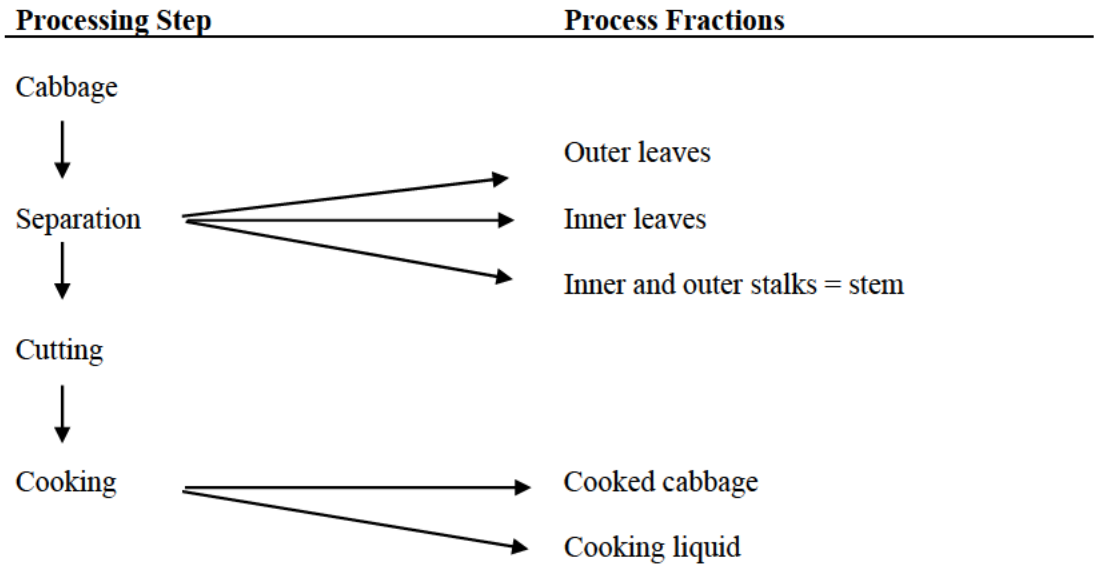

Table 2.14.2-4: Mass balance of the production of cooked cabbage - DocID 2002/1004078

	control		treated		
weight of process fraction (kg)	7-G01N063R	7-G01N063R	11-G01N063R	5-G01N064R	11-G01N065R
cabbage	11.22	6.99	11.99	13.10	6.70
outer leaves	0.97	0.64	0.83	1.28	0.75
inner leaves	9.26	5.69	10.28	10.74	5.51
stem	0.73	0.66	0.69	0.77	0.41
cut cabbage	8.09	4.74	8.97	9.89	4.76
cut cabbage to cooking	4.15	4.74	4.82	4.55	4.76
water added for cooking	2.46	2.44	2.40	2.41	2.44
cooked cabbage	3.49	4.56	4.13	3.66	4.41
cooking liquid	2.56	2.06	2.62	2.62	2.04
treated = received 3 x 100 g a.s./ha 21 ± 1, 14 ± 1, and 7 ± 1 days before sampling.					

Figure 2.14.2-4: Cabbage processing procedure flowchart - sauerkraut production - DocID 2002/1004078

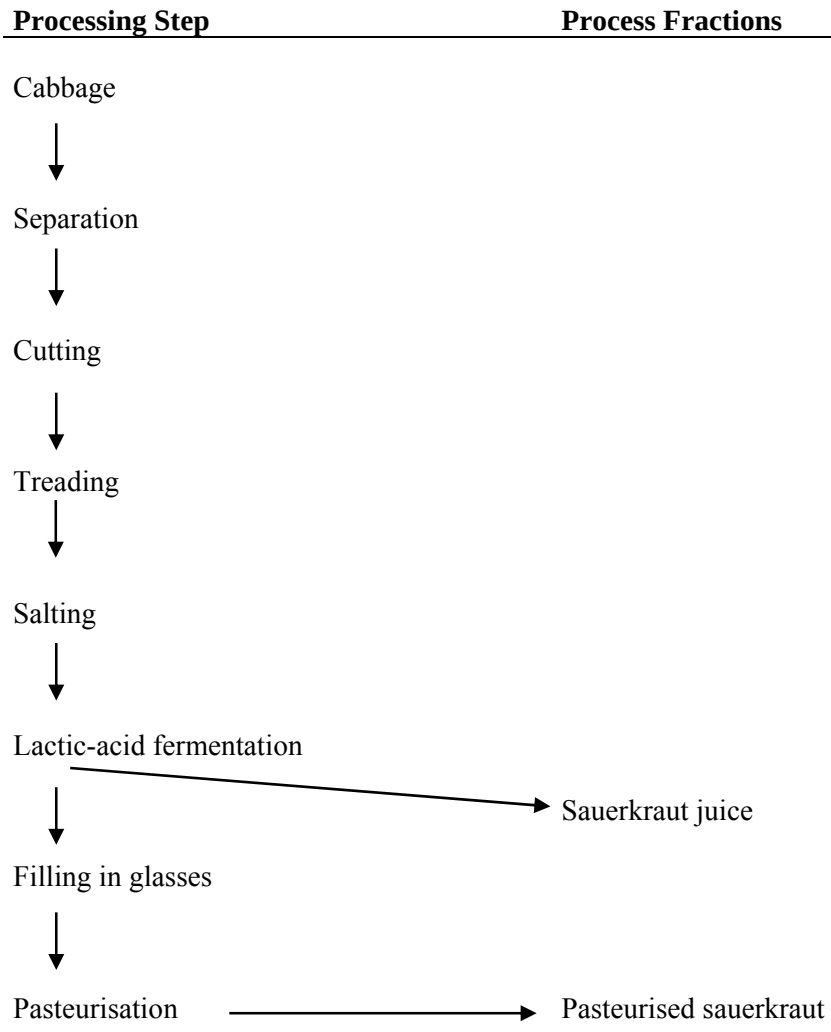

Table 2.14.2-5: Mass balance of the production of sauerkraut - DocID 2002/1004078

	Control		Treated		
Weight of process fraction (kg)	7-G01N063R	7-G01N063R	11-G01N063R	5-G01N064R	11-G01N065R
Cabbage	29.76	45.50	34.74	35.16	36.80
Outer leaves	2.79	3.40	2.68	3.98	1.80
Cabbage - outer leaves	26.92	42.00	32.00	31.09	35.00
Cut cabbage	26.00	41.60	31.80	30.80	34.90
Cut cabbage to processing	23.48	15.50	21.00	20.20	19.40
Salt solution added	5.20	8.85	5.20	5.20	5.20
Sauerkraut	21.00	15.30	16.40	17.20	19.90
Sauerkraut juice	6.50	9.05	9.15	7.95	4.10
treated = received 3 x 100 g a.s./ha 21 ± 1, 14 ± 1, and 7 ± 1 days before sampling.					

Table 2.14.2-6: Transfer factors for cooked cabbage and sauerkraut - DocID 2002/1004078

	alpha-cypermethrin residue found (mg/kg)			Transfer Factor		
	11-G01N063R	5-G01N064R	11-G01N065R	11-G01N063R	5-G01N064R	11-G01N065R
Cabbage (RAC)*	<0.05	<0.05	<0.05	1	1	1
Sauerkraut	<0.05	<0.05	<0.05	<1	<1	<1
Sauerkraut juice	<0.05	<0.05	<0.05	<1	<1	<1
Cabbage (RAC)*	<0.05	<0.05	<0.05	1	1	1
Outer leaves	0.17	0.27	0.15	cannot be calculated**		
Inner leaves	<0.05	<0.05	<0.05	<1	<1	<1
Stems	<0.05	<0.05	<0.05	<1	<1	<1
Cooked cabbage	<0.05	<0.05	<0.05	<1	<1	<1
Cooking liquid	<0.05	<0.05	<0.05	<1	<1	<1

* The treated cabbage samples used for the residue determination (RAC) were: 9-G01N063R, 3-G01N064R, and 9-G01N065R, respectively.

* No transfer factor could be determined--no detectable residue in the RAC cabbage. However, these cabbage plants received 30 times the maximum single seasonal application, therefore adjusting for the exaggeration would result in no detectable (<0.05 mg/kg) alpha-cypermethrin residues in the outer cabbage leaf fraction.

Conclusion:

The data from this processing study demonstrate clearly that there is little or no likelyhood of detectable alpha-cypermethrin residues resulting in the edible process fractions or processing waste steams from cabbage treated at the GAP proposed rate.

2.14.3 Estimation of MRL, HR and STMR for head cabbage

For *head cabbage*, the following residue studies were considered (BASF DocIDs): 2004/1006470, 2006/1026852 and 2007/1013148.

The following residue values (PHI=7±1 days for EU-N, PHI=3±1 days for EU-S. or later if higher residues occured) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2004/1006470 (field)	<0.05 (4x)	-
2006/1026852 (field)	<0.01, 0.014, 0.018, 0.104	<0.01, 0.013
2007/1013148 (field)	<0.01 (3x), 0.105	0.010, 0.013
OECD-MRL-calculation	**0.2 (n=12, STMR=0.034, HR=0.105)**	**0.03 (4, STMR=0.013, HR=0.012)**

_ underlined values were used for risk assessment purposes

2.15 Leafy brassica

Besidues the new studies presented in Section M-CA 6.3, no additional residue data were considered for leafy brassica.

2.16 Lettuce

In the framework of the original inclusion of alpha-cypermethrin into AnnexI of Directive 91/414; residue studies in lettuce were peer-reviewed. Below, peer reviewed data supporting the current GAP and considered for MRLproposal and risk assessment are summarized. New trials are presented inSection M-CA 6.3.3; the MRL calculations are shown in Section M-CA 6.7. Additional glasshouse and field data are also presented below.

Table 2.16-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Lettuce	1981	-	-	2 (field)	FR	2	AL-726-004
Lettuce	1992	2 (indoor)	FR	-	-	2	AL-726-012
Lettuce	2003	4	DE	-	-	4	2004/1006469
Lettuce	2005	4 (glasshouse)	BE, DE, DK, FR	4 (glasshouse)	ES, FR, GR, IT		2006/1037555
Lettuce	2008	2	DE, NL	2	FR, IT	4	BASF DocID 2009/1090703
Total number of trials per region		**6 (indoor) 6 (field)**	**-**	**4 (field) 4 (glasshouse)**	**Total number of trials**	**4**	

2.16.1 Supervised residue trials in lettuce

In the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414; residue studies in lettuce were peer-reviewed. Below, residue data from peer-reviewed trials supporting the current GAP and considered for MRL proposal and risk assessment are summarized. New trials are presented in Section M-CA 6.3.4; the MRL calculations are shown in Section M-CA 6.7.

Lamb´s lettuce

Two indoor trials were performed in lamb´s lettuce in France (Northern European region) in 1992. A 50 g a.s./L formulation of alpha-cypermethrin was applied twice at a target rate of 10 g a.s./ha. Lettuce leaves were sampled 7 days after the last application. Residues in leaves ranged between 0.283 and 0.294 mg/kg.

The trial data and residue results are summarised in Table 2.16.1-2.

Lettuce

Two open field trials were carried out in France (Southern European region) in 1981 using one application of alpha-cypermethrin (100 g a.s./L EC) at rates of 9 or 15 g a.s./ha. Lettuce was sampled immediately after application as well as 3 and 10 days afterwards.
After 9 g a.s./ha, residues in the lettuce immediately after application were 0.31 mg/kg falling to 0.21 and 0.12 mg/kg after 3 and 10 days, respectively. After 15 g a.s./ha, residues in the lettuce immediately after application were 0.41 mg/kg falling to 0.28 and 0.17 mg/kg after 3 and 10 days, respectively.

During the 2003 growing season, a total of 4 trials was conducted in head lettuce. BAS 310 03 I and BAS 310 QC I, both soluble concentrate formulations of alpha-cypermethrin (SC; 100 g a.s./L) were foliar applied once at a rate of 9 g a.s./ha to head lettuce plants on separate plots. The growth stages of the plants were between 45 (50% of the expected head size reached) and 49 (typical size, form and firmness of heads reached) at application.
Head lettuce specimens were collected directly after the application from each plot at all locations as well as 3 and 7 days thereafter.
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS with BASF analytical method No. 546/0 which has a limit of quantitation of 0.05 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.16.1-1

Eight glasshouse trials were performed in the year 2005 in Belgium, Denmark, France (Southern European region), Germany, Greece, Italy and Spain to determine the magnitude of the residue after application of a 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC; BAS 310 40 I). The product was foliar applied once at a rate of 40 g a.s./ha. The growth stage of the plants at application was between 41 (heads begin to form: the two youngest leaves do not unfold) and 47 (70% of the expected head size reached).
Samples of lettuce heads were collected 0, 2-4, 6-8 and 13-14 days after application. The specimens were analysed for total cypermethrin residues by means of HPLC-MS/MS with BASF analytical method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.16.1-1

Table 2.16.1-1: Residues in head lettuce – open field trials

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF RDI No. AL-726-004 BEGR.82.035 Trial No. W/FR/E 81/446 Study not to GLP Study carried out in 1981	Lettuce (variety Capitan) outdoor	France 33 – Eysines (South of EU)	9 g a.s./ha 23.10.81 Fastac EC 100 g a.s./L	near harvest	0 3 10	lettuce 0.31 lettuce 0.21 lettuce 0.12	SAMS 233-1 lettuces: mean recovery 120% at 0.10 mg/kg GC/ECD Residues analysed as underivatised analyte
BASF RDI No. AL-726-004 BEGR.82.035 Trial No. W/FR/E 81/446 Study not to GLP Study carried out in 1981	Lettuce (variety Capitan) outdoor	France 33 – Eysines (South of EU)	15 g a.s./ha 23.10.81 Fastac EC 100 g a.s./L	near harvest	0 3 10	lettuce 0.41 lettuce 0.28 lettuce 0.17	
BASF Doc ID 2004/1006469 Trial No. ACK/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Nadine)	Germany 16818 Wustrau Brandenburg	9 g a.s./ha 10.06.03 BAS 310 03 I	45	0 3 7	head 0.189 head 0.099 head <0.05	BASF analytical method No. 546/0 lettuce head: mean recovery = 101.5 %; SD: +/- 4.7; CV: 4.7; n=4; fortification range 0.05-0.5 mg/kg Residue analysed as total

Table 2.16.1-1: Residues in head lettuce – open field trials

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/1006469 Trial No. AGR/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Jesina)	Germany 47638 Straelen Nordrhein-Westfalen	9 g a.s./ha 03.06.03 BAS 310 03 I	48	0 3 7	head 0.441 head 0.068 head 0.054	cypermethrin
BASF Doc ID 2004/1006469 Trial No. DU2/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Ponchito)	Germany 69121 Handschuhs-heim Baden-Württemberg	9 g a.s./ha 23.06.03 BAS 310 03 I	48	0 3 7	head 0.073 head <0.05 head 0.060	
BASF Doc ID 2004/1006469 Trial No. DU4/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Nadine)	Germany 67376 Harthausen Rheinland-Pfalz	9 g a.s./ha 23.06.03 BAS 310 03 I	49	0 3 7	head 0.232 head 0.149 head 0.063	
BASF Doc ID 2004/1006469 Trial No. ACK/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Nadine)	Germany 16818 Wustrau Brandenburg	9 g a.s./ha 10.06.03 BAS 310 QC I	45	0 3 7	head 0.174 head 0.108 head <0.05	
BASF Doc ID 2004/1006469 Trial No. AGR/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Jesina)	Germany 47638 Straelen Nordrhein-Westfalen	9 g a.s./ha 03.06.03 BAS 310 QC I	48	0 3 7	head 0.287 head 0.091 head 0.054	
BASF Doc ID 2004/1006469 Trial No. DU2/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Ponchito)	Germany 69121 Handschuhs-heim Baden-Württemberg	9 g a.s./ha 23.06.03 BAS 310 QC I	48	0 3 7	head 0.155 head 0.058 head <0.05	
BASF Doc ID 2004/1006469 Trial No. DU4/03/03 Study to GLP Study carried out in 2003	Head lettuce (variety Nadine)	Germany 67376 Harthausen Rheinland-Pfalz	9 g a.s./ha 23.06.03 BAS 310 QC I	49	0 3 7	head 0.105 head 0.104 head 0.082	

Table 2.16.1-2: Residues in head lettuce – glasshouse trials

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF RDI No. AL-726-012 RMACH 9201/05 Trial No. MA/44/05/1 Study not to GLP Study carried out in 1992	Corn salad (variety Verte de Cambrai)	France Machecoul Pays de la Loire (North of EU) indoor	10 g a.s./ha 2 treatm. last date 06.10.92 Fastac 50 g a.s./ha	-	7	leaves 0.294	Mean recovery 86% GC-ECD Residues analysed as underivatised analyte
BASF RDI No. AL-726-012 RMACH 9201/05 Trial No. MA/44/05/2 Study not to GLP Study carried out in 1992	Corn salad (variety Verte de Cambrai)	France Machecoul Pays de la Loire (North of EU) indoor	10 g a.s./ha 2 treatm. last date 06.10.92 Fastac 50 g a.s./ha	-	7	leaves 0.283	
BASF Doc ID 2006/1037555 Trial No. 05 I CL FR P33 Study to GLP Study carried out in 2005	Head lettuce (variety Mathilda)	France 37700 Saint Pierre des Corps, Centre France (North of EU)	40 g a.s./ha 25.10.05 glasshouse	41	0 3 6 14	heads 0.930 heads 1.095 heads 0.685 heads 0.427	BASF analytical method No. 567/0 lettuce heads: mean recovery = 112.0% (110.3-113.8%); SD: n/a; CV: n/a; n=2; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1037555 Trial No. AC/05/044 Study to GLP Study carried out in 2005	Head lettuce (variety Ponchito)	Germany 16833 Fehrbellin Brandenburg	40 g a.s./ha 08.09.05 glasshouse	44	0 4 7 13	heads 1.054 heads 0.206 heads 0.091 heads 0.018	
BASF Doc ID 2006/1037555 Trial No. AGR/52/05 Study to GLP Study carried out in 2005	Head lettuce (variety Delta F1 Omega)	Belgium 3454 Rummen Limburg	40 g a.s./ha 16.06.05 glasshouse	47	0 3 8 14	heads 1.039 heads 0.445 heads 0.212 heads 0.049	
BASF Doc ID 2006/1037555 Trial No. ALB/190509-01 Study to GLP Study carried out in 2005	Head lettuce (variety Hawaii)	Denmark 7000 Fredericia	40 g a.s./ha 17.06.05 glasshouse	44	0 3 7 14	heads 0.613 heads 0.432 heads 0.274 heads 0.177	
BASF Doc ID 2006/1037555 Trial No. 05 I CL FR P37 Study to GLP Study carried out in 2005	Head lettuce (variety Boreal)	France 13550 Noves Provence (South of EU)	40 g a.s./ha 29.11.05 glasshouse	47	0 3 7 14	heads 0.928 heads 0.768 heads 0.682 heads 0.454	
BASF Doc ID 2006/1037555 Trial No. IR05BASI11LG01 Study to GLP Study carried out in 2005	Head lettuce (variety Limax)	Italy 45020 Lusia (RO) Rovigo	40 g a.s./ha 29.06.05 glasshouse	47	0 3 7 14	heads 0.948 heads 0.853 heads 0.300 heads 0.153	BASF analytical method No. 567/0 lettuce heads: mean recovery = 112.0% (110.3-113.8%); SD: n/a; CV: n/a; n=2; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1037555 Trial No. 05ES/084R Study to GLP Study carried out in 2005	Head lettuce (variety Filipus)	Spain 41710 Utrera Andalucia Sevilla	40 g a.s./ha 07.10.05 glasshouse	47	0 3 7 13	heads 0.800 heads 0.560 heads 0.299 heads 0.201	

Table 2.16.1-2: Residues in head lettuce – glasshouse trials

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1037555 Trial No. 05RF045 Study to GLP Study carried out in 2005	Head lettuce (variety Atraxion)	Greece Profitis Thessaloniki Central Macedonia GR 57200	40 g a.s./ha 12.10.05 glasshouse	47	0 2 7 13	heads 1.648 heads 0.765 heads 0.571 heads 0.154	

Report:	**Klaas P.,Ziske J., 2009d**
Title:	Study on the residue behaviour of Alpha-Cypermethrin in lettuce after treatment with BAS 310 51 I and BAS 310 40 I under field conditions in Germany, The Netherlands, Southern France and Italy, 2008
Document No:	BASF DocID 2009/1090703
Guidelines:	EEC 91/414 Annex III (Part A Section 8), EEC 91/414 Annex II (Part A Section 6), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 1607/VI/97 rev. 2 10.06.1999
GLP	yes

Executive Summary

During the growing season of 2008, four field trials with lettuce (field conditions) were conducted in Northern and Southern Europe, in order to determine the magnitude of the residues of BAS 310 I (alpha-cypermethrin) after treatment with BAS 310 51 I and BAS 310 40 I.
Each field trial consisted of three plots:
Trials L080177 (Germany) and L080178 (The Netherlands):
Plot 1: Control
Plot 2: Treated twice with 0.3 L/ha of BAS 310 51 I (15.0 g/ha of BAS 310 I, ME formulation)
Plot 3: Treated twice with 0.15 L/ha of BAS 310 40 I (15.0 g/ha of BAS 310 I, EC formulation)
Trials L080179 (France-South) and L080180 (Italy):
Plot 1: Control
Plot 2: Treated once with 0.6 L/ha of BAS 310 51 I (30.0 g/ha of BAS 310 I, ME formulation)
Plot 3: Treated once with 0.3 L/ha of BAS 310 40 I (30.0 g/ha of BAS 310 I, EC formulation)
The applications took place 13-15 days before harvest (DBH) and 7 DBH on trials L080177 and L080178 and 6-7 DBH on trials L080179 and L080180 with a spray volume of 400 L/ha of spray.
Alpha-cypermethrin was extracted from specimens according to an adaption of BASF Method No. 567/0 (L0020/01) with a limit of quantitation (LOQ) of 0.01 mg/kg. The analytical results demonstrate that the treatment with BAS 310 51 I and BAS 310 40 I did not lead to significantly different residue levels in the raw agricultural commodity at harvest.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 51 I
BAS 310 40 I
Lot/Batch #: BAS 310 51 I, 101156, alpha-cypermethrin: 50 g a.s./L
BAS 310 40 I, 1209, alpha-cypermethrin: 100 g a.s./L
Purity:
CAS#: 67375-30-8
Development code:
Spiking levels: 0.01-1.00 mg/kg

2. Test Commodity:

Crop: Lettuce
Type: Leaf vegetable
Variety: Tizian, Santoro, Fortina, Noisette
Botanical name: *Lactuca sativa*
Crop parts(s) or processed
Commodity: Head
Sample size: 12 units/1.0-2.0 kg

B. STUDY DESIGN

1. Test procedure

During the 2008 growing season, a total of four open field trials were conducted in lettuce in Germany, The Netherlands, Southern France and Italy.
The objective of the study was to determine the magnitude of alpha-cypermethrin residues in lettuce after application of the formulations BAS 310 51 I (ME) and BAS 310 40 I (EC). The additional objective was the comparison of the residue levels after the application of the EC and the ME formulation.
Each field trial consisted of three plots (one control plot, plot 1), one plot treated with BAS 310 51 I (plot 2) and another plot treated with BAS 310 40 I (plot 3). In two of four trials (L080177 and L080178) on the treated plot 2, BAS 310 51 I was applied twice at a rate targeting 0.3 L product/ha, equal to 15 g alpha-cypermethrin/ha and on the treated plot 3, BAS 310 40 I was applied twice at a rate targeting 0.15 L product/ha, equal to 15 g alpha-cypermethrin/ha. The applications were made targeting 14±1 days and 7±1 days before harvest.
For the other two trials trials (L080179 and L080180) on the treated plot 2, BAS 310 51 I was applied once at a rate targeting 0.6 L product/ha, equal to 30 g alpha-cypermethrin/ha and on the treated plot 3, BAS 310 40 I was applied once at a rate targeting 0.3 L product/ha, equal to 30 g alpha-cypermethrin/ha. The applications were made targeting 7±1 days before harvest. Both formulations were applied with the same GAP.

0 DALA (days after last application) sampling in the untreated plot was performed. The treated lettuce specimens (head) were taken from both plots immediately after the last application (0 DALA). Untreated and treated specimens were additionally collected 3±1, 7±1 and 14±1 DALA.

Table 2.16.1-3: Target application rates and timings for lettuce

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (g a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2008	4	2	F	BAS 310 51 I	alpha-cypermethrin	15	400	7±1, 14±1 days before harvest
		1				30		
	4	2	F	BAS 310 40 I	alpha-cypermethrin	15	400	7±1, 14±1 days before harvest
		1				30		

2. Description of analytical procedures

Alpha-cypermethrin was extracted from specimens according to an adaption of BASF Method No. 567/0 (L0020/01) with a limit of quantitation (LOQ) of 0.01 mg/kg.
The residues of alpha-cypermethrin in lettuce specimens were extracted from plant matrices using a mixture of methanol, water and HCl 2 mol/L. For clean-up a liquid/liquid partition against cyclohexane was used. The final determination of alpha-cypermethrin was performed by LC-MS/MS.

Table 2.16.1-4: Summary of recoveries for alpha-cypermethrin in lettuce

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
BASF Method No. 567/0		**alpha-cypermethrin (BAS 310 I)**		
lettuce (head)		8	100	7

II. RESULTS AND DISCUSSION

Directly after the last application (0 DALA) of BAS 310 51 I (plot 2), alpha-cypermethrin residues were 0.29-0.66 mg/kg. At 3±1 days after the last application, residues were 0.10-0.35 mg/kg, at 7±1 days after the last application residues were 0.07-0.33 mg/kg, and at 14±1 days after the last application, residues were 0.02-0.10 mg/kg.
Directly after the last application (0 DALA) of BAS 310 40 I (plot 3), alpha-cypermethrin residues were 0.36-0.46 mg/kg. At 3±1 days after the last application, residues were 0.14-0.22 mg/kg, at 7±1 days after the last application residues were 0.13-0.15 mg/kg, and at 14±1 days after the last application, residues were 0.03-0.12 mg/kg.
An overall summary of the residues is given in the table below.

Table 2.16.1-5: Summary of residues in lettuce after application of BAS 310 51 I or BAS 310 40 I

Crop	Year	No. of Appl.	Application	DALA[1)]	BBCH[2)]	Range of Residues (mg/kg)	
						Matrix	alpha-cypermethrin
Lettuce	2008	1	BAS 310 51 I (ME)	0 2-3 6-7 13-14	41-47	head	0.42-0.56 0.17-0.20 0.07-0.11 0.02-0.07
		2		0 3 7 13-15	47-48	head	0.29-0.66 0.10-0.35 0.10-0.33 0.06-0.10
		1	BAS 310 40 I (EC)	0 2-3 6-7 13-14	41-47	head	0.40-0.46 0.19-0.21 0.15 0.03-0.09
		2		0 3 7 13-15	47-48	head	0.36-0.39 0.14-0.22 0.13-0.15 0.06-0.12

1) DALA Days after last application
2) BBCH stage at respective sampling

III. CONCLUSION

The analytical results demonstrate that the treatment with BAS 310 51 I and BAS 310 40 I did not lead to significantly different residue levels in the raw agricultural commodity at harvest.

Table 2.16.1-6: Residues of alpha-cypermethrin after application of the formulations BAS 310 51 I and BAS 310 40 I in Northern Europe

Study Details		Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
							Matrix	Alpha-Cypermethrin
Study code:	319698	Lettuce	Germany	2 x BAS 310 51 I 0.0150	48	0	head	0.66
Doc ID:	2009/1090703					3	head	<u>0.35</u>
Trial No.:	L080177					7	head	0.33
GLP:	yes					13	head	0.10
Year	2008			2 x BAS 310 40 I 0.0150		0	head	0.36
						3	head	0.22
						7	head	0.15
						13	head	0.12
Study code:	319698	Lettuce	The Netherlands	2 x BAS 310 51 I 0.0150	47	0	head	0.29
Doc ID:	2009/1090703					3	head	0.10
Trial No.:	L080178.					7	head	0.10
GLP:	yes					15	head	0.06
Year	2008			2 x BAS 310 40 I 0.0150		0	head	0.39
						3	head	<u>0.14</u>
						7	head	0.13
						15	head	0.06

DALA = Days after last application
_ underlined values were used for MRL calculation

Table 2.16.1-7: Residues of alpha-cypermethrin after application of the formulations BAS 310 51 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin
Study code: 319698 **Doc ID:** 2009/1090703 **Trial No.:** L080179 **GLP:** yes **Year** 2008	Lettuce	France	1 x BAS 310 51 I 0.0300	41	0	head	0.42
					2	head	<u>0.20</u>
					6	head	0.11
					13	head	0.07
			1 x BAS 310 40 I 0.0300		0	head	0.46
					2	head	0.19
					6	head	0.15
					13	head	0.09
Study code: 319698 **Doc ID:** 2009/1090703 **Trial No.:** L080180 **GLP:** yes **Year** 2008	Lettuce	Italy	1 x BAS 310 51 I 0.0300	47	0	head	0.56
					3	head	0.17
					7	head	0.07
					14	head	0.02
			1 x BAS 310 40 I 0.0300		0	head	0.40
					3	head	<u>0.21</u>
					7	head	0.15
					14	head	0.03

DALA = Days after last application
_ underlined values were used for MRL calculation

2.17 Spinach

Residue data from supervised trials in spinach were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.17-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Spinach (field)	2005	1	NL	-	-	1	2006/1026849
Spinach (field)	2006	3	DE, FR	-	-	3	2007/1007944 2008/1037135
Spinach (field)	2006	4	DE	-	-	4	2007/1035745
Total number of trials per region		**8**			**Total number of trials**	**8**	

2.17.1 Supervised residue trials in spinach

North L (2006)

Full study reference

North L (2006): Study on the residue behaviour of BAS 310 I in Spinach after treatment with BAS 310 40 I under field conditions in Northern Europe during 2005; BASF DocID 2006/1026849

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of alpha-cypermethrin in Spinach after treatment with BAS 310 40 I under field conditions in Northern Europe during 2006; BASF DocID 2007/1007944

Report:
North L., 2007e
Study on the residue behaviour of Alpha-Cypermethrin in spinach after treatment with BAS 310 40 I under field conditions in Northern Europe during 2006
2007/1007944

Guidelines: none

GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Report:
North L., 2008a
Amendment No. 1: Study on the residue behaviour of Alpha-Cypermethrin in spinach after treatment with BAS 310 40 I under field conditions in Northern Europe during 2006
2008/1037135

Guidelines: none

GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Material and Methods:

In the years 2005 and 2006 a residue program on spinach was conducted in the field in France (Northern European region), Germany and the Netherlands.
During the 2005 growing season, one field trial in spinach was conducted in the Netherlands. BAS 310 40 I, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L), was foliar applied to separate plots of spinach either once or twice at a target rate of 12.5 g a.s./ha. The last treatment was performed at growth stage 13 (3rd true leaf unfolded). Specimens of spinach leaves were collected immediately after the last application from each plot as well as 3, 7, and 14 days thereafter. The specimens were analysed using BASF method no. 567/0, which quantifies the residues of underivatised cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

In the year 2006, three residue trials were conducted in France (Northern European region) and Germany. A 100 g a.s./L emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I) was foliar applied to separate plots of spinach plants either as a single application at a target rate of 12.5 g a.s./ha or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants was between 35 (leaf rosette has reached 50% of the expected diameter typical for the variety) and 49 (typical leaf mass reached) at the last application. Samples of spinach foliage were collected 0, 3, 7 and 14 days after the last application in all trials.
Residue analysis was performed according to BASF analytical method No. 567/0 which determines total cypermethrin residues by means of HPLC-MS/MS with a LOQ of 0.01 mg/kg.

The trial data and residue results are summarised in Table 2.17.1-1.

Table 2.17.1-1: Residues in spinach

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026849 Trial No. AF/8819/BA/1 Study to GLP Study carried out in 2005	Spinach (variety Monza)	The Netherlands 3286 bb Klaaswaal, Zuid Holland	12.5 g a.s./ha 16.08.05	13	0 3 7 14	foliage 0.698 foliage 0.220 foliage 0.179 foliage 0.037	BASF analytical method No. 567/0 foliage: mean recovery = 93.9 %; SD: +/- 14.6; CV: 15.6; n=3; fortification range 0.01-3.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007944 Trial No. AF/10498/BA/1 Study to GLP Study carried out in 2006	Spinach (variety Laska)	France Saint Lambert de Levées 49400 (North of EU)	12.5 g a.s./ha 21.11.06	49	0 3 7 14	foliage 1.097 foliage 0.720 foliage 0.545 foliage 0.212	
BASF Doc ID 2007/1007944 Trial No. AF/10498/BA/2 Study to GLP Study carried out in 2006	Spinach (variety Elan)	Germany Frisdo Notel 30982 Patternsen-Jeinsen Lower Saxony	12.5 g a.s./ha 13.09.06	35	0 3 7 14	foliage 0.548 foliage 0.374 foliage 0.188 foliage 0.068	BASF method No. 567/0 foliage: mean recovery = 88.3%; SD: +/- 7.0; CV: 7.9; n=6; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007944 Trial No. AF/10498/BA/6 Study to GLP Study carried out in 2006	Spinach (variety Boeing)	France 91160 Saulx les Chartreux North of EU	12.5 g a.s./ha 08.12.06	47-49	0 3 7 14	foliage 0.687 foliage 0.808 foliage 0.711 foliage 0.839	
BASF Doc ID 2006/1026849 Trial No. AF/8819/BA/1 Study to GLP Study carried out in 2005	Spinach (variety Monza)	The Netherlands 3286 bb Klaaswaal, Zuid Holland	12.5 g a.s./ha 2 treatm. last date 16.08.05	13 at last treatm.	0 3 7 14	foliage 0.950 foliage 0.394 foliage <u>0.135</u> foliage 0.033	BASF analytical method No. 567/0 foliage: mean recovery = 93.9 %; SD: +/- 14.6; CV: 15.6; n=3; fortification range 0.01-3.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007944 Trial No. AF/10498/BA/1 Study to GLP Study carried out in 2006	Spinach (variety Laska)	France Saint Lambert de Levées 49400 (North of EU)	12.5 g a.s./ha 2 treatm. last date 21.11.06	49 at last treatm.	0 3 7 14	foliage 1.854 foliage 1.340 foliage <u>0.891</u> foliage 0.391	BASF method No. 567/0 foliage: mean recovery = 88.3%; SD: +/- 7.0; CV: 7.9; n=6; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin

Table 2.17.1-1: Residues in spinach

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007944 Trial No. AF/10498/BA/2 Study to GLP Study carried out in 2006	Spinach (variety Elan)	Germany Frisdo Notel 30982 Patternsen-Jeinsen Lower Saxony	12.5 g a.s./ha 2 treatm. last date 13.09.06	35 at last treatm.	0 3 7 14	foliage 0.612 foliage 0.331 foliage <u>0.185</u> foliage 0.086	
BASF Doc ID 2007/1007944 Trial No. AF/10498/BA/6 Study to GLP Study carried out in 2006	Spinach (variety Boeing)	France 91160 Saulx les Chartreux North of EU	12.5 g a.s./ha 2 treatm. last date 08.12.06	47-49 at last treatm.	0 3 7 14	foliage 1.133 foliage 1.420 foliage <u>1.113</u> foliage 0.965	BASF method No. 567/0 foliage: mean recovery = 88.3%; SD: +/- 7.0; CV: 7.9; n=6; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin

_ underlined values were used for MRL calculation

Findings:

The residue studies in spinach presented in the alpha-cypermethrin dossier were carried out in 3 different EU countries and provide data relevant to conditions in the Northern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-4 trials including both variants, all conducted in the Northern European region

After one application at 12.5 g a.s./ha, initial residues of alpha-cypermethrin in spinach foliage ranged between 0.55 mg/kg and 1.1 mg/kg. Residues declined to levels between 0.220-0.81 mg/kg, 0.179-0.71 mg/kg and 0.037-0.84 mg/kg 3, 7 and 14 days after application.

After two applications at 12.5 g a.s./ha, initial residues spinach foliage ranged between 0.61 mg/kg and 1.9 mg/kg. Residues declined to levels between 0.33-1.4 mg/kg, 0.135-1.11 mg/kg and 0.033-0.96 mg/kg 3, 7 and 14 days after application.

Conclusion:

In the years 2005 and 2006 a residue program on spinach was conducted in the field in France (Northern European region), Germany and the Netherlands.
In 4 trials supporting the GAP for the Northern European region, residues in spinach ranged between 0.135-1.11 mg/kg at the target PHI of 7 days.

Report:	**Anonymous, 2007a**
Title:	Residue behaviour of Alpha-Cypermethrin in/on fennel (bulbs formed by leafs stems), curly kale (leaves), blanched celery (stems) and spinach (leaves) outdoor after application of Fastac SC Super Contact (SC, 100 g/L) in Germany, 2006
Document No:	BASF DocID 2007/1035745
Guidelines:	none
GLP	yes

Executive Summary

During the growing season 2006 residue decline, shortened decline and harvest trials, were performed in fennel (bulbs formed by leaf stems), curly kale (leaves), blanched celery (stems) and spinach (leaves) as field trials on several sites in Germany.
Fastac SC Super Contact (alpha-cypermethrin, SC 100) was applied foliar 2 times at rates of 0.125 L product per hectare (12.5 g/ha alpha-cypermethrin) during growing season. The trials were conducted in order to determine the magnitude of the residues of the active ingredients in or on fennel (bulbs formed by leaf stems), curly kale (leaves), blanched celery (stems) and spinach (leaves). The samples were collected 0 - 7 -10 -14 (decline curves), 0 - 7 -14 (shortened decline study) and 14 days (harvest trials) after last application, respectively.
The samples were analysed for alpha-cypermethrin with modified method DFG S19. The limit of quantification (LOQ) was 0.010 mg/kg (alpha-cypermethrin).
Average recoveries from untreated samples, fortified with alpha-cypermethrin were 69 - 105 % in fennel and 74 - 92 % in curly kale at fortification levels between 0.01 mg/kg and 0.10 mg/kg. In blanched celery the recoveries were 75 - 110 % at fortification levels between 0.01 mg/kg and 0.5 mg/kg. In spinach the recoveries were 90 -100 % at fortification levels between 0.01 mg/kg and 1.0 mg/kg.
In context of this summary, only the results of alpha-cypermethrin for spinach are reported.
Residues of alpha-cypermethrin were at 7 DALA in spinach leaves in the range of 0.36 to 0.48 mg/kg.

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:**

Description:	BAS 310 41 I
Lot/Batch #:	not reported (BAS 310 41 I, 100 g/L alpha-cypermethrin, SC)
Purity:	not relevant
CAS#:	alpha-cypermethrin: 67375-30-8
Development code:	not applicable
Spiking levels:	0.01-1.0 mg/kg

2. **Test Commodity:**

Crop:	Spinach
Type:	Leaf vegetables & fresh herbs
Variety:	Puma F1, Stamm 1146, Rhino RZ, El Paso
Botanical name:	*Spinacia oleracea* L.
Crop parts(s) or processed commodity:	Leaves
Sample size:	min. 0.5 kg

B. STUDY DESIGN

1. Test procedure

During the growing season 2006 residue decline, shortened decline and harvest trials, were performed in fennel (bulbs formed by leaf stems), curly kale (leaves), blanched celery (stems) and spinach (leaves) as field trials on several sites in Germany.

Fastac SC Super Contact (alpha-cypermethrin, SC 100) was applied foliar 2 times at rates of 0.125 L product per hectare (12.5 g/ha alpha-cypermethrin) during growing season. The trials were conducted in order to determine the magnitude of the residues of the active ingredients in or on fennel (bulbs formed by leaf stems), curly kale (leaves), blanched celery (stems) and spinach (leaves). The samples were collected 0 - 7 -10 -14 (decline curves), 0 - 7 -14 (shortened decline study) and 14 days (harvest trials) after last application, respectively.

Table 2.17.1-2: Target application rates and timings for spinach

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2006	4	2	F	BAS 310 41 I (SC)	alpha-cypermethrin	0.0125	300-600	n r.

n r. not reported

2. Description of analytical procedures

The samples were analysed for alpha-cypermethrin with modified method DFG S19. The limit of quantification (LOQ) was 0.010 mg/kg (alpha-cypermethrin).
Average recoveries from untreated samples, fortified with alpha-cypermethrin were 69 - 105 % in fennel and 74 - 92 % in curly kale at fortification levels between 0.01 mg/kg and 0.10 mg/kg. In blanched celery the recoveries were 75 - 110 % at fortification levels between 0.01 mg/kg and 0.5 mg/kg. In spinach the recoveries were 90 - 100 % at fortification levels between 0.01 mg/kg and 1.0 mg/kg.
Alpha-cypermethrin: the samples were analysed for alpha-cypermethrin by the method DFG S 19 modified. The samples are extracted with acetone/water 2+1 (v+v) with subsequent extraction with cyclohexane/ethyl acetate 1+1 (v+v) and partition into acetone/cyclohexane/ethyl acetate. The extracts are cleaned up by gel permeation cbromatography on a Bio Beads SX-3 column followed by a silica gel fractionation. The residues are determined by gas chromatography using an electron capture detector. The quantification is done by external standardisation. The residues are not corrected for the recovery rates.

Table 2.17.1-3: Summary of recoveries of alpha-cypermethrin in spinach leaves

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
DFG S 19 (modified)		alpha-cypermethrin			
spinach leaves	0.01 / 0.1 / 1.0	7	94.1	3.9	3.7

II. RESULTS AND DISCUSSION

In context of this summary, only the results of alpha-cypermethrin for spinach are reported.
Residues of alpha-cypermethrin were at 7 DALA in spinach leaves in the range of 0.36 to 0.48 mg/kg.

Table 2.17.1-4: Summary of residues of BAS 310 I in spinach from trials according to critical GAP after application of BAS 310 41 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin (BAS 310 I)
Spinach	2006	2	BAS 310 41 I (SC)	7±1	16-49	leaves	0.36 - 0.48

DALA = days after last application
BBCH = growth stage at respective sampling

III. CONCLUSION

After 2 applications of SC-formulation BAS 310 41 I at rates of 0.0125 kg alpha-cypermethrin/ha, residues in curly kale leaves were at the day of the last application in the range of 0.24 to 0.60 mg/kg. Fourteen days later (at 14 DALA), residues were still ranging between 0.05 and 0.19 mg/kg.

Table 2.17.1-5: Residues of BAS 310 I after two applications of the formulation BAS 310 41 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: not reported **Doc ID:** 2007/1035745 **Trial No.:** RU-I-18 06 SN DD 1/1 **GLP:** yes (only analytical phase) **Year** 2006	Spinach	Germany (N)	BAS 310 41 I 2 x 0.0125	14-16	7	leaves	0.46
Study code: not reported **Doc ID:** 2007/1035745 **Trial No.:** RU-I-18 06 BB FO 1/1 **GLP:** yes (only analytical phase) **Year** 2006	Spinach	Germany (N)	BAS 310 41 I 2 x 0.0125	37	7	leaves	0.47
Study code: not reported **Doc ID:** 2007/1035745 **Trial No.:** RU-I-18 06 NW BN 1/1 **GLP:** yes (only analytical phase) **Year** 2006	Spinach	Germany (N)	BAS 310 41 I 2 x 0.0125	17-18	7	leaves	0.36
Study code: not reported **Doc ID:** 2007/1035745 **Trial No.:** RU-I-18 06 NW BN 1/2 **GLP:** yes (only analytical phase) **Year** 2006	Spinach	Germany (N)	BAS 310 41 I 2 x 0.0125	43-45	7	leaves	0.48

DALA = days after last application
BBCH = growth stage at respective sampling
_ underlined values were used for MRL calculation

2.17.2 Estimation of MRL, HR and STMR for spinach

For *spinach*, the following residue studies were considered (BASF DocIDs): 2006/1026849, 2007/1007944 and 2007/1035745.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026849	0,135	-
2007/1007944	0.185, 0.891, 1.113	-
2007/1035745	0.36, 0.46, 0.47, 0.48	-
OECD-MRL-calculation	**2.0 (n=8, STMR=0.465, HR=1.113)**	**-**

_ underlined values were used for risk assessment purposes

2.18 Green beans with pods

Residue data from supervised trials in green beans with pods were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.18-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Green beans with pods (field)	2005	4	DE, FR, NL, UK	4	ES, FR, GR, IT	8	2006/1026857
Green beans with pods (field)	2006	4	BE, FR, NL, UK	4	ES, FR, GR, IT	8	2007/1007950
Total number of trials per region		**8**		**8**	**Total number of trials**	**16**	

2.18.1 Supervised residue trials in green beans with pods

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of alpha-cypermethrin in green beans (Phaseolus sp.) after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026857

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alpha-cypermethrin in green beans *(Phaseolus sp.)* after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007950

Material and Methods:

In the years 2005 and 2006 a residue program on green beans was conducted in Belgium, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom under open field conditions.

During the 2005 growing season, eight field trials were performed in green beans in France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to bean plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 69 (end of flowering) and 79 (pods: individual beans easily visible).
In four trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to bean plants once at a target rate of 25 g a.s./ha at growth stages between 71 (10% of pods have reached typical length/beginning of pot development) and 78 (80% of pods have reached typical length).
Specimens of beans with pods and remaining plant were collected immediately after the last application from each plot, as well as approximately 3, 7 and 14 days thereafter.
Specimens were analysed using BASF analytical method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, eight field trials were conducted in green beans in Belgium, France (Northern and Southern European region), Greece, Italy, Spain, the Netherlands and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to bean plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 76 (60% of pods have reached typical length) and 79 (Pods: individual beans easily visible).
In four trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to bean plants once at a target rate of 25 g a.s./ha. The growth stage of the plants at application was between 77 (70% of pods have reached typical length, pods still break cleanly) and 86 (60% of pods ripe (beans hard)).
Specimens of beans with pods and remaining plant were collected immediately after the last application from each plot, as well as 3-4, 7-8 and 14 days thereafter.
Specimens were analysed using BASF analytical method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.18.1-1.

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/1 Study to GLP Study carried out in 2005	Fresh beans (variety Paulista)	United Kingdom Woodfield farm Pershore Birmingham WR10 3AG	12.5 g a.s./ha 21.07.05	69-79	0 0 3 3 7 7 14 14	beans with pods 0.052 remaining plant 0.519 beans with pods 0.028 remaining plant 0.249 beans with pods 0.021 remaining plant 0.138 beans with pods 0.011 remaining plant 0.034	
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/2 Study to GLP Study carried out in 2005	Fresh beans (variety Booster)	France Rue Gabriel Jeanton Lacrost 71700 (North of EU)	12.5 g a.s./ha 19.07.05	77	0 0 3 3 7 7 14 14	beans with pods 0.019 remaining plant 0.284 beans with pods 0.013 remaining plant 0.237 beans with pods 0.011 remaining plant 0.067 beans with pods <0.01 remaining plant 0.041	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/3 Study to GLP Study carried out in 2005	Fresh beans (variety Sigma)	Germany 68623 Lampertheim	12.5 g a.s./ha 22.07.05	78	0 0 3 3 7 7 14 14	beans with pods 0.016 remaining plant 0.861 beans with pods 0.011 remaining plant 0.192 beans with pods <0.01 remaining plant 0.034 beans with pods <0.01 remaining plant 0.032	

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/4 Study to GLP Study carried out in 2005	Fresh beans (variety Mention)	The Netherlands Leembaan, 6595 Ottersum Limburg	12.5 g a.s./ha 19.08.05	76-77	0 0 3 3 7 7 14 14	beans with pods 0.017 remaining plant 0.341 beans with pods 0.014 remaining plant 0.131 beans with pods 0.031 remaining plant 0.128 beans with pods 0.020 remaining plant 0.071	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/1 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Angers)	France Les Serres de Moulin a Vent Le Moulin a Vent, 49680 (North of EU)	12.5 g a.s./ha 28.07.06	78	0 0 3 3 7 7 14 14	beans with pods <0.01 remaining plant without roots 0.418 beans with pods <0.01 remaining plant without roots 0.076 beans with pods <0.01 remaining plant without roots 0.071 beans with pods <0.01 remaining plant without roots 0.038	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg, beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/2 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Minidor)	The Netherlands Zelder 1 6595 NW Ottersum Limburg	12.5 g a.s./ha 22.08.06	76	0 0 3 3 7 7 14 14	beans with pods <0.01 remaining plant without roots 0.248 beans with pods 0.011 remaining plant without roots 0.149 beans with pods 0.010 remaining plant without roots 0.081 beans with pods <0.01 remaining plant without roots 0.083	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg, beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/3 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Polder)	Belgium Rue de la Chapelle, 9 6210 Villers-Perwin	12.5 g a.s./ha 25.07.06	77	0 0 3 3 7 7 14 14	beans with pods 0.011 remaining plant without roots 0.392 beans with pods 0.011 remaining plant without roots 0.202 beans with pods 0.013 remaining plant without roots 0.078 beans with pods <0.01 remaining plant without roots 0.046	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg, beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/4 Study to GLP Study carried out in 2006	Fresh beans with pods	United Kingdom Woodfield farm Birligham Pershore WR10 3AG	12.5 g a.s./ha 07.09.06	77-79	0 0 4 4 8 8 14 14	beans with pods 0.015 remaining plant without roots 0.254 beans with pods 0.019 remaining plant without roots 0.197 beans with pods 0.010 remaining plant without roots 0.100 beans with pods <0.01 remaining plant without roots <0.01	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg, beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/1 Study to GLP Study carried out in 2005	Fresh beans (variety Paulista)	United Kingdom Woodfield Fram, Pershore Birmingham WR10 3AG	12.5 g a.s./ha 2 treatm. last date 21.07.05	69-79 at last treatm.	0 0 3 3 7 7 14 14	beans with pods 0.048 remaining plant 0.394 beans with pods 0.027 remaining plant 0.132 beans with pods 0.026 remaining plant 0.087 beans with pods <0.01 remaining plant 0.055	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/2 Study to GLP Study carried out in 2005	Fresh beans (variety Booster)	France Rue Gabriel Jeanton Lacrost 71700 (North of EU)	12.5 g a.s./ha 2 treatm. last date 19.07.05	77 at last treatm.	0 0 3 3 7 7 14 14	beans with pods 0.025 remaining plant 0.373 beans with pods 0.016 remaining plant 0.184 beans with pods <u>0.011</u> remaining plant 0.144 beans with pods <0.01 remaining plant 0.077	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/3 Study to GLP Study carried out in 2005	Fresh beans (variety Sigma)	Germany 68623 Lampertheim	12.5 g a.s./ha 2 treatm. last date 22.07.05	78 at last treatm.	0 0 3 3 7 7 14 14	beans with pods 0.025 remaining plant 0.816 beans with pods 0.020 remaining plant 0.253 beans with pods <0.01 remaining plant 0.077 beans with pods <u>0.014</u> remaining plant 0.092	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/4 Study to GLP Study carried out in 2005	Fresh beans (variety Mention)	The Netherlands Leembaan, 6595 Ottersum Limburg	12.5 g a.s./ha 2 treatm. last date 19.08.05	76-77 at last treatm.	0 0 3 3 7 7 14 14	beans with pods 0.023 remaining plant 0.339 beans with pods 0.031 remaining plant 0.305 beans with pods <u>0.015</u> remaining plant 0.177 beans with pods 0.014 remaining plant 0.107	

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/1 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Angers)	France Les Serres de Moulin a Vent Le Moulin a Vent, 49680 (North of EU)	12.5 g a.s./ha 2 treatm. last date 28.07.06	78 at last treatm.	0 0 3 3 7 7 14 14	beans with pods <0.01 remaining plant without roots 0.535 beans with pods 0.011 remaining plant without roots 0.123 beans with pods <u><0.01</u> remaining plant without roots 0.089 beans with pods <0.01 remaining plant without roots 0.059	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg, beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/2 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Minidor)	The Netherlands Zelder 1 6595 NW Ottersum Limburg	12.5 g a.s./ha 2 treatm. last date 22.08.06	76 at last treatm.	0 0 3 3 7 7 14 14	beans with pods 0.024 remaining plant without roots 0.328 beans with pods 0.012 remaining plant without roots 0.199 beans with pods <u>0.017</u> remaining plant without roots 0.172 beans with pods 0.012 remaining plant without roots 0.135	

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/3 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Polder)	Belgium Rue de la Chapelle, 9 6210 Villers-Perwin	12.5 g a.s./ha 2 treatm. last date 25.07.06	77 at last treatm.	0 0 3 3 7 7 14 14	beans with pods 0.028 remaining plant without roots 0.357 beans with pods 0.019 remaining plant without roots 0.344 beans with pods <u>0.019</u> remaining plant without roots 0.127 beans with pods 0.013 remaining plant without roots 0.073	
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/4 Study to GLP Study carried out in 2006	Fresh beans with pods	United Kingdom Woodfield farm Birligham Pershore WR10 3AG	12.5 g a.s./ha 2 treatm. last date 07.09.06	77-79 at last treatm.	0 0 4 4 8 8 14 14	beans with pods 0.020 remaining plant without roots 0.525 beans with pods 0.018 remaining plant without roots 0.313 beans with pods <u>0.018</u> remaining plant without roots 0.123 beans with pods <0.01 remaining plant without roots 0.074	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/5 Study to GLP Study carried out in 2005	Fresh beans (variety Adagio)	France Les Saignes Montromant 69610 (South of EU)	25 g a.s./ha 26.07.05	77	0 0 3 3 7 7 14 14	beans with pods 0.047 remaining plant 0.504 beans with pods <u>0.028</u> remaining plant 0.256 beans with pods 0.017 remaining plant 0.105 beans with pods <0.01 remaining plant 0.050	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/6 Study to GLP Study carried out in 2005	Fresh beans (variety Avalon)	Italy Viabastia 6609 Lugo (RA) Emilia Romana 48022	25 g a.s./ha 05.07.05	71-75	0 0 3 3 7 7 14 14	beans with pods 0.054 remaining plant 0.888 beans with pods <u>0.063</u> remaining plant 0.796 beans with pods 0.033 remaining plant 0.305 beans with pods 0.021 remaining plant 0.160	

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/7 Study to GLP Study carried out in 2005	Fresh beans (variety Antea)	Spain Carretera San Adrian No. 1 Milagro 31320	25 g a.s./ha 26.09.05	72-73	0 0 3 3 7 7 14 14	beans with pods 0.042 remaining plant 0.856 beans with pods <u>0.020</u> remaining plant 0.455 beans with pods 0.013 remaining plant 0.273 beans with pods <0.01 remaining plant 0.139	BASF analytical method N 567/0 beans with pods: mean recovery = 87.6%; SD: +/- 9.7; CV: 11.1; n=6; fortification range 0.01-0.5 mg/kg remaining plant: mean recovery = 77.8%; SD: 11.0 +/-; CV: 14.2; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026857 Trial No. AF/8825/BA/8 Study to GLP Study carried out in 2005	Fresh beans (variety Plati)	Greece Thessaloniki Central Macedonia GR-56224	25 g a.s./ha -05.09.05	78	0 0 3 3 7 7 14 14	beans with pods 0.023 remaining plant 0.921 beans with pods 0.013 remaining plant 0.594 beans with pods <u>0.018</u> remaining plant 0.364 beans with pods 0.010 remaining plant 0.186	

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/5 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Booster)	France 26 Rue Neuve St Caprais 31330 (South of EU)	25 g a.s./ha 05.09.06	77-79	0 0 3 3 8 8 14 14	beans with pods 0.064 remaining plant without roots 1.551 / 1.504* beans with pods <u>0.045</u> remaining plant without roots 1.013 / 0.978* beans with pods <0.01 remaining plant without roots 0.040 beans with pods <0.01 remaining plant without roots 0.021	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin *two measurements: undiluted/diluted

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/6 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Aneto)	Spain C/Neuva No. 8 Melida Navarra 31382	25 g a.s./ha 22.09.06	77	0 0 3 3 7 7 14 14	beans with pods 0.015 remaining plant without roots 0.911 beans with pods <u>0.028</u> remaining plant without roots 0.634 beans with pods 0.0185 remaining plant without roots 0.426 beans with pods 0.013 remaining plant without roots 0.245	BASF analytical method N 567/0 remaining plant without roots: mean recovery = 94.7%; SD: +/- 6.4; CV: 6.7%; n=10; fortification range 0.01-2.0 mg/kg beans with pods: mean recovery = 88.1%; SD: +/- 5.0; CV: 5.7%; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin *two measurements: undiluted/diluted
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/7 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Rosana)	Greece Giannitsa Pella Central Macedonia	25 g a.s./ha 11.09.06	77	0 0 3 3 7 7 14 14	beans with pods 0.014 remaining plant without roots 0.938 / 1.376* beans with pods <u>0.050</u> remaining plant without roots 0.704 beans with pods 0.026 remaining plant without roots 0.351 beans with pods 0.011 remaining plant without roots 0.142	

Table 2.18.1-1: Residues in green beans with pods

GAP for EU-N is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007950 Trial No. AF/10492/BA/8 Study to GLP Study carried out in 2006	Fresh beans with pods (variety Masai)	Italy Az. Agr. Rilli Via Boncellino, 57 Bagnacavallo (RA) 48012	25 g a.s./ha 25.09.06	85-86	0 0 3 3 7 7 14 14	beans with pods 0.021 remaining plant without roots 0.979 beans with pods <u>0.039</u> remaining plant without roots 0.198 beans with pods 0.021 remaining plant without roots 0.073 beans with pods <0.01 remaining plant without roots 0.051	

_ underlined values were used for MRL calculation

Findings:

The residue studies in green beans presented in the alpha-cypermethrin dossier were carried out in 8 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-8 trials including both variants, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions-8 trials conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues in bean plant material ranged between 0.248 mg/kg and 0.861 mg/kg. Residues declined to levels between 0.076-0.249 mg/kg, 0.034-0.138 mg/kg and <0.01-0.083 mg/kg at 3-4, 7-8 and 14 days after application. In beans with pods, initial residues ranged between <0.01 mg/kg and 0.052 mg/kg. Residues declined to levels between <0.01-0.028 mg/kg, <0.01-0.031 mg/kg and <0.01-0.020 mg/kg at 3-4, 7-8 and 14 days after application.

After two applications at 12.5 g a.s./ha, initial residues in bean plant material ranged between 0.328 mg/kg and 0.816 mg/kg. Residues declined to levels between 0.123-0.344 mg/kg, 0.077-0.177 mg/kg and 0.055-0.135 mg/kg at 3-4, 7-8 and 14 days after application. In beans with pods, initial residues in ranged between <0.01 mg/kg and 0.048 mg/kg. Residues declined to levels between 0.011-0.031 mg/kg, <0.01-0.026 mg/kg and <0.01-0.014 mg/kg at 3-4, 7-8 and 14 days after application.

After one application at 25 g a.s./ha, initial residues in bean plant material ranged between 0.504 mg/kg and 1.551/1.504 mg/kg. Residues declined to levels between 0.198-1.013 / 0.978 mg/kg, 0.040-0.426 mg/kg and 0.021-0.245 mg/kg at 3, 7-8 and 14 days after application. In beans with pods, initial residues in ranged between 0.014 mg/kg and 0.064 mg/kg. Residues declined to levels between 0.013-0.063 mg/kg, <0.01-0.033 mg/kg and <0.01-0.021 mg/kg at 3, 7-8 and 14 days after application.

Conclusion:

In the years 2005 and 2006 a residue program on green beans was conducted in Belgium, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom under open field conditions.
In 8 trials supporting the proposed field GAP for the Southern European region, residues in bean plant material ranged between 0.198-1.013 / 0.978 mg/kg at the target PHI of 3 days and between 0.040-0.426 mg/kg at the target PHI of 7±1 days. Residues in beans with pods ranged between 0.013-0.063 mg/kg at the target PHI of 3 days and between <0.01-0.033 mg/kg at the target PHI of 7±1 days.

2.18.2 Estimation of MRL, HR and STMR for green beans with pods

For *green beans with pods*, the following residue studies were considered (BASF DocIDs): 2006/1026857 and 2007/1007950.

The following residue values (PHI=7±1 days for EU-N or PHI=3±1 days for EU-S, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026857	0.11, 0.014, 0.015, 0.026	0.018, 0.020. 0.028, 0.063
2007/1007950	<0.01, 0.017, 0.018, 0.019	0.028, 0.039, 0.045, 0.050
OECD-MRL-calculation	**0.05 (n=8, STMR=0.016, HR=0.026)**	**<u>0.15</u> (n=8, STMR=0.034, HR=0.063)**

_ underlined values were used for risk assessment purposes

2.19 Green peas without pods

Residue data from supervised trials in green peas without pods were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.19-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Green peas without pods (field)	2005	4	DE, FR, NL, UK	2	GR, IT	6	2006/1026856
Green peas without pods (field)	2006	4	DE, DK, FR, UK	2	ES, FR	6	2007/1007951
Total number of trials per region		**8**		**4**	**Total number of trials**	**12**	

2.19.1 Supervised residue trials in green peas without pods

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of alpha-cypermethrin in green peas (field) after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026856

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alpha-cypermethrin in fresh green peas (field) after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007951

Material and Methods:

In the years 2005 and 2006 a residue program on peas was conducted in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom under open field conditions.

During the 2005 growing season, six field trials were conducted in fresh peas in France (Northern European region), Germany, Greece, Italy, the Netherlands and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 76 (60% of pods have reached typical length; juice exudes if pressed. Tenderometer value: 115 TE) and 79 (Pods have reached typical size (green ripe); peas fully formed).
In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea plants once at a target rate of 25 g a.s./ha at growth stages between 74 (40% of pods have reached typical length; juice exudes if pressed. Tenderometer value: 95 TE) and 79.
Specimens of peas and remaining plant were collected immediately after the last application from each plot, as well as approximately 3, 7 and 14 days thereafter.
Specimens were analysed using BASF analytical method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, six field trials were conducted in fresh peas in Denmark, France (Southern European region), Germany, Spain and the United Kingdom.
In four trials, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 67 (flowering declining) and 79 (pods have reached typical size (green ripe); peas fully formed).

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea plants once at a target rate of 25 g a.s./ha.
Specimens of peas and remaining plant were collected immediately after the last application from each plot, as well as 2-4, 7 and 14 days thereafter.
Specimens were analysed using BASF analytical method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.19.1-1.

Table 2.19.1-1: Residues in green peas without pods

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 1-3 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/2 Study to GLP Study carried out in 2005	Fresh peas	France Le Moulin Avent 49680 Vivy (North of EU)	12.5 g a.s./ha 20.08.05	77	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.070 peas <0.01 remaining plant 0.050 peas <0.01 remaining plant 0.064 peas <0.01 remaining plant 0.048	
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/3 Study to GLP Study carried out in 2005	Fresh peas	Germany 67126 Hochdorf-Assenheim	12.5 g a.s./ha 21.06.05	76	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.056 peas <0.01 remaining plant 0.038 peas <0.01 remaining plant 0.023 peas <0.01 remaining plant 0.012	BASF analytical method N 567/0 peas: mean recovery = 78.7%; SD: +/- 8.0; CV: 10.1; n=4; fortification range 0.01-0.1 mg/kg remaining plant: mean recovery = 81.8%; SD: 11.2 +/-; CV: 13.7; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/4 Study to GLP Study carried out in 2005	Fresh peas	The Netherlands 6598 Heijen, Limburg	12.5 g a.s./ha 03.08.05	78-79	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.825 peas <0.01 remaining plant 0.245 peas <0.01 remaining plant 0.254 peas <0.01 remaining plant 0.191	

Table 2.19.1-1: Residues in green peas without pods

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 1-3 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/7 Study to GLP Study carried out in 2005	Fresh peas	United Kingdom Bishop Burton, Yorkshire HU17 8QA	12.5 g a.s./ha 27.07.05	76-77	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.353 peas <0.01 remaining plant 0.060 peas <0.01 remaining plant 0.099 peas <0.01 remaining plant 0.094	
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/1 Study to GLP Study carried out in 2006	Field pea (green) (variety Merveille de Kelvedon)	France Clery St Audre 46370 (South of EU)	12.5 g a.s./ha 19.06.06	77-79	0 0 3 3 7 7 14 14	peas without pods <0.01 remaining plant 0.159 peas without pods <0.01 remaining plant 0.037 peas without pods <0.01 remaining plant 0.047 peas without pods <0.01 remaining plant 0.033	BASF analytical method N 567/0 peas: mean recovery = 79.9%; SD: +/- 21.9; CV: 27.4; n=6; fortification range 0.01-1.0 mg/kg remaining plant: mean recovery = 83.3%; SD: 8.7 +/-; CV: 10.4; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/2 Study to GLP Study carried out in 2006	Field pea (green) (variety Progress No. 9)	Denmark Rojleskovvej 18 DK-5500 Middelfart	12.5 g a.s./ha 12.07.06	77	0 0 2 2 7 7 14 14	peas without pods <0.01 remaining plant 0.514 peas without pods <0.01 remaining plant 0.361 peas without pods <0.01 remaining plant 0.226 peas without pods <0.01 remaining plant 0.281	BASF analytical method N 567/0 peas: mean recovery = 79.9%; SD: +/- 21.9; CV: 27.4; n=6; fortification range 0.01-1.0 mg/kg remaining plant: mean recovery = 83.3%; SD: 8.7 +/-; CV: 10.4; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/3 Study to GLP Study carried out in 2006	Field pea (green) (variety Geisha)	United Kingdom Holbeach Hurn Lincolnshire PE12 8LR	12.5 g a.s./ha 31.07.06	77	0 0 3 3 7 7 14 14	peas without pods <0.01 remaining plant 0.254 peas without pods <0.01 remaining plant 0.087 peas without pods <0.01 remaining plant 0.062 peas without pods <0.01 remaining plant 0.066	

Table 2.19.1-1: Residues in green peas without pods

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 1-3 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/4 Study to GLP Study carried out in 2006	Field pea (green) (variety Santana)	Germany Vor den Hofen 31303 Burgdorf-Hulptingsen Lower Saxony	12.5 g a.s./ha 26.10.06	67	0 0 4 4 7 7 14 14	peas without pods <0.01 remaining plant 0.289 peas without pods <0.01 remaining plant 0.266 peas without pods <0.01 remaining plant 0.185 peas without pods <0.01 remaining plant 0.180	BASF analytical method N 567/0 peas: mean recovery = 79.9%; SD: +/- 21.9; CV: 27.4; n=6; fortification range 0.01-1.0 mg/kg remaining plant: mean recovery = 83.3%; SD: 8.7 +/-; CV: 10.4; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/2 Study to GLP Study carried out in 2005	Fresh peas	France Le Moulin Avent 49680 Vivy	12.5 g a.s./ha 2 treatm. last date 20.08.05	77 at last treatm.	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.104 peas <u><0.01</u> remaining plant 0.063 peas <0.01 remaining plant 0.076 peas <0.01 remaining plant 0.054	BASF analytical method N 567/0 peas: mean recovery = 78.7%; SD: +/- 8.0; CV: 10.1; n=4; fortification range 0.01-0.1 mg/kg remaining plant: mean recovery = 81.8%; SD: 11.2 +/-; CV: 13.7; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/3 Study to GLP Study carried out in 2005	Fresh peas	Germany 67126 Hochdorf-Assenheim	12.5 g a.s./ha 2 treatm. last date 21.06.05	76 at last treatm.	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.068 peas <u><0.01</u> remaining plant 0.057 peas <0.01 remaining plant 0.028 peas <0.01 remaining plant 0.010	BASF analytical method N 567/0 peas: mean recovery = 78.7%; SD: +/- 8.0; CV: 10.1; n=4; fortification range 0.01-0.1 mg/kg remaining plant: mean recovery = 81.8%; SD: 11.2 +/-; CV: 13.7; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/4 Study to GLP Study carried out in 2005	Fresh peas	The Netherlands 6598 Heijen, Limburg	12.5 g a.s./ha 2 treatm. last date 03.08.05	78-79 at last treatm.	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.859 peas <u><0.01</u> remaining plant 0.354 peas <0.01 remaining plant 0.233 peas <0.01 remaining plant 0.345	

Table 2.19.1-1: Residues in green peas without pods

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 1-3 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/7 Study to GLP Study carried out in 2005	Fresh peas	United Kingdom Bishop Burton, Yorkshire HU17 8QA	12.5 g a.s./ha 2 treatm. last date 27.07.05	76-77 at last treatm.	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.420 peas <u>≤0.01</u> remaining plant 0.213 peas <0.01 remaining plant 0.301 peas <0.01 remaining plant 0.292	BASF analytical method N 567/0 peas: mean recovery = 78.7%; SD: +/- 8.0; CV: 10.1; n=4; fortification range 0.01-0.1 mg/kg remaining plant: mean recovery = 81.8%; SD: 11.2 +/-; CV: 13.7; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/1 Study to GLP Study carried out in 2006	Field pea (green) (variety Merveille de Kelvedon)	France Clery St Audre 46370 (North of EU)	12.5 g a.s./ha 2 treatm. last date 19.06.06	77-79 at last treatm.	0 0 3 3 7 7 14 14	peas without pods <0.01 remaining plant 0.225 peas without pods <u>≤0.01</u> remaining plant 0.057 peas without pods <0.01 remaining plant 0.104 peas without pods <0.01 remaining plant 0.068	BASF analytical method N 567/0 peas: mean recovery = 79.9%; SD: +/- 21.9; CV: 27.4; n=6; fortification range 0.01-1.0 mg/kg remaining plant: mean recovery = 83.3%; SD: 8.7 +/-; CV: 10.4; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/2 Study to GLP Study carried out in 2006	Field pea (green) (variety Progress No. 9)	Denmark Rojleskovvej 18 DK-5500 Middelfart	12.5 g a.s./ha 2 treatm. last date 12.07.06	77 at last treatm	0 0 2 2 7 7 14 14	peas without pods <0.01 remaining plant 0.333 peas without pods <u>≤0.01</u> remaining plant 0.430 peas without pods <0.01 remaining plant 0.200 peas without pods <0.01 remaining plant 0.166	BASF analytical method N 567/0 peas: mean recovery = 79.9%; SD: +/- 21.9; CV: 27.4; n=6; fortification range 0.01-1.0 mg/kg remaining plant: mean recovery = 83.3%; SD: 8.7 +/-; CV: 10.4; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/3 Study to GLP Study carried out in 2006	Field pea (green) (variety Geisha)	United Kingdom Holbeach Hurn Lincolnshire PE12 8LR	12.5 g a.s./ha 2 treatm. last date 31.07.06	77 at last treatm	0 0 3 3 7 7 14 14	peas without pods <0.01 remaining plant 0.275 peas without pods <u>≤0.01</u> remaining plant 0.190 peas without pods <0.01 remaining plant 0.199 peas without pods <0.01 remaining plant 0.226	

Table 2.19.1-1: Residues in green peas without pods

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 1-3 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/4 Study to GLP Study carried out in 2006	Field pea (green) (variety Santana)	Germany Vor den Hofen 31303 Burgdorf-Hulptingsen Lower Saxony	12.5 g a.s./ha 2 treatm. last date 26.10.06	71 at last treatm	0 0 4 4 7 7 14 14	peas without pods <0.01 remaining plant 0.635 peas without pods <u><0.01</u> remaining plant 0.230 peas without pods <0.01 remaining plant 0.346 peas without pods <0.01 remaining plant 0.259	
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/5 Study to GLP Study carried out in 2005	Fresh peas	Italy S. Pietro in Casole, 40018 Bologna	25 g a.s./ha 20.06.05	77-79	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.243 peas <u><0.01</u> remaining plant 0.267 peas <0.01 remaining plant 0.091 peas <0.01 remaining plant 0.105	BASF analytical method N 567/0 peas: mean recovery = 78.7%; SD: +/- 8.0; CV: 10.1; n=4; fortification range 0.01-0.1 mg/kg remaining plant: mean recovery = 81.8%; SD: 11.2 +/-; CV: 13.7; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026856 Trial No. AF/8826/BA/6 Study to GLP Study carried out in 2005	Fresh peas	Greece Komara, Evros, Thrace, GR-68200	25 g a.s./ha 24.06.05	74	0 0 3 3 7 7 14 14	peas <0.01 remaining plant 0.568 peas <u><0.01</u> remaining plant 0.201 peas <0.01 remaining plant 0.282 peas <0.01 remaining plant 0.259	BASF analytical method N 567/0 peas: mean recovery = 78.7%; SD: +/- 8.0; CV: 10.1; n=4; fortification range 0.01-0.1 mg/kg remaining plant: mean recovery = 81.8%; SD: 11.2 +/-; CV: 13.7; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/5 Study to GLP Study carried out in 2006	Field pea (green) (variety Milan)	France St Nicholas de la G 82210 (South of EU)	25 g a.s./ha 22.05.06	69-71	0 0 2 2 7 7 14 14	peas without pods <0.01 remaining plant 0.721 peas without pods <u><0.01</u> remaining plant 0.165 peas without pods <0.01 remaining plant 0.068 peas without pods <0.01 remaining plant 0.140	BASF analytical method N 567/0 peas: mean recovery = 79.9%; SD: +/- 21.9; CV: 27.4; n=6; fortification range 0.01-1.0 mg/kg remaining plant: mean recovery = 83.3%; SD: 8.7 +/-; CV: 10.4; n=8; fortification range 0.01-2.0 mg/kg Residue analysed as total cypermethrin

Table 2.19.1-1: Residues in green peas without pods

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 1-3 days (outdoor)
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days (outdoor)

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007951 Trial No. AF/10491/BA/6 Study to GLP Study carried out in 2006	Field pea (green)	Spain Zaragoza	25 g a.s./ha		0 0 3 3 7 7 14 14	peas without pods <0.01 remaining plant 1.023 peas without pods <u><0.01</u> remaining plant 0.431 peas without pods <0.01 remaining plant 0.297 peas without pods <0.01 remaining plant 0.071	

_ underlined values were used for MRL calculation

Findings:

The residue studies in peas presented in the alpha-cypermethrin dossier were carried out in 6 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 1-3 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-8 trials including both variants, 7 conducted in the Northern European region and one conducted in the Southern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions-4 trials conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues in pea plant material ranged between 0.056 mg/kg and 0.825 mg/kg. Residues declined to levels between 0.037-0.361 mg/kg, 0.023–0.254 mg/kg and 0.012-0.281 mg/kg at 2-4, 7 and 14 days after application. Treated pea samples were free of residues above the limit of quantitation of the analytical method at all sampling intervals (all <0.01 mg/kg).

After two applications at 12.5 g a.s./ha, initial residues in pea plant material ranged between 0.068 mg/kg and 0.859 mg/kg. Residues declined to levels between 0.057-0.430 mg/kg, 0.028-0.346 mg/kg and 0.010-0.345 mg/kg at 2-4, 7 and 14 days after application. Treated pea samples were free of residues above the limit of quantitation of the analytical method at all sampling intervals (all <0.01 mg/kg).
After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in pea plant material ranged between 0.243 mg/kg and 1.023 mg/kg. Residues declined to levels between 0.165-0.431 mg/kg, 0.068-0.297 mg/kg and 0.071-0.259 mg/kg at 2-3, 7 and 14 days after application. Treated pea samples were free of residues above the limit of quantitation of the analytical method at all sampling intervals (all <0.01 mg/kg).

Conclusion:

In the years 2005 and 2006 a residue program on peas was conducted in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom under open field conditions.
In 8 trials supporting the proposed field GAP for the Northern European region, residues in pea plant material ranged between 0.068 mg/kg and 0.859 mg/kg at the target PHI of 0 days and between 0.057-0.430 mg/kg at the target PHI of 3±1 days. Treated pea samples were free of residues above the limit of quantitation of the analytical method at all sampling intervals (all <0.01 mg/kg).
In 4 trials supporting the proposed field GAP for the Southern European region, residues in pea plant material ranged between 0.165-0.431 mg/kg at the target PHI of 3±1 days. Treated pea samples were free of residues above the limit of quantitation of the analytical method at all sampling intervals (all <0.01 mg/kg).

2.19.2 Estimation of MRL, HR and STMR for green peas without pods

For *green peas without pods*, the following residue studies were considered (BASF DocIDs): 2006/1026856 and 2007/1007951.

The following residue values (PHI=3±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026856	<0.01 (4x)	<0.01 (2x)
2007/1007951	<0.01 (4x)	<0.01 (2x)
OECD-MRL-calculation	**0.01 (n=8, STMR=0.01, HR=0.01)**	**0.01 (n=4, STMR=0.01, HR=0.01)**

_ underlined values were used for risk assessment purposes

2.20 Asparagus

The Good Agricultural Practice (GAP) for use of BAS 310 55 I on asparagus is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at growth stage BBCH 11-93 after the harvest period of the sprouts has ended (PHI F)

The green asparagus plant parts that are treated with BAS 310 55 I do not develop before the harvest of the subterraneous sprouts has ended (BBCH 11-93). Therefore, there is no risk of residues occurring in the asparagus stems that are consumed and a MRL at the limit of quantitation of the analytical method (0.01 mg/kg) is considered appropriate.

2.21 Artichoke

Residue data from supervised trials in artichoke were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.21-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Artichoke (field)	2005	-	-	2	ES, IT	2	2006/1026845
Artichoke (field)	2006	-	-	2	FR, GR	2	2007/1007948
Total number of trials per region				**4**	**Total number of trials**	**4**	

2.21.1 Supervised residue trials in artichoke

Jones G (2006)

Full study reference

Jones G (2006): Study on the residue behaviour of alpha-cypermethrin in artichoke after treatment with BAS 310 40 I under field conditions in Southern Europe, 2005; BASF DocID 2006/1026845

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alpha-cypermethrin in Artichoke (field) after treatment with BAS 310 40 I under field conditions in Southern Europe during 2006; BASF DocID 2007/1007948

Material and Methods:

In the years 2005 and 2006 a residue program on artichokes was conducted in the field in France (Southern European region), Greece, Italy and Spain under open field conditions.
Alpha-cypermethrin, formulated as an emulsifiable concentrate (EC 100 g a.s./L; BAS 310 40 I) was foliar applied to artichokes plants once at a target rate of 25 g a.s./ha.
The growth stage of the plants at application was between 47 (harvestable vegetative plant parts have reached 70% of final size) and 51 (inflorescence or flower buds visible).
Artichoke flower heads were collected 0, 3, 7 and 14 days after application.
Residue analysis was performed according to BASF analytical method No. 567/0 which determines total cypermethrin residues by means of HPLC-MS/MS with a LOQ of 0.01 mg/kg.
The trial data and residue results are summarized inTable 2.21.1-1.

Table 2.21.1-1: Residues in artichokes

GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026845 Trial No. AF/8828/BA/1 Study to GLP Study carried out in 2005	Artichoke (variety Chata de Tudela)	Spain Finca la Hincosa, Camino Bollulus, Bollulus del Candado	25 g a.s./ha 13.12.05	51	0 3 7 14	flower heads 0.038 flower heads 0.019 flower heads <u>0.021</u> flower heads 0.012	BASF analytical method No. 567/0 foliage: mean recovery = 91.9 %; SD: +/- 15.3; CV: 16.6; n=3; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026845 Trial No. AF/8828/BA/2 Study to GLP Study carried out in 2005	Artichoke (variety Violetto Toscano)	Italy Ag Agr Pistolesi, Podere Pappasous, Via Aurelia, Toscana	25 g a.s./ha 29.04.05	47	0 3 7 14	flower heads 0.060 flower heads 0.060 flower heads <u>0.040</u> flower heads 0.014	
BASF Doc ID 2007/1007948 Trial No. AF/10494/BA/1 Study to GLP Study carried out in 2006	Artichoke (variety Macau)	France Darles, St. Caprais, Garosses, 31330 (South of EU)	25 g a.s./ha 11.05.06	47	0 3 7 14	flower heads 0.035 flower heads 0.026 flower heads <u>0.019</u> flower heads 0.015	BASF analytical method No. 567/0 flower heads: mean recovery = 77% (66.7-86.8%); SD: +/- n/a; CV: n/a; n=2; fortification range 0.01-0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007948 Trial No. AF/10494/BA/2 Study to GLP Study carried out in 2006	Artichoke (variety Wild Artichoke)	Greece Kato Souli Attiki Central Greece	25 g a.s./ha 05.05.06	47-49	0 3 7 14	flower heads 0.039 flower heads 0.042 flower heads <u>0.025</u> flower heads <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in artichoke presented in the alpha-cypermethrin dossier were carried out in 4 different EU countries and provide data relevant to conditions in the Southern European region.
The proposed GAP is

- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days

The following residue studies were presented:

- one treatment at a target rate of 25 g a.s./ha under open field conditions-4 trials conducted in the Southern European region

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in artichoke flower heads ranged between 0.035 mg/kg and 0.060 mg/kg. Residues declined to levels between 0.019–0.060 mg/kg, 0.019-0.040 mg/kg and <0.01-0.015 mg/kg at 3, 7 and 14 days after application.

Conclusion:

In the years 2005 and 2006 a residue program on artichokes was conducted in the field in France (Southern European region), Greece, Italy and Spain under open field conditions.
In 4 trials supporting the GAP for the Southern European region, residues in artichoke flower heads ranged between 0.019-0.040 mg/kg at the target PHI of 7 days.

2.21.2 Estimation of MRL, HR and STMR for artichoke

For *artichoke*, the following residue studies were considered (BASF DocIDs): 2006/1026845 and 2007/1007948.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026845	-	0.021, 0.040
2007/1007948	-	0.019, 0.025
OECD-MRL-calculation		**<u>0.08</u> (n=4, STMR=0.023, HR=0.04)**

_ underlined values were used for risk assessment purposes

2.22 Celery

Residue data from supervised trials in celery were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.22-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Celery (field)	2005	2	DE	-	-	2	2006/1029533
Celery (field)	2006	3	DE	-	-	3	2007/1035745
Total number of trials per region		**5**			**Total number of trials**	**5**	

2.22.1 Supervised residue trials in celery

Report:	**Anonymous, 2006a**
Title:	Residue behaviour of Alpha-Cypermethrin in/on fennel, curly cale, cucumbers, carrots, radies, celery, celeriac and bunshing or green onions after outdoor application of Fastac SC (SC 100) in Germany, 2005
Document No:	BASF DocID 2006/1029533
Guidelines:	EEC 91/414 Annex III (Part A Section 8), EEC 91/414 Annex II (Part A Section 6), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 1607/VI/97 rev. 2 10.06.1999
GLP	No, not subject to GLP regulations

Executive Summary

During the 2005 growing season two open field trials were conducted in celery in Germany to determine the magnitude of alpha-cypermethrin residues in celery after application of the formulation Fastac SC (100 g a.s./L, SC). Each field trial consisted of two plots: one control plot and one plot treated with alpha-cypermethrin. Fastac SC was applied twice at a single rate equivalent to 0.012-0.013 kg a.s./ha. The applications were made targeting 14±1 days and 7±1 days before harvest. The treated celery specimens (stalks) were taken immediately after the last application (0 DALA) and 7, 10 and 14 days thereafter.
Alpha-cypermethrin was analysed by Method No. SAA/C/PSM 023, an LC-MS/MS method with an LOQ of 0.005 mg/kg. At 7 days after the last application, alpha-cypermethrin residues were 0.1443-0.1822 mg/kg.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: Fastac SC
Lot/Batch #: Fastac SC, alpha-cypermethrin: 100 g a.s./L
Purity:
CAS#: 67375-30-8
Development code:
Spiking levels: Not reported

2. Test Commodity:

Crop: Celery
Type: Stalk vegetable
Variety: Tango
Botanical name: *Apium graveolens*
Crop parts(s) or processed Commodity: Stalks
Sample size: >1.1-1.9 kg

B. STUDY DESIGN

1. Test procedure

During the 2005 growing season two open field trials were conducted in celery in Germany to determine the magnitude of alpha-cypermethrin residues in celery after application of the formulation Fastac SC (100 g a.s./L, SC).
Each field trial consisted of two plots: one control plot and one plot treated with alpha-cypermethrin. Fastac SC was applied twice at a single rate equivalent to 0.012-0.013 kg a.s./ha. The applications were made targeting 14±1 days and 7±1 days before harvest.
0 and 7 DALA (days after last application) sampling in the untreated plot was performed. The treated celery specimens (stalks) were taken immediately after the last application (0 DALA) and 7, 10 and 14 days thereafter. Samples were stored frozen until analysis.

Table 2.22.1-1: Target application rates and timings for celery

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (g a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2005	2	2	F	Fastac SC	alpha-cypermethrin	12-13	590-640	7±1, 14±1 days before harvest

2. Description of analytical procedures

Alpha-cypermethrin was analysed by Method No. SAA/C/PSM 023. Samples were extracted with methanol/water/HCl. The final determination of alpha-cypermethrin was performed by LC-MS/MS with an LOQ of 0.005 mg/kg.

Table 2.22.1-2: Summary of recoveries for alpha-cypermethrin in celery

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
SAA/C/PSM 023		alpha-cypermethrin (BAS 310 I)		
celery	n r.	n r.	92-97	n.r.

n r. not reported

II. RESULTS AND DISCUSSION

Directly after the last application (0 DALA) of Fastac SC, alpha-cypermethrin residues were 0.2120-0.3381 mg/kg. At 7 days after the last application, residues were 0.1443-0.1822 mg/kg. They declined to 0.0658-0.1045 mg/kg at 14 DALA. No residues were found in the untreated specimens.
An overall summary of the residues is given in the table below.

Table 2.22.1-3: Summary of residues in celery after application of Fastac SC

Crop	Year	No. of Appl.	Application	DALA[1)]	BBCH[2)]	Range of Residues (mg/kg)	
						Matrix	Alpha-Cypermethrin
Celery	2005	2	Fastac SC (SC)	0	n r.	stalk	0.2120-0.3381
				7	ripe for harvesting	stalk	0.1443-0.1822
				10		stalk	0.1212-0.1360
				14		stalk	0.0658-0.1045

1) DALA Days after last application
2) BBCH stage at respective sampling

III. CONCLUSION

At 7 days after the last application, alpha-cypermethrin residues were 0.1443-0.1822 mg/kg.

Table 2.22.1-4: Residues of alpha-cypermethrin after application of the formulation Fastac SC in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin
Study code: -	Celery	Germany	2 x Fastac SC 0.012-0.013	n r.	0	stalk	0.2120
Doc ID: 2006/1029533					7	stalk	<u>0.1443</u>
Trial No.: RU-I-1605 RPNW 2/1					10	stalk	0.1360
GLP: yes (analytical part)					14	stalk	0.0658
Year 2005							
Study code: -	Celery	Germany	2 x Fastac SC 0.013	n r.	0	stalk	0.3381
Doc ID: 2006/1029533					7	stalk	<u>0.1822</u>
Trial No.: RU-I-1605 RPNW 2/2					10	stalk	0.1212
GLP: yes (analytical part)					14	stalk	0.1045
Year 2005							

DALA = Days after last application
_ underlined values were used for MRL calculation
n r. not reported

Report:	**Anonymous, 2007b**
Title:	Residue behaviour of Alpha-Cypermethrin in/on fennel (bulbs formed by leafs stems), curly kale (leaves), blanched celery (stems) and spinach (leaves) outdoor after application of Fastac SC Super Contact (SC, 100 g/L) in Germany, 2006
Document No:	BASF DocID 2007/1035745
Guidelines:	EEC 91/414 Annex III (Part A Section 8), EEC 91/414 Annex II (Part A Section 6), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 1607/VI/97 rev. 2 10.06.1999
GLP	Yes

Executive Summary

During the 2006 growing season three open field trials were conducted in celery in Germany to determine the magnitude of alpha-cypermethrin residues in celery after application of the formulation Fastac SC (100 g a.s./L, SC). Each field trial consisted of two plots: one control plot and one plot treated with alpha-cypermethrin. Fastac SC was applied twice at a single rate equivalent to 0.0125-0.014 kg a.s./ha. The applications were made targeting 14±1 days and 7±1 days before harvest. The treated celery specimens (stalks) were taken at 7 and 14 DALA.
Alpha-cypermethrin was analysed by a modification of Method No. DFG S-19, an GC-ECD method with an LOQ of 0.01 mg/kg. At 7 days after the last application, alpha-cypermethrin residues were 0.22-0.30 mg/kg.

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:**
   Description: Fastac SC
   Lot/Batch #: Fastac SC, alpha-cypermethrin: 100 g a.s./L
   Purity:
   CAS#: 67375-30-8
   Development code:
   Spiking levels: Not reported

2. **Test Commodity:**
   Crop: Celery
   Type: Stalk vegetable
   Variety: Tango, Utah
   Botanical name: *Apium graveolens*
   Crop parts(s) or processed Commodity: Stalks
   Sample size: ≥2.0 kg / 12 units

B. STUDY DESIGN

1. Test procedure

During the 2006 growing season three open field trials were conducted in celery in Germany to determine the magnitude of alpha-cypermethrin residues in celery after application of the formulation Fastac SC (100 g a.s./L, SC).
Each field trial consisted of two plots: one control plot and one plot treated with alpha-cypermethrin. Fastac SC was applied twice at a single rate equivalent to 0.0125-0.014 kg a.s./ha. The applications were made targeting 14±1 days and 7±1 days before harvest.
7 DALA (days after last application) sampling in the untreated plot was performed. The treated celery specimens (stalks) were taken at 7 and 14 DALA. Samples were stored frozen until analysis.

Table 2.22.1-5: Target application rates and timings for celery

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (g a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2006	3	2	F	Fastac SC	alpha-cypermethrin	12.5-14	424-640	7±1, 14±1 days before harvest

2. Description of analytical procedures

Alpha-cypermethrin was analysed by a modification of Method No. DFG S-19. Samples were extracted with acetone/water and purified with a silicagel column. The final determination of alpha-cypermethrin was performed by GC-ECD with an LOQ of 0.01 mg/kg.

Table 2.22.1-6: Summary of recoveries for alpha-cypermethrin in celery

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
DFG S-19		alpha-cypermethrin (BAS 310 I)		
celery	0.01-0.5	n r.	97	n.r.

n r. not reported

II. RESULTS AND DISCUSSION

At 7 days after the last application, alpha-cypermethrin residues were 0.22-0.30 mg/kg. They declined to 0.11-0.17 mg/kg at 14 DALA. No residues were found in the untreated specimens.
An overall summary of the residues is given in the table below.

Table 2.22.1-7: Summary of residues in celery after application of Fastac SC

Crop	Year	No. of Appl.	Application	DALA[1)]	BBCH[2)]	Range of Residues (mg/kg)	
						Matrix	Alpha-Cypermethrin
Celery	2006	2	Fastac SC (SC)	7 14	ripe for harvesting / 49	stalk stalk	0.22-0.30 0.11-0.17

1) DALA Days after last application
2) BBCH stage at respective sampling

III. CONCLUSION

At 7 days after the last application, alpha-cypermethrin residues were 0.22-0.30 mg/kg.

Table 2.22.1-8: Residues of alpha-cypermethrin after application of the formulation Fastac SC in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin
Study code: - **Doc ID:** 2007/1035745 **Trial No.:** RU-I-1706 RPNW 1/1 **GLP:** yes (analytical part) **Year** 2006	Celery	Germany	2 x Fastac SC 0.013-0.014	n r.	7 14	stalk stalk	<u>0.30</u> 0.17
Study code: - **Doc ID:** 2007/1035745 **Trial No.:** RU-I-1706 RPNW 1/2 **GLP:** yes (analytical part) **Year** 2006	Celery	Germany	2 x Fastac SC 0.012-0.013	n r.	7 14	stalk stalk	<u>0.23</u> 0.13
Study code: - **Doc ID:** 2007/1035745 **Trial No.:** RU-I-1706 NWBN 1/1 **GLP:** yes (analytical part) **Year** 2006	Celery	Germany	2 x Fastac SC 0.013	48	7 14	stalk stalk	<u>0.22</u> 0.11

DALA = Days after last application
_ underlined values were used for MRL calculation
n r. not reported

2.22.2 Estimation of MRL, HR and STMR for celery

For *celery*, the following residue studies were considered (BASF DocIDs): 2006/1029533 and 2007/1035745.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1029533	0.1443, 0.1822	-
2007/1035745	0.22,0.23,0.30	-
OECD-MRL-calculation	**0.7 (n=5, STMR=0.22, HR=0.30)**	**-**

_ underlined values were used for risk assessment purposes

2.23 Leek

Residue data from supervised trials in leek were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.23-1: Number of residue trials conducted in leek per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Leek (field)	2005	4	DE, FR, NL, UK	2	FR, IT	6	2006/1026850
Leek (field)	2006	4	BE, DE, FR, UK	2	ES, FR	6	2007/1008498
Total number of trials per region					**Total number of trials**		

2.23.1 Supervised residue trials in leek

Evans L (2007)

Full study reference

Evans L (2007): Study on the residue behaviour of alpha-cypermethrin in Leek after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026850

Diehl M (2007)

Full study reference

Diehl M (2007): Study on the Residue Behaviour of BAS 310 I in Leek after Treatment with BAS 310 40 I under open field Conditions in Southern and Northern Europe, 2006; BASF DocID 2007/1008498

Material and Methods:

In the years 2005 and 2006 a residue program on leek was conducted in Belgium, France (Northern and Southern European region), Germany, Italy, Spain, the Netherlands and the United Kingdom under open field conditions.

During the 2005 growing season, six field trials were conducted in leek in France (Northern and Southern European region), Germany, Italy, the Netherlands and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to leek plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 42 and 49 (growth complete; length and stem diameter typical for variety reached).

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to leek plants once at a target rate of 25 g a.s./ha at growth stages between 41 (leaf bases begin to thicken or extend) and 47 (70% of the expected shaft length and diameter reached).
Specimens of leek plants without roots were collected immediately after the last application from each plot, as well as approximately 3, 7 and 14 days thereafter.
Specimens were analysed using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 2006 growing season, six field trials were conducted in leek in Belgium, France (Northern and Southern European region), Germany, Spain, and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to leek plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 44 and 48. In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to leek plants once at a target rate of 25 g a.s./ha at growth stages between 45 (50% of the expected shaft diameter reached) and 47 (70% of the expected shaft length and diameter reached).
Specimens of leek plants without roots were collected immediately after the last application from each plot, as well as 2-4, 7 and 13-15 days thereafter.
Specimens were analysed using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.23.1-1.

Table 2.23.1-1: Residues in leek

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/1 Study to GLP Study carried out in 2005	Leek	United Kingdom Ravenshead Nottinghamshire NG5 8PB	12.5 g a.s./ha 27.09.05	43	0 3 7 14	plant without root 0.183 plant without root 0.078 plant without root 0.047 plant without root 0.055	BASF method N 567/0 plant without root: mean recovery = 92.8%; SD: +/- 8.9; CV: 9.6; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/2 Study to GLP Study carried out in 2005	Leek	France 45730 Dan Pierre en Burly (North of EU)	12.5 g a.s./ha 20.09.05	47	0 3 7 14	plant without root 0.059 plant without root 0.052 plant without root 0.037 plant without root 0.017	
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/3 Study to GLP Study carried out in 2005	Leek	Germany 67125 Dannstadt	12.5 g a.s./ha 19.04.05	49	0 3 7 14	plant without root 0.056 plant without root 0.055 plant without root 0.046 plant without root 0.033	
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/4 Study to GLP Study carried out in 2005	Leek	The Netherlands 5375 KR Reek Brabant	12.5 g a.s./ha 23.09.05	42	0 3 7 14	plant without root 0.026 plant without root 0.026 plant without root 0.014 plant without root 0.014	
BASF Doc ID 2007/1008498 Trial No. A/NF/I/06/155 Study to GLP Study carried out in 2006	Leek (variety Pasteur)	France Mardeuil Marne (North of EU)	13 g a.s./ha 31.08.06	45	0 3 7 13	plant without root 0.026 plant without root 0.024 plant without root 0.016 plant without root <0.01	BASF method N 567/0 plant: mean recovery = 97.9%; SD: +/- 7.5; CV: 7.6; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008498 Trial No. A/BE/I/06/157 Study to GLP Study carried out in 2006	Leek (variety Ashton)	Belgium Sint-Kateljine-Waver Antwerpen	12 g a.s./ha 25.09.06	44-45	0 3 7 14	plant without root 0.021 plant without root 0.029 plant without root 0.020 plant without root 0.015	
BASF Doc ID 2007/1008498 Trial No. A/GE/I/06/158 Study to GLP Study carried out in 2006	Leek (variety Kenton)	Germany Erfurt (Thüringen)	13 g a.s./ha 30.10.06	48	0 2 7 14	plant without root 0.018 plant without root 0.025 plant without root 0.023 plant without root 0.017	

Table 2.23.1-1: Residues in leek

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008498 Trial No. A/UK/I/06/156 Study to GLP Study carried out in 2006	Leek (variety Pancho)	United Kingdom Pershore Worcestershire	13 g a.s./ha 11.09.06	45	0 4 7 15	plant without root 0.063 plant without root 0.052 plant without root 0.037 plant without root <0.01	BASF method N 567/0 plant: mean recovery = 97.9%; SD: +/- 7.5; CV: 7.6; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/1 Study to GLP Study carried out in 2005	Leek	United Kingdom Ravenshead Nottinghamshire NG5 8PB	12.5 g a.s./ha 2 treatm. last date 04.10.05	43 at last treatm.	0 3 7 14	plant without root 0.144 plant without root 0.082 plant without root 0.056 plant without root 0.058	BASF method N 567/0 plant without root: mean recovery = 92.8%; SD: +/- 8.9; CV: 9.6; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/2 Study to GLP Study carried out in 2005	Leek	France 45730 Dan Pierre en Burly (North of EU)	12.5 g a.s./ha 2 treatm. last date 27.09.05	47-49 at last treatm.	0 3 7 14	plant without root 0.083 plant without root 0.070 plant without root 0.053 plant without root 0.034	
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/3 Study to GLP Study carried out in 2005	Leek	Germany 67125 Dannstadt	12.5 g a.s./ha 2 treatm. last date 26.04.05	49 at last treatm.	0 3 7 14	plant without root 0.084 plant without root 0.069 plant without root 0.071 plant without root 0.064	
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/4 Study to GLP Study carried out in 2005	Leek	The Netherlands 5375 KR Reek Brabant	12.5 g a.s./ha 2 treatm. last date 04.10.05	44 at last treatm.	0 3 7 14	plant without root 0.055 plant without root 0.031 plant without root 0.033 plant without root 0.021	
BASF Doc ID 2007/1008498 Trial No. A/NF/I/06/155 Study to GLP Study carried out in 2006	Leek (variety Pasteur)	France Mardeuil Marne (North of EU)	13 g a.s./ha 2 treatm. last date 31.08.06	45 at last treatment	0 3 7 13	plant without root 0.032 plant without root 0.029 plant without root 0.018 plant without root 0.010	BASF method N 567/0 plant: mean recovery = 97.9%; SD: +/- 7.5; CV: 7.6; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008498 Trial No. A/BE/I/06/157 Study to GLP Study carried out in 2006	Leek (variety Ashton)	Belgium Sint-Kateljine-Waver Antwerpen	12 g a.s./ha 2 treatm. last date 25.09.06	44-45 at last treatment	0 3 7 14	plant without root 0.048 plant without root 0.038 plant without root 0.039 plant without root 0.026	

Table 2.23.1-1: Residues in leek

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 2 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1008498 Trial No. A/GE/I/06/158 Study to GLP Study carried out in 2006	Leek (variety Kenton)	Germany Erfurt (Thüringen)	14 /13 g a.s./ha 2 treatm. last date 30.10.06	48 at last treatment	0 2 7 14	plant without root 0.042 plant without root 0.036 plant without root <u>0.032</u> plant without root 0.032	BASF method N 567/0 plant: mean recovery = 97.9%; SD: +/- 7.5; CV: 7.6; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008498 Trial No. A/UK/I/06/156 Study to GLP Study carried out in 2006	Leek (variety Pancho)	United Kingdom Pershore Worcestershire	13 g a.s./ha 2 treatm. last date 11.09.06	45 at last treatment	0 4 7 15	plant without root 0.130 plant without root 0.078 plant without root <u>0.048</u> plant without root <0.01	
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/5 Study to GLP Study carried out in 2005	Leek	France 31840 Seilh (South of EU)	25 g a.s./ha 10.10.05	41-43	0 3 7 14	plant without root 0.093 plant without root <u>0.062</u> plant without root 0.045 plant without root 0.030	BASF method N 567/0 plant without root: mean recovery = 92.8%; SD: +/- 8.9; CV: 9.6; n=8; fortification range 0.01-0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026850 Trial No. AF/8821/BA/6 Study to GLP Study carried out in 2005	Leek	Italy Lusia-Rovigo 45020	25 g a.s./ha 21.09.05	47	0 3 7 14	plant without root 0.149 plant without root <u>0.105</u> plant without root 0.105 plant without root 0.054	
BASF Doc ID 2007/1008498 Trial No. A/SP/I/06/159 Study to GLP Study carried out in 2006	Leek (variety Stal)	Spain Masanasa Valencia	27 g a.s./ha 29.09.06	45	0 3 7 14	plant without root 0.042 plant without root <u>0.017</u> plant without root 0.011 plant without root 0.014	BASF method N 567/0 plant: mean recovery = 97.9%; SD: +/- 7.5; CV: 7.6; n=7; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1008498 Trial No. A/SF/I/06/160 Study to GLP Study carried out in 2006	Leek (variety Selecta)	France Noves Bouches de Rhone (South of EU)	25 g a.s./ha 11.09.06	47	0 3 7 14	plant without root 0.144 plant without root <u>0.032</u> plant without root <0.01 plant without root <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue studies in leek presented in the alpha-cypermethrin dossier were carried out in 7 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 2 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions-8 trials including both variants, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions-4 trials conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues of alpha-cypermethrin in leek plants ranged between 0.018 mg/kg and 0.183 mg/kg. Residues declined to levels between 0.024–0.078 mg/kg, 0.014-0.047 mg/kg and <0.01-0.055 mg/kg at 2-4, 7 and 13-15 days after application.

After two applications at 12.5 g a.s./ha, initial residues in leek plants ranged between 0.032 mg/kg and 0.144 mg/kg. Residues declined to levels between 0.029-0.082 mg/kg, 0.018–0.071 mg/kg and <0.01-0.064 mg/kg at 2-4, 7 and 13-15 days after application.

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in leek plants ranged between 0.042 mg/kg and 0.149 mg/kg. Residues declined to levels between 0.017–0.105 mg/kg, <0.01-0.105 mg/kg and <0.01-0.054 mg/kg at 3, 7 and 14 days after application.

Conclusion:

In the years 2005 and 2006 a residue program on leek was conducted in Belgium, France (Northern and Southern European region), Germany, Italy, Spain, the Netherlands and the United Kingdom under open field conditions.
In 8 trials supporting the proposed field GAP for the Northern European region, residues in leek ranged between 0.018-0.071 mg/kg at the target PHI of 7 days.
In 4 trials supporting the proposed field GAP for the Southern European region, residues in leek ranged between 0.017-0.105 mg/kg at the target PHI of 3 days.

2.23.2 Estimation of MRL, HR and STMR for leek

For *leek*, the following residue studies were considered (BASF DocIDs): 2006/1026850 and 2007/1008498.

The following residue values (PHI=7±1 days for EU-N or PHI=3±1 days for EU-S, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026850	0.033, 0.053, 0.058, 0.071	0.062, 0.105
2007/1008498	0.018, 0.032, 0.039, 0.048	0.017, 0.032
OECD-MRL-calculation	**<u>0.15</u> (n=8, STMR=0.044, HR=0.071)**	**<u>0.2</u> (n=4, STMR=0.047, HR=0.105)**

_ underlined values were used for risk assessment purposes

2.24 Pulses (dry beans and peas)

Residue data from supervised trials in pulses (dry beans and peas) were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.24-1: Number of residue trials conducted in pulses (dry beans and peas) per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Pulses (field)	2005	4	FR, UK	4	ES, IT	8	2006/1026858
Pulses (field)	2006	4	DE, FR, NL	4	ES, FR, GR	8	2007/1007949
Total number of trials per region					**Total number of trials**		

2.24.1 Supervised residue trials in pulses (dry beans and peas)

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of alpha-cypermethrin in dry beans (*Vicia faba*) and dry peas (*Pisum sativum*) after treatment with either one or two applications of BAS 310 40 I under field conditions in Northern and Southern Europe during 2005; BASF DocID 2006/1026858

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alpha-cypermethrin in dry beans (*Vicia faba*) and dry peas (*Pisum sativum*) after treatment with either one or two applications of BAS 310 40 I under field conditions in Northern and Southern Europe during 2006; BASF DocID 2007/1007949

Material and Methods:

In the years 2005 and 2006 a residue program in dry peas and beans was conducted in France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.

During the 2005 growing season, eight field trials were conducted dry peas and beans in France (Northern European region), Italy, Spain and the United Kingdom.

In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea or bean plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the pea plants at the last application was between 75 (50% of pods have reached typical length; juice exudes if pressed.Tenderometer value: 105 TE) and 79 (pods have reached typical size (green ripe); peas fully formed), the beans were at stages 83 (30% of pods ripe (beans hard)) to 86 (60% of pods ripe (beans hard)).

In four trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea and bean plants once at a target rate of 25 g a.s./ha. The growth stage of the pea plants at application was between 73 (30% of pods have reached typical length; juice exudes if pressed.Tenderometer value: 80 TE) and 89 (fully ripe: all pods dry and brown. Seeds dry and hard (dry ripe)), the beans were at stages 79 (pods: individual beans easily visible) to 87 (70% of pods ripe (beans hard)).

Specimens of peas and remaining plant were collected immediately after the last application from each plot, as well as approximately 14, 21 and 28 days thereafter.

Specimens were analysed using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2006 growing season, eight field trials were conducted in dry peas and beans in France (Northern and Southern European region), Germany, Greece, Spain and the Netherlands.

In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea or bean plants on separate plots either once or twice at a target rate of 12.5 g a.s./ha. The growth stage of the pea plants at the last application was between 71 (10% of pods have reached typical length; juice exudes if pressed) and 87 (70% of pods ripe, seeds final colour, dry and hard), the beans were at stages 73 (30% of pods have reached typical length) to 81 (10% of pods ripe (beans hard) Seeds beginning to mature).

In four trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to pea and bean plants once at a target rate of 25 g a.s./ha. The growth stage of the pea plants at application was between 87 (70% of pods ripe, seeds final colour, dry and hard) and 89 (fully ripe: all pods dry and brown. Seeds dry and hard (dry ripe)), the beans were at stages 79 (pods: individual beans easily visible) to 93.

Specimens of peas and remaining plant were collected immediately after the last application from each plot, as well as 13-14, 20-22 and 27-28 days thereafter.

Specimens were analysed using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantification of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.24.1-1 for dry beans and in Table 2.24.1-2 for dry peas.

Table 2.24.1-1: Residues in dry beans

GAP for EU-N is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 3 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/5 Study to GLP Study carried out in 2005	Dry bean	United Kingdom Chapel Lane Aslochton Nottingham NG13 9AR	12.5 g as/ha 29.07.05	83	0 0 14 14 21 21 28 28	seed <0.01 straw 0.167 seed <0.01 straw 0.310 seed <0.01 straw 0.158 seed <0.01 straw 0.089	BASF method N 567/0 seed: mean recovery = 70.7%; SD: +/- 11.6; CV: 16.5; n=4; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 84.3%; SD: 17.4 +/-; CV: 20.7; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/6 Study to GLP Study carried out in 2005	Dry bean (variety Castel)	France Mavry, St Georges de Layon, 49700 (North of EU)	12.5 g as/ha 27.06.05	86	0 0 14 14 21 21 28 28	seed <0.01 straw 0.436 seed <0.01 straw 0.205 seed <0.01 straw 0.236 seed <0.01 straw 0.210	
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/6 Study to GLP Study carried out in 2006	Dry field bean (variety Cebeco 5930)	The Netherlands Lingestraat 6662 NN Elst Gelderland	12.5 g as/ha 11.07.06	73	0 0 14 14 22 22 28 28	seed <0.01 straw 0.488 seed <0.01 straw 0.107 seed <0.01 straw 0.077 seed <0.01 straw 0.050	BASF method N 567/0 seed: mean recovery = 78.2%; SD: +/- 8.7; CV: 11.2; n=6; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 83.0%; SD: 12.6 +/-; CV: 15.2; n=8; fortification range 0.01 – 2.0

Table 2.24.1-1: Residues in dry beans

GAP for EU-N is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 3 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/9 Study to GLP Study carried out in 2006	Dry field bean (variety Irena)	France Gaec du Pre de Chene Pre du Chene Boce 49150 (North of EU)	12.5 g as/ha 29.06.06	81	0 0 14 14 21 21 28 28	seed <0.01 straw 0.584 seed <0.01 straw 0.079 seed <0.01 straw 0.107 seed <0.01 straw 0.084	mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/5 Study to GLP Study carried out in 2005	Dry bean (variety TBA)	United Kingdom Chapel Lane Aslochton Nottingham NG13 9AR	12.5 g as/ha 2 treatm. last date 29.07.05	83 at last treatm.	0 0 14 14 21 21 28 28	seed <0.01 straw 0.278 seed <u><0.01</u> straw 0.295 seed <0.01 straw 0.396 seed <0.01 straw 0.109	BASF method N 567/0 seed: mean recovery = 70.7%; SD: +/- 11.6; CV: 16.5; n=4; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 84.3%; SD: 17.4 +/-; CV: 20.7; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/6 Study to GLP Study carried out in 2005	Dry bean (variety Castel)	France Mavry, St Georges de Layon, 49700 (North of EU)	12.5 g as/ha 2 treatm. last date 27.06.05	86 at last treatm.	0 0 14 14 21 21 28 28	seed <0.01 straw 0.574 seed <u><0.01</u> straw 0.459 seed <0.01 straw 0.453 seed <0.01 straw 0.593	
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/6 Study to GLP Study carried out in 2006	Dry field bean (variety Cebeco 5930)	The Netherlands Lingestraat 6662 NN Elst Gelderland	12.5 g as/ha 2 treatm. last date 11.07.06	73 at last treatm.	0 0 14 14 22 22 28 28	seed <0.01 straw 0.638 seed <u><0.01</u> straw 0.167 seed <0.01 straw 0.075 seed <0.01 straw 0.101	BASF method N 567/0 seed: mean recovery = 78.2%; SD: +/- 8.7; CV: 11.2; n=6; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 83.0%; SD: 12.6 +/-; CV: 15.2; n=8; fortification range 0.01 – 2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/9 Study to GLP Study carried out in 2006	Dry field bean (variety Irena)	France Gaec du Pre de Chene Pre du Chene Boce 49150 (North of EU)	12.5 g as/ha 2 treatm. last date 29.06.06	81 at last treatm.	0 0 14 14 21 21 28 28	seed <0.01 straw 0.536 seed <u><0.01</u> straw 0.095 seed <0.01 straw 0.182 seed <0.01 straw 0.203	
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/7 Study to GLP Study carried out in 2005	Dry bean (variety Agua Dulce)	Spain Rey de Los Moros Cortes 31530	25 g as/ha 01.06.05	79	0 0 14 14 21 21 28 28	seed <0.01 straw 0.490 seed <u><0.01</u> straw 0.095 seed <0.01 straw 0.077 seed <0.01 straw 0.092	BASF method N 567/0 seed: mean recovery = 70.7%; SD: +/- 11.6; CV: 16.5; n=4; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 84.3%; SD: 17.4 +/-; CV: 20.7; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/8 Study to GLP Study carried out in 2005	Dry bean (variety Polo)	Italy Az Agr Cantaglia Via Poniticelli 2 Pepola di Malalbergo 40058	25 g as/ha 13.06.05	87	0 0 14 14 21 21 28 28	seed <0.01 straw 0.760 seed <u><0.01</u> straw 0.289 seed <0.01 straw 0.153 seed <0.01 straw 0.265	

Table 2.24.1-1: Residues in dry beans

GAP for EU-N is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 3 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/8 Study to GLP Study carried out in 2006	Dry field bean (variety Reina Bianca)	Spain Plaza Mayor Cortes 31530	25 g as/ha 01.06.06	93	0 0 14 14 21 21 28 28	seed <0.01 straw 0.731 seed <u><0.01</u> straw 0.424 seed <0.01 straw 0.245 seed <0.01 straw 0.261	BASF method N 567/0 seed: mean recovery = 78.2%; SD: +/- 8.7; CV: 11.2; n=6; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 83.0%; SD: 12.6 +/-; CV: 15.2; n=8; fortification range 0.01 – 2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/11 Study to GLP Study carried out in 2006	Dry field bean (variety Linex)	France La Guille 11150 Villasavary South of EU	25 g as/ha 02.08.06	79	0 0 14 14 21 21 28 28	seed <0.01 straw 1.077 seed <u><0.01</u> straw 0.107 seed <0.01 straw 0.057 seed <0.01 straw 0.045	

_ underlined values were used for MRL calculation

Table 2.24.1-2: Residues in dry peas

GAP for EU-N is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 3 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/1 Study to GLP Study carried out in 2005	Dry pea (variety Kobleackie)	United Kingdom Smithy Fram Main Road Morley Ilkeston Derbyshire DE7 6DF	12.5 g as/ha 15.07.05	75-77	0 0 14 14 21 21 28 28	seed <0.01 straw 0.190 seed <0.01 straw 0.122 seed <0.01 straw 0.251 seed <0.01 straw 0.274	BASF method N 567/0 seed: mean recovery = 70.7%; SD: +/- 11.6; CV: 16.5; n=4; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 84.3%; SD: 17.4 +/-; CV: 20.7; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/2 Study to GLP Study carried out in 2005	Dry pea (variety Hardy)	France 10 Rue d´Enjanville Rouvres St Jean 45300 (North of EU)	12.5 g as/ha 21.06.05	79	0 0 14 14 21 21 28 28	seed <0.01 straw 0.121 seed <0.01 straw 0.119 seed <0.01 straw 0.237 seed <0.01 straw 0.203	
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/1 Study to GLP Study carried out in 2006	Dry pea (variety Arthur)	France 1 Rue d´Enzanville 45300 Rouvres-St Jean (North of EU)	12.5 g as/ha 29.06.06	85-87	0 0 14 14 21 21 28 28	seed <0.01 straw 0.127 seed <0.01 straw 0.246 seed <0.01 straw 0.577 seed <0.01 straw 0.295	BASF method N 567/0 seed: mean recovery = 78.2%; SD: +/- 8.7; CV: 11.2; n=6; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 83.0%; SD: 12.6 +/-; CV: 15.2; n=8; fortification range 0.01 – 2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/2 Study to GLP Study carried out in 2006	Dry pea (variety Santana)	Germany Vor den Hofen 31303 Burgdorf-Hulptingsen Lower Saxony	12.5 g as/ha 27.10.06	71	0 0 13 13 20 20 28 28	seed <0.01 straw 0.345 seed <0.01 straw 0.244 seed <0.01 straw 0.249 seed <0.01 straw 0.212	

Table 2.24.1-2: Residues in dry peas

GAP for EU-N is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 3 days

GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/1 Study to GLP Study carried out in 2005	Dry pea (variety Kobleackie)	United Kingdom Smithy Fram Main Road Morley Ilkeston Derbyshire DE7 6DF	12.5 g as/ha 2 treatm. last date 15.07.05	75-77 at last treatm.	0 0 14 14 21 21 28 28	seed <0.01 straw 0.562 seed <u><0.01</u> straw 0.439 seed <0.01 straw 0.524 seed <0.01 straw 0.519	BASF method N 567/0 seed: mean recovery = 70.7%; SD: +/- 11.6; CV: 16.5; n=4; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 84.3%; SD: 17.4 +/-; CV: 20.7; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/2 Study to GLP Study carried out in 2005	Dry pea (variety Hardy)	France 10 Rue d´Enjanville Rouvres St Jean 45300 (North of EU)	12.5 g as/ha 2 treatm. last date 21.06.05	79 at last treatm.	0 0 14 14 21 21 28 28	seed <0.01 straw 0.209 seed <u><0.01</u> straw 0.299 seed <0.01 straw 0.365 seed <0.01 straw 0.362	
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/1 Study to GLP Study carried out in 2006	Dry pea (variety Arthur)	France 1 Rue d´Enzanville 45300 Rouvres-St Jean (North of EU)	12.5 g as/ha 2 treatm. last date 29.06.06	85-87 at last treatm.	0 0 14 14 21 21 28 28	seed <0.01 straw 0.305 seed <u><0.01</u> straw 0.249 seed <0.01 straw 1.008 seed <0.01 straw 0.665	BASF method N 567/0 seed: mean recovery = 78.2%; SD: +/- 8.7; CV: 11.2; n=6; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 83.0%; SD: 12.6 +/-; CV: 15.2; n=8; fortification range 0.01 – 2.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/2 Study to GLP Study carried out in 2006	Dry pea (variety Santana)	Germany Vor den Hofen 31303 Burgdorf-Hulptingsen Lower Saxony	12.5 g as/ha 2 treatm. last date 27.10.06	71 at last treatm.	0 0 13 13 20 20 28 28	seed <0.01 straw 0.389 seed <u><0.01</u> straw 0.376 seed <0.01 straw 0.268 seed <0.01 straw 0.332	
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/3 Study to GLP Study carried out in 2005	Dry pea (variety Ideal)	Spain Rey de los Moros Cortes, 31530	25 g as/ha 01.06.05	73-74	0 0 14 14 21 21 28 28	seed <0.01 straw 0.230 seed <u><0.01</u> straw 0.323 seed <0.01 straw 0.673 seed not determined straw not determined	BASF method N 567/0 seed: mean recovery = 70.7%; SD: +/- 11.6; CV: 16.5; n=4; fortification range 0.01 – 0.1 mg/kg straw: mean recovery = 84.3%; SD: 17.4 +/-; CV: 20.7; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026858 Trial No. AF/8824/BA/4 Study to GLP Study carried out in 2005	Dry pea (variety Corallo)	Italy Az Agr Cantaglia Via Poniticelli 2 Pepola di Malalbergo 40058	25 g as/ha 22.06.05	87-89	0 0 14 14 21 21 28 28	seed <0.01 straw 1.213 seed <0.01 straw 0.154 seed 0.022 straw 0.477 seed <u>0.042</u> straw 0.355	
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/3 Study to GLP Study carried out in 2006	Dry pea (variety Baceaua)	France St Martin Castelsanasian 82100 South of EU	25 g as/ha 06.06.06	87	0 0 13 13 21 21	seed <0.01 straw 1.503 seed <u><0.01</u> straw 0.849 seed <0.01 straw 0.997	BASF method N 567/0 seed: mean recovery = 78.2%; SD: +/- 8.7; CV: 11.2; n=6; fortification range 0.01 – 0.1 mg/kg

Table 2.24.1-2: Residues in dry peas

GAP for EU-N is 2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 7 days GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 3 days							
GLP and trial details	Crop	Country	Application rate g as/ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007949 Trial No. AF/10493/BA/4 Study to GLP Study carried out in 2006	Dry pea (variety Lotus)	Greece Apollonia Thessaloniki GR-57015 Central Macedonia	25 g as/ha 26.05.06	87-89	0 0 13 13 21 21 27 27	seed <0.01 straw 1.097 seed <u><0.01</u> straw 0.410 seed <0.01 straw 0.290 seed <0.01 straw 0.505	straw: mean recovery = 83.0%; SD: 12.6 +/-; CV: 15.2; n=8; fortification range 0.01 – 2.0 mg/kg Residue analysed as total cypermethrin

_ underlined values were used for MRL calculation

Findings:

The residue studies in dry peas and beans presented in the alpha-cypermethrin dossier were carried out in 7 different EU countries and provide data relevant to conditions in the Northern and Southern European region.

The proposed GAP is

- for the Northern European region: 2 applications with 15 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 7 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray under open field conditions with a PHI of 3 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha under open field conditions – 8 trials including both variants, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions – 8 trials conducted in the Southern European region

After one application at 12.5 g a.s./ha, initial residues in pea or bean straw ranged between 0.121 mg/kg and 0.584 mg/kg. Residues in straw ranged between 0.079 – 0.310 mg/kg, 0.077 – 0.577 mg/kg and 0.050 – 0.519 mg/kg at 13-14, 20-22 and 28 days after application. Treated pea or bean seed samples were free of residues above the limit of quantification of the analytical method at all sampling intervals (all <0.01 mg/kg).

After two applications at 12.5 g a.s./ha, initial residues in pea or bean straw ranged between 0.209 mg/kg and 0.638 mg/kg. Residues in straw ranged between 0.095 – 0.459 mg/kg, 0.075 – 1.008 mg/kg and 0.101 – 0.665 mg/kg at 13-14, 20-22 and 28 days after application. Treated pea or bean seed samples were free of residues above the limit of quantification of the analytical method at all sampling intervals (all <0.01 mg/kg).

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in pea or bean straw ranged between 0.230 mg/kg and 1.503 mg/kg. Residues declined to levels between 0.095 – 0.849 mg/kg, 0.057 – 0.997 mg/kg and 0.045 – 0.505 mg/kg at 13-14, 21 and 27-28 days after application. Total residues of cypermethrin were below the LOQ (<0.01 mg/kg) in all seed specimens collected 0, 13-14, 21 and 27-28 days after application, except for specimens collected 21 and 28 days after application at Trial AF/8824/BA/4 that had residue levels of 0.022 mg/kg and 0.042 mg/kg respectively. As the treated seed samples from this trial were free of residues above the LOQ at earlier sampling intervals, this finding is most probably caused by a contamination.

Conclusion:

The residue studies presented demonstrate that dry peas or beans are free of residues above the limit of quantification of the analytical method (0.01 mg/kg).

2.24.2 Estimation of MRL, HR and STMR for pulses (dry beans and peas)

For *pulses (dry beans and peas)*, the following residue studies were considered (BASF DocIDs): 2006/1026858 and 2007/1007949.

The following residue values (PHI=14±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026858 (open field)	<0.01 (4x)	<0.01 (3x), 0.042
2007/1007949 (open field)	<0.01 (4x)	<0.01 (4x)
OECD-MRL-calculation (open field)	**0.01 (n=8, STMR=0.01, HR=0.01)**	**<u>0.06</u> (n=8, STMR=0.01, HR=0.042)**

_ underlined values were used for risk assessment purposes

2.25 Cotton

Residue data from supervised trials in cotton were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below

Table 2.25-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Cotton	2004	-	-	2	ES, GR	2	2005/1007583
Cotton	2005	-	-	4	ES, GR	4	2006/1026847
Cotton	2006	-	-	4	ES, GR	4	2007/1007947
Total number of trials per region				**10**	**Total number of trials**	**10**	

2.25.1 Supervised residue trials in cotton

Schroth E (2005a)

Full study reference

Schroth E (2005): Study on the residue behavior of Alpha-cypermethrin on cotton after application of BAS 310 41 I under field conditions in Spain and Greece, 2004; BASF DocID 2005/1007583

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of alpha-cypermethrin in Cotton after treatment with BAS 310 40 I under field conditions in Southern Europe during 2005; BASF DocID 2006/1026847

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of alpha-cypermethrin in Cotton after treatment with BAS 310 40 I under field conditions in Southern Europe during 2006; BASF DocID 2007/1007947

Material and Methods:

In the years 2004, 2005 and 2006 a residue program on cotton was conducted in Greece and Spain.

During the 2004 growing season, 2 field trials were conducted in different representative cotton growing areas in Greece and Spain to determine the residue level of alpha-cypermethrin. The soluble concentrate formulation BAS 310 41 I (SC; 100 g a.s./L) was applied once or twice to different plots of cotton plants a target application rate of 15 g a.s./ha. Specimens of plants and seeds were collected at the day of the last application as well as 6-7 and 14 days thereafter.
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS with BASF method No. 567/0 which has a limit of quantification of 0.01 mg/kg.

During the 2005 growing season, four field trials were conducted in representative cotton growing areas in Spain and Greece.
An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L, BAS 310 40 I), was foliar applied once at a target rate of 25 g a.s./ha to cotton at growth stages between BBCH 85-87 (about 50 – 70% of bolls open). Specimens of seed and rest of plant without roots were collected immediately after application from each plot as well as 2-3, 6-7 and 13-14 days thereafter.
The specimens were analysed for total cypermethrin residues using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantification of 0.01 mg/kg in all sample materials.

During the 2006 growing season, four field trials were conducted in representative cotton growing areas in Spain and Greece.
An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L, BAS 310 40 I), was foliar applied once at a target rate of 25 g a.s./ha to cotton at growth stages up to 87.
Specimens of seed and rest of plant without roots were collected immediately after application from each plot as well as 3, 7 and 13-14 days thereafter.
The specimens were analysed for total cypermethrin residues using BASF method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS with a limit of quantification of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarized in Table 2.25.1-1.

Table 2.25.1-1: Residues in cotton

GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2005/1007583 Trial No. ALO/06/04 Study to GLP Study carried out in 2004	Cotton (variety Hermes)	Spain E-41729 Trajano, Sevilla Andalucia	15 g a.s./ha 29.09.04	85	0 0 6 6 14 14	seeds <0.01 plants 0.37 seeds <0.01 plants 0.21 seeds <0.01 plants 0.05	BASF analytical method N 567/0 plant (without roots), seeds: mean recovery = 77.1 %; SD: +/- 7.2; CV: 9.4%; n=6; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2005/1007583 Trial No. GRE/01/04 Study to GLP Study carried out in 2004	Cotton (variety Sandra)	Greece G-59100 Imathia Veria, Macedonia	15 g a.s./ha 18.10.04	86	0 0 7 7 14 14	seeds 0.02 plants 0.06 seeds <0.01 plants 0.06 seeds <0.01 plants 0.07	
BASF Doc ID 2005/1007583 Trial No. ALO/06/04 Study to GLP Study carried out in 2004	Cotton (variety Hermes)	Spain E-41729 Trajano, Sevilla Andalucia	15 g a.s./ha 2 treatm. last date 29.09.04	85 at last treatm.	0 0 6 6 14 14	seeds <0.01 plants 0.49 seeds <0.01 plants 0.32 seeds <0.01 plants 0.05	
BASF Doc ID 2005/1007583 Trial No. GRE/01/04 Study to GLP Study carried out in 2004	Cotton (variety Sandra)	Greece G-59100 Imathia Veria Macedonia	15 g a.s./ha 2 treatm. last date 18.10.04	86 at last treatm.	0 0 7 7 14 14	seeds 0.04 plants 0.09 seeds <0.01 plants 0.11 seeds 0.01 plants 0.20	
BASF Doc ID 2006/1026847 Trial No. AF/8822/BA/1 Study to GLP Study carried out in 2005	Cotton (variety Flora)	Spain 41740 Lebrija, Sevilla	25 g a.s./ha 26.09.05	85	0 0 3 3 7 7 14 14	seeds 0.011 rest of plant without root 0.704 seeds 0.010 rest of plant without root 0.203 seeds <u>≤0.01</u> rest of plant without root 0.196 seeds <0.01 rest of plant without root 0.078	BASF analytical method N 567/0 seeds: mean recovery = 104.7%; SD: +/- 7.6; CV: 7.3%; n=4; fortification range 0.01 – 1.0 mg/kg rest of plant: mean recovery = 81.1%; SD: +/- 12.4; CV: 15.2; n=4; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026847 Trial No. AF/8822/BA/2 Study to GLP Study carried out in 2005	Cotton (variety Pandora)	Spain 41100 Coria del Rio Sevilla	25 g a.s./ha 27.09.05	86	0 0 2 2 7 7 13 13	seeds 0.011 rest of plant without root 0.781 seeds <0.01 rest of plant without root 0.279 seeds <0.01 rest of plant without root 0.455 seeds <u>0.018</u> rest of plant without root 0.194	BASF analytical method N 567/0 seeds: mean recovery = 104.7%; SD: +/- 7.6; CV: 7.3%; n=4; fortification range 0.01 – 1.0 mg/kg rest of plant: mean recovery = 81.1%; SD: +/- 12.4; CV: 15.2; n=4; fortification range 0.01 – 1.0 mg/kg Residue analysed as total cypermethrin

Table 2.25.1-1: Residues in cotton

GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026847 Trial No. AF/8822/BA/3 Study to GLP Study carried out in 2005	Cotton (variety Flora)	Spain La Base, Dos Hermanas 41820 Sevilla	25 g a.s./ha 04.10.05	85	0 0 3 3 6 6 14 14	seeds <0.01 rest of plant without root 0.274 seeds <0.01 rest of plant without root 0.382 seeds <u>≤0.01</u> rest of plant without root 0.153 seeds <0.01 rest of plant without root 0.143	
BASF Doc ID 2006/1026847 Trial No. AF/8822/BA/4 Study to GLP Study carried out in 2005	Cotton (variety Velos)	Greece Gefya, Thessaloniki, Central Macedonia GR-57011	25 g a.s./ha 07.10.05	87	0 0 3 3 7 7 14 14	seeds 0.013 rest of plant without root 0.986 seeds <0.01 rest of plant without root 0.359 seeds <u>≤0.01</u> rest of plant without root 0.548 seeds <0.01 rest of plant without root 0.187	
BASF Doc ID 2007/1007947 Trial No. AF/10495/BA/1 Study to GLP Study carried out in 2006	Cotton	Greece	25 g a.s./ha 13.10.06		0 0 3 3 7 7 13 13	seeds <0.01 rest of plant without root 0.836 seeds <0.01 rest of plant without root 0.614 seeds <u>≤0.01</u> rest of plant without root 0.380 seeds <0.01 rest of plant without root 0.202	BASF analytical method N 567/0 rest of plant without roots: mean recovery = 76.4%; SD: +/- 11.8; CV: 15.4%; n=5; fortification range 0.01 – 1.0 mg/kg seeds: mean recovery = 87.3%; SD: +/- 12.6; CV: 14.4%; n=4; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007947 Trial No. AF/10495/BA/2 Study to GLP Study carried out in 2006	Cotton	Greece	25 g a.s./ha 13.10.06		0 0 3 3 7 7 13 13	seeds <0.01 rest of plant without root 0.570 seeds <0.01 rest of plant without root 0.942 seeds <u>≤0.01</u> rest of plant without root 0.340 seeds <0.01 rest of plant without root 0.383	BASF analytical method N 567/0 rest of plant without roots: mean recovery = 76.4%; SD: +/- 11.8; CV: 15.4%; n=5; fortification range 0.01 – 1.0 mg/kg seeds: mean recovery = 87.3%; SD: +/- 12.6; CV: 14.4%; n=4; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin

Table 2.25.1-1: Residues in cotton

GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007947 Trial No. AF/10495/BA/3 Study to GLP Study carried out in 2006	Cotton (variety Viky)	Spain Finca El Vergel Burquillos 41220	25 g a.s./ha 26.09.06	85-86	0 0 3 3 7 7 14 14	seeds 0.016 rest of plant without root 1.097 seeds <0.01 rest of plant without root 0.753 seeds <u><0.01</u> rest of plant without root 0.479 seeds <0.01 rest of plant without root 0.384	
BASF Doc ID 2007/1007947 Trial No. AF/10495/BA/4 Study to GLP Study carried out in 2006	Cotton (variety Celia)	Spain C/Guadalete, 16 Coto de Bornes Cadiz 11640	25 g a.s./ha 25.08.06	86	0 0 3 3 7 7 14 14	seeds <0.01* rest of plant without root 0.578 seeds <0.01 rest of plant without root 0.342 seeds <u><0.01</u> rest of plant without root 0.215 seeds <0.01 rest of plant without root 0.153 *0.009	BASF analytical method N 567/0 rest of plant without roots: mean recovery = 76.4%; SD: +/- 11.8; CV: 15.4%; n=5; fortification range 0.01 – 1.0 mg/kg seeds: mean recovery = 87.3%; SD: +/- 12.6; CV: 14.4%; n=4; fortification range 0.01 – 0.1 mg/kg Residue analysed as total cypermethrin

_ underlined values were used for MRL calculation

Findings:

The residue studies in cotton presented in the alpha-cypermethrin dossier were carried out in 2 different EU countries and provide data relevant to conditions in the Southern European region.
The proposed GAP is

- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days

The following residue studies were presented:

- one or two treatments at a target rate of 15 g a.s./ha under open field conditions – 2 trials including both variants, conducted in the Southern European region
- one treatment at a target rate of 25 g a.s./ha under open field conditions – 8 trials conducted in the Southern European region

After one application at 15 g a.s./ha, initial residues of alpha-cypermethrin in cotton plants ranged between 0.06 mg/kg and 0.37 mg/kg. Residues in plants declined to 0.06 – 0.21 mg/kg and 0.05 – 0.07 mg/kg 6-7 and 14 days after application. In cotton seed, a residue of 0.02 mg/kg was found immediately after application in one out of two trials. All other seed samples did not show residues above the LOQ (all <0.01 mg/kg).

After two applications at 15 g a.s./ha, initial residues of alpha-cypermethrin in cotton plants ranged between 0.09 mg/kg and 0.49 mg/kg. Residues in plants declined to 0.11 – 0.32 mg/kg and 0.05 – 0.20 mg/kg 6-7 and 14 days after application. In cotton seed, no residues above the LOQ were found in one out of two trials (all <0.01 mg/kg). Seeds taken in the second trial showed initial residues of 0.04 mg/kg which declined to <0.01 mg/kg and 0.01 mg/kg, at 7 and 14 days after the last application, respectively.

After one application at 25 g a.s./ha, initial residues of alpha-cypermethrin in cotton plants ranged between 0.274 mg/kg and 1.097 mg/kg. Residues declined to levels between 0.203 – 0.942 mg/kg, 0.153 – 0.548 mg/kg and 0.078 – 0.384 mg/kg at 2-3, 6-7 and 13-14 days after application.
Initial residues in seeds ranged between <0.01 – 0.016 mg/kg and declined to <0.01 – 0.01 mg/kg at 2-3 days after application. No residues in seeds above the LOQ (0.01 mg/kg) were found at 6-7 and 13-14 days after application in 7 trials, an isolated finding of 0.018 mg/kg occurred in one trial (AF/8822/BA/2) 13 days after application.

Conclusion:

In 8 trials supporting the proposed field GAP for the Southern European region, residues in cotton plants ranged between 0.153 – 0.548 mg/kg at the target PHI of 7 ±1 days. No residues in seeds above the LOQ (0.01 mg/kg) were found 6-7 and 13-14 days after application in 7 trials, an isolated finding of 0.018 mg/kg occurred in one trial (AF/8822/BA/2) 13 days after application.

2.25.2 Estimation of MRL, HR and STMR for cotton

For *cotton*, the following residue studies were considered (BASF DocIDs): 2006/1026847 and 2007/1007947.

The following residue values (PHI=7±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Southern Europe (S-EU) [mg/kg]
2006/1026847 (open field)	<0.01 (3x), 0.018
2007/1007949 (open field)	<0.01 (4x)
OECD-MRL-calculation (open field)	**<u>0.03</u> (n=8, STMR=0.01, HR=0.018**

_ underlined values were used for risk assessment purposes

2.26 Oilseed rape

In the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414; residue studies in oilseed rape were peer-reviewed. Below, residue data from peer-reviewed trials supporting the current GAP and considered for MRL proposal and risk assessment are summarized. New trials are presented in Section M-CA 6.3.4; the MRL calculations are shown in Section M-CA 6.7. An overview on the peer-reviewed studies considered is given below.

Table 2.26-1: Number of peer-reviewed residue trials in oilseed rape considered for risk assessment per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Oilseed rape	1983	5	UK	-	-	5	AL-750-004
Oilseed rape	1992	-	-	2	FR	2	AL-750-021
Oilseed rape	2001	-	-	2	ES, FR	2	AL-750-041 / 2002/1004087
Total number of trials per region		**5**		**4**	**Total number of trials**	**9**	

Table 2.26-2: Processing studies available for oilseed rape

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Oilseed rape	2000	-	-	2	FR	2	AL-750-038 2001/7001649
Oilseed rape	2001	-	-	2	FR	2	AL-750-042 2002/7005182
Total number of trials per region					Total number of trials	4	

2.26.1 Supervised residue trials in oilseed rape

Forbes S., MacKay C, (1984)
Full study reference
Forbes S., MacKay C, (1984): Residues of WL85871 In Oil Seed Rape From UK SRC Sittingbourne SBGR.84.014
BASF RDI-No.AL-750-004

Carlon R. (1993)
Full study reference
Carlon R. (1993): Residues of Alphacypermethrin in Rapeseeds From France Treated with Fastac PVP 1992 Trials BEGR.93.012A
BASF RDI-No.AL-750-021

Trewhitt J., Zimmermann U. (2002)
Full study reference
Trewhitt J., Zimmermann U. (2002): Study on the Residue Behavior of Alphacypermethrin (AC 900049) in Winter Oil Seed Rape After Treatment with BAS 310 08 I Under Field Conditions in France (S) and Spain, 2001
BASF Agricultural Center Limburgerhof, Germany Report No. 4807
BASF RDI-No.AL-750-041
BASF DocID 2002/1004087

Material and Methods:

Five trials were carried out in the United Kingdom in 1983 in which alpha-cypermethrin was applied to oil seed rape. The compound was applied on a single occasion in each trial, as a diluted EC formulation (100 g a.s./L), at a dosage rate of 20 g a.s./ha. The growth stage of the plants at application was between 61 (10% of flowers on main raceme open, main raceme elongating) and 75 (50% of pods have reached final size).
Rape seed was harvested at 61, 69, 70, 73 or 75 days after application.
In treated seed from all trials residues of alpha-cypermethrin were found to be below the limit of determination (all <0.01 mg/kg).

Two trials in winter oilseed rape were performed in France (Southern European region) in 1992.
Alpha-cypermethrin EC 50 g a.s./L and alpha-cypermethrin PVP 50 g a.s./kg were applied to rape, on a single occasion, at dosage rates of 10 and 20 g a.s./ha.
Rapeseeds were sampled at maturity 64 or 75 days after treatment, and analysed for residues of alpha-cypermethrin.
No detectable residues were found in any of the seed samples (all <0.01 mg/kg).

Two oil seed rape decline trials were established in 2001, one in France (Southern European region) and one in Spain. Oil seed rape was treated with a 150 g a.s./kg WG alpha-cypermethrin formulation. In these decline study trials, the plants received either a single or two foliar applications at 10 g a.s./ha.
The last application was performed between growth stages BBCH 69 (end of flowering) and BBCH 77 (70% the pods have reached final size).
In the Spanish trial, whole plant specimens were collected immediately after the last application. Whole plant samples with pods removed and the removed pods were taken after 29 days. Seed specimens were sampled 42 days after application.
In the French trial, whole plant specimens were collected immediately after the last application. Whole plant samples with pods removed and removed pods were taken after 29 days. Seed specimens were sampled 43, 50 (commercial harvest), and 57 days after application.
After a single application at 10 g a.s./ha, residues in whole plant ranged between 0.07 – 0.11 mg/kg. Residues in plant without pods sampled after 29 days ranged between <0.05 – 0.11 mg/kg, in pods taken at the same time points, residues between <0.05 – 0.11 mg/kg were found. No residues above the LOQ (0.05 mg/kg) were found in any of the treated seed samples.
After two applications at 10 g a.s./ha, residues in whole plant ranged between 0.06 – 0.28 mg/kg. Residues in plant without pods sampled after 29 days ranged between <0.05 – 0.24 mg/kg, in pods taken at the same time points, residues between <0.05 – 0.36 mg/kg were found.
In the Spanish trial, a trace residue of 0.06 mg/kg was detected in seed after 42 days.
No residues above the LOQ (0.05 mg/kg) were found in any of the treated seed samples from the French trial.

Table 2.26.1-1: Results of residue trials with alpha-cypermethrin conducted in oilseed rape

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-750-004 SBGR.84.014 Trial No. not reported Study not to GLP Study carried out in 1983	Oilseed rape (variety Jet Neuf)	United Kingdom Castle Howard Farms	20 g a.s./ha 31.05.83 Fastac EC 100 g a.s./L	-	61	seed <u><0.01</u>	SAMS 351-1 rape seed: mean recovery 81.7%; SD +/- 10.4; CV: 12.7% n=3; fortification level 0.1 mg/kg GC/ECD Residues analyzed as underivatized analyte
BASF RDI No. AL-750-004 SBGR.84.014 Trial No. WROSR 3 Study not to GLP Study carried out in 1983	Oilseed rape (variety Jet Neuf)	United Kingdom Stratford Davies	20 g a.s./ha 18.05.83 Fastac EC 100 g a.s./L	-	69	seed <u><0.01</u>	
BASF RDI No. AL-750-004 SBGR.84.014 Trial No. 83/307 SCUK Study not to GLP Study carried out in 1983	Oilseed rape (variety Jet Neuf)	United Kingdom S. Smart	20 g a.s./ha 17.05.83 Fastac EC 100 g a.s./L	-	73	seed <u><0.01</u>	

Table 2.26.1-1: Results of residue trials with alpha-cypermethrin conducted in oilseed rape

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-750-004 SBGR.84.014 Trial No. 83/306 SCUK Study not to GLP Study carried out in 1983	Oilseed rape (variety Jet Neuf)	United Kingdom P. Gordon	20 g a.s./ha 16.05.83 Fastac EC 100 g a.s./L	-	70	seed <0.01	
BASF RDI No. AL-750-004 SBGR.84.014 Trial No. 83/305 SCUK Study not to GLP Study carried out in 1983	oilseed rape (variety Jet Neuf)	United Kingdom J. Kidston	20 g a.s./ha 16.05.83 Fastac EC 100 g a.s./L	-	75	seed <0.01	SAMS 351-1 rape seed: mean recovery 81.7%; SD +/- 10.4; CV: 12.7% n=3; fortification level 0.1 mg/kg GC/ECD Residues analyzed as underivatized analyte
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/244 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 01 – Massieux (South of EU)	10 g a.s./ha 04.05.92 Fastac EC 50 g a.s./L	63	64	seed <0.01	SAMS 351-02 seed: 100% at 0.10 mg/kg GC/ECD Residues analyzed as underivatized analyte
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/244 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 01 – Massieux (South of EU)	20 g a.s./ha 04.05.92 Fastac EC 50 g a.s./L	63	64	seed <0.01	
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/244 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 01 – Massieux (South of EU)	10 g a.s./ha 04.05.92 Fastac PVP 50 g a.s./kg	63	64	seed <0.01	SAMS 351-02 seed: 100% at 0.10 mg/kg GC/ECD Residues analyzed as underivatized analyte
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/244 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 01 – Massieux (South of EU)	20 g a.s./ha 04.05.92 Fastac PVP 50 g a.s./kg	63	64	seed <0.01	
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/246 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 69 – Genay (South of EU)	10 g a.s./ha 23.04.92 Fastac EC 50 g a.s./L	-	75	seed <0.01	
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/246 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 69 – Genay (South of EU)	20 g a.s./ha 23.04.92 Fastac EC 50 g a.s./L	-	75	seed <0.01	

Table 2.26.1-1: Results of residue trials with alpha-cypermethrin conducted in oilseed rape

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/246 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 69 – Genay (South of EU)	10 g a.s./ha 23.04.92 Fastac PVP 50 g a.s./kg	-	75	seed <0.01	
BASF RDI No. AL-750-021 BEGR.93.012 Trial No. W/FR/R/92/246 Study not to GLP Study carried out in 1992	Winter oilseed rape (variety Samourai)	France 69 – Genay (South of EU)	20 g a.s./ha 23.04.92 Fastac PVP 50 g a.s./kg	-	75	seed <u><0.01</u>	
BASF Doc ID 2002/1004087 Trial No. ALO/10/01 Study to GLP Study carried out in 2001	Winter oil seed rape (variety Kabel)	Spain E-41410 Carmona Andalucia	10 g a.s./ha 05.04.01 150 g a.s./kg WG	77	0 29 29 42	plants 0.11 plants minus pods 0.11 pods 0.11 seed <0.05	BASF analytical method N RLA 12594.02V (989/0) plants: mean recovery = 81.6 %; SD: +/- 3.6 ; CV: 4.4 %; n= 4; fortification range 0.05 – 0.5 mg/kg plants without pods: mean recovery = 83.6 %; SD: +/- 9.1; CV: 10.9%; n=4; fortification range 0.05 – 0.5 mg/kg pods: mean recovery = 81.5 %; SD: +/- 8.9 ; CV: 11.0 %; n=4 ; fortification range 0.05 – 0.5 mg/kg seeds: mean recovery = 84.8 %; SD: +/- 9.2 ; CV: 10.8%; n= 8; fortification range 0.05 – 0.5 mg/kg Residue analysed as BAS 310 I
BASF Doc ID 2002/1004087 Trial No. FTL/07/01 Study to GLP Study carried out in 2001	Winter oil seed rape (variety Cheyenne)	France 82170 Pompignan Midi-Pyrenees (South of EU)	10 g a.s./ha 09.05.01 150 g a.s./kg WG	69	0 29 29 43 50 57	plants 0.07 plants minus pods <0.05 pods <0.05 seed <0.05 seed <0.05 seed <0.05	
BASF Doc ID 2002/1004087 Trial No. ALO/10/01 Study to GLP Study carried out in 2001	Winter oil seed rape (variety Kabel)	Spain E-41410 Carmona Andalucia	10 g a.s./ha 2 treatm. last date 05.04.01 150 g a.s./kg WG	77 at last applicati on	0 29 29 42	plants 0.28 plants minus pods <u>0.24</u> pods 0.36 seed <u>0.06</u>	
BASF Doc ID 2002/1004087 Trial No. FTL/07/01 Study to GLP Study carried out in 2001	Winter oil seed rape (variety Cheyenne)	France 82170 Pompignan Midi-Pyrenees (South of EU)	12/10 g a.s./ha 2 treatm. last date 09.05.01 150 g a.s./kg WG	69	0 29 29 43 50 57	plants 0.06 plants minus pods <u><0.05</u> pods <0.05 seed <u><0.05</u> seed <0.05 seed <0.05	

2.26.2 Processing studies in oilseed rape

Within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 this processing study in oilseed rape (BASF RDI-No. AL-750-038) was submitted. The study was assessed as not being required (3rd Addendum to the Monograph: Addendum Chapter B-7 (Residue data), January 2003). Therefore, a review was not performed at this time. The study was not included in the application as it had been submitted already with the alpha-cypermethrin addendum dossier. Below it is summarized for the reviewer´s convenience.

Report:
Grolleau G. 2001b
Alphacypermethrin (AC 900049) 150 g a.s./kg WG (RLM 11203): At harvest residue study on alphacypermethrin in oil seed rape and oil, South France, 2000
BASF RDI-No.: AL-750-038
BASF DocID 2001/7001649

Guidelines: EEC 91/414 Annex II 6,EEC 91/414 Annex III 8,EEC 96/68,EEC 7029/VI/95 rev. 5,EEC 1999/11/EC,Decret No. 98-1312 (31-12-1998)

GLP: yes
(certified by Groupe Interministeriel des Produits Chimiques, France)

Materials and Methods:

In 2000, two oilseed rape (OSR) residue trials were established in Southern France. OSR [variety: Expert (trial 00-602-649) and variety: Bristol (00-602-65)] was treated with the 150 g a.s./kg WG (RLM 11203) alpha-cypermethrin formulation. In each of these two residue trials, OSR received either a single 10 g a.s./ha or a single 100 g a.s./ha foliar application 50 days prior to typical commercial harvest. In separate plots, OSR also received three 10 g a.s./ha or three 100 g a.s./ha foliar applications 79, 59, and 50 days prior to typical commercial harvest.

Actual applications were within 10% of nominal, ranging from 9.5 g a.s./ha to 10.6 g a.s./ha (10 g a.s./ha) or 94 g a.s./ha to 98 g a.s./ha (100 g a.s./ha). The single applications occurred at the BBCH 67 (flowering declining, majority of the petals fallen) growth stage. The last of the three applications also occurred at BBCH 67. Application volumes ranged from 374 L/ha to 405 L/ha. Oil seed rape specimens [seeds after harvester thrashing] were sampled at typical commercial harvest, between BBCH 89 - 92 (00-602-649) and at BBCH 92 (00-602-650).

Field samples for residue analysis from both trials were frozen within 24 hours of sampling and remained frozen (including during transportation) until analysis. These specimens were shipped frozen to BASF Agro Research, BASF plc, Unit 60, 156 Fareham Road, Gosport, Hampshire, PO13 0AU, UK, where they were stored until shipment to the analytical laboratory.

Bulk OSR samples from the 1 x 100 g a.s./ha and the 3 x 100 g a.s./ha plots from both trials (00-602-649 and 00-602-650), sufficient for processing, were shipped unfrozen to the processor [Viticulture Recherhhe et Developpement (VITI R & D).

The processing phase was conducted at VITI R&D, 101 impasse des Capitelles, VILLETELLE, France, according to VITI R&D Processing Phase Plan GRA 0006 EUR.

Specimen analysis was conducted at CEM Analytical Services Ltd. (CEMAS), Glendale Park, Fernbank Road, North Ascot, Berkshire, UK. OSR seeds were ground prior to analysis while all other specimen were homogenized with dry ice. The OSR specimens were analyzed using BASF Agro Research analytical method RLA 12594.02V entitled:

"Analysis of Alphacypermethrin (AC 900049) in Olives and Olive Oil."

A method performance check was included with study AL-GE-00-999 (CEMR-1429) and validated the 0.05 mg/kg alpha-cypermethrin LOQ in OSR substrates.

Concurrent procedural recoveries at the LOQ [0.05 mg/kg in seeds, crude oil, and press cake)] and 10 times the LOQ [in seeds, refined oil, and press cake] were within the acceptable range of 70% to 110%.

Findings:

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any untreated harvest seed specimens.

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any harvest OSR seeds taken 50 days after the single 10 g a.s./ha or the last of the three 10 g a.s./ha applications from either trial (00-602-649 or 00-602-650). Alpha-cypermethrin residues ranged from <0.05 mg/kg (00-602-649) to 0.07 mg/kg (00-602-650) in harvest OSR seeds taken 50 days after the single 100 g a.s./ha application; from 0.07 mg/kg (00-602-650) to 0.10 mg/kg (00-602-649) in harvest OSR seeds taken 50 days after the third 100 g a.s./ha application.

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any untreated OSR processed fractions (press cake, crude oil, and refined oil).

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any OSR processed fraction (press cake, crude oil, and refined oil) resulting from seed taken 50 days after the single 100g a.s./ha or the last of three 100 g a.s./ha applications.

Figure 2.26.2-1: OSR Processing procedure flowchart - DocID AL-750-038

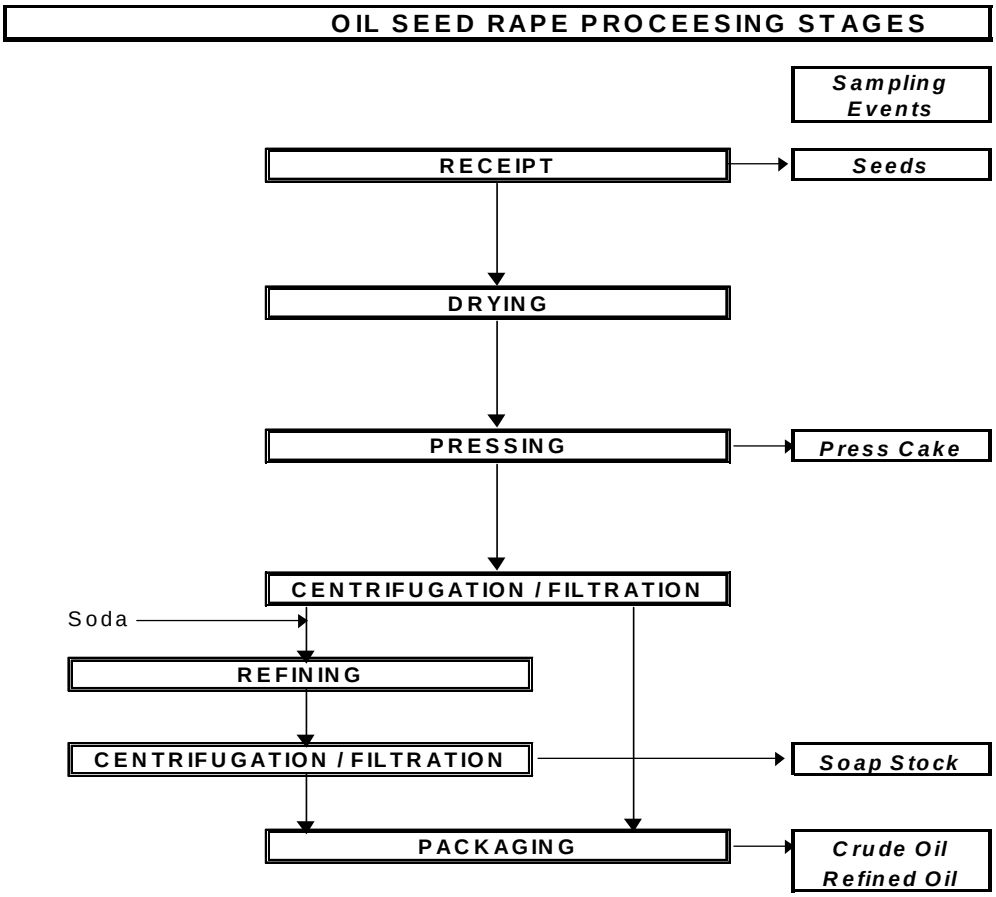

Table 2.26.2-1: Mass balance of OSR processing - DocID AL-750-038

	00-602-649			00-602-650		
Process Step	U1	T11	T21	U2	T12	T22
Seed prior to cleaning (kg)	5.75	7.55	6.30	5.55	6.70	7.00
Seed after cleaning (kg)	4.90	7.30	6.10	5.50	6.60	6.90
Loss on cleaning (%)	15	3	3	1	1	1
Seed prior to drying (kg)	4.90	7.30	6.10	5.50	6.60	6.90
Seed after drying (kg)	4.50	6.70	5.60	5.00	6.00	6.50
Loss on drying (%)	8	8	8	9	10	6
Seed to pressing (kg)	4.50	6.70	5.60	5.00	6.00	6.50
Press cake (kg)	2.90	4.25	3.55	3.20	3.75	3.90
Crude oil (g)	1540	2340	1900	1860	2150	2370
Accountability (%) (1)	99	98	97	101	98	96
Accountability (%) (2)	77	87	87	91	88	90
Crude oil (g)	1540	2340	1900	1860	2150	2370
Filtered crude oil (g)	1480	2230	1800	1798	2058	2300
Centrifuged/filtered crude oil (g)	1366	2142	1748	1690	1943	2201
Accountability (%) (3)	88	92	92	91	90	93
Crude oil to refining (mL)	800	800	800	800	800	800
Filtered refined oil (mL)	720	710	710	750	750	750
Centrifuged/filtered refined oil (mL)	685	665	660	605	655	630
Accountability (%) (4)	86	83	83	76	82	79
U1 and U2 are the control OSR seeds from trial 00-602-649 and 00-602-650, respectively.						
T11 and T12 are the OSR seeds from from the 00-602-649 1 x 100 g a.s./ha and 3 x 100 g a.s./ha applications, respectively						
T12 and T22 are the OSR seeds from from the 00-602-650 1 x 100 g a.s./ha and 3 x 100 g a.s./ha applications, respectively						
all percentages are rounded to the nearest whole number						
accountability (%) (1) = pressing efficiency = [press cake weight + crude oil weight/seed to pressing] x 100						
accountability (%) (2) = [press cake weight + crude oil weight/beginning seed weight] x 100						
accountability (%) (3) = [centrifuged/filtered crude oil weight/initial crude oil weight] x 100						
accountability (%) (4) = [centrifuged/filtered refined oil volume/initial crude oil volume to refining] x 100						

On average, approximately 55% of the initial seed weight (before cleaning and drying) is accounted for in the press cake and approximately 29% is accounted for in the centrifuged/filtered crude oil.

Based on the alpha-cypermethrin residue (ranging from < 0.05 mg/kg to 0.07 mg/kg) found in the OSR seed treated at an exaggerated rate (1 x 100 g a.s./ha), there would be no reasonable expectation of alpha-cypermethrin residue above the method LOQ (0.05 mg/kg) in any processed fraction (press cake, crude oil, refined oil). The results of this processing study support this conclusion.

Table 2.26.2-2: Transfer factors of OSR processing - DocID AL-750-038

	1 x 100 g a.s./ha application rate				3 x 100 g a.s./ha application rate			
OSR	**BAS 310 I Found (mg/kg)**		**Transfer Factor**		**BAS 310 I Found (mg/kg)***		**Transfer Factor**	
Process Fraction	00-602-649	00-602-650	00-602-649	00-602-650	00-602-649	00-602-650	00-602-649	00-602-650
seed	<0.05	0.07	1	1	<0.05	0.10	1	1
press cake	<0.05	<0.05	<1	<1	<0.05	<0.05	<1	<1
crude oil	<0.05	<0.05	<1	<1	<0.05	<0.05	<1	<1
refined oil	<0.05	<0.05	<1	<1	<0.05	<0.05	<1	<1

Within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 this processing study in oilseed rape (BASF RDI-No. AL-750-042) was submitted. The study was assessed as not being required (3rd Addendum to the Monograph: Addendum Chapter B-7 (Residue data), January 2003). Therefore, a review was not performed at this time. The study was not included in the application as it had been submitted already with the alpha-cypermethrin addendum dossier. Below it is summarized for the reviewer´s convenience.

Report:	Trewhitt J. and Zimmermann U. 2002b Processing study on the residue behavior of alphacypermethrin in oil seed rape after application of BAS 310 08 I under field conditions in France (S), 2001. BASF RDI No.: AL-750-042 BASF DocID 2002/7005182
Guidelines:	EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8),EEC 1607/VI/97 rev. 2 10.06.1999,EEC 7029/VI/95 rev. 5,EEC 7525/VI/95 rev. 2
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Materials and Methods:

In 2001, two winter oilseed rape (OSR) residue trials were established in southern France. OSR [variety: Elite (FBD/07/01) and variety: Cheyenne (FTL/08/01)] was treated with the 150 g a.s./kg WG (RLM 11203) alpha-cypermethrin formulation. In each of these two residue trials, OSR received two 100 g a.s./ha foliar applications 63 days (FTL/08/01)/64 days (FBD/07/01) and 49 days (FTL/08/01)/50 days (FBD/07/01) prior to typical commercial harvest.

Actual applications were within 10% of nominal, ranging from 99 g a.s./ha to 109 g a.s./ha. In both trials, the last applications occurred at the BBCH 71 (10% of pods have reached final size). Application volumes ranged from 298 L/ha to 327 L/ha. Oil seed rape specimens (seeds) were sampled at typical commercial harvest BBCH 89 (fully ripe: nearly all pods ripe, seeds black and hard).

Field samples for residue analysis from both trials were frozen within 24 hours of sampling and remained frozen (including during transportation) until analysis. These specimens were shipped frozen to BASF Akteingesellschaft, BASF Agricultural Center Limburgerhof, Crop Protection Division, Limburgerhof, Germany, where they were stored until shipment to the analytical laboratory.

Bulk OSR samples from both trials, sufficient for processing, were shipped unfrozen to the processor [Viticulture Recherche et Developpement (VITI R & D).

The processing phase was conducted at VITI R&D, 101 impasse des Capitelles, VILLETELLE, France.

Specimen analysis was conducted at the BASF Agricultural Research Center, Limburgerhof, Germany. The OSR specimens were analyzed using BASF method 989/0 (BASF Agro Research analytical method RLA 12594.02V) entitled:

"Analysis of Alphacypermethrin (AC 900049) in Olives and Olive Oil."

A method performance check was included with study AL-GE-00-999 (CEMR-1429) and validated the 0.05 mg/kg alpha-cypermethrin LOQ in OSR substrates.

Concurrent procedural recoveries at the LOQ (0.05 mg/kg) and 10 times the LOQ in seeds, refined oil, and press cake were reasonable given the complexity of the matrices, but not always within the 70% to 110% range.

Findings:

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in any untreated harvest seed specimens.

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in the press cake from untreated OSR seeds. Apparent alpha-cypermethrin residues were found in both the crude oil [and the refined oil [0.05 mg/kg (FBD/07/01) and 0.05 mg/kg (FTL/08/01)] from untreated OSR seeds.

Detectable alpha-cypermethrin residues were found in the treated OSR seeds from both trials [0.07 mg/kg (FBD/07/01) and 0.16 mg/kg (FTL/08/01)].

No detectable alpha-cypermethrin residues were found above the validated method LOQ (0.05 mg/kg) in the press cake from OSR seeds receiving two 100 g a.s./ha and sampled at typical commercial harvest (49 DAT2/50 DAT2; BBCH 89).

Detectable alpha-cypermethrin residues were found in both the crude oil [0.11 mg/kg (FBD/07/01) and 0.13 mg/kg (FTL/08/01)] and refined oil [0.09 mg/kg (FBD/07/01) and 0.16 mg/kg (FTL/08/01)] from OSR seeds receiving two applications 100 g a.s./ha and sampled at typical commercial harvest (49 DAT2/50 DAT2; BBCH 89).

Table 2.26.2-3: OSR processing procedure flowchart - DocID AL-750-042

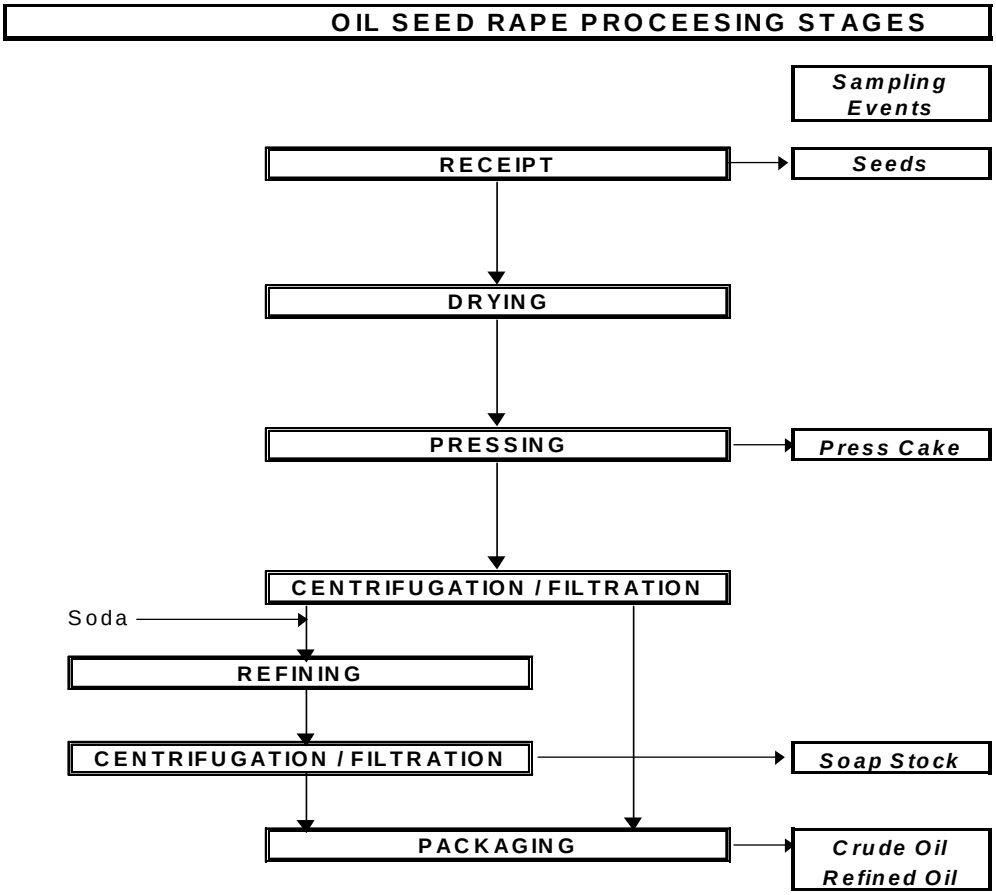

Table 2.26.2-4: Mass balance of OSR processing - DocID AL-750-042

	FBD/07/01		FTL/08/01	
Process Step	**Untreated**	**Treated**	**Untreated**	**Treated**
Seed to pressing (kg)	6.15	6.05	6.10	5.85
Press cake (kg)	3.10	2.95	3.35	3.10
Crude oil (g)	1457	1579	1702	1879
Accountability (%) (1)	74	75	83	85
Crude oil to refining (g)	715	711	713	721
Centrifuged/filtered refined oil (g)	547	540	542	556
Accountability (%) (2)	77	76	76	77
Untreated = the control OSR seeds from each trial.				
Treated = OSR seeds from each trial treated twice at 100 g a.s./ha.				
all percentages are rounded to the nearest whole number				
accountability (%) (1) = pressing efficiency = [press cake weight + crude oil weight/seed to pressing] x 100				
accountability (%) (2) = [centrifuged/filtered refined oil weight/initial crude oil weight to refining] x 100				

Based on the alpha-cypermethrin residue (ranging from < 0.05 mg/kg to 0.07 mg/kg) found in the OSR seed treated at an exaggerated rate (1 x 100 g a.s./ha), there would be no reasonable expectation of alpha-cypermethrin residue above the method LOQ (0.05 mg/kg) in any processed fraction (press cake, crude oil, refined oil). The results of this processing study support this conclusion.

Table 2.26.2-5: Transfer Factors of OSR processing - DocID AL-750-042

OSR	BAS 310 I Found (mg/kg)		Transfer Factor		BAS 310 I Found (mg/kg)*		Transfer Factor	
Process Fraction	FBD/07/01	FTL/08/01	FBD/07/01	FTL/08/01	FBD/07/01	FTL/08/01	FBD/07/01	FTL/08/01
Seed	0.07	0.16	1	1	0.07	0.16	1	1
Press cake	<0.05	<0.05	<1	<1	<0.05	<0.05	<1	<1
Crude oil	0.11	0.13	1.6	<1	0.06	<0.05	<1	<1
Refined oil	0.09	0.16	1.3	1	<0.05	0.11	<1	<1
* CRUDE OIL/REFINED OIL RESIDUE (AND TRANSFER FACTOR) CORRECTED FOR APPARENT ALPHA-CYPERMETHRIN IN CONTROLS.								

2.27 Barley

2.27.1 Supervised residue trials in barley

In the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414; residue studies in barley were peer-reviewed. Below, residue data from peer-reviewed trials supporting the current GAP and considered for MRL proposal and risk assessment are summarized. New trials are presented in Section M-CA 6.3.5; the MRL calculations are shown in Section M-CA 6.7. An overview on the peer-reviewed studies considered is given below.

Table 2.27.1-1: Number of peer-reviewed residue trials in barley considered for risk assessment per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Barley	1992	-	-	1	FR	1	AL-730-029
Total number of trials per region					**Total number of trials**	**1**	

Carlon R. (2007)

Full study reference
Carlon R. (2007): Residues of Alpha-cypermethrin in cereals from France treated with Fastac PVP – 1992 trials
BASF RDI-No.AL-730-029

Material and Methods:

One trial in spring barley was performed in France (Southern European region) during the year 1992. Two different formulation of aplha-cypermethrin, a 50 g a.s./L EC and a 50 g a.s./kg PVP, were applied once to separate plots each at rates of 15 and 30 g a.s./ha. Samples of grain and straw were taken 48 days after application. Residues in straw ranged between 0.05 – 0.10 mg/kg. No residues above the LOQ (0.010 mg/kg) were found in treated grain samples.

The trial data and residue results are summarized in Table 2.27.1-2.

Table 2.27.1-2: Results of residue trials with alpha-cypermethrin conducted in barley

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/238 Study to GLP Study carried out in 1992	Summer barley (variety Delta)	France 69 – Quincieux (South of EU)	15 g a.s./ha 04.06.92 Fastac PVP 50 g a.s./kg	-	48 48	grain <0.01 straw 0.05	SAMS 351-02 wheat straw: 98.5% at 0.10 mg/kg wheat grain: mean recovery 108.8% (97.5/120%); SD: n/a; CV: n/a; n=2; fortification range 0.05 - 0.10 mg/kg barley straw: 97.5 – 110% at 0.10 mg/kg barley grain: 113% at 0.10 mg/kg Residues analysed as total cypermethrin
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/238 Study to GLP Study carried out in 1992	Summer barley (variety Delta)	France 69 – Quincieux (South of EU)	30 g a.s./ha 04.06.92 Fastac PVP 50 g a.s./kg	-	48 48	grain <0.01 straw 0.10	SAMS 351-02 wheat straw: 98.5% at 0.10 mg/kg wheat grain: mean recovery 108.8% (97.5/120%); SD: n/a; CV: n/a; n=2; fortification range 0.05 - 0.10 mg/kg barley straw: 97.5 – 110% at 0.10 mg/kg barley grain: 113% at 0.10 mg/kg Residues analysed as total cypermethrin
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/238 Study to GLP Study carried out in 1992	Summer barley (variety Delta)	France 69 – Quincieux (South of EU)	15 g a.s./ha 04.06.92 Fastac EC 50 g a.s./L	-	48 48	grain <u>≤0.01</u> straw <u>0.08</u>	
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/238 Study to GLP Study carried out in 1992	Summer barley (variety Delta)	France 69 – Quincieux (South of EU)	30 g a.s./ha 04.06.92 Fastac EC 50 g a.s./L	-	48 48	grain <0.01 straw 0.08	

2.28 Maize

Table 2.28-1: Number of residue trials conducted in maize per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Maize	1981	1	FR	1	FR	2	AL-730-013
Maize	1989	-	-	2	FR	2	AL-730-019
Maize	1990	-	-	3	FR	3	AL-730-027
Maize	1986	1	DE	-	-	1	AL-730-057
Maize	1986	1	DE	-	-	1	AL-730-058
Maize	1986	1	DE	-	-	1	AL-730-059
Maize	1986	1	DE	-	-	1	AL-730-060
Maize	1987	1	DE	-	-	1	AL-730-062
Maize	1987	1	DE	-	-	1	AL-730-063
Maize	1987	1	DE	-	-	1	AL-730-064
Maize	1987	1	DE	-	-	1	AL-730-065
Maize	1982	(1)	South Africa			(1)	AL-730-014
Maize	1983	(1)	Brazil			(1)	AL-730-015
Maize	2009	2	DE, FR	1	ES	3	2009/1125196
Total number of trials per region		**11 (13)**		**7**	**Total number of trials**	**18 (20)**	

2.28.1 Supervised residue trials in maize

Report:	**Oxspring S., 2010c**
Title:	Study on the behaviour of alpha-cypermethrin in maize after treatment with BAS 310 40 I in Northern and Southern Europe during 2009
Document No:	BASF DocID 2009/1125196
Guidelines:	EEC 91/414 (1607/VI/97), EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EU Directive 96/68/EC
GLP	Yes

Executive Summary

During the growing season of 2009, a total of three trials were conducted in representative maize growing areas in Germany, France and Spain, to determine the magnitude and decline of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC).
Therefore BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g/L) was applied twice or once to the treated plot at a rate equivalent to 0.0150 kg a.s./ha or 0.0300 kg a.s./ha. In all trials the applications were made at crop stages BBCH 83 and 87.
For the analysis samples of maize were taken directly after the last application (0 DALA) and at 7±1, 14±1, 21±1 and 32±1 DALA, where specimens of cobs without husks, rest of plant with husks, grain and rest of plant without roots were taken.
The maize specimens were analysed for alpha-cypermethrin according to BASF Method No. 567/0, which determines the analytes by means of LC-MS/MS with a limit of quantitation (LOQ) of 0.01 mg/kg.
In all trials, residues of alpha-cypermethrin (BAS 310 I) in treated maze grain and cobs were not detectable. The treated maize whole plant specimens collected directly after the last application (0 DALA) of BAS 310 40 I contained a residue of 0.177-0.290 mg/kg. Residues in maize rest of plant ranged from 0.118-0.318 mg/kg.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 40 I
Lot/Batch #: 1209 (BAS 310 40 I, 100 g/L alpha-cypermethrin)
Purity:
CAS#: 52315-07-8 (alpha-cypermethrin)
Development code:
Spiking levels: 0.01-1.0 mg/kg

2. Test Commodity:

Crop: Maize
Type: Cereals
Variety: Bredao, Zidane, Sancia
Botanical name: *Zea mays*
Crop parts(s) or processed
Commodity: Whole plant w/o roots, cobs, rest of plant, grain
Sample size: 1.0 kg

B. STUDY DESIGN

1. Test procedure

During the growing season of 2009, a total of three trials were conducted in representative maize growing areas in Germany, France and Spain, to determine the magnitude and decline of residues of BAS 310 I (alpha-cypermethrin) in or on raw agricultural commodities (RAC).
Each field trial consisted of two plots, one control plot (plot 1) and one plot treated with BAS 310 40 I (plot 2).
In two of three trials (L090227 and L090228) BAS 310 40 I was applied twice at a rate equivalent to 0.015 kg alpha-cypermethrin/ha and for the other trial (L090229) the application was made once at a rate equivalent to 0.0300 kg alpha-cypermethrin/ha. The applications took place at BBCH 83-87 with an application rate of 200 L/ha.
For the analysis samples of maize were taken directly after the last application (0 DALA) and at 7±1, 14±1, 21±1 and 32±1 DALA, where specimens of cobs without husks, rest of plant with husks, grain and rest of plant w/o roots were taken.

Table 2.28.1-1: Target application rates and timings for maize

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2009	3	2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0150	200	1st appl.: 83-87 BBCH 2nd appl.: 85-87 BBCH
		1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0300	200	1st appl.: 83-85 BBCH

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).

The residues of alpha-cypermethrin in maize specimens were extracted from plant matrices using a mixture of methanol/water/hydrochloric acid. A liquid/liquid partition against cyclohexane was used for clean up. The final determination of alpha-cypermethrin was performed by LC-MS/MS. The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery averaged 79±9% for alpha-cypermethrin at fortification levels of 0.01 mg/kg and 1.0 mg/kg.

Table 2.28.1-2: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in maize

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
whole plant w/o root	0.01-0.5	12	77	11	14
rest of plant	0.01-1.0	4	81	11	13
grain	0.01-0.1	8	82	5	6
cobs	0.01-0.1	4	76	3	4

II. RESULTS AND DISCUSSION

Directly after the last application (0 DALA) of BAS 310 40 I, alpha-cypermethrin residues ranged between 0.177-0.290 mg/kg in whole plant specimens. At 7±1 days after the last application residues were 0.189-0.318 mg/kg in rest of plant and <0.01 mg/kg in maize cobs and grain respectively. At 14±1 DALA, residues were 0.137-0.266 mg/kg in rest of plant and <0.01 mg/kg in cobs and grain respectively. After a longer PHI (20-33 DALA) residues in maize rest of plant ranged between 0.118-0.182 mg/kg.

An overall summary of the residues is given in the table below.

Table 2.28.1-3: Summary of residues of BAS 310 I in maize from trials according to critical GAP after application of BAS 310 51 I and BAS 310 40 I

Crop	Year	No. of Appl.	Application	DALA	BBCH	Residues Found (mg/kg)	
						Matrix	alpha-cypermethrin (BAS 310 I)
Maize	2009	1	BAS 310 40 I (EC)	0	83-85	whole plant	0.177
				7	85	cobs w/o husks	<0.01
				13	85	cobs w/o husks	<0.01
				7	85	rest of plant with husks*	0.318
				13	85	rest of plant with husks*	0.266
				21	87	grain	<0.01
				33	89	grain	<0.01
				21	87	rest of plant*	0.168
				33	89	rest of plant*	0.118
		2		0	85-87	whole plant	0.267-0.290
				6	86	cobs w/o husks	<0.01
				13	87	cobs w/o husks	<0.01
				6	86	rest of plant with husks*	0.287
				13	87	rest of plant with husks*	0.137
				7	87	rest of plant*	<0.01
				14	89	rest of plant*	<0.01
				20-21	88-89	rest of plant*	<0.01
				32	89	rest of plant*	<0.01
				7	87	grain	0.189
				14	89	grain	0.214
				20-21	88-89	grain	0.134-0.182
				32	89	grain	0.134

DALA = days after last application
BBCH = growth stage at respective sampling
* = without roots

III. CONCLUSION

Directly after the last application (0 DALA) of BAS 310 40 I, alpha-cypermethrin residues ranged between 0.177-0.290 mg/kg in whole plant specimens. At 14±1 DALA, the residues of alpha-cypermethrin found in rest of plant were 0.137-0.266 mg/kg and <0.01 mg/kg in grain. In all trials, residues of alpha-cypermethrin in treated maize grain and cobs were not detectable.

Table 2.28.1-4: Residues of BAS 310 I after applications of BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 357886 **Doc ID:** 2009/1125196 **Trial No.:** L090227 **GLP:** yes **Year** 2009	Maize	Germany	BAS 310 40 I 2 x 0.0151-0.0174	85	0	whole plant*	0.290
					6	cobs w/o husks	<0.01
					6	rest of plant with husks*	0.287
					13	cobs w/o husks	<0.01
					13	rest of plant with husks*	0.137
					20	grain	<u><0.01</u>
					20	rest of plant*	<u>0.134</u>
					32	grain	<0.01
					32	rest of plant*	0.134
Study code: 357886 **Doc ID:** 2009/1125196 **Trial No.:** L090228 **GLP:** yes **Year** 2009	Maize	France	BAS 310 40 I 2 x 0.0150	87	0	whole plant	0.267
					7	grain	<0.01
					7	rest of plant*	0.287
					14	grain	<u><0.01</u>
					14	rest of plant*	<u>0.214</u>
					21	grain	<0.01
					21	rest of plant*	0.182

DALA = days after last application
BBCH = growth stage at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 2.28.1-5: Residues of BAS 310 I after application of BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 357886	Maize	Spain	BAS 310 40 I 1 x 0.0323	83-85	0	whole plant*	0.177
Doc ID: 2009/1125196					7	cobs w/o husks	<0.01
Trial No.: L090229					7	rest of plant with husks*	0.318
GLP: yes					13	cobs w/o husks	<0.01
Year 2009					13	rest of plant with husks*	0.266
					21	grain	<u><0.01</u>
					21	rest of plant*	<u>0.168</u>
					33	grain	<0.01
					33	rest of plant*	0.118

DALA = days after last application
BBCH = growth stage at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Bosio P G (1982)
Full study reference
Bosio P G (1982): Residues of WL 85871 in maize from France; Shell Chemie Berre BEGR.82.036, BASF RDI No. AL-730-013

Carlon R (1990)
Full study reference
Carlon R (1990): Residues of alphacypermethrin in maize from France treated with FASTAC – 1989 trials, Shell Chemie Berre BETR.90.009; BASF RDI No. AL-730-019

Bosio P G (1990)
Full study reference
Bosio P G (1990): Residues of alphacypermethrin in maize from France treated with FASTAC – 1990 trials, Shell Chemie Berre BETR.90.019; BASF RDI No. AL-730-027

Undeutsch (1987)
Full study reference
Undeutsch (1987): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-058

Undeutsch (1987)
Full study reference
Undeutsch (1987): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-057

Undeutsch (1987)
Full study reference
Undeutsch (1987): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-059

Undeutsch (1987)
Full study reference
Undeutsch (1987): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-060

Undeutsch (1988)
Full study reference
Undeutsch (1988): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-062

Undeutsch (1988)
Full study reference
Undeutsch (1988): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-063

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Undeutsch (1988): Berichtsbogen für Rückstandsuntersuchungen mit Pflanzenbehandlungs-mitteln, BASF RDI No. AL-730-065

Archer S M, Forbes S (1982)
Full study reference
Archer S M, Forbes S (1982): Residues of WL 85871 in maize from South Africa, SRC Sittingbourne SBGR.82.387; BASF RDI No. AL-730-014

Bosio P G (1983)
Full study reference
Bosio P G (1983): Residues of WL 85871 in maize grain from Brazil-1983 trials Shell Chemie Berre BEGR.83.055; BASF RDI No. AL-730-015

Material and Methods:

In the years 1981-1990 a field program on maize was conducted in France (Northern and Southern European region), Germany, South Africa and Brazil to investigate the residue behaviour of alpha-cypermethrin.

During the 1981 growing season, two field trials were conducted in maize in France (Northern and Southern European region). An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; WL 85871 100 g a.s./L EC) was foliar applied to maize plants on two different plots in each trial. In one variant, one treatment was done at a target rate of 30 g a.s./ha. In the second variant, a single treatment at a target rate of 50 g a.s./ha was applied. The application took place at a plant height between 0.85–1.3 m. Specimens of maize plant and grain were collected 65, 95 and 100 days after application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 233-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 1989 growing season, two field trials were conducted in maize in France (Southern European region). An emulsifiable concentrate formulation of alpha-cypermethrin (50 g a.s./L; FASTAC® 50 g a.s./L EC) was foliar applied to maize plants twice at a target rate of 30 g a.s./ha. The last application took place at the beginning drying of the plants. Specimens of silage and grain were collected 13, 29, 42 and 50 days after the last application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 383-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 1990 growing season, three field trials were conducted in maize in France (Southern European region). An emulsifiable concentrate formulation of alpha-cypermethrin (50 g a.s./L; FASTAC® 50 g a.s./L EC) was foliar applied to maize plants twice at a target rate of 40 g a.s./ha. The last application took place between flowering and ripening of cobs. Specimens of grain were collected 13, 22 and 40 days after the last application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 351-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 1986 growing season, four field trials were conducted in maize in Germany. A suspension concentrate formulation of alpha-cypermethrin (SC 100 g a.s./L) was foliar applied to maize plants once at a target rate of 17.5 g a.s./ha. The application took place approximately two months before harvest. Specimens of grain were collected 0, 7, 14, 21 days (plant) and 57-62 days (cobs and straw) after application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 383-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 1987 growing season, four field trials were conducted in maize in Germany. A suspension concentrate formulation of alpha-cypermethrin (SC 100 g a.s./L) was foliar applied to maize plants once at a target rate of 17.5 g a.s./ha. The application took place at growth stage 57 (tassel emergence). Specimens of grain were collected 0, 7, 14, 21 days (plant) and 56-69 days (grain) after application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 351-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

One trial was performed in maize during the growing season 1982 in South Africa. An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; WL 85871 100 g a.s./L EC) was foliar applied to maize plants once at a target rate of 30 g a.s./ha. Specimens of maize grain were collected 0, 2, 7 and 14 days after application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 351-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

During the 1983 growing season, one field trial was conducted in maize in Brazil. An emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L; WL 85871 100 g a.s./L EC) was foliar applied to maize plants on two different plots in each trial. In one variant, two treatments were done at a target rate of 12 g a.s./ha. In the second variant, two treatments at a target rate of 24 g a.s./ha were applied. The last application took place at ripening of the cobs. Specimens of maize grain were collected 22 days after the last application.
The specimens were analysed for alpha-cypermethrin by means of GC-ECD with method No. SAMS 351-1 which has a limit of quantitation of 0.01 mg/kg in all sample materials.

The trial data and residue results are summarised in Table 2.28.1-6.

Table 2.28.1-6: Residues in maize

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID AL-730-013 Trial No. W/FR/E81/221 Study not to GLP Study carried out in 1981	Maize (variety Stella)	France 01 - Loyettes (South of EU)	30 g a.s./ha 01.07.81	Plant height 0.85 m	65 100	plant <0.01 grain <0.01	
BASF Doc ID AL-730-013 Trial No. W/FR/E81/883 Study not to GLP Study carried out in 1981	Maize (variety Fontenac)	France 28 - Chateaudun (North of EU)	30 g a.s./ha 16.07.81	Plant height 1.3	95 95	plant <0.01 grain <0.01	Analytical method No. SAMS 233-1 plant: recovery = 93%; SD: n/a; CV: n/a%; fortification level 0.2 mg/kg grain: recovery = 91%; SD: n/a; CV: n/a%; fortification level 0.2 mg/kg Residue analysed as total cypermethrin
BASF Doc ID AL-730-013 Trial No. W/FR/E81/221 Study not to GLP Study carried out in 1981	Maize (variety Stella)	France 01 - Loyettes (South of EU)	50 g a.s./ha 01.07.81	Plant height 0.85 m	65 100	plant <0.01 grain <0.01	
BASF Doc ID AL-730-013 Trial No. W/FR/E81/883 Study not to GLP Study carried out in 1981	Maize (variety Fontenac)	France 28 - Chateaudun (North of EU)	50 g a.s./ha 16.07.81	Plant height 1.3	95 95	plant <0.01 grain <0.01	
BASF Doc ID AL-730-019 Trial No. S/FR/E89/161 Study not to GLP Study carried out in 1989	Maize (variety Sabrina)	France 31 - Muret (South of EU)	30 g a.s./ha 2 treatm. last date 17.08.89	Begin drying	29 50	silage 0.19 grain <0.01	Analytical method No. SAMS 383-1 silage: recovery = 102%; SD: n/a; CV: n/a%; fortification level 0.2 mg/kg grain: recovery = 91%; SD: n/a; CV: n/a%; fortification level 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID AL-730-019 Trial No. S/FR/E89/416 Study not to GLP Study carried out in 1989	Maize (variety Sabrina)	France 47 - Bouglon (South of EU)	30 g a.s./ha 2 treatm. last date 16.08.89	P0	13 42	silage 0.32 grain <0.01	
BASF Doc ID AL-730-027 Trial No. S/FR/E90/160 Study to GLP Study carried out in 1990	Maize (variety Jubilé)	France 82340 St Michel (South of EU)	40 g a.s./ha 2 treatm. last date 23.08.90	82 at last treatm.	13	grain <u>≤0.01</u>	
BASF Doc ID AL-730-027 Trial No. S/FR/E90/424 Study to GLP Study carried out in 1990	Maize (variety Ibisco)	France Lot-et-Garonne (South of EU)	40 g a.s./ha 2 treatm. last date 09.08.90	82 at last treatm.	40	grain <0.01	Analytical method No. SAMS 351-1 grain: recovery = 70/95%; SD: n/a; CV: n/a%; n=2; fortification level 0.20 mg/kg Residue analysed as total cypermethrin
BASF Doc ID AL-730-027 Trial No. S/FR/E90/447 Study to GLP Study carried out in 1990	Maize (variety Jubilé)	France 33 - Lubbon (South of EU)	40 g a.s./ha 2 treatm. last date 13.08.90	flowering	22	grain <0.01	

Table 2.28.1-6: Residues in maize

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID AL-730-057 Trial No. R32-86 Study not to GLP Study carried out in 1986	Maize (variety Limac)	Germany 64521Groß-Gerau (old postal code: 6080)	17.5 g a.s./ha 05.08.86	2 months before harvest	0 7 14 21 57 57	plant 0.52 plant 0.16 plant 0.33 plant 0.10 cob <0.01 straw 0.08	Analytical method No. SAMS 383-1 plant: recovery = 95%; SD: n/a; CV: n/a% cob: recovery = 98%; SD: n/a; CV: n/a% straw: recovery = 95%; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-058 Trial No. R31-86 Study not to GLP Study carried out in 1986	Maize (variety Limac)	Germany 64625 Bensheim (old postal code: 6140)	17.5 g a.s./ha 06.08.86	2 months before harvest	0 7 14 21 61 61	plant 0.4 plant 0.25 plant 0.12 plant 0.07 cob <0.01 straw 0.05	Analytical method No. SAMS 383-1 plant: recovery = 80%; SD: n/a; CV: n/a% cob: recovery = 95%; SD: n/a; CV: n/a% straw: recovery = 98%; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-059 Trial No. R33-86 Study not to GLP Study carried out in 1986	Maize (variety Limac)	Germany 63500 Seligenstadt (old postal code: 6453)	17.5 g a.s./ha 05.08.86	2 months before harvest	0 7 14 21 59 59	plant 0.14 plant 0.10 plant 0.18 plant 0.14 cob <0.01 straw 0.08	Analytical method No. SAMS 383-1 plant: recovery = 96%; SD: n/a; CV: n/a% cob: recovery = 98%; SD: n/a; CV: n/a% straw: recovery = 95%; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-060 Trial No. R34-86 Study not to GLP Study carried out in 1986	Maize (variety Limac)	Germany 23795 Bad Segeberg (old postal code: 2360)	17.5 g a.s./ha 30.07.86	2 months before harvest	0 7 14 21 59 59	plant 1.19 plant 0.86 plant 0.32 plant 0.15 cob <0.01 straw 0.23	Analytical method No. SAMS 383-1 plant: recovery = 92%; SD: n/a; CV: n/a% cob: recovery = 98%; SD: n/a; CV: n/a% straw: recovery = 95%; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-062 Trial No. R94/87 Study not to GLP Study carried out in 1987	Maize (variety Limac)	Germany 64625 Bensheim (old postal code: 6140)	17.5 g a.s./ha 06.08.87	57	0 7 14 21 63	plant 0.02 plant 0.01 plant 0.01 plant 0.02 grain <0.01	Analytical method No. SAMS 351-1 plant: recovery = 80%; SD: n/a; CV: n/a% grain: recovery = 115%; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-063 Trial No. R95/87 Study not to GLP Study carried out in 1987	Maize (variety Limac)	Germany 64521 Groß-Gerau (old postal code: 6080)	17.5 g a.s./ha 05.08.87	57	0 7 14 21 56	plant 0.83 plant 0.20 plant 0.29 plant 0.29 grain <0.01	Analytical method No. SAMS 351-1 plant: recovery = 86%; SD: n/a; CV: n/a% grain: recovery = 115%; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-064 Trial No. R96/87 Study not to GLP Study carried out in 1987	Maize (variety Tau)	Germany 85395 Billingsdorf/ Freising (old postal code: 8053)	17.5 g a.s./ha 10.08.87	57	0 7 14 21 60	plant 1.0 plant 0.38 plant 0.11 plant 0.18 grain <0.01	Analytical method No. SAMS 351-1 plant: recovery = 90%; SD: n/a; CV: n/a% grain: recovery = 88 %; SD: n/a; CV: n/a% Residue analysed as total cypermethrin

Table 2.28.1-6: Residues in maize

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 14 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID AL-730-065 Trial No. R97/87 Study not to GLP Study carried out in 1987	Maize (variety Limac)	Germany 63500 Seligenstadt (old postal code: 6453)	17.5 g a.s./ha 06.08.87	57	0 7 14 21 69	plant 1.74 plant 0.69 plant 0.46 plant 0.34 grain <0.01	Analytical method No. SAMS 351-1 plant: recovery = 86%; SD: n/a; CV: n/a% grain: recovery = 88 %; SD: n/a; CV: n/a% Residue analysed as total cypermethrin
BASF Doc ID AL-730-014 Trial No. S/SA/E81/443 Study not to GLP Study carried out in 1982	Maize (variety Pioneer 473)	South Africa A.J. Friedman Settlers	30 g a.s./ha 05.05. 82	not reported	0 2 7 14	grain 0.01 grain <0.01 grain <0.01 grain <0.01	Analytical method No. SAMS 351-1 grain: recovery = 110 %; SD: n/a; CV: n/a% Fortification level: 0.1 mg/kg Residue analysed as total cypermethrin
BASF Doc ID AL-730-015 Trial No. I/RE/MI-01/82 Study not to GLP Study carried out in 1983	Maize (variety Agroceres)	Brazil Ribeiraro Preto - SP	12 g a.s./ha 2 treatm. last date 15.03.83	consistent	22	grain <0.01	Analytical method No. SAMS 351-1 grain: recovery = 85 %; SD: n/a; CV: n/a% Fortification level: 0.10 mg/kg Residue analysed as total cypermethrin
BASF Doc ID AL-730-015 Trial No. I/RE/MI-01/82 Study not to GLP Study carried out in 1983	Maize (variety Agroceres)	Brazil Ribeiraro Preto - SP	24 g a.s./ha 2 treatm. last date 15.03.83	consistent	22	grain <0.01	

_ underlined values were used for MRL determination

Findings:

The residue studies in maize presented in the alpha-cypermethrin dossier were carried out in 4 different EU and Non-EU countries and provide data relevant to conditions in the Northern and Southern European region.

The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 14 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 14 days

The following residue studies were presented:

- one treatment at a target rate of either 30 or 50 g a.s./ha-2 trials including both variants, one conducted in the Northern European Region and one conducted in the Southern European Region
- one treatment at a target rate of 30 g a.s./ha-one trial conducted in South Africa
- two treatments at a target rate of 30 g a.s./ha-2 trials, both conducted in the Southern European Region
- two treatments at a target rate of 40 g a.s./ha-3 trials, all conducted in the Southern European Region
- one treatment at a target rate of 17.5 g a.s./ha-8 trials, all conducted in the Northern European Region
- two treatments at a target rate of either 12 or 24 g a.s./ha-one trial conducted in Brazil

Two trials were performed in the year 1981 in France with a single treatment at a target rate of either 30 or 50 g a.s./ha. No residues above the limit of quantitation (LOQ; 0.01 mg/kg) were found in treated maize plant samples collected 65 or 95 days after application. Mature grain specimens collected 95 or 100 days after application were free of residues above the LOQ (all <0.01 mg/kg).

One trial was performed in South Africa in the year 1982 with a single application at a target rate of 30 g a.s./ha. Immediately after application, a residue at the LOQ (0.01 mg/kg) was found in treated maize grain. No residues above the LOQ were found at later sampling points 2, 7 and 14 days after application.

Two trials were performed in the year 1989 in France with two applications at a target rate of 30 g a.s./ha. Residues in silage ranged between 0.19 mg/kg at 29 days after the last application and 0.32 mg/kg at 13 days after the last application. Residues in grain were below the LOQ of the analytical method 42 and 50 days after the last application, respectively (<0.01 mg/kg).

Three trials were performed in the year 1990 in France with two applications at a target rate of 40 g a.s./ha. No residues above the LOQ of the analytical method (0.01 mg/kg) were found in treated maize grain samples collected 13, 22 or 40 days after the last application.

Eight trials were performed in the years 1986 and 1987 in Germany with a single treatment at 17.5 g a.s./ha. Residues in plant material immediately after application ranged between 0.02–1.74 mg/kg and declined to 0.01-0.86 mg/kg, 0.01-0.46 mg/kg and 0.02-0.34 mg/kg at 7, 14 and 21 days thereafter.
In the 1986 trials, cob and straw samples were collected 57-62 days after application. In the 1987 trials, grain samples were taken 56-69 days after application. All treated samples of whole cobs or maize grain were free of residues above the LOQ of the analytical methods used (0.01 mg/kg) in all trials. In straw, residues between 0.08-0.23 mg/kg were found.

One trial was performed in the year 1983 in Brazil with two treatments at a target rate of either 12 or 24 g a.s./ha. No residues above the limit of quantitation (LOQ; 0.01 mg/kg) were found in treated maize grain samples collected 22 days after the last application.

Conclusion:

17 residue trials in maize are presented which demonstrate the absence of residues in maize grain. Even after two applications at 40 g a.s./ha, which is considerably higher than the proposed GAP, no residues were found in maize grain. This is due to the fact that alpha-cypermethrin is a non-systemic compound and therefore not translocated in the plant. Residues can only arise from direct contact with the spray. Maize cobs are protected from the spray by the hull leaves which prevent contamination of the kernels. No residues are to be expected in the grain, a situation which is confirmed by the residue studies presented.

2.28.2 Estimation of MRL, HR and STMR for maize

For *maize*, the following residue studies were considered (BASF DocIDs): 2009/1125196, AL-730-013, AL-730-057, AL-730-058, AL-730-059, AL-730-060, AL-730-062, AL-730-063, AL-730-064, AL-730-065, AL-730-013, AL-730-019 and AL-730-027.

The following residue values (PHI=13-100 days) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2009/1125196	<0.01 (2x)	<0.01
AL-730-013	<0.01	-
AL-730-057	<0.01	-
AL-730-058	<0.01	-
AL-730-059	<0.01	-
AL-730-060	<0.01	-
AL-730-062	<0.01	-
AL-730-063	<0.01	-
AL-730-064	<0.01	-
AL-730-065	<0.01	-
AL-730-013	-	<0.01
AL-730-019	-	<0.01 (2x)
AL-730-027	-	<0.01 (3x)
OECD-MRL-calculation	**<u>0.01</u> (n=11, STMR=0.01, HR=0.01)**	**<u>0.01</u> (n=7, STMR=0.01, HR=0.01)**

_ underlined values were used for risk assessment purposes

2.29 Rice

Residue data from supervised trials in rice were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below

Table 2.29-1: Number of residue trials conducted per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Rice	2005	-	-	4	ES, FR, GR, IT	4	2006/1026848
Rice	2006	-	-	4	ES, FR, GR, IT	4	2007/1007946
Total number of trials per region				**8**	**Total number of trials**	**8**	

2.29.1 Supervised residue trials in rice

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of BAS 310 I in Rice after treatment with BAS 310 40 I under field conditions in Southern Europe during 2005; BASF DocID 2006/1026848

North L (2007)

Full study reference

North L (2007): Study on the residue behaviour of alpha-cypermethrin in Rice after treatment with BAS 310 40 I under field conditions in Southern Europe during 2006; BASF DocID 2007/1007946

Material and Methods:

During the 2005 and 2006 growing seasons, eight field trials in rice were conducted in representative rice growing areas in France (Southern European region), Spain, Italy and Greece.

BAS 310 40 I, an emulsifiable concentrate formulation of alpha-cypermethrin (100 g a.s./L), was foliar applied either once or twice to separate plots of rice at a rate of 12.5 g a.s./ha. The growth stage of the plants at the last application was between 65 (full flowering: anthers visible on most spikelets) and 87 (hard dough: grain content solid, fingernail impression held).
Rice plant (without root) specimens were collected immediately after the last application. Panicles and the rest of plant (without root) were collected 13–15 days after the last application. Rice grain and straw specimens were collected at 20-22 and 27–29 days after the last application.
The specimens were analysed using BASF method no. 567/0 which quantifies the residues of underivatised cypermethrin by means of HPLC-MS/MS with a limit of quantitation of 0.01 mg/kg in all sample materials

The trial data and residue results are summarised in Table 2.29.1-1.

Table 2.29.1-1: Residues in rice

GAP for EU-S is 1-2 applications with 12.5 g a.s./ha at infestation as an overall spray, PHI = 21 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/1 Study to GLP Study carried out in 2005	Rice (variety Puntal)	Spain C/Nueva No. 31 Coria del Rio Sevilla	12.5 g as/ha 12.09.05	77	0 14 14 21 21 28 28	whole plant (no roots) 0.181 panicles 0.097 rest of plant w/o roots 0.091 straw 0.077 grain 0.059 straw 0.052 grain 0.050	BASF analytical method No. 567/0 whole plant: mean recovery = 77.9% (76.1-79.7%); SD: +/- n/a; CV: n/a; n=2; fortification range 0.01-3.0 mg/kg panicle: mean recovery = 83.8%; SD: +/- 18.0; CV: 21.5%; n=3; fortification range 0.01-1.0 mg/kg grain: mean recovery = 71.3%; SD: +/- 4.3; CV: 6.0%; n=5; fortification range 0.01-0.2 mg/kg straw: mean recovery = 103.0%; SD:18.5 +/-; CV: 18.0%; n=4; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 97.8% (80.8-114.9%); SD: n/a; CV: n/a; n=2; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/2 Study to GLP Study carried out in 2005	Rice (variety Ligalon)	France Domaine Saint Gabriel 11800 (South of EU)	12.5 g as/ha 25.08.05	73-75	0 13 13 21 21	whole plant (no roots) 0.265 panicles 0.160 rest of plant w/o roots 0.088 straw 0.072 grain 0.015	
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/3 Study to GLP Study carried out in 2005	Rice (variety Lido)	Italy Az. Agr. Cantaglia Via Ponticelli 2 Malalbergo Bologna 40058	12.5 g as/ha 29.08.05	75-77	0 14 14 21 21 28 28	whole plant (no roots) 0.511 panicles 0.144 rest of plant w/o roots 0.134 straw 0.349 grain 0.176 straw 0.330 grain 0.206	BASF analytical method No. 567/0 whole plant: mean recovery = 77.9% (76.1-79.7%); SD: +/- n/a; CV: n/a; n=2; fortification range 0.01-3.0 mg/kg panicle: mean recovery = 83.8%; SD: +/- 18.0; CV: 21.5%; n=3; fortification range 0.01-1.0 mg/kg grain: mean recovery = 71.3%; SD: +/- 4.3; CV: 6.0%; n=5; fortification range 0.01-0.2 mg/kg straw: mean recovery = 103.0%; SD:18.5 +/-; CV: 18.0%; n=4; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 97.8% (80.8-114.9%); SD: n/a; CV: n/a; n=2; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/4 Study to GLP Study carried out in 2005	Rice (variety Thai Bonnet)	Nea Malgara Thessaloniki Central Macedonia GR-57300	12.5 g as/ha 26.08.05	75	0 14 14 21 21 28 28	whole plant (no roots) 0.416 panicles 0.102 rest of plant w/o roots 0.093 straw 0.044 grain 0.039 straw 0.056 grain 0.020	
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/1 Study to GLP Study carried out in 2006	Rice (variety Cigalon)	France Domaine Saint Gabriel Marseilette 11800 (South of EU)	12.5 g as/ha 17.08.06	65-69	0 14 14 21 21 27 27	plant without root 0.270 panicles 0.063 rest of plant w/o root 0.12 grain 0.049 straw 0.060 grain 0.027 straw 0.053	BASF analytical method No. 567/0 whole plant (without roots): mean recovery = 83.8%; SD: +/- 7.8; CV: 9.3%; n=6; fortification range 0.01-1.0 mg/kg panicle: mean recovery = 77.4%; SD: +/- 7.5; CV: 9.7%; n=5; fortification range 0.01-

Table 2.29.1-1: Residues in rice

GAP for EU-S is 1-2 applications with 12.5 g a.s./ha at infestation as an overall spray, PHI = 21 days							
GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/2 Study to GLP Study carried out in 2006	Rice (variety Puntal)	Spain Cerro del Parlade Cita 9024 Poligono Industrial Isla Menor-Los Palacios	12.5 g as/ha 10.10.06	83	0 13 13 20 20 28 28	plant without root 0.250 panicles 0.036 rest of plant w/o root 0.068 grain 0.064 straw 0.049 grain 0.046 straw 0.052	1.0 mg/kg grain: mean recovery = 70.6%; SD:8.8 +/-; CV: 12.5%; n=5; fortification range 0.01-1.0 mg/kg straw: mean recovery = 81.4%; SD:10.8 +/-; CV: 13.3%; n=5; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 83.0%; SD: +/- 7.7; CV: 9.3%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/3 Study to GLP Study carried out in 2006	Rice (variety Cadet)	Italy Az. Agr. Cantaglia Via Ponticelli 2 Malalbergo Bologna 40058	12.5 g as/ha 20.09.06	87	0 15 15 22 22 29 29	plant without root 0.063 panicles 0.028 rest of plant w/o root 0.015 grain 0.028 straw 0.064 grain 0.020 straw 0.048	BASF analytical method No. 567/0 whole plant (without roots): mean recovery = 83.8%; SD: +/- 7.8; CV: 9.3%; n=6; fortification range 0.01-1.0 mg/kg panicle: mean recovery = 77.4%; SD: +/- 7.5; CV: 9.7%; n=5; fortification range 0.01-1.0 mg/kg
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/4 Study to GLP Study carried out in 2006	Rice (variety Claudio)	Greece Nea Malgara Thessaloniki Central Macedonia GR-57300	12.5 g as/ha 09.08.06	72-74	0 14 14 21 21 27 27	plant without root 0.26 panicles 0.085 rest of plant w/o root 0.084 grain 0.040 straw 0.049 grain 0.023 straw 0.021	grain: mean recovery = 70.6%; SD:8.8 +/-; CV: 12.5%; n=5; fortification range 0.01-1.0 mg/kg straw: mean recovery = 81.4%; SD:10.8 +/-; CV: 13.3%; n=5; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 83.0%; SD: +/- 7.7; CV: 9.3%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin

Table 2.29.1-1: Residues in rice

GAP for EU-S is 1-2 applications with 12.5 g a.s./ha at infestation as an overall spray, PHI = 21 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/1 Study to GLP Study carried out in 2005	Rice (variety Puntal)	Spain C/Nueva No. 31 Coria del Rio Sevilla	12.5 g as/ha 2 treatm. last date 12.09.05	77 at last treatm.	0 14 14 21 21 28 28	whole plant (no roots) 0.335 panicles 0.140 rest of plant w/o roots 0.091 straw 0.131 grain 0.148 straw 0.082 grain 0.011	BASF analytical method No. 567/0 whole plant: mean recovery = 77.9% (76.1-79.7%); SD: +/- n/a; CV: n/a; n=2; fortification range 0.01-3.0 mg/kg panicle: mean recovery = 83.8%; SD: +/- 18.0; CV: 21.5%; n=3; fortification range 0.01-1.0 mg/kg grain: mean recovery = 71.3%; SD: +/- 4.3; CV: 6.0%; n=5; fortification range 0.01-0.2 mg/kg straw: mean recovery = 103.0%; SD:18.5 +/-; CV: 18.0%; n=4; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 97.8% (80.8-114.9%); SD: n/a; CV: n/a; n=2; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/2 Study to GLP Study carried out in 2005	Rice (variety Ligalon)	France Domaine Saint Gabriel 11800 (South of EU)	12.5 g as/ha 2 treatm. last date 25.08.05	73-75 at last treatm.	0 13 13 21 21	whole plant (no roots) 0.452 panicles 0.200 rest of plant w/o roots 0.123 straw 0.194 grain 0.011	
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/3 Study to GLP Study carried out in 2005	Rice (variety Lido)	Italy Az. Agr. Cantaglia Via Ponticelli 2 Malalbergo Bologna 40058	12.5 g as/ha 2 treatm. last date 29.08.05	75-77 at last treatm.	0 14 14 21 21 28 28	whole plant (no roots) 0.463 panicles 0.357 rest of plant w/o roots 0.194 straw 0.354 grain 0.205 straw 0.351 grain 0.151	BASF analytical method No. 567/0 whole plant: mean recovery = 77.9% (76.1-79.7%); SD: +/- n/a; CV: n/a; n=2; fortification range 0.01-3.0 mg/kg panicle: mean recovery = 83.8%; SD: +/- 18.0; CV: 21.5%; n=3; fortification range 0.01-1.0 mg/kg grain: mean recovery = 71.3%; SD: +/- 4.3; CV: 6.0%; n=5; fortification range 0.01-0.2 mg/kg straw: mean recovery = 103.0%; SD:18.5 +/-; CV: 18.0%; n=4; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 97.8% (80.8-114.9%); SD: n/a; CV: n/a; n=2; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026848 Trial No. AF/8823/BA/4 Study to GLP Study carried out in 2005	Rice (variety Thai Bonnet)	Nea Malgara Thessaloniki Central Macedonia GR-57300	12.5 g as/ha 2 treatm. last date 26.08.05	75 at last treatm.	0 14 14 21 21 28 28	whole plant (no roots) 0.291 panicles 0.207 rest of plant w/o roots 0.086 straw 0.141 grain <0.01 straw 0.077 grain 0.039	

Table 2.29.1-1: Residues in rice

GAP for EU-S is 1-2 applications with 12.5 g a.s./ha at infestation as an overall spray, PHI = 21 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/1 Study to GLP Study carried out in 2006	Rice (variety Cigalon)	France Domaine Saint Gabriel Marseilette 11800 (South of EU)	12.5 g as/ha 2 treatm. last date 17.08.06	65-69 at last treatm.	0 14 14 21 21 27 27	plant without root 0.380 panicles 0.092 rest of plant w/o root 0.14 grain <u>0.066</u> straw <u>0.11</u> grain 0.036 straw 0.061	BASF analytical method No. 567/0 whole plant (without roots): mean recovery = 83.8%; SD: +/- 7.8; CV: 9.3%; n=6; fortification range 0.01-1.0 mg/kg panicle: mean recovery = 77.4%; SD: +/- 7.5; CV: 9.7%; n=5; fortification range 0.01-1.0 mg/kg grain: mean recovery = 70.6%; SD:8.8 +/-; CV: 12.5%; n=5; fortification range 0.01-1.0 mg/kg straw: mean recovery = 81.4%; SD:10.8 +/-; CV: 13.3%; n=5; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 83.0%; SD: +/- 7.7; CV: 9.3%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/2 Study to GLP Study carried out in 2006	Rice (variety Puntal)	Spain Cerro del Parlade Cita 9024 Poligono Industrial Isla Menor-Los Palacios	12.5 g as/ha 2 treatm. last date 10.10.06	83 at last treatm.	0 13 13 20 20 28 28	plant without root 0.350 panicles 0.120 rest of plant w/o root 0.120 grain <u>0.130</u> straw <u>0.091</u> grain 0.076 straw 0.075	
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/3 Study to GLP Study carried out in 2006	Rice (variety Cadet)	Italy Az. Agr. Cantaglia Via Ponticelli 2 Malalbergo Bologna 40058	12.5 g as/ha 2 treatm. last date 20.09.06	87 at last treatm.	0 15 15 22 22 29 29	plant without root 0.320 panicles 0.180 rest of plant w/o root 0.086 grain <u>0.110</u> straw <u>0.190</u> grain 0.084 straw 0.190	BASF analytical method No. 567/0 whole plant (without roots): mean recovery = 83.8%; SD: +/- 7.8; CV: 9.3%; n=6; fortification range 0.01-1.0 mg/kg panicle: mean recovery = 77.4%; SD: +/- 7.5; CV: 9.7%; n=5; fortification range 0.01-1.0 mg/kg grain: mean recovery = 70.6%; SD:8.8 +/-; CV: 12.5%; n=5; fortification range 0.01-1.0 mg/kg straw: mean recovery = 81.4%; SD:10.8 +/-; CV: 13.3%; n=5; fortification range 0.01-1.0 mg/kg rest of plant: mean recovery = 83.0%; SD: +/- 7.7; CV: 9.3%; n=5; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007946 Trial No. AF/10496/BA/4 Study to GLP Study carried out in 2006	Rice (variety Claudio)	Greece Nea Magara Thessaloniki Central Macedonia GR-57300	12.5 g as/ha 2 treatm. last date 09.08.06	72-74	0 14 14 21 21 27 27	plant without root 0.59 panicles 0.14 rest of plant w/o root 0.19 grain <u>0.073</u> straw 0.065 grain 0.054 straw <u>0.13</u>	

_ underlined values were used for MRL calculation

Findings:

The residue studies in rice presented in the alpha-cypermethrin dossier were carried out in 4 different countries and provide data relevant to conditions in the Southern European region.

The proposed GAP is

- for the Southern European region: 1-2 applications with 12.5 g a.s./ha applied at infestation as an overall spray with a PHI of 21 days

The following residue studies were presented:

- one or two treatments at a target rate of 12.5 g a.s./ha-8 trials including both variants, all conducted in the Southern European Region

During the 2005 and 2006 growing seasons, eight field trials were conducted in representative rice growing areas in France (Southern European region), Spain, Italy and Greece.

After one application at 12.5 g a.s./ha, total residues of cypermethrin were 0.063-0.511 mg/kg in rice plant specimens collected immediately after the last application. Total residues were 0.028-0.160 mg/kg and 0.015-0.134 mg/kg, respectively, in rice panicles and the rest of the plant taken 13–15 days after the last application. Total residues were 0.015–0.176 mg/kg and 0.044–0.349 mg/kg, respectively, in grain and straw specimens collected 20-22 days after the last application and 0.020–0.206 mg/kg and 0.021–0.330 mg/kg, respectively, in grain and straw collected after 27–29 days.
After two applications at 12.5 g a.s./ha, total residues of cypermethrin were 0.291–0.590 mg/kg in rice plant specimens collected immediately after the last application. Total residues were 0.092–0.357 mg/kg and 0.086–0.194 mg/kg, respectively, in rice panicles and the rest of the plant taken 13–15 days after the last application. Total residues were <0.01–0.205 mg/kg and 0.065–0.354 mg/kg, respectively, in grain and straw specimens collected 20-22 days after the last application and 0.011–0.151 mg/kg and 0.061–0.351 mg/kg, respectively, in grain and straw collected after 27–29 days.

Conclusion:

During the 2005 and 2006 growing seasons, eight field trials in rice were conducted in representative rice growing areas in France (Southern European region), Spain, Italy and Greece.
In 8 trials supporting the GAP for the Southern European region, total residues were <0.01–0.205 mg/kg and 0.065–0.354 mg/kg, respectively, in grain and straw specimens collected 20-22 days after the last application.

2.29.2 Estimation of MRL, HR and STMR for rice

For *rice*, the following residue studies were considered (BASF DocIDs): 2006/1026848 and 2007/1007946.

The following residue values (PHI=14-21±1 days, or later if higher residue values occurred) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2006/1026848	-	0.011, 0.039, 0.148, 0.206
2007/1007946	-	0,066, 0.073, 0.11, 0.13,
OECD-MRL-calculation	**-**	**<u>0.4</u> (n=8, STMR=0.092, HR=0.206)**

_ underlined values were used for risk assessment purposes

2.30 Wheat

2.30.1 Supervised residue trials in wheat

In the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414; residue studies in wheat were peer-reviewed. Below, residue data from peer-reviewed trials supporting the current GAP and considered for MRL proposal and risk assessment are summarized. New trials are presented in Section M-CA 6.3.6; the MRL calculations are shown in Section M-CA 6.7. An overview on the peer-reviewed studies considered is given below.

Table 2.30.1-1: Number of peer-reviewed residue trials in wheat considered for risk assessment per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Wheat	1983	1	FR	2	FR	3	AL-730-001
Wheat	1984	4	FR	4	FR	8	AL-730-003
Wheat	1992	-	-	1	FR	1	AL-730-029
Wheat	1992	-	-	2	FR	2	AL-730-030
Wheat	1992	1	DE	-	-	1	AL-730-031
Total number of trials per region		**6**		**9**	**Total number of trials**	**15**	

Bosio P. (1983)
Full study reference
Bosio P. (1983): Residues of WL85871 in Wheat From France 1983 Trials
Shell Chemie Berre BEGR.83.061
BASF RDI-No.AL-730-001

Bosio P. (1985)
Full study reference
Bosio P. (1983): Residues of WL85871 in Wheat From France 1984 Trials
Shell Chemie Berre BEGR.85.004
BASF RDI-No.AL-730-003

Carlon R. (1992)
Full study reference
Carlon R. (1992): Residues of Alpha-cypermethrin in cereals from France treated with Fastac PVP – 1992 trials
BASF RDI-No.AL-730-029

Carlon R. (1992)
Full study reference
Carlon R. (1992): Residues of Alpha-cypermethrin in cereals from France treated with Fastac PVP – 1992 trials
BASF RDI-No.AL-730-030

Carlon R. (1993)
Full study reference
Carlon R. (1992): Residues of Alpha-cypermethrin in wheat from Germany treated with Fastac PVP – 1992 trials
BASF RDI-No.AL-730-031

Materials and methods:

Three trials were performed in France (Northern and Southern European region) during the year 1983. Alpha-cypermethrin 100 g a.s./L EC was sprayed on wheat, on a single occasion, diluted with water at a dosage rate of 17.5 g a.s./ha.
Wheat was sampled nearly 38 – 46 days after treatment and analysed for residues of alpha-cypermethrin. No detectable residues were found in any of the grain samples (LOQ 0.01 mg/kg) while residues in straw ranged between 0.17 and 0.44 mg/kg.

Eight trials were performed in France (Northern and Southern European region) in the year 1984.
Alpha-cypermethrin 50 g a.s./L EC was sprayed on wheat, on a single occasion, diluted with water at a concentration in spray of 3 g a.s./hL, corresponding to a dosage rate of 15 g a.s./ha.

Wheat grain and straw was sampled at intervals up to 10 weeks after treatment and analysed for residues of alpha-cypermethrin.
Results showed that in grain no detectable residues were found in any of the samples (all <0.01 mg/kg).
In straw, on average, residues declined from 0.5 mg/kg immediately after treatment to 0.15 mg/kg after 2 weeks and below 0.1 mg/kg at harvest (PHI 6 to 10 weeks).

Three trials in wheat were performed in France (Southern European region) during the year 1992.
Alpha-cypermethrin EC 50 g a.s./L and alpha-cypermethrin PVP 50 g a.s./kg were applied to wheat, on a single occasion, at dosage rates of 15 and 30 g a.s./ha.
Straw and grain were sampled at maturity, 38 - 50 days after treatment and analysed for residues of alpha-cypermethrin.
No detectable residues were found in any of the grain samples (all <0.01 mg/kg).
Residue levels in straw after application of 15 g a.s./ha ranged between 0.02 – 0.75 mg/kg with the EC formulation and between 0.05 – 0.60 mg/kg with the PVP formulation.
One trial in wheat was performed during the year 1992 in Germany. Alpha-cypermethrin EC (100 g a.s./l and alpha-cypermethrin PVP 150 g a.s./kg were applied to wheat, on a single occasion, each formulation at a dosage rate of 15 g a.s./ha. Straw and grain samples were taken at maturity, 34 days after treatment, and analysed for residues of alpha-cypermethrin.
No detectable residues were found in any of the grain samples from both formulations (all <0.01 mg/kg). In straw, low levels of residues were found from both formulations (0.03 mg/kg for PVP and 0.01 mg/kg for EC).
The trial data and residue results are summarized in Table 2.30.1-2.

Table 2.30.1-2: Results of residue trials with alpha-cypermethrin conducted in wheat

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-730-001 BEGR.83.061 Trial No. S/FR/E83/220 Study not to GLP Study carried out in 1983	Winter wheat (variety Talent)	France 01 – Nievroz (South of EU)	17.5 g a.s./ha 07.06.83 Fastac EC 100 g a.s./L	10.51	38 38	grain <u>≤0.01</u> straw <u>0.44</u>	
BASF RDI No. AL-730-001 BEGR.83.061 Trial No. S/FR/E83/419 Study not to GLP Study carried out in 1983	Winter wheat (variety Hardi)	France 24 – Cherval (South of EU)	17.5 g a.s./ha 09.06.83 Fastac EC 100 g a.s./L	10.2	43 43	grain <u>≤0.01</u> straw <u>0.29</u>	SAMS 351-1 Recovery: Wheat grain 100% at 0.10 mg/kg Wheat straw 100% at 0.25 mg/kg
BASF RDI No. AL-730-001 BEGR.83.061 Trial No. S/FR/E83/878 Study not to GLP Study carried out in 1983	Winter wheat (variety Caton)	France 27 – Breuilpont (North of EU)	17.5 g a.s./ha 06.06.83 Fastac EC 100 g a.s./L	10.52	46 46	grain <u>≤0.01</u> straw <u>0.17</u>	
BASF RDI No. AL-730-003 BEGR.85.004 Trial No. S/FE/E84/217 Study not to GLP Study carried out in 1984	Winter wheat (variety talent)	France 69 – Taponas (South of EU)	15 g a.s./ha 05.06.84 Fastac EC 50 g a.s./L	Zd 64	14 28 42 73 0 7 14 28 42 73	grain <0.01 grain <u>≤0.01</u> grain <0.01 grain <0.01 straw 0.66 straw 0.23 straw 0.09 straw 0.08 straw <u>0.12</u> straw 0.12	
BASF RDI No. AL-730-003 BEGR.85.004 Trial No. S/FR/E84/218 Study not to GLP Study carried out in 1984	Winter wheat (variety Talent)	France 69 – Drace (South of EU)	15 g a.s./ha 26.06.84 Fastac EC 50 g a.s./L	Zd 83	0 7 14 21 28 62 0 7 14 21 28 62	grain <0.01 grain <0.01 grain <0.01 grain <0.01 grain <u>≤0.01</u> grain <0.01 straw 0.16 straw 0.12 straw 0.09 straw 0.05 straw 0.06 straw <u>0.13</u>	SAMS 233-1 AL-730-003 is a supplement to document AL-730-004. It reports the same trials, but the residue results are calculated based on dry weight.
BASF RDI No. AL-730-003 BEGR.85.004 Trial No. S/FR/E84/347 Study not to GLP Study carried out in 1984	Winter wheat (variety Top)	France 33 – Salleboeuf (South of EU)	15 g a.s./ha 25.05.84 Fastac EC 50 g a.s./L	Zd 68	28 43 ~71 0 7 14 28 43 ~71	grain <u>≤0.01</u> grain <0.01 grain <0.01 straw 1.1 straw 0.62 straw 0.18 straw <u>0.16</u> straw 0.09 straw 0.03	

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-730-003 BEGR.85.004 Trial No. S/FR/E84/348 Study not to GLP Study carried out in 1984	Winter wheat (variety Top)	France 33 – Salleboeuf (South of EU)	15 g a.s./ha 22.06.84 Fastac EC 50 g a.s./L	Zd 76	7 14 21 28 ~42 0 7 14 21 28 ~42	grain <0.01 grain <0.01 grain <0.01 grain <u>≤0.01</u> grain <0.01 straw 0.15 straw 0.07 straw 0.05 straw 0.04 straw 0.03 straw <u>0.06</u>	SAMS 233-1 AL-730-003 is a supplement to document AL-730-004. It reports the same trials, but the residue results are calculated based on dry weight.
BASF RDI No. AL-730-003 BEGR.85.003 Trial No. S/FR/E84/890 Study not to GLP Study carried out in 1984	Winter wheat (variety Caton)	France 27 – Le Plessis Hebert (North of EU)	15 g a.s./ha 06.06.84 Fastac EC 50 g a.s./L	Zd 60	28 41 0 7 14 28 41	grain <u>≤0.01</u> grain <0.01 straw 0.58 straw 0.25 straw 0.04 straw 0.06 straw <u>0.07</u>	
BASF RDI No. AL-730-003 BEGR.85.003 Trial No. S/FR/E84/891 Study not to GLP Study carried out in 1984	Winter wheat (variety Caton)	France 27 – Le Plessis Hebert (North of EU)	15 g a.s./ha 05.07.84 Fastac EC 50 g a.s./L	Zd 60	7 13 20 0 7 13 20	grain <0.01 grain <0.01 grain <u>≤0.01</u> straw 0.54 straw 0.40 straw 0.36 straw <u>0.29</u>	
BASF RDI No. AL-730-003 BEGR.85.004 Trial No. S/FR/E84/892 Study not to GLP Study carried out in 1984	Winter wheat (variety Rivoli)	France 27 – Le Plessis Hebert (North of EU)	15 g a.s./ha 14.06.84 Fastac EC 50 g a.s./L	Zd 68	28 41 62 0 7 14 28 41 62	grain <u>≤0.01</u> grain <0.01 grain <0.01 straw 0.38 straw 0.17 straw 0.21 straw <u>0.19</u> straw 0.13 straw 0.10	
BASF RDI No. AL-730-003 BEGR.85.004 Trial No. S/FR/E84/893 Study not to GLP Study carried out in 1984	Winter wheat (variety Rivoli)	France 27 – Le Plessis Hebert (North of EU)	15 g a.s./ha 05.07.84 Fastac EC 50 g a.s./L	Zd 80	0 7 13 20 27 42 0 7 13 20 27 42	grain <0.01 grain <0.01 grain <0.01 grain <0.01 grain <u>≤0.01</u> grain <0.01 straw 0.50 straw 0.28 straw 0.18 straw 0.14 straw <u>0.12</u> straw 0.10	
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/239 Study to GLP Study carried out in 1992	Summer wheat (variety Prinkal)	France 69 – Quincieux (South of EU)	15 g a.s./ha 04.06.92 Fastac EC 50 g a.s./L	-	48 48	grain <0.01 straw 0.02	SAMS 351-02 wheat straw: 98.5% at 0.10 mg/kg wheat grain: mean recovery 108.8% (97.5/120%); SD: n/a; CV: n/a; n=2; fortification range 0.05 - 0.10 mg/kg barley straw: 97.5 – 110% at 0.10 mg/kg barley grain: 113% at 0.10 mg/kg Residues analysed as total cypermethrin
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/239 Study to GLP Study carried out in 1992	Summer wheat (variety Prinkal)	France 69 – Quincieux (South of EU)	30 g a.s./ha 04.06.92 Fastac EC 50 g a.s./L	-	48 48	grain <0.01 straw 0.08	

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/239 Study to GLP Study carried out in 1992	Summer wheat (variety Prinkal)	France 69 – Quincieux (South of EU)	15 g a.s./ha 04.06.92 Fastac PVP 50 g a.s./kg	-	48 48	grain ≤0.01 straw 0.05	
BASF RDI No. AL-730-029 BEGR.93.009 Trial No. W/FR/E/92/239 Study to GLP Study carried out in 1992	Summer wheat (variety Prinkal)	France 69 – Quincieux (South of EU)	30 g a.s./ha 04.06.92 Fastac PVP 50 g a.s./kg	-	48 48	grain <0.01 straw 0.06	
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/209 Study to GLP Study carried out in 1992	Winter wheat (variety Darius)	France 38 – Montalieu (South of EU)	15 g a.s./ha 01.06.92 Fastac EC 50 g a.s./L	68	50 50	grain ≤0.01 straw 0.09	SAMS 351-02 wheat straw: 93% at 0.10 mg/kg wheat grain: 98% at 0.10 mg/kg GC/ECD Residues analysed as total cypermethrin
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/209 Study to GLP Study carried out in 1992	Winter wheat (variety Darius)	France 38 – Montalieu (South of EU)	30 g a.s./ha 01.06.92 Fastac EC 50 g a.s./L	68	50 50	grain <0.01 straw 0.30	
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/209 Study to GLP Study carried out in 1992	Winter wheat (variety Darius)	France 38 – Montalieu (South of EU)	15 g a.s./ha 01.06.92 Fastac PVP 50 g a.s./kg	68	50 50	grain <0.01 straw 0.06	
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/209 Study to GLP Study carried out in 1992	Winter wheat (variety Darius)	France 38 – Montalieu (South of EU)	30 g a.s./ha 01.06.92 Fastac PVP 50 g a.s./kg	68	50 50	grain <0.01 straw 0.19	SAMS 351-02 wheat straw: 93% at 0.10 mg/kg wheat grain: 98% at 0.10 mg/kg GC/ECD Residues analysed as total cypermethrin
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/869 Study to GLP Study carried out in 1992	Winter wheat (variety Artaban)	France 01 – Parcay - Meslay (South of EU)	15 g a.s./ha 09.06.92 Fastac EC 50 g a.s./L	74	38 38	grain ≤0.01 straw 0.75	
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/869 Study to GLP Study carried out in 1992	Winter wheat (variety Artaban)	France 01 – Parcay - Meslay (South of EU)	30 g a.s./ha 09.06.92 Fastac EC 50 g a.s./L	74	38 38	grain <0.01 straw 2.00	

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Remarks
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/869 Study to GLP Study carried out in 1992	Winter wheat (variety Artaban)	France 01 – Parcay - Meslay (South of EU)	15 g a.s./ha 09.06.92 Fastac PVP 50 g a.s./kg	74	38 38	grain <0.01 straw 0.60	
BASF RDI No. AL-730-030 BEGR.93.010 Trial No. W/FR/E/92/869 Study to GLP Study carried out in 1992	Winter wheat (variety Artaban)	France 01 – Parcay - Meslay (South of EU)	30 g a.s./ha 09.06.92 Fastac PVP 50 g a.s./kg	74	38 38	grain <0.01 straw 1.60	
BASF RDI No. AL-730-031 BEGR.93.013 Trial No. BE61 Study to GLP Study carried out in 1992	Summer wheat (variety Star)	Germany D-7775 Bermatingen	15 g a.s./ha 03.07.92 Fastac EC 100 g a.s./L	BBCH 75	34 34	grain <u>≤0.01</u> straw 0.01	SAMS 351-02 wheat straw: 87% at 0.10 mg/kg wheat grain: 97% at 0.10 mg/kg GC/ECD Residues analysed as total cypermethrin
BASF RDI No. AL-730-031 BEGR.93.013 Trial No. BE61 Study to GLP Study carried out in 1992	Summer wheat (variety Star)	Germany D-7775 Bermatingen	15 g a.s./ha 03.07.92 Fastac PVP 150 g a.s./kg	BBCH 75	34 34	grain <0.01 straw <u>0.03</u>	SAMS 351-02 wheat straw: 87% at 0.10 mg/kg wheat grain: 97% at 0.10 mg/kg GC/ECD Residues analysed as total cypermethrin

2.31 Sugar Beet

Residue data from supervised trials in sugar beet were used for risk assessment of alpha-cypermethrin. An overview on the studies is given below.

Table 2.31-1: Number of residue trials conducted in sugar beets per geographical region and vegetation period

Crop	Vegetation Period	Number of Trials					Report-No.
		EU North	Country	EU South	Country	Total	
Sugar beet	2002	4	FR	-	-	4	2003/1021716
Sugar beet	2003	4	DE	-	-	4	2004/1006468
Sugar beet	2005	-	-	4	ES, GR, IT	4	2006/1026851
Sugar beet	2006	-	-	4	FR, GR, IT	4	2007/1007941
Total number of trials per region		**8**		**8**	**Total number of trials**	**16**	

2.31.1 Supervised residue trials in sugar Beet

Harrison C (2003)

Full study reference

Harrison C (2003): To determine the magnitude of alpha-cypermethrin residues at harvest in the raw agricultural commodity sugar beet resulting from sequential directed application of Mageos MD, in Northern France 2002-2003; BASF DocID 2003/1021716

Raunft E, Rabe U, Mackenroth C (2004)

Full study reference

Raunft E, Rabe U, Mackenroth C (2004): Study on the residue behaviour of alpha-cypermethrin in sugar beets after application of BAS 310 031 and BAS 310 QC I under field conditions in Germany, 2003; BASF DocID 2004/1006468

Jones G (2007)

Full study reference

Jones G (2007): Study on the residue behaviour of alpha-cypermethrin in sugar beet after treatment with BAS 310 40 I under field conditions in Southern Europe during 2005; BASF DocID 2006/1026851

Oxspring S (2007)

Full study reference

Oxspring S (2007): Study on the residue behaviour of alphacypermethrin in sugar beet after treatment with BAS 310 40 I under field conditions in Southern Europe during 2006; BASF DocID 2007/1007941

Material and Methods:

A field program on sugar beets was conducted in the years 2002, 2003, 2005 and 2006 in France (Northern and Southern European region), Germany, Greece, Italy, and Spain.

Four residue trials were conducted on sugar beet during 2002, in Northern France. Two applications of alpha-cypermethrin, formulated as a wettable granule (WG 150 g/kg; BAS 310 08 I) were applied at a target rate of 7.5 g a.s./ha diluted with water immediately prior to application to a spray volume of 200 L/ha. The treatments were performed at growth stage 39 (Crop cover complete: leaves cover 90% of ground). Specimens of crop from the untreated and treated plots were taken by hand at normal commercial harvest.
Crop specimens were analysed for residues of alpha-cypermethrin by means of GC-MS using Agrisearch Method 'Alphacypermethrin/Crops/KLS/03/1', corresponding to the Cyanamid method SAMS 351-2 with a limit of quantitation (LOQ) of 0.01 mg/kg.

During the 2003 growing season, a total of 4 trials was conducted in sugar beets. BAS 310 03 I and BAS 310 QC I, both soluble concentrate formulations of alpha-cypermethrin (SC; 100 g a.s./L) were foliar applied once at a rate of 10 g a.s./ha to sugar beets. The treatment was done at growth stage 18 (8 leaves unfolded). Sugar beet whole plant specimens were collected directly after the last application from each plot at all locations. Sugar beet leaves (with tops) and roots were taken at growth stages 39 (crop cover complete, 14-30 days after application) and 49 (beet-root has reached harvestable size, 98-140 days after application).
The specimens were analysed for alpha-cypermethrin by means of HPLC-MS/MS with BASF analytical method No. 546/0 which has a LOQ of 0.05 mg/kg in all sample materials.

In the years 2005 and 2006, eight residue trials were performed in the Southern European region to investigate the residue behaviour of alpha-cypermethrin after application of a 100 g a.s./L emulsifiable concentrate formulation (EC; BAS 310 40 I). The product was foliar applied at a target rate of 25 g a.s./ha 14±1 days before harvest at growth stage 38 (leaves cover 80% of ground) to 49 (beet root has reached harvestable size). Samples of roots and leaves with tops were collected 0, 7, 14 and 21 days after application.
Residue analysis was performed by means HPLC-MS/MS according to BASF analytical method No. 567/0 with a LOQ of 0.01 mg/kg.

The trial data and residue results are summarised in Table 2.31.1-1.

Table 2.31.1-1: Residues in sugar beets

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2003/1021716 Trial No. AF/6792/BA/1 Study to GLP Study carried out in 2002	Sugar beet (variety Angelina)	France 45300 Manchecourt Dossainville (North of EU)	7.5/7.7 g a.s./ha 2 treatm. last date 27.06.02	39 at last treatm.	92	roots <0.01 tops <0.01	Agrisearch analytical method: Alphacypermethrin/Crops/KLS/03/1 (corresponding to method SAMS 351-2) roots, tops: mean recovery = 74%; SD: +/- 21.1; CV: 28.5%; n=4; fortification range 0.01-0.25 mg/kg
BASF Doc ID 2003/1021716 Trial No. AF/6792/BA/2 Study to GLP Study carried out in 2002	Sugar beet (variety Crocodile)	France 45300 Douzonville, Manchecourt (North of EU)	7.6/7.7 g a.s./ha 2 treatm. last date 27.06.02	39 at last treatm	92	roots <0.01 tops <0.01	
BASF Doc ID 2003/1021716 Trial No. AF/6792/BA/3 Study to GLP Study carried out in 2002	Sugar beet (variety Angelina)	France 45300 Yevre-la-Ville (North of EU)	7.5/7.4 g a.s./ha 2 treatm. last date 27.06.02	39 at last treatm	88	roots <0.01 tops <0.01	
BASF Doc ID 2003/1021716 Trial No. AF/6792/BA/4 Study to GLP Study carried out in 2002	Sugar beet (variety Rafole)	France 45300 Thignonville (North of EU)	7.8/7.3 g a.s./ha 2 treatm. last date 27.06.02	39 at last treatm	84	roots <0.01 tops <0.01	

Table 2.31.1-1: Residues in sugar beets

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2004/1006468 Trial No. ACK/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Milan)	Germany 16833 Brunne Brandenburg	10 g a.s./ha 30.05.03 100 g a.s./L SC 310 03 I	18	0 25 25 108 108	Plants 0.446 leaves w.tops <u>≤0.05</u> roots <u>≤0.05</u> leaves w.tops <0.05 roots <0.05	
BASF Doc ID 2004/1006468 Trial No. AGR/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Dorena)	Germany 47574 Goch-Nierswalde Nordrhein-Westfalen	10 g a.s./ha 06.06.03 100 g a.s./L SC 310 03 I	18	0 14 14 98 98	Plants 0.273 leaves w.tops <u>≤0.05</u> roots <u>≤0.05</u> leaves w.tops <0.05 roots <0.05	BASF analytical method No. 546/0: plants, roots, leaves (with tops): mean recovery = 94.1%; SD: +/- 9.0; CV: 9.6%; n=6; fortification range 0.05-0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2004/1006468 Trial No. DU2/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Tatjana)	Germany 74193 Stetten a. H. Baden-Württemberg	10 g a.s./ha 23.05.03 100 g a.s./L SC 310 03 I	18	0 21 21 131 131	Plants 0.138 leaves w.tops <u>0.292</u> roots <u>0.228</u> leaves w.tops <0.05 roots <0.05	
BASF Doc ID 2004/1006468 Trial No. DU4/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Tatjana)	Germany 67229 Gerolsheim Rheinland-Pfalz	10 g a.s./ha 14.05.03 100 g a.s./L SC 310 03 I	18	0 30 30 140 140	Plants 0.208 leaves w.tops <u>≤0.05</u> roots <u>≤0.05</u> leaves w.tops <0.05 roots <0.05	BASF analytical method No. 546/0: plants, roots, leaves (with tops): mean recovery = 94.1%; SD: +/- 9.0; CV: 9.6%; n=6; fortification range 0.05-0.5 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2004/1006468 Trial No. ACK/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Milan)	Germany 16833 Brunne Brandenburg	10 g a.s./ha 30.05.03 100 g a.s./L SC 310 QC I	18	0 25 25 108 108	Plants 0.312 leaves w.tops <0.05 roots <0.05 leaves w.tops <0.05 roots <0.05	
BASF Doc ID 2004/1006468 Trial No. AGR/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Dorena)	Germany 47574 Goch-Nierswalde Nordrhein-Westfalen	10 g a.s./ha 06.06.03 100 g a.s./L SC 310 QC I	18	0 14 14 98 98	Plants 0.334 leaves w.tops 0.070 roots <0.05 leaves w.tops <0.05 roots <0.05	
BASF Doc ID 2004/1006468 Trial No. DU2/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Tatjana)	Germany 74193 Stetten a. H. Baden-Württemberg	10 g a.s./ha 23.05.03 100 g a.s./L SC 310 QC I	18	0 21 21 131 131	Plants 0.098 leaves w.tops <0.05 roots <0.05 leaves w.tops <0.05 roots <0.05	
BASF Doc ID 2004/1006468 Trial No. DU4/02/03 Study to GLP Study carried out in 2003	Sugar beet (variety Tatjana)	Germany 67229 Gerolsheim Rheinland-Pfalz	10 g a.s./ha 14.05.03 100 g a.s./L SC 310 QC I	18	0 30 30 140 140	Plants 0.335 leaves w.tops <0.05 roots <0.05 leaves w.tops <0.05 roots <0.05	

Table 2.31.1-1: Residues in sugar beets

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2006/1026851 Trial No. AF/8832/BA/1 Study to GLP Study carried out in 2005	Sugar beet (variety)	Italy Budrio, 40054 Bologna	25 g a.s./ha 26.07.05	39-49	0 0 7 7 14 14 21 21	roots <0.01 leaves with tops 0.724 roots <u><0.01</u> leaves with tops <u>0.227</u> roots <0.01 leaves with tops 0.159 roots <0.01 leaves with tops 0.045	BASF analytical method No. 567/0 roots: mean recovery = 94.4% (87.1/101.6%); SD: n/a; CV: n/a; n=2; fortification range 0.01-0.1 mg/kg tops (leaves and stem): mean recovery = 94.5%; SD: +/- 25.9; CV: 27.4%; n=3; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026851 Trial No. AF/8832/BA/2 Study to GLP Study carried out in 2005	Sugar beet (variety)	Spain Coris del Rio 41100 Seville	25 g a.s./ha 23.06.05	49	0 0 7 7 14 14 21 21	roots <0.01 leaves with tops 0.482 roots <u><0.01</u> leaves with tops <u>0.079</u> roots <0.01 leaves with tops 0.059 roots <0.01 leaves with tops 0.020	
BASF Doc ID 2006/1026851 Trial No. AF/8832/BA/3 Study to GLP Study carried out in 2005	Sugar beet (variety)	Spain Las Marismas de Lebrija 30188	25 g a.s./ha 28.06.05	49	0 0 7 7 14 14 21 21	roots <0.01 leaves with tops 0.462 roots <u><0.01</u> leaves with tops 0.035 roots <0.01 leaves with tops <u>0.048</u> roots <0.01 leaves with tops <0.01	BASF analytical method No. 567/0 roots: mean recovery = 94.4% (87.1/101.6%); SD: n/a; CV: n/a; n=2; fortification range 0.01-0.1 mg/kg tops (leaves and stem): mean recovery = 94.5%; SD: +/- 25.9; CV: 27.4%; n=3; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2006/1026851 Trial No. AF/8832/BA/4 Study to GLP Study carried out in 2005	Sugar beet (variety)	Greece Thessaloniki Central Macedonia GR-57300	25 g a.s./ha 18.08.05	47	0 0 7 7 14 14 21 21	roots <0.01 leaves with tops 0.220 roots <u><0.01</u> leaves with tops <u>0.152</u> roots <0.01 leaves with tops 0.073 roots <0.01 leaves with tops 0.052	
BASF Doc ID 2007/1007941 Trial No. AF/10501/BA/1 Study to GLP Study carried out in 2006	Sugar beet (variety Lactitiae)	France, La Creuze Mizerieux; Loire (South of EU)	25 g a.s./ha 12.09.06	39	0 0 7 7 14 14 21 21	tops 0.767 roots 0.012 tops <u>0.103</u> roots <u><0.01</u> tops 0.055 roots <0.01 tops 0.045 roots <0.01	BASF analytical method No. 567/0 roots: mean recovery = 104.3%; SD: +/- 8.9; CV: 8.5%; n=4; fortification range 0.01-0.1 mg/kg tops: mean recovery = 81 5%; SD: +/- 14.8; CV: 18.2%; n=4; fortification range 0.01-1.0 mg/kg Residue analysed as total cypermethrin
BASF Doc ID 2007/1007941 Trial No. AF/10501/BA/2 Study to GLP Study carried out in 2006	Sugar beet (variety Rizor)	Italy Castel S. Pietro Poggio Piccoco	25 g a.s./ha 07.08.06	38	0 0 7 7 14 14 21 21	tops 0.777 roots <0.01 tops <u>0.173</u> roots <u><0.01</u> tops 0.072 roots <0.01 tops 0.053 roots <0.01	

Table 2.31.1-1: Residues in sugar beets

GAP for EU-N is 1-2 applications with 15 g a.s./ha at infestation as an overall spray, PHI = 14 days
GAP for EU-S is 1 application with 30 g a.s./ha at infestation as an overall spray, PHI = 7 days

GLP and trial details	Crop	Country	Application rate g a.s./ha	Crop growth stage	PHI (days)	Residues found (mg/kg)	Recovery data
BASF Doc ID 2007/1007941 Trial No. AF/10501/BA/3 Study to GLP Study carried out in 2006	Sugar beet (variety Ornelia)	Italy, Budrio Prunaro; Bologna	25 g a.s./ha 04.08.06	39	0 0 7 7 14 14 21 21	tops 0.487 roots <0.01 tops <u>0.236</u> roots <u><0.01</u> tops 0.093 roots <0.01 tops 0.058 roots <0.01	
BASF Doc ID 2007/1007941 Trial No. AF/10501/BA/4 Study to GLP Study carried out in 2006	Sugar beet (variety Doria)	Greece Galatades Pella Central Macedonia	25 g a.s./ha 03.08.06	43-44	0 0 7 7 14 14 21 21	tops 0.376 roots <0.01 tops <u>0.075</u> roots <u><0.01</u> tops 0.026 roots <0.01 tops 0.022 roots <0.01	

_ underlined values were used for MRL calculation

Findings:

The residue data presented in the alpha-cypermethrin dossier were carried out in 5 different EU countries and provide data relevant to conditions in the Northern and Southern European region.
The proposed GAP is

- for the Northern European region: 1-2 applications with 15 g a.s./ha applied at infestation as an overall spray with a PHI of 14 days
- for the Southern European region: a single application with 30 g a.s./ha applied at infestation as an overall spray with a PHI of 7 days

The following residue studies were presented:

- two treatments at a target rate of 7.5 g a.s./ha-4 trials conducted in the Northern European region
- one treatment at a target rate of 10 g a.s./ha-4 trials each including two variants with different SC formulations, all conducted in the Northern European region
- one treatment at a target rate of 25 g a.s./ha-8 trials, all conducted in the Southern European region

No residues above the limit of quantitation of the analytical method (LOQ) were found after two treatments at a target rate of 7.5 g a.s./ha, applied at growth stage 18 in tops or roots sampled at normal harvest (PHI 84-92 days). Residues in all samples were <0.01 mg/kg.

Directly after application of BAS 310 I at a target rate of 10 g a.s./ha, alpha-cypermethrin residues in whole plant samples ranged between 0.138 and 0.446 mg/kg in case of BAS 310 03 I and between 0.098 and 0.335 mg/kg with BAS 310 QC I.
At the second sampling which took place after 14 to 30 days, alpha-cypermethrin was found in sugar beet tops at a level of <0.05 in three trials of variant BAS 310 03 I and between <0.05 and 0.070 mg/kg in variant BAS 310 QC I. In one trial of variant 310 03 I, residues in sugar beet tops were 0.292 mg/kg 21 days after application (trial DU2/02/03). At the same sampling date, residues in roots were 0.228 mg/kg in the same trial, while all other treated root samples did not show residues above the LOQ of the analytical method (all <0.05 mg/kg), regardless of sampling interval. Residues in sugar beet tops were below the LOQ at the last sampling, 98–131 days after application.

In eight trials that were performed in the Southern European region, one treatment at a target rate of 25 g a.s./ha was applied. Initial residues in sugar beet leaves with tops ranged between 0.220–0.777 mg/kg. Residues declined rapidly to 0.035–0.236 mg/kg, 0.026–0.159 mg/kg and <0.01–0.058 mg/kg 7, 14 and 21 days after application, respectively.
In sugar beet roots, there was one isolated finding of a residue slightly above the LOQ immediately after application (0.012 mg/kg; trial AF/10501/BA/1). In all other treated root samples no residues above the LOQ were found (all <0.01 mg/kg).

Conclusion:

A field program on sugar beets was conducted in the years 2002, 2003, 2005 and 2006 in France (Northern and Southern region), Germany, Greece, Italy, and Spain. In eight trials that support the proposed GAP for the Southern European region, residues in sugar beet leaves with tops ranged between 0.035–0.236 mg/kg at the target PHI of 7 days, while no residues above the LOQ (0.01 mg/kg) were found in sugar beet roots at the same time point.

2.31.2 Estimation of MRL, HR and STMR for sugar beet

For *sugar beet*, the following residue studies were considered (BASF DocIDs): 2003/1021716, 2004/1006468, 2006/1026851 and 2007/1007941.

The following residue values (PHI=14-92 days for EU-N and PHI=7 days for EU-S) were taken for STMR/HR/MRL calculation using the OECD MRL calculator:

Study report (BASF DocID)	Northern Europe (N-EU) [mg/kg]	Southern Europe (S-EU) [mg/kg]
2003/1021716	<0.01 (4x)	-
2004/1006468	<0.05 (3x), 0,228	-
2006/1026851	-	<0.01 (4x)
2007/1007941	-	<0.01 (4x)
OECD-MRL-calculation	**0.4 (n=8, STMR=0.03, HR=0.228)**	**<u>0.01</u> (n=8, STMR=0.01, HR=0.01)**

_ underlined values were used for risk assessment purposes

3 Dietary exposure assessment

3.1 Alpha-cypermethrin

The relevant calculations for parent alpha-cypermethrin are included in chapter CA 06.09.

3.2 Metabolites (TTC)

The following metabolites of alpha-cypermethrin were considered to be addressed in the dietary exposure calculations (see also Document N3):

Table 3.2-1: Overview: Relevant Metabolites of Alpha-Cypermethrin for Risk Assessment

Metabolite
M310I001
M310I003
M310I004
M310I005
M310I006
M310I007
M310I008
M310I009
M310I010
M310I011
M310I013
M310I017
M310I018
M310I019
M310I021
M310I024
M310I025
M310I026

In a first step the parent/metabolite ratios were derived from the respective metabolism studies as described in chapter CA 06.09.

Plant metabolism studies (see chapter CA 06.02 → cabbage: DocID AL-640-001, lettuce: DocID 2012/1084223, wheat: DocID AL-640-004)

Cabbage

The following TRR values were reported:

Table 3.2-2: Metabolism Study Cabbage: TRR Values (Stalk - B = worst case)

Metabolite	TRR [mg/kg]
Parent	0.1
M310I011	0.05
M310I013	0.05
M310I017	0.05
M310I024	0.05

Based on the molecular weights, the following parent/metabolite ratios were obtained:

Table 3.2-3: Cabbage: Parent/Metabolite Ratios Considering the Molecular Weights

Metabolite	Parent/Metabolite Ratio
Parent	1
M310I011	0.26
M310I013	0.28
M310I017	0.52
M310I024	0.24

When now assuming, that a cabbage head consists of about 45% outer leaves (old), about 45% outer leaves (new) and about 10% stalk, the following refinement can be made:

Considering the worst case parent/metabolite ratios (see Table 4 of DocID AL-640-001 → outer leaves (old) - B: **8.6 mg/kg** vs. **0.6 mg/kg**, outer leaves (new) - A: **0.4 mg/k**g vs. **<0.05** mg/kg, stalk - B: **0.1 mg/kg** vs. **0.05 mg/kg**) leads to a combined "cabbage refinement factor" of 0.14 ((45 x 0.07 + 45 x 0.125 + 10 x 0.5)/100).

When now also considering the molecular weights, the following parent/metabolite ratios were calculated:

Table 3.2-4: Cabbage: Parent/Metabolite Ratios (Based on a Combined "Cabbage Refinement Factor" of 0.14) Considering the Molecular Weights

Metabolite	Parent/Metabolite Ratio
Parent	1
M310I011	0.07
M310I013	0.08
M310I017	0.15
M310I024	0.07

Lettuce

The following TRR values were reported:

Table 3.2-5: Metabolism Study Lettuce: TRR Values

Metabolite	TRR [%]			
	3 DALA (Benzyl)	**3 DALA (Cyclo)**	**7 DALA (Benzyl)**	**7 DALA (Cyclo)**
Parent	**93.3**	**98.0**	**88.8**	**92.6**
M310I005	**3.1**		2.3	
M310I006	**3.1**		2.3	
M310I007	2.4		**4.9**	
M310I008		3.1		**5.9**
M310I009		3.1		**5.9**

TRR values being relevant for the most critical ratio were highlighted in bold

Based on the molecular weights, the following parent/metabolite ratios were obtained:

Table 3.2-6: Lettuce: Parent/Metabolite Ratios Considering the Molecular Weights

Metabolite	Parent/Metabolite Ratio
Parent	1
M310I005	0.031
M310I006	0.032
M310I007	0.057
M310I008	0.057
M310I009	0.070

Wheat

The following TRR values were reported:

Table 3.2-7: Metabolism Study Wheat: TRR Values

Metabolite	TRR [%]					
	Wheat forage (0DAT-1) (Benzyl)	**Wheat forage (0DAT-1) (Cyclo)**	**Wheat forage (7DAT-1) (Benzyl)**	**Wheat forage (7DAT-1) (Cyclo)**	**Wheat forage (0DAT-2) (Benzyl)**	**Wheat forage (0DAT-2) (Cyclo)**
Parent	96.2	98.7	91.6	95.8	87.6	87.8
M310I017					2.3	3.8
M310I011					0.5	
M310I018					**1.2**	
M310I013						
M310I026						
M310I024						
M310I010						
M310I001				1.0		0.5
trans-DCVA						

continued

Metabolite	TRR [%]					
	Hay (21DAT-2) (Benzyl)	**Hay (21DAT-2) (Cyclo)**	**Straw (42DAT-2) (Benzyl)**	**Straw (42DAT-2) (Cyclo)**	**Grain (42DAT-2) (Benzyl)**	**Grain (42DAT-2) (Cyclo)**
Parent	56.7	65.3	58.0	71.1	60.3	22.5
M310I017	3.3	3.7	3.5	**4.8**		
M310I011	**0.7**		0.7			
M310I018	0.7		0.3			
M310I013	0.3		**0.6**			
M310I026			**0.2**			
M310I024			**0.2**			
M310I010			**0.3**			
M310I001		0.6		**2.3**		
trans-DCVA				**1.5**		

TRR values being relevant for the most critical ratio were highlighted in bold

Based on the molecular weights, the following parent/metabolite ratios were obtained:

Table 3.2-8: Wheat: Parent/Metabolite Ratios Considering the Molecular Weights

Metabolite	Parent/Metabolite Ratio					
	Wheat forage (0DAT-1) (Benzyl)	Wheat forage (0DAT-1) (Cyclo)	Wheat forage (7DAT-1) (Benzyl)	Wheat forage (7DAT-1) (Cyclo)	Wheat forage (0DAT-2) (Benzyl)	Wheat forage (0DAT-2) (Cyclo)
Parent	1.0	1.0	1.0	1.0	1.0	1.0
M310I017						
M310I011						
M310I018					0.007	
M310I013						
M310I026						
M310I024						
M310I010						
M310I001						
trans-DCVA (assigned to M310I001)						

continued

Metabolite	Parent/Metabolite Ratio					
	Hay (21DAT-2) (Benzyl)	Hay (21DAT-2) (Cyclo)	Straw (42DAT-2) (Benzyl)	Straw (42DAT-2) (Cyclo)	Grain (42DAT-2) (Benzyl)	Grain (42DAT-2) (Cyclo)
Parent	1.0	1.0	1.0	1.0	1.0	1.0
M310I017				0.07*		
M310I011		0.006*				
M310I018						
M310I013			0.006*			
M310I026			0.003			
M310I024			0.002*			
M310I010			0.003			
M310I001				0.016		
trans-DCVA (assigned to M310I001)				0.011		

* in context of the cabbage metabolism study a higher ratio was determined

Animal metabolism studies (see chapter CA 06.02 → laying hens: DocID 2014/1140293, lactating ruminants: DocID 2014/1083330)

Laying hens

The following TRR values were reported:

Table 3.2-9: Metabolism Study Laying Hens: TRR Values

Metabolite	TRR [%]					
	Breast muscle (Benzyl)	Breast muscle (Cyclo)	Thigh muscle (Benzyl)	Thigh muscle (Cyclo)	Egg yolk (Benzyl)	Egg yolk (Cyclo)
Parent	45.2	**9.0**	60.5	20.9	41.6	34.3
M310I019						
M310I001		**23.7**		16.8	3.2	
M310I003						
continued						
Metabolite	**TRR [%]**					
	Egg white (Benzyl)	Egg white (Cyclo)	Liver (Benzyl)	Liver (Cyclo)	Poolet fat (Benzyl)	Pooled fat (Cyclo)
Parent		**9.9**	**7.4**		62.6	45.4
M310I019			**15.2**			
M310I001		**18.1**		**57.7**		
M310I003		**9.2**		**2.9**		

TRR values being relevant for the most critical ratio were highlighted in bold

The following parent/metabolite ratios were obtained (molecular weights were **not** considered - this represents a worst case scenario as molecular weights of all three metabolites are lower than the one of the parent molecule, which would lead to even lower ratios):

Table 3.2-10: Laying hens: Parent/Metabolite Ratios

Metabolite	Parent/Metabolite Ratio					
	Breast muscle (Benzyl)	Breast muscle (Cyclo)	Thigh muscle (Benzyl)	Thigh muscle (Cyclo)	Egg yolk (Benzyl)	Egg yolk (Cyclo)
Parent	1.0	1.0	1.0	1.0	1.0	1.0
M310I019						
M310I001		**2.63**		0.8	0.08	
M310I003						
continued						
Metabolite	**Parent/Metabolite Ratio**					
	Egg white (Benzyl)	Egg white (Cyclo)	Liver (Benzyl)	Liver (Cyclo)	Poolet fat (Benzyl)	Pooled fat (Cyclo)
Parent		1.0	1.0		1.0	1.0
M310I019			**2.05**			
M310I001		**1.83**		**0.01***		
M310I003		**0.93**		**0.00054****		

Most critical ratios were highlighted in bold

* as no parent value is available, as worst case for M310I001 (DCVA) the current feed burden level was compared to the feeding levels of the hen residue transfer study (DocID CY-870-018, Table 3) and the obtained residue values for DCVA. Due to a current feed burden level close to the 0.4 mg/kg-dose rate, a DCVA residue value of 0.01 mg/kg liver is considered to be relevant (this is an absolute value which will not be multiplied with STMR or HR values)

** this value was calculated by applying the TRR ratio of M310I001/M310I003 on the M310I001 residue value of 0.01 mg/kg liver considering the different molecular weights (this is an absolute value which will not be multiplied with STMR or HR values).

Lactating ruminants

The following TRR values were reported:

Table 3.2-11: Metabolism Study Lactating ruminants: TRR Values

Metabolite	TRR [%] - Benzyl					
	Milk	Omental Fat	Subcut. Fat	Renal Fat	Kidney	Liver
Parent	**51.9**	77.3	74.5	83.8	**4.9**	**4.8**
M310I010	**29.6**				**38.0**	**3.2**
M310I011					**16.6**	**1.8**
M310I021						**7.4**
continued						
Metabolite	**TRR [%] - Cyclo**					
	Milk 55-96h	Milk 103-127h	Fat Goat 3	Fat Goat 4	Muscle	Kidney	Liver
Parent	80.50	74.00	**71.20**	89.80	**65.60**	**1.20**	**2.70**
M310I004						**16.80**	**2.50**
M310I001			**2.90**	1.90	**4.10**	**9.60**	**4.20**
M310I003						**3.50**	
M310I021							**4.70**

TRR values being relevant for the most critical ratio were highlighted in bold

Based on the molecular weights, the following parent/metabolite ratios were obtained:

Table 3.2-12: Lactating Ruminants: Parent/Metabolite Ratios Considering the Molecular Weights

Metabolite	Parent/Metabolite Ratio - Benzyl					
	Milk	Omental Fat	Subcut. Fat	Renal Fat	Kidney	Liver
Parent	1.0	1.0	1.0	1.0	1.0	1.0
M310I010	**0.37**				**5.06**	**0.44**
M310I011					**1.75**	**0.19**
M310I021						**2.25**

continued

Metabolite	Parent/Metabolite Ratio - Cyclo						
	Milk 55-96h	Milk 103-127h	Fat Goat 3	Fat Goat 4	Muscle	Kidney	Liver
Parent	1.0	1.0	1.0	1.0	1.0	1.0	1.0
M310I004						**12.95**	**0.86**
M310I001			**0.02**	0.01	**0.03**	**4.03**	**0.78**
M310I003						**1.58**	
M310I021							**2.55**

Most critical ratios were highlighted in bold

Summarizing, the following parent/metabolite ratios for plant and animal commodities were used for calculation purposes in context of the chronic and acute assessment.

Table 3.2-13: Final Parent/Metabolite Ratios for the Calculation of Chronic and Acute Dietary Risk Assessment

Metabolite	Input value								
	all plants	ruminant muscle	ruminant liver	ruminant kidney	ruminant fat	poultry muscle	poultry liver	milk	eggs
M310I001	0.016	0.03	0.78	4.03	0.02	2.63	0.01*		1.83
M310I003				1.58			0.00054*		0.93
M310I004			0.86	12.95					
M310I005	0.031								
M310I006	0.032								
M310I007	0.057								
M310I008	0.057								
M310I009	0.07								
M310I010	0.003		0.44	5.06				0.37	
M310I011	0.07		0.19	1.75					
M310I013	0.006								
M310I017	0.15								
M310I018	0.007								
M310I019							2.05		
M310I021			2.55						
M310I024	0.07								
M310I025	0.08								
M310I026	0.003								

* absolute value which will not be multiplied with STMR or HR values

In a second step, the most critical STMR/HR of Table 6.9-2 of chapter CA 06.09 (resp. Table 5-1 of this document) per relevant crop were identified to obtain the final STMRs/HRs to be multiplied with the input value from Table 3.2-13.

To facilitate replicability, in the following table the identified most critical STMR/HR are shown:

Table 3.2-14: Final STMR/HR Values for the Calculation of Chronic and Acute Dietary Risk Assessment

Commodity	STMRs [mg/kg]	HRs [mg/kg]
Table grapes	0.049	0.069
Wine grapes	0.049	0.069
Strawberries	0.020	0.054
Table olives	0.050	0.260
Potatoes	0.010	0.010
Carrots	0.010	0.010
Celeriac	0.010	0.010
Horseradish	0.010	0.010
Parsley root	0.010	0.010
Radishes	0.010	0.010
Salsify	0.010	0.010
Swedes	0.010	0.010
Turnips	0.010	0.010
Onions	0.010	0.010
Shallots	0.010	0.010
Tomatoes	0.016	0.037
Peppers	0.018	0.033
Aubergines (egg plants)	0.016	0.037
Cucumbers	0.014	0.037
Courgettes	0.014	0.037
Melons	0.014	0.048
Pumpkins	0.014	0.048
Broccoli	0.030	0.047
Cauliflower	0.010	0.085
Brussels sprouts	0.018	0.046
Head cabbage	0.034	0.105
Leafy brassica	0.367	0.590
Chinese cabbage	0.367	0.590
Kale	0.367	0.590
Other leafy brassica	0.367	0.590
Lettuce and other salad plants including Brassicacea	0.195	0.722
Lamb's lettuce	0.195	0.722
Lettuce	0.195	0.722
Scarole (broad-leaf endive)	0.195	0.722
Cress	0.195	0.722
Land cress	0.195	0.722
Rocket, Rucola	0.195	0.722
Red mustard	0.195	0.722

Commodity	STMRs [mg/kg]	HRs [mg/kg]
Leaves and sprouts of Brassica spp	0.195	0.722
Other lettuce and other salad plants	0.195	0.722
Spinach	0.465	1.110
Purslane	0.465	1.110
Beet leaves (chard)	0.465	1.110
Other spinach and similar	0.465	1.110
Beans (with pods)	0.034	0.063
Beans (without pods)	0.010	0.010
Peas (with pods)	0.034	0.063
Peas (without pods)	0.010	0.010
Asparagus	0.010	0.010
Celery	0.220	0.300
Globe artichokes	0.023	0.040
Leek	0.047	0.105
PULSES, DRY	0.010	0.042
Beans	0.010	0.042
Lentils	0.010	0.042
Peas	0.010	0.042
Lupins	0.010	0.042
Other pulses, dry	0.010	0.042
Rape seed	0.010	0.060
Mustard seed	0.010	0.060
Cotton seed	0.010	0.018
Safflower	0.000	0.050
Borage	0.000	0.050
Olives for oil production	0.050	0.260
Barley	0.035	0.083
Maize	0.010	0.010
Oats	0.035	0.083
Rice	0.092	0.206
Rye	0.010	0.010
Wheat	0.010	0.010
Sugar beet (root)	0.030	0.228
Swine: Meat	0.050	0.050
Swine: Fat free of lean meat	0.050	0.050
Swine: Liver	0.050	0.050
Swine: Kidney	0.050	0.050
Swine: Edible offal	0.050	0.050
Other swine products	0.050	0.050
Bovine: Meat	0.050	0.050
Bovine: Fat	0.100	0.100
Bovine: Liver	0.050	0.050
Bovine: Kidney	0.050	0.050
Bovine: Edible offal	0.050	0.050
Other bovine products	0.050	0.050
Sheep: Meat	0.050	0.050
Sheep: Fat	0.100	0.100
Sheep: Liver	0.050	0.050

Commodity	STMRs [mg/kg]	HRs [mg/kg]
Sheep: Kidney	0.050	0.050
Sheep: Edible offal	0.050	0.050
Other sheep products	0.050	0.050
Goat: Meat	0.050	0.050
Goat: Fat	0.100	0.100
Goat: Liver	0.050	0.050
Goat: Kidney	0.050	0.050
Goat: Edible offal	0.050	0.050
Other goat products	0.050	0.050
Horse: Meat	0.050	0.050
Horse: Fat	0.100	0.100
Horse: Liver	0.050	0.050
Horse: Kidney	0.050	0.050
Horse: Edible offal	0.050	0.050
Other horse products	0.050	0.050
Poultry: Meat	0.050	0.050
Poultry: Fat	0.050	0.050
Poultry: Liver	0.050	0.050
Poultry: Kidney	0.050	0.050
Poultry: Edible offal	0.050	0.050
Other poultry products	0.050	0.050
Other farm animals	0.050	0.050
Other farm animals: Meat	0.050	0.050
Other farm animals: Fat	0.050	0.050
Other farm animals: Liver	0.050	0.050
Other farm animals: Kidney	0.050	0.050
Other farm animals: Edible offal	0.050	0.050
Other other farm animal products	0.050	0.050
Milk and cream, not concentrated, nor containing added sugar or sweetening matter, butter and other fats derived from milk, cheese and curd	0.010	0.010
Milk and milk products: Cattle	0.010	0.010
Milk and milk products: Sheep	0.010	0.010
Milk and milk products: Goat	0.010	0.010
Milk and milk products: Horse	0.010	0.010
Other milk and milk products	0.010	0.010
Birds' eggs	0.010	0.010
Eggs: Chicken	0.010	0.010

Commodity	STMRs [mg/kg]	HRs [mg/kg]
Eggs: Duck	0.010	0.010
Eggs: Goose	0.010	0.010
Eggs: Quail	0.010	0.010
Other eggs	0.010	0.010

3.2.1 Chronic Assessments

All chronic assessments were conducted using the EFSA PRIMo model (version 2). Chronic assessments were run against the trigger values 0.0000025 mg/kg bw (genotoxic), 0.0003 mg/kg bw (neurotoxic) and 0.0015 mg/kg bw (Cramer class III).

3.2.1.1 Chronic assessments applying the genotox-trigger value

The input values (parent/metabolite ratios) of Table 3.2-13 were multiplied with the STMRs of Table 3.2-14 and run against the trigger value 0.0000025 mg/kg bw using the EFSA PRIMo model (version 2).

In Table 3.2-15 to Table 3.2-32 the results of the EFSA PRIMo calculations are shown.

3.2.1.2 Chronic assessments applying the neurotox-trigger value

The input values (parent/metabolite ratios) of Table 3.2-13 were multiplied with the STMRs of Table 3.2-14 and run against the trigger value 0.0003 mg/kg bw using the EFSA PRIMo model (version 2).

In Table 3.2-33 to Table 3.2-50 the results of the EFSA PRIMo calculations are shown.

3.2.1.3 Chronic assessments applying the Cramer class III-trigger value

The input values (parent/metabolite ratios) of Table 3.2-13 were multiplied with the STMRs of Table 3.2-14 and run against the trigger value 0.0015 mg/kg bw using the EFSA PRIMo model (version 2).

In Table 3.2-51 to Table 3.2-68 the results of the EFSA PRIMo calculations are shown.

Table 3.2-15: Chronic Assessment of Metabolite M310I001 (genotox trigger)

M310I001

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
68	7835
No of diets exceeding ADI:	26

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
7835.1	ES child	6761.7	Poultry: Meat	530.0	Birds' eggs	99.3	Swine: Liver	
6783.9	WHO Cluster diet B	5137.3	Poultry: Meat	591.1	Bovine: Kidney	362.3	Birds' eggs	
6157.7	WHO cluster diet E	5338.9	Poultry: Meat	461.2	Birds' eggs	50.3	Wine grapes	
5659.3	FR toddler	4416.4	Poultry: Meat	745.8	Birds' eggs	210.6	Spinach	
5508.5	WHO regional European diet	4646.3	Poultry: Meat	457.5	Birds' eggs	75.8	Swine: Meat	
5027.5	NL child	3722.0	Poultry: Meat	432.8	Birds' eggs	119.1	Bovine: Liver	
4225.8	DE child	3126.7	Poultry: Meat	815.9	Birds' eggs	60.8	Spinach	
4040.8	ES adult	3331.2	Poultry: Meat	341.0	Birds' eggs	66.8	Lettuce and other salad plants	
3683.3	FR all population	3173.5	Poultry: Meat	219.6	Birds' eggs	125.4	Wine grapes	
3599.7	FR infant	2988.6	Poultry: Meat	324.4	Birds' eggs	131.9	Spinach	
3155.7	WHO Cluster diet F	2393.3	Poultry: Meat	334.3	Birds' eggs	80.6	Bovine: Kidney	
2861.1	WHO cluster diet D	2095.2	Poultry: Meat	298.9	Birds' eggs	120.9	Bovine: Kidney	
2823.2	IE adult	1888.6	Poultry: Meat	354.0	Sheep: Liver	199.5	Birds' eggs	
2384.6	NL general	1786.7	Poultry: Meat	214.1	Birds' eggs	55.6	Swine: Meat	
2116.4	LT adult	1659.9	Poultry: Meat	239.3	Birds' eggs	58.0	Swine: Meat	
1633.6	UK Infant	984.4	Birds' eggs	193.5	Sugar beet (root)	185.3	Bovine: Kidney	
1315.4	UK Toddler	651.8	Birds' eggs	439.1	Sugar beet (root)	55.2	Bovine: Kidney	
993.6	DK child	632.2	Birds' eggs	199.3	Bovine: Liver	35.2	Wheat	
908.4	SE general population 90th percentile	646.6	Birds' eggs	58.7	Leafy brassica	50.3	Lettuce and other salad plants	
650.5	UK vegetarian	255.7	Birds' eggs	189.3	Poultry: Meat	72.5	Sugar beet (root)	
506.6	DK adult	268.1	Birds' eggs	84.6	Bovine: Liver	43.7	Wine grapes	
460.0	UK Adult	224.4	Birds' eggs	76.7	Sugar beet (root)	34.0	Wine grapes	
244.3	PT General population	78.0	Wine grapes	46.2	Rice	34.1	Potatoes	
237.7	FI adult	165.2	Birds' eggs	9.7	Lettuce and other salad plants	9.6	Wine grapes	
217.4	IT adult	69.7	Lettuce and other salad plants	28.4	Spinach	26.5	Wheat	
202.3	IT kids/toddler	51.4	Lettuce and other salad plants	42.5	Wheat	18.0	Beet leaves (chard)	
67.7	**PL general population**	**22.0**	**Potatoes**	**10.0**	**Table grapes**	**9.0**	**Tomatoes**	

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 67.7 % to 7835 % of the ADI.
For 26 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-16: Chronic Assessment of Metabolite M310I003 (genotox trigger)

M310I003

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
573

No of diets exceeding ADI: 22

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
572.9	UK Infant	500.3	Birds' eggs	72.6	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
415.9	WHO Cluster diet B	231.7	Bovine: Kidney	184.1	Birds' eggs		FRUIT (FRESH OR FROZEN)	
414.6	DE child	414.6	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
379.0	FR toddler	379.0	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
352.9	UK Toddler	331.2	Birds' eggs	21.6	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
328.6	SE general population 90th percentile	328.6	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
321.3	DK child	321.3	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
276.0	ES child	269.4	Birds' eggs	6.6	Swine: Kidney		FRUIT (FRESH OR FROZEN)	
257.0	NL child	219.9	Birds' eggs	18.5	Swine: Kidney	18.5	Swine: Kidney	
243.1	WHO regional European diet	232.5	Birds' eggs	10.5	Bovine: Kidney	0.1	Poultry: Liver	
234.4	WHO cluster diet E	234.4	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
201.5	WHO Cluster diet F	169.9	Birds' eggs	31.6	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
199.3	WHO cluster diet D	151.9	Birds' eggs	47.4	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
175.8	ES adult	173.3	Birds' eggs	2.5	Swine: Kidney		FRUIT (FRESH OR FROZEN)	
164.9	FR infant	164.9	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
136.2	DK adult	136.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
129.9	UK vegetarian	129.9	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
122.4	UK Adult	114.0	Birds' eggs	8.3	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
121.6	LT adult	121.6	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
111.6	FR all population	111.6	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
108.8	NL general	108.8	Birds' eggs	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)	
101.4	IE adult	101.4	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
84.0	**FI adult**	**84.0**	**Birds' eggs**		**FRUIT (FRESH OR FROZEN)**		**FRUIT (FRESH OR FROZEN)**	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 0 % to 573 % of the ADI.
For 22 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-17: Chronic Assessment of Metabolite M310I004 (genotox trigger)

M310I004			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
2025

No of diets exceeding ADI: 9

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2025.0	WHO Cluster diet B	1899.3	Bovine: Kidney	125.7	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
753.0	UK Infant	595.4	Bovine: Kidney	157.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
556.5	NL child	151.5	Swine: Kidney	151.5	Swine: Kidney	130.3	Bovine: Liver	
414.2	WHO cluster diet D	388.5	Bovine: Kidney	25.7	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
387.5	IE adult	387.5	Sheep: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
276.1	WHO Cluster diet F	259.0	Bovine: Kidney	17.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
218.1	DK child	218.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
212.6	UK Toddler	177.4	Bovine: Kidney	35.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
190.1	ES child	108.7	Swine: Liver	54.4	Swine: Kidney	27.1	Bovine: Liver	
97.8	WHO regional European diet	86.3	Bovine: Kidney	11.4	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
92.6	DK adult	92.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
90.7	UK Adult	68.2	Bovine: Kidney	22.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
74.8	NL general	43.5	Swine: Liver	31.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
68.7	ES adult	29.5	Swine: Liver	20.7	Swine: Kidney	18.5	Bovine: Liver	
64.9	LT adult	36.7	Swine: Liver	28.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
28.6	WHO cluster diet E	28.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 0 % to 2025 % of the ADI.
For 9 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-18: Chronic Assessment of Metabolite M310I005 (genotox trigger)

M310I005

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identifed (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 131 – 1117

No of diets exceeding ADI: 27

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1116.5	UK Toddler	850.8	Sugar beet (root)	65.6	Rice	48.6	Wheat	
887.0	WHO Cluster diet B	118.8	Olives for oil production	108.9	Wine grapes	105.8	Wheat	
769.0	FR toddler	408.0	Spinach	62.8	Potatoes	46.5	Beans (with pods)	
719.0	NL child	214.5	Spinach	82.8	Leafy brassica	74.7	Lettuce and other salad plants	
619.8	UK Infant	375.0	Sugar beet (root)	72.1	Rice	40.3	Potatoes	
601.7	IE adult	91.4	Leafy brassica	76.0	Wine grapes	72.1	Spinach	
513.2	WHO cluster diet D	166.9	Leafy brassica	80.6	Wheat	63.1	Rice	
507.3	SE general population 90th percentile	113.8	Leafy brassica	97.5	Lettuce and other salad plants	51.7	Potatoes	
480.0	FR infant	255.5	Spinach	51.3	Potatoes	35.5	Beans (with pods)	
473.3	PT General population	151.2	Wine grapes	89.6	Rice	66.1	Potatoes	
468.5	FR all population	243.0	Wine grapes	77.4	Lettuce and other salad plants	40.8	Wheat	
462.6	WHO cluster diet E	97.5	Wine grapes	48.9	Wheat	48.0	Lettuce and other salad plants	
458.1	DE child	117.8	Spinach	77.1	Table grapes	51.0	Wheat	
443.4	ES child	100.9	Lettuce and other salad plants	55.1	Rice	55.0	Wheat	
421.8	ES adult	129.5	Lettuce and other salad plants	46.3	Beet leaves (chard)	42.7	Spinach	
421.2	IT adult	135.0	Lettuce and other salad plants	55.1	Spinach	51.3	Wheat	
413.1	NL general	81.8	Spinach	57.9	Lettuce and other salad plants	51.5	Leafy brassica	
406.0	WHO regional European diet	99.5	Lettuce and other salad plants	49.8	Potatoes	36.8	Wheat	
398.3	UK vegetarian	140.5	Sugar beet (root)	49.4	Wine grapes	43.6	Rice	
392.0	IT kids/toddler	99.7	Lettuce and other salad plants	82.4	Wheat	34.8	Beet leaves (chard)	
390.3	WHO Cluster diet F	72.5	Lettuce and other salad plants	57.6	Leafy brassica	44.6	Wheat	
375.4	UK Adult	148.7	Sugar beet (root)	65.8	Wine grapes	41.7	Rice	
314.1	DK child	68.3	Wheat	54.8	Rye	34.1	Lettuce and other salad plants	
233.7	DK adult	84.7	Wine grapes	31.4	Lettuce and other salad plants	25.0	Wheat	
154.0	LT adult	39.4	Potatoes	24.4	Rice	16.8	Head cabbage	
140.5	FI adult	18.9	Lettuce and other salad plants	18.5	Wine grapes	16.9	Leafy brassica	
131.2	PL general population	42.6	Potatoes	19.4	Table grapes	17.5	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 131.2 % to 1117 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-19: Chronic Assessment of Metabolite M310I006 (genotox trigger)

M310I006

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
135	1153
No of diets exceeding ADI:	27

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1152.5	UK Toddler	878.2	Sugar beet (root)	67.8	Rice	50.1	Wheat	
915.6	WHO Cluster diet B	122.7	Olives for oil production	112.4	Wine grapes	109.2	Wheat	
793.8	FR toddler	421.1	Spinach	64.8	Potatoes	48.0	Beans (with pods)	
742.2	NL child	221.4	Spinach	85.4	Leafy brassica	77.1	Lettuce and other salad plants	
639.8	UK Infant	387.1	Sugar beet (root)	74.4	Rice	41.6	Potatoes	
621.1	IE adult	94.3	Leafy brassica	78.4	Wine grapes	74.4	Spinach	
529.8	WHO cluster diet D	172.2	Leafy brassica	83.2	Wheat	65.2	Rice	
523.7	SE general population 90th percentile	117.4	Leafy brassica	100.7	Lettuce and other salad plants	53.3	Potatoes	
495.5	FR infant	263.8	Spinach	52.9	Potatoes	36.6	Beans (with pods)	
488.6	PT General population	156.1	Wine grapes	92.4	Rice	68.3	Potatoes	
483.6	FR all population	250.9	Wine grapes	79.9	Lettuce and other salad plants	42.1	Wheat	
477.5	WHO cluster diet E	100.7	Wine grapes	50.5	Wheat	49.5	Lettuce and other salad plants	
472.9	DE child	121.6	Spinach	79.6	Table grapes	52.6	Wheat	
457.7	ES child	104.2	Lettuce and other salad plants	56.9	Rice	56.8	Wheat	
435.4	ES adult	133.7	Lettuce and other salad plants	47.8	Beet leaves (chard)	44.1	Spinach	
434.8	IT adult	139.4	Lettuce and other salad plants	56.8	Spinach	52.9	Wheat	
426.5	NL general	84.5	Spinach	59.8	Lettuce and other salad plants	53.2	Leafy brassica	
419.1	WHO regional European diet	102.8	Lettuce and other salad plants	51.4	Potatoes	38.0	Wheat	
411.1	UK vegetarian	145.0	Sugar beet (root)	51.0	Wine grapes	45.0	Rice	
404.7	IT kids/toddler	102.9	Lettuce and other salad plants	85.1	Wheat	35.9	Beet leaves (chard)	
402.9	WHO Cluster diet F	74.9	Lettuce and other salad plants	59.5	Leafy brassica	46.1	Wheat	
387.5	UK Adult	153.5	Sugar beet (root)	67.9	Wine grapes	43.1	Rice	
324.2	DK child	70.5	Wheat	56.6	Rye	35.2	Lettuce and other salad plants	
241.2	DK adult	87.4	Wine grapes	32.4	Lettuce and other salad plants	25.8	Wheat	
159.0	LT adult	40.6	Potatoes	25.2	Rice	17.3	Head cabbage	
145.1	FI adult	19.5	Lettuce and other salad plants	19.1	Wine grapes	17.4	Leafy brassica	
135.4	PL general population	44.0	Potatoes	20.1	Table grapes	18.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 135.4 % to 1153 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-20: Chronic Assessment of Metabolite M310I007 (genotox trigger)

M310I007

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identifed (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 241 – 2053

No of diets exceeding ADI: 27

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2052.9	UK Toddler	1564.3	Sugar beet (root)	120.7	Rice	89.3	Wheat	
1630.9	WHO Cluster diet B	218.5	Olives for oil production	200.2	Wine grapes	194.6	Wheat	
1413.9	FR toddler	750.1	Spinach	115.5	Potatoes	85.6	Beans (with pods)	
1322.1	NL child	394.3	Spinach	152.2	Leafy brassica	137.3	Lettuce and other salad plants	
1139.7	UK Infant	689.5	Sugar beet (root)	132.6	Rice	74.2	Potatoes	
1106.3	IE adult	168.0	Leafy brassica	139.7	Wine grapes	132.5	Spinach	
943.7	WHO cluster diet D	306.8	Leafy brassica	148.3	Wheat	116.1	Rice	
932.8	SE general population 90th percentile	209.2	Leafy brassica	179.3	Lettuce and other salad plants	95.0	Potatoes	
882.6	FR infant	469.9	Spinach	94.3	Potatoes	65.2	Beans (with pods)	
870.2	PT General population	278.0	Wine grapes	164.7	Rice	121.6	Potatoes	
861.5	FR all population	446.9	Wine grapes	142.3	Lettuce and other salad plants	75.0	Wheat	
850.6	WHO cluster diet E	179.3	Wine grapes	89.9	Wheat	88.2	Lettuce and other salad plants	
842.3	DE child	216.6	Spinach	141.8	Table grapes	93.7	Wheat	
815.3	ES child	185.6	Lettuce and other salad plants	101.3	Rice	101.1	Wheat	
775.6	ES adult	238.1	Lettuce and other salad plants	85.1	Beet leaves (chard)	78.6	Spinach	
774.5	IT adult	248.3	Lettuce and other salad plants	101.2	Spinach	94.3	Wheat	
759.6	NL general	150.4	Spinach	106.5	Lettuce and other salad plants	94.7	Leafy brassica	
746.6	WHO regional European diet	183.0	Lettuce and other salad plants	91.5	Potatoes	67.6	Wheat	
732.3	UK vegetarian	258.3	Sugar beet (root)	90.9	Wine grapes	80.2	Rice	
720.8	IT kids/toddler	183.2	Lettuce and other salad plants	151.5	Wheat	64.0	Beet leaves (chard)	
717.7	WHO Cluster diet F	133.4	Lettuce and other salad plants	106.0	Leafy brassica	82.1	Wheat	
690.3	UK Adult	273.4	Sugar beet (root)	121.0	Wine grapes	76.7	Rice	
577.5	DK child	125.5	Wheat	100.7	Rye	62.6	Lettuce and other salad plants	
429.7	DK adult	155.7	Wine grapes	57.7	Lettuce and other salad plants	45.9	Wheat	
283.2	LT adult	72.4	Potatoes	44.9	Rice	30.9	Head cabbage	
258.4	FI adult	34.7	Lettuce and other salad plants	34.1	Wine grapes	31.0	Leafy brassica	
241.2	PL general population	78.3	Potatoes	35.8	Table grapes	32.2	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 241.2 % to 2053 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-21: Chronic Assessment of Metabolite M310I008 (genotox trigger)

M310I008

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum		241	2053
No of diets exceeding ADI:		27	

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2052.9	UK Toddler	1564.3	Sugar beet (root)	120.7	Rice	89.3	Wheat	
1630.9	WHO Cluster diet B	218.5	Olives for oil production	200.2	Wine grapes	194.6	Wheat	
1413.9	FR toddler	750.1	Spinach	115.5	Potatoes	85.6	Beans (with pods)	
1322.1	NL child	394.3	Spinach	152.2	Leafy brassica	137.3	Lettuce and other salad plants	
1139.7	UK Infant	689.5	Sugar beet (root)	132.6	Rice	74.2	Potatoes	
1106.3	IE adult	168.0	Leafy brassica	139.7	Wine grapes	132.5	Spinach	
943.7	WHO cluster diet D	306.8	Leafy brassica	148.3	Wheat	116.1	Rice	
932.8	SE general population 90th percentile	209.2	Leafy brassica	179.3	Lettuce and other salad plants	95.0	Potatoes	
882.6	FR infant	469.9	Spinach	94.3	Potatoes	65.2	Beans (with pods)	
870.2	PT General population	278.0	Wine grapes	164.7	Rice	121.6	Potatoes	
861.5	FR all population	446.9	Wine grapes	142.3	Lettuce and other salad plants	75.0	Wheat	
850.6	WHO cluster diet E	179.3	Wine grapes	89.9	Wheat	88.2	Lettuce and other salad plants	
842.3	DE child	216.6	Spinach	141.8	Table grapes	93.7	Wheat	
815.3	ES child	185.6	Lettuce and other salad plants	101.3	Rice	101.1	Wheat	
775.6	ES adult	238.1	Lettuce and other salad plants	85.1	Beet leaves (chard)	78.6	Spinach	
774.5	IT adult	248.3	Lettuce and other salad plants	101.2	Spinach	94.3	Wheat	
759.6	NL general	150.4	Spinach	106.5	Lettuce and other salad plants	94.7	Leafy brassica	
746.6	WHO regional European diet	183.0	Lettuce and other salad plants	91.5	Potatoes	67.6	Wheat	
732.3	UK vegetarian	258.3	Sugar beet (root)	90.9	Wine grapes	80.2	Rice	
720.8	IT kids/toddler	183.2	Lettuce and other salad plants	151.5	Wheat	64.0	Beet leaves (chard)	
717.7	WHO Cluster diet F	133.4	Lettuce and other salad plants	106.0	Leafy brassica	82.1	Wheat	
690.3	UK Adult	273.4	Sugar beet (root)	121.0	Wine grapes	76.7	Rice	
577.5	DK child	125.5	Wheat	100.7	Rye	62.6	Lettuce and other salad plants	
429.7	DK adult	155.7	Wine grapes	57.7	Lettuce and other salad plants	45.9	Wheat	
283.2	LT adult	72.4	Potatoes	44.9	Rice	30.9	Head cabbage	
258.4	FI adult	34.7	Lettuce and other salad plants	34.1	Wine grapes	31.0	Leafy brassica	
241.2	PL general population	78.3	Potatoes	35.8	Table grapes	32.2	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 241.2 % to 2053 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-22: Chronic Assessment of Metabolite M310I009 (genotox trigger)

M310I009

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

			TMDI (range) in % of ADI minimum - maximum						
			296	2521					
		No of diets exceeding ADI:	27						
Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2521.1	UK Toddler	1921.1	Sugar beet (root)	148.2	Rice	109.7	Wheat	
2002.8	WHO Cluster diet B	268.3	Olives for oil production	245.8	Wine grapes	239.0	Wheat	
1736.3	FR toddler	921.2	Spinach	141.8	Potatoes	105.1	Beans (with pods)	
1623.6	NL child	484.3	Spinach	186.9	Leafy brassica	168.6	Lettuce and other salad plants	
1399.7	UK Infant	846.8	Sugar beet (root)	162.9	Rice	91.1	Potatoes	
1358.7	IE adult	206.3	Leafy brassica	171.5	Wine grapes	162.8	Spinach	
1158.9	WHO cluster diet D	376.8	Leafy brassica	182.1	Wheat	142.5	Rice	
1145.5	SE general population 90th percentile	256.9	Leafy brassica	220.2	Lettuce and other salad plants	116.7	Potatoes	
1083.9	FR infant	577.0	Spinach	115.8	Potatoes	80.1	Beans (with pods)	
1068.7	PT General population	341.4	Wine grapes	202.2	Rice	149.3	Potatoes	
1058.0	FR all population	548.8	Wine grapes	174.7	Lettuce and other salad plants	92.1	Wheat	
1044.6	WHO cluster diet E	220.2	Wine grapes	110.4	Wheat	108.3	Lettuce and other salad plants	
1034.4	DE child	266.0	Spinach	174.2	Table grapes	115.1	Wheat	
1001.2	ES child	227.9	Lettuce and other salad plants	124.4	Rice	124.2	Wheat	
952.5	ES adult	292.4	Lettuce and other salad plants	104.6	Beet leaves (chard)	96.5	Spinach	
951.2	IT adult	304.9	Lettuce and other salad plants	124.3	Spinach	115.8	Wheat	
932.9	NL general	184.8	Spinach	130.8	Lettuce and other salad plants	116.3	Leafy brassica	
916.9	WHO regional European diet	224.8	Lettuce and other salad plants	112.4	Potatoes	83.1	Wheat	
899.3	UK vegetarian	317.2	Sugar beet (root)	111.7	Wine grapes	98.5	Rice	
885.2	IT kids/toddler	225.0	Lettuce and other salad plants	186.1	Wheat	78.5	Beet leaves (chard)	
881.4	WHO Cluster diet F	163.8	Lettuce and other salad plants	130.2	Leafy brassica	100.8	Wheat	
847.7	UK Adult	335.8	Sugar beet (root)	148.6	Wine grapes	94.2	Rice	
709.2	DK child	154.1	Wheat	123.7	Rye	76.9	Lettuce and other salad plants	
527.7	DK adult	191.2	Wine grapes	70.8	Lettuce and other salad plants	56.4	Wheat	
347.8	LT adult	88.9	Potatoes	55.1	Rice	37.9	Head cabbage	
317.3	FI adult	42.6	Lettuce and other salad plants	41.9	Wine grapes	38.1	Leafy brassica	
296.3	PL general population	96.2	Potatoes	43.9	Table grapes	39.6	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 296.3 % to 2521 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-23: Chronic Assessment of Metabolite M310I010 (genotox trigger)

M310I010

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
13	6103
No of diets exceeding ADI:	23

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
6103.0	UK Infant	5729.5	Milk and cream,	232.6	Bovine: Kidney	80.9	Bovine: Liver	49.8
5938.6	FR toddler	5864.2	Milk and cream,	39.5	Spinach	6.1	Potatoes	19.5
4657.7	NL child	4339.5	Milk and cream,	66.9	Bovine: Liver	63.3	Swine: Liver	18.9
3855.8	FR infant	3809.3	Milk and cream,	24.7	Spinach	5.0	Potatoes	14.3
3252.8	UK Toddler	3057.3	Milk and cream,	82.3	Sugar beet (root)	69.3	Bovine: Kidney	96.0
2159.4	DE child	2115.1	Milk and cream,	11.4	Spinach	7.5	Table grapes	16.4
2011.9	DK child	1869.5	Milk and cream,	112.0	Bovine: Liver	6.6	Wheat	22.0
1985.6	ES child	1851.8	Milk and cream,	55.8	Swine: Liver	21.3	Swine: Kidney	11.9
1880.6	SE general population 90th percentile	1831.5	Milk and cream,	11.0	Leafy brassica	9.4	Lettuce and other salad plants	15.3
1362.6	WHO Cluster diet B	742.1	Bovine: Kidney	470.1	Milk and cream,	64.5	Bovine: Liver	30.8
1049.6	NL general	971.2	Milk and cream,	22.3	Swine: Liver	16.1	Bovine: Liver	9.2
961.1	WHO cluster diet D	746.4	Milk and cream,	151.8	Bovine: Kidney	16.1	Leafy brassica	19.0
864.2	DK adult	794.0	Milk and cream,	47.6	Bovine: Liver	8.2	Wine grapes	7.8
853.2	FI adult	839.6	Milk and cream,	1.8	Lettuce and other salad plants	1.8	Wine grapes	5.6
806.3	ES adult	732.8	Milk and cream,	15.1	Swine: Liver	12.5	Lettuce and other salad plants	7.6
792.5	WHO regional European diet	713.6	Milk and cream,	33.7	Bovine: Kidney	9.6	Lettuce and other salad plants	14.8
734.6	WHO Cluster diet F	586.8	Milk and cream,	101.2	Bovine: Kidney	8.8	Bovine: Liver	13.7
669.1	IE adult	411.9	Milk and cream,	198.9	Sheep: Liver	8.8	Leafy brassica	16.6
634.4	LT adult	586.2	Milk and cream,	18.9	Swine: Liver	14.5	Bovine: Liver	8.7
520.7	UK vegetarian	482.2	Milk and cream,	13.6	Sugar beet (root)	4.8	Wine grapes	21.5
518.3	UK Adult	443.8	Milk and cream,	26.6	Bovine: Kidney	14.4	Sugar beet (root)	20.7
502.4	WHO cluster diet E	443.0	Milk and cream,	14.7	Bovine: Liver	9.4	Wine grapes	15.5
441.7	FR all population	396.4	Milk and cream,	23.5	Wine grapes	7.5	Lettuce and other salad plants	8.2
45.8	**PT General population**	**14.6**	**Wine grapes**	**8.7**	**Rice**	**6.4**	**Potatoes**	**16.3**
40.8	**IT adult**	**13.1**	**Lettuce and other salad plants**	**5.3**	**Spinach**	**5.0**	**Wheat**	**9.8**
37.9	**IT kids/toddler**	**9.6**	**Lettuce and other salad plants**	**8.0**	**Wheat**	**3.4**	**Beet leaves (chard)**	**13.8**
12.7	**PL general population**	**4.1**	**Potatoes**	**1.9**	**Table grapes**	**1.7**	**Tomatoes**	**7.4**

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 12.7 % to 6103 % of the ADI.
For 23 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-24: Chronic Assessment of Metabolite M310I011 (genotox trigger)

M310I011

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum		296	2553
No of diets exceeding ADI:		27	

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2552.9	UK Toddler	1921.1	Sugar beet (root)	148.2	Rice	109.7	Wheat	
2287.4	WHO Cluster diet B	268.3	Olives for oil production	256.7	Bovine: Kidney	245.8	Wine grapes	
1736.3	FR toddler	921.2	Spinach	141.8	Potatoes	105.1	Beans (with pods)	
1720.8	NL child	484.3	Spinach	186.9	Leafy brassica	168.6	Lettuce and other salad plants	
1515.1	UK Infant	846.8	Sugar beet (root)	162.9	Rice	91.1	Potatoes	
1444.6	IE adult	206.3	Leafy brassica	171.5	Wine grapes	162.8	Spinach	
1217.1	WHO cluster diet D	376.8	Leafy brassica	182.1	Wheat	142.5	Rice	
1145.5	SE general population 90th percentile	256.9	Leafy brassica	220.2	Lettuce and other salad plants	116.7	Potatoes	
1083.9	FR infant	577.0	Spinach	115.8	Potatoes	80.1	Beans (with pods)	
1068.7	PT General population	341.4	Wine grapes	202.2	Rice	149.3	Potatoes	
1058.0	FR all population	548.8	Wine grapes	174.7	Lettuce and other salad plants	92.1	Wheat	
1051.0	WHO cluster diet E	220.2	Wine grapes	110.4	Wheat	108.3	Lettuce and other salad plants	
1038.7	ES child	227.9	Lettuce and other salad plants	124.4	Rice	124.2	Wheat	
1034.4	DE child	266.0	Spinach	174.2	Table grapes	115.1	Wheat	
965.9	ES adult	292.4	Lettuce and other salad plants	104.6	Beet leaves (chard)	96.5	Spinach	
951.2	IT adult	304.9	Lettuce and other salad plants	124.3	Spinach	115.8	Wheat	
949.4	NL general	184.8	Spinach	130.8	Lettuce and other salad plants	116.3	Leafy brassica	
931.1	WHO regional European diet	224.8	Lettuce and other salad plants	112.4	Potatoes	83.1	Wheat	
920.2	WHO Cluster diet F	163.8	Lettuce and other salad plants	130.2	Leafy brassica	100.8	Wheat	
899.3	UK vegetarian	317.2	Sugar beet (root)	111.7	Wine grapes	98.5	Rice	
885.2	IT kids/toddler	225.0	Lettuce and other salad plants	186.1	Wheat	78.5	Beet leaves (chard)	
861.9	UK Adult	335.8	Sugar beet (root)	148.6	Wine grapes	94.2	Rice	
757.5	DK child	154.1	Wheat	123.7	Rye	76.9	Lettuce and other salad plants	
548.2	DK adult	191.2	Wine grapes	70.8	Lettuce and other salad plants	56.4	Wheat	
362.2	LT adult	88.9	Potatoes	55.1	Rice	37.9	Head cabbage	
317.3	FI adult	42.6	Lettuce and other salad plants	41.9	Wine grapes	38.1	Leafy brassica	
296.3	PL general population	96.2	Potatoes	43.9	Table grapes	39.6	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 296.3 % to 2553 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-25: Chronic Assessment of Metabolite M310I013 (genotox trigger)

M310I013			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	25 216
No of diets exceeding ADI:	6

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
216.1	UK Toddler	164.7	Sugar beet (root)	12.7	Rice	9.4	Wheat	25.7
171.7	WHO Cluster diet B	23.0	Olives for oil production	21.1	Wine grapes	20.5	Wheat	53.2
148.8	FR toddler	79.0	Spinach	12.2	Potatoes	9.0	Beans (with pods)	33.0
139.2	NL child	41.5	Spinach	16.0	Leafy brassica	14.5	Lettuce and other salad plants	34.5
120.0	UK Infant	72.6	Sugar beet (root)	14.0	Rice	7.8	Potatoes	25.2
116.5	IE adult	17.7	Leafy brassica	14.7	Wine grapes	14.0	Spinach	29.0
99.3	WHO cluster diet D	32.3	Leafy brassica	15.6	Wheat	12.2	Rice	36.9
98.2	SE general population 90th percentile	22.0	Leafy brassica	18.9	Lettuce and other salad plants	10.0	Potatoes	28.1
92.9	FR infant	49.5	Spinach	9.9	Potatoes	6.9	Beans (with pods)	23.9
91.6	PT General population	29.3	Wine grapes	17.3	Rice	12.8	Potatoes	31.6
90.7	FR all population	47.0	Wine grapes	15.0	Lettuce and other salad plants	7.9	Wheat	15.4
89.5	WHO cluster diet E	18.9	Wine grapes	9.5	Wheat	9.3	Lettuce and other salad plants	28.7
88.7	DE child	22.8	Spinach	14.9	Table grapes	9.9	Wheat	28.3
85.8	ES child	19.5	Lettuce and other salad plants	10.7	Rice	10.6	Wheat	22.7
81.6	ES adult	25.1	Lettuce and other salad plants	9.0	Beet leaves (chard)	8.3	Spinach	14.0
81.5	IT adult	26.1	Lettuce and other salad plants	10.7	Spinach	9.9	Wheat	18.6
80.0	NL general	15.8	Spinach	11.2	Lettuce and other salad plants	10.0	Leafy brassica	16.8
78.6	WHO regional European diet	19.3	Lettuce and other salad plants	9.6	Potatoes	7.1	Wheat	27.1
77.1	UK vegetarian	27.2	Sugar beet (root)	9.6	Wine grapes	8.4	Rice	14.4
75.9	IT kids/toddler	19.3	Lettuce and other salad plants	16.0	Wheat	6.7	Beet leaves (chard)	26.6
75.5	WHO Cluster diet F	14.0	Lettuce and other salad plants	11.2	Leafy brassica	8.6	Wheat	26.2
72.7	UK Adult	28.8	Sugar beet (root)	12.7	Wine grapes	8.1	Rice	11.5
60.8	DK child	13.2	Wheat	10.6	Rye	6.6	Lettuce and other salad plants	42.2
45.2	DK adult	16.4	Wine grapes	6.1	Lettuce and other salad plants	4.8	Wheat	14.8
29.8	LT adult	7.6	Potatoes	4.7	Rice	3.3	Head cabbage	17.2
27.2	FI adult	3.7	Lettuce and other salad plants	3.6	Wine grapes	3.3	Leafy brassica	10.6
25.4	PL general population	8.2	Potatoes	3.8	Table grapes	3.4	Tomatoes	14.3

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 25.4 % to 216 % of the ADI.
For 6 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-26: Chronic Assessment of Metabolite M310I017 (genotox trigger)

M310I017

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 635 - 5402

No of diets exceeding ADI: 27

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
5402.4	UK Toddler	4116.6	Sugar beet (root)	317.6	Rice	235.1	Wheat	
4291.8	WHO Cluster diet B	575.0	Olives for oil production	526.8	Wine grapes	512.1	Wheat	
3720.7	FR toddler	1974.1	Spinach	304.0	Potatoes	225.2	Beans (with pods)	
3479.2	NL child	1037.7	Spinach	400.5	Leafy brassica	361.3	Lettuce and other salad plants	
2999.3	UK Infant	1814.5	Sugar beet (root)	349.0	Rice	195.2	Potatoes	
2911.4	IE adult	442.2	Leafy brassica	367.5	Wine grapes	348.8	Spinach	
2483.4	WHO cluster diet D	807.4	Leafy brassica	390.2	Wheat	305.4	Rice	
2454.7	SE general population 90th percentile	550.5	Leafy brassica	471.9	Lettuce and other salad plants	250.0	Potatoes	
2322.7	FR infant	1236.6	Spinach	248.2	Potatoes	171.5	Beans (with pods)	
2290.1	PT General population	731.6	Wine grapes	433.3	Rice	320.0	Potatoes	
2267.1	FR all population	1176.0	Wine grapes	374.4	Lettuce and other salad plants	197.3	Wheat	
2238.5	WHO cluster diet E	471.9	Wine grapes	236.6	Wheat	232.1	Lettuce and other salad plants	
2216.6	DE child	570.1	Spinach	373.2	Table grapes	246.7	Wheat	
2145.4	ES child	488.4	Lettuce and other salad plants	266.7	Rice	266.1	Wheat	
2041.0	ES adult	626.7	Lettuce and other salad plants	224.0	Beet leaves (chard)	206.7	Spinach	
2038.2	IT adult	653.3	Lettuce and other salad plants	266.4	Spinach	248.1	Wheat	
1999.0	NL general	395.9	Spinach	280.2	Lettuce and other salad plants	249.2	Leafy brassica	
1964.8	WHO regional European diet	481.7	Lettuce and other salad plants	240.8	Potatoes	178.0	Wheat	
1927.1	UK vegetarian	679.8	Sugar beet (root)	239.3	Wine grapes	211.0	Rice	
1896.8	IT kids/toddler	482.2	Lettuce and other salad plants	398.8	Wheat	168.3	Beet leaves (chard)	
1888.6	WHO Cluster diet F	351.0	Lettuce and other salad plants	278.9	Leafy brassica	216.0	Wheat	
1816.5	UK Adult	719.5	Sugar beet (root)	318.4	Wine grapes	201.9	Rice	
1519.6	DK child	330.3	Wheat	265.1	Rye	164.9	Lettuce and other salad plants	
1130.7	DK adult	409.6	Wine grapes	151.8	Lettuce and other salad plants	120.8	Wheat	
745.3	LT adult	190.4	Potatoes	118.1	Rice	81.3	Head cabbage	
679.9	FI adult	91.3	Lettuce and other salad plants	89.7	Wine grapes	81.6	Leafy brassica	
634.9	PL general population	206.1	Potatoes	94.1	Table grapes	84.8	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 634.9 % to 5402 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-27: Chronic Assessment of Metabolite M310I018 (genotox trigger)

PbAld

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	30 - 252
No of diets exceeding ADI:	**14**

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
252.1	UK Toddler	192.1	Sugar beet (root)	14.8	Rice	11.0	Wheat	27.3
200.3	WHO Cluster diet B	26.8	Olives for oil production	24.6	Wine grapes	23.9	Wheat	46.9
173.6	FR toddler	92.1	Spinach	14.2	Potatoes	10.5	Beans (with pods)	35.1
162.4	NL child	48.4	Spinach	18.7	Leafy brassica	16.9	Lettuce and other salad plants	37.4
140.0	UK Infant	84.7	Sugar beet (root)	16.3	Rice	9.1	Potatoes	27.7
135.9	IE adult	20.6	Leafy brassica	17.2	Wine grapes	16.3	Spinach	30.7
115.9	WHO cluster diet D	37.7	Leafy brassica	18.2	Wheat	14.3	Rice	38.4
114.6	SE general population 90th percentile	25.7	Leafy brassica	22.0	Lettuce and other salad plants	11.7	Potatoes	29.1
108.4	FR infant	57.7	Spinach	11.6	Potatoes	8.0	Beans (with pods)	27.2
106.9	PT General population	34.1	Wine grapes	20.2	Rice	14.9	Potatoes	32.8
105.8	FR all population	54.9	Wine grapes	17.5	Lettuce and other salad plants	9.2	Wheat	16.0
104.5	WHO cluster diet E	22.0	Wine grapes	11.0	Wheat	10.8	Lettuce and other salad plants	31.1
103.4	DE child	26.6	Spinach	17.4	Table grapes	11.5	Wheat	28.7
100.1	ES child	22.8	Lettuce and other salad plants	12.4	Rice	12.4	Wheat	22.1
95.2	ES adult	29.2	Lettuce and other salad plants	10.5	Beet leaves (chard)	9.6	Spinach	12.6
95.1	IT adult	30.5	Lettuce and other salad plants	12.4	Spinach	11.6	Wheat	16.0
93.3	NL general	18.5	Spinach	13.1	Lettuce and other salad plants	11.6	Leafy brassica	17.6
91.7	WHO regional European diet	22.5	Lettuce and other salad plants	11.2	Potatoes	8.3	Wheat	26.5
89.9	UK vegetarian	31.7	Sugar beet (root)	11.2	Wine grapes	9.8	Rice	13.9
88.5	IT kids/toddler	22.5	Lettuce and other salad plants	18.6	Wheat	7.9	Beet leaves (chard)	24.2
88.1	WHO Cluster diet F	16.4	Lettuce and other salad plants	13.0	Leafy brassica	10.1	Wheat	27.5
84.8	UK Adult	33.6	Sugar beet (root)	14.9	Wine grapes	9.4	Rice	11.4
70.9	DK child	15.4	Wheat	12.4	Rye	7.7	Lettuce and other salad plants	46.9
52.8	DK adult	19.1	Wine grapes	7.1	Lettuce and other salad plants	5.6	Wheat	15.3
34.8	LT adult	8.9	Potatoes	5.5	Rice	3.8	Head cabbage	17.2
31.7	FI adult	4.3	Lettuce and other salad plants	4.2	Wine grapes	3.8	Leafy brassica	10.4
29.6	PL general population	9.6	Potatoes	4.4	Table grapes	4.0	Tomatoes	12.7

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 29.6 % to 252 % of the ADI.
For 14 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-28: Chronic Assessment of Metabolite M310I019 (genotox trigger)

M310I019

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
24

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
24.0	NL child	24.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
20.5	WHO regional European diet	20.5	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
3.9	NL general	3.9	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I019 is unlikely to present a public health concern.

Table 3.2-29: Chronic Assessment of Metabolite M310I021 (genotox trigger)

M310I021

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
1153

No of diets exceeding ADI: 11

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1152.9	IE adult	1152.9	Sheep: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
754.6	NL child	387.7	Bovine: Liver	366.8	Swine: Liver		FRUIT (FRESH OR FROZEN)	
649.1	DK child	649.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
469.0	UK Infant	469.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
403.9	ES child	323.3	Swine: Liver	80.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
374.0	WHO Cluster diet B	374.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
275.7	DK adult	275.7	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
222.6	NL general	129.5	Swine: Liver	93.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
193.1	LT adult	109.3	Swine: Liver	83.8	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
142.8	ES adult	87.7	Swine: Liver	55.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
104.8	UK Toddler	104.8	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
85.0	WHO cluster diet E	85.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
76.5	WHO cluster diet D	76.5	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
67.1	UK Adult	67.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
51.0	WHO Cluster diet F	51.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
34.0	WHO regional European diet	34.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 0 % to 1153 % of the ADI.
For 11 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-30: Chronic Assessment of Metabolite M310I024 (genotox trigger)

PbAlc

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	296 2521
No of diets exceeding ADI:	27

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2521.1	UK Toddler	1921.1	Sugar beet (root)	148.2	Rice	109.7	Wheat	
2002.8	WHO Cluster diet B	268.3	Olives for oil production	245.8	Wine grapes	239.0	Wheat	
1736.3	FR toddler	921.2	Spinach	141.8	Potatoes	105.1	Beans (with pods)	
1623.6	NL child	484.3	Spinach	186.9	Leafy brassica	168.6	Lettuce and other salad plants	
1399.7	UK Infant	846.8	Sugar beet (root)	162.9	Rice	91.1	Potatoes	
1358.7	IE adult	206.3	Leafy brassica	171.5	Wine grapes	162.8	Spinach	
1158.9	WHO cluster diet D	376.8	Leafy brassica	182.1	Wheat	142.5	Rice	
1145.5	SE general population 90th percentile	256.9	Leafy brassica	220.2	Lettuce and other salad plants	116.7	Potatoes	
1083.9	FR infant	577.0	Spinach	115.8	Potatoes	80.1	Beans (with pods)	
1068.7	PT General population	341.4	Wine grapes	202.2	Rice	149.3	Potatoes	
1058.0	FR all population	548.8	Wine grapes	174.7	Lettuce and other salad plants	92.1	Wheat	
1044.6	WHO cluster diet E	220.2	Wine grapes	110.4	Wheat	108.3	Lettuce and other salad plants	
1034.4	DE child	266.0	Spinach	174.2	Table grapes	115.1	Wheat	
1001.2	ES child	227.9	Lettuce and other salad plants	124.4	Rice	124.2	Wheat	
952.5	ES adult	292.4	Lettuce and other salad plants	104.6	Beet leaves (chard)	96.5	Spinach	
951.2	IT adult	304.9	Lettuce and other salad plants	124.3	Spinach	115.8	Wheat	
932.9	NL general	184.8	Spinach	130.8	Lettuce and other salad plants	116.3	Leafy brassica	
916.9	WHO regional European diet	224.8	Lettuce and other salad plants	112.4	Potatoes	83.1	Wheat	
899.3	UK vegetarian	317.2	Sugar beet (root)	111.7	Wine grapes	98.5	Rice	
885.2	IT kids/toddler	225.0	Lettuce and other salad plants	186.1	Wheat	78.5	Beet leaves (chard)	
881.4	WHO Cluster diet F	163.8	Lettuce and other salad plants	130.2	Leafy brassica	100.8	Wheat	
847.7	UK Adult	335.8	Sugar beet (root)	148.6	Wine grapes	94.2	Rice	
709.2	DK child	154.1	Wheat	123.7	Rye	76.9	Lettuce and other salad plants	
527.7	DK adult	191.2	Wine grapes	70.8	Lettuce and other salad plants	56.4	Wheat	
347.8	LT adult	88.9	Potatoes	55.1	Rice	37.9	Head cabbage	
317.3	FI adult	42.6	Lettuce and other salad plants	41.9	Wine grapes	38.1	Leafy brassica	
296.3	PL general population	96.2	Potatoes	43.9	Table grapes	39.6	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 296.3 % to 2521 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-31: Chronic Assessment of Metabolite M310I025 (genotox trigger)

M310I025

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
339	2881
No of diets exceeding ADI:	27

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2881.3	UK Toddler	2195.5	Sugar beet (root)	169.4	Rice	125.4	Wheat	
2289.0	WHO Cluster diet B	306.7	Olives for oil production	280.9	Wine grapes	273.1	Wheat	
1984.4	FR toddler	1052.8	Spinach	162.1	Potatoes	120.1	Beans (with pods)	
1855.6	NL child	553.4	Spinach	213.6	Leafy brassica	192.7	Lettuce and other salad plants	
1599.6	UK Infant	967.7	Sugar beet (root)	186.1	Rice	104.1	Potatoes	
1552.8	IE adult	235.8	Leafy brassica	196.0	Wine grapes	186.0	Spinach	
1324.5	WHO cluster diet D	430.6	Leafy brassica	208.1	Wheat	162.9	Rice	
1309.2	SE general population 90th percentile	293.6	Leafy brassica	251.7	Lettuce and other salad plants	133.3	Potatoes	
1238.8	FR infant	659.5	Spinach	132.4	Potatoes	91.5	Beans (with pods)	
1221.4	PT General population	390.2	Wine grapes	231.1	Rice	170.7	Potatoes	
1209.1	FR all population	627.2	Wine grapes	199.7	Lettuce and other salad plants	105.2	Wheat	
1193.9	WHO cluster diet E	251.7	Wine grapes	126.2	Wheat	123.8	Lettuce and other salad plants	
1182.2	DE child	304.0	Spinach	199.0	Table grapes	131.6	Wheat	
1144.2	ES child	260.5	Lettuce and other salad plants	142.2	Rice	141.9	Wheat	
1088.6	ES adult	334.2	Lettuce and other salad plants	119.5	Beet leaves (chard)	110.3	Spinach	
1087.0	IT adult	348.4	Lettuce and other salad plants	142.1	Spinach	132.3	Wheat	
1066.1	NL general	211.2	Spinach	149.5	Lettuce and other salad plants	132.9	Leafy brassica	
1047.9	WHO regional European diet	256.9	Lettuce and other salad plants	128.4	Potatoes	94.9	Wheat	
1027.8	UK vegetarian	362.6	Sugar beet (root)	127.6	Wine grapes	112.6	Rice	
1011.6	IT kids/toddler	257.2	Lettuce and other salad plants	212.7	Wheat	89.8	Beet leaves (chard)	
1007.3	WHO Cluster diet F	187.2	Lettuce and other salad plants	148.8	Leafy brassica	115.2	Wheat	
968.8	UK Adult	383.7	Sugar beet (root)	169.8	Wine grapes	107.7	Rice	
810.5	DK child	176.1	Wheat	141.4	Rye	87.9	Lettuce and other salad plants	
603.1	DK adult	218.5	Wine grapes	81.0	Lettuce and other salad plants	64.4	Wheat	
397.5	LT adult	101.6	Potatoes	63.0	Rice	43.4	Head cabbage	
362.6	FI adult	48.7	Lettuce and other salad plants	47.9	Wine grapes	43.5	Leafy brassica	
338.6	PL general population	109.9	Potatoes	50.2	Table grapes	45.2	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 338.6 % to 2881 % of the ADI.
For 27 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-32: Chronic Assessment of Metabolite M310I026 (genotox trigger)

M310I026

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0000025	ARfD (mg/kg bw):	0.005
Source of ADI:	Genotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
13	108
No of diets exceeding ADI:	1

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
108.0	UK Toddler	82.3	Sugar beet (root)	6.4	Rice	4.7	Wheat	96.0
85.8	WHO Cluster diet B	11.5	Olives for oil production	10.5	Wine grapes	10.2	Wheat	30.8
74.4	FR toddler	39.5	Spinach	6.1	Potatoes	4.5	Beans (with pods)	19.5
69.6	NL child	20.8	Spinach	8.0	Leafy brassica	7.2	Lettuce and other salad plants	18.9
60.0	UK Infant	36.3	Sugar beet (root)	7.0	Rice	3.9	Potatoes	49.8
58.2	IE adult	8.8	Leafy brassica	7.4	Wine grapes	7.0	Spinach	16.6
49.7	WHO cluster diet D	16.1	Leafy brassica	7.8	Wheat	6.1	Rice	19.0
49.1	SE general population 90th percentile	11.0	Leafy brassica	9.4	Lettuce and other salad plants	5.0	Potatoes	15.3
46.5	FR infant	24.7	Spinach	5.0	Potatoes	3.4	Beans (with pods)	14.3
45.8	PT General population	14.6	Wine grapes	8.7	Rice	6.4	Potatoes	16.3
45.3	FR all population	23.5	Wine grapes	7.5	Lettuce and other salad plants	3.9	Wheat	8.2
44.8	WHO cluster diet E	9.4	Wine grapes	4.7	Wheat	4.6	Lettuce and other salad plants	15.5
44.3	DE child	11.4	Spinach	7.5	Table grapes	4.9	Wheat	16.4
42.9	ES child	9.8	Lettuce and other salad plants	5.3	Rice	5.3	Wheat	11.9
40.8	ES adult	12.5	Lettuce and other salad plants	4.5	Beet leaves (chard)	4.1	Spinach	7.6
40.8	IT adult	13.1	Lettuce and other salad plants	5.3	Spinach	5.0	Wheat	9.8
40.0	NL general	7.9	Spinach	5.6	Lettuce and other salad plants	5.0	Leafy brassica	9.2
39.3	WHO regional European diet	9.6	Lettuce and other salad plants	4.8	Potatoes	3.6	Wheat	14.8
38.5	UK vegetarian	13.6	Sugar beet (root)	4.8	Wine grapes	4.2	Rice	21.5
37.9	IT kids/toddler	9.6	Lettuce and other salad plants	8.0	Wheat	3.4	Beet leaves (chard)	13.8
37.8	WHO Cluster diet F	7.0	Lettuce and other salad plants	5.6	Leafy brassica	4.3	Wheat	13.7
36.3	UK Adult	14.4	Sugar beet (root)	6.4	Wine grapes	4.0	Rice	20.7
30.4	DK child	6.6	Wheat	5.3	Rye	3.3	Lettuce and other salad plants	22.0
22.6	DK adult	8.2	Wine grapes	3.0	Lettuce and other salad plants	2.4	Wheat	7.8
14.9	LT adult	3.8	Potatoes	2.4	Rice	1.6	Head cabbage	8.7
13.6	FI adult	1.8	Lettuce and other salad plants	1.8	Wine grapes	1.6	Leafy brassica	5.6
12.7	PL general population	4.1	Potatoes	1.9	Table grapes	1.7	Tomatoes	7.4

Conclusion:

The estimated Theoretical Maximum Daily Intakes based on MS and WHO diets and pTMRLs were in the range of 12.7 % to 108 % of the ADI.
For 1 diets the ADI is exceeded. Further refinements of the dietary intake estimates have not been performed. A public health risk can not be excluded at the moment.

Table 3.2-33: Chronic Assessment of Metabolite M310I001 (neurotox trigger)

M310I001

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
1	65
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
65.3	ES child	56.3	Poultry: Meat	4.4	Birds' eggs	0.8	Swine: Liver	
56.5	WHO Cluster diet B	42.8	Poultry: Meat	4.9	Bovine: Kidney	3.0	Birds' eggs	
51.3	WHO cluster diet E	44.5	Poultry: Meat	3.8	Birds' eggs	0.4	Wine grapes	
47.2	FR toddler	36.8	Poultry: Meat	6.2	Birds' eggs	1.8	Spinach	
45.9	WHO regional European diet	38.7	Poultry: Meat	3.8	Birds' eggs	0.6	Swine: Meat	
41.9	NL child	31.0	Poultry: Meat	3.6	Birds' eggs	1.0	Bovine: Liver	
35.2	DE child	26.1	Poultry: Meat	6.8	Birds' eggs	0.5	Spinach	
33.7	ES adult	27.8	Poultry: Meat	2.8	Birds' eggs	0.6	Lettuce and other salad plants	
30.7	FR all population	26.4	Poultry: Meat	1.8	Birds' eggs	1.0	Wine grapes	
30.0	FR infant	24.9	Poultry: Meat	2.7	Birds' eggs	1.1	Spinach	
26.3	WHO Cluster diet F	19.9	Poultry: Meat	2.8	Birds' eggs	0.7	Bovine: Kidney	
23.8	WHO cluster diet D	17.5	Poultry: Meat	2.5	Birds' eggs	1.0	Bovine: Kidney	
23.5	IE adult	15.7	Poultry: Meat	3.0	Sheep: Liver	1.7	Birds' eggs	
19.9	NL general	14.9	Poultry: Meat	1.8	Birds' eggs	0.5	Swine: Meat	
17.6	LT adult	13.8	Poultry: Meat	2.0	Birds' eggs	0.5	Swine: Meat	
13.6	UK Infant	8.2	Birds' eggs	1.6	Sugar beet (root)	1.5	Bovine: Kidney	
11.0	UK Toddler	5.4	Birds' eggs	3.7	Sugar beet (root)	0.5	Bovine: Kidney	
8.3	DK child	5.3	Birds' eggs	1.7	Bovine: Liver	0.3	Wheat	
7.6	SE general population 90th percentile	5.4	Birds' eggs	0.5	Leafy brassica	0.4	Lettuce and other salad plants	
5.4	UK vegetarian	2.1	Birds' eggs	1.6	Poultry: Meat	0.6	Sugar beet (root)	
4.2	DK adult	2.2	Birds' eggs	0.7	Bovine: Liver	0.4	Wine grapes	
3.8	UK Adult	1.9	Birds' eggs	0.6	Sugar beet (root)	0.3	Wine grapes	
2.0	PT General population	0.7	Wine grapes	0.4	Rice	0.3	Potatoes	
2.0	FI adult	1.4	Birds' eggs	0.1	Lettuce and other salad plants	0.1	Wine grapes	
1.8	IT adult	0.6	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.7	IT kids/toddler	0.4	Lettuce and other salad plants	0.4	Wheat	0.1	Beet leaves (chard)	
0.6	PL general population	0.2	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I001 is unlikely to present a public health concern.

Table 3.2-34: Chronic Assessment of Metabolite M310I003 (neurotox trigger)

M310I003

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 5

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
4.8	UK Infant	4.2	Birds' eggs	0.6	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
3.5	WHO Cluster diet B	1.9	Bovine: Kidney	1.5	Birds' eggs		FRUIT (FRESH OR FROZEN)	
3.5	DE child	3.5	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
3.2	FR toddler	3.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
2.9	UK Toddler	2.8	Birds' eggs	0.2	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
2.7	SE general population 90th percentile	2.7	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
2.7	DK child	2.7	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
2.3	ES child	2.2	Birds' eggs	0.1	Swine: Kidney		FRUIT (FRESH OR FROZEN)	
2.1	NL child	1.8	Birds' eggs	0.2	Swine: Kidney	0.2	Swine: Kidney	
2.0	WHO regional European diet	1.9	Birds' eggs	0.1	Bovine: Kidney	0.0	Poultry: Liver	
2.0	WHO cluster diet E	2.0	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.7	WHO Cluster diet F	1.4	Birds' eggs	0.3	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
1.7	WHO cluster diet D	1.3	Birds' eggs	0.4	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
1.5	ES adult	1.4	Birds' eggs	0.0	Swine: Kidney		FRUIT (FRESH OR FROZEN)	
1.4	FR infant	1.4	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.1	DK adult	1.1	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.1	UK vegetarian	1.1	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.0	UK Adult	1.0	Birds' eggs	0.1	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
1.0	LT adult	1.0	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.9	FR all population	0.9	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.9	NL general	0.9	Birds' eggs	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)	
0.8	IE adult	0.8	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.7	FI adult	0.7	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I003 is unlikely to present a public health concern.

Table 3.2-35: Chronic Assessment of Metabolite M310I004 (neurotox trigger)

M310I004

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
17

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
16.9	WHO Cluster diet B	15.8	Bovine: Kidney	1.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
6.3	UK Infant	5.0	Bovine: Kidney	1.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
4.6	NL child	1.3	Swine: Kidney	1.3	Swine: Kidney	1.1	Bovine: Liver	
3.5	WHO cluster diet D	3.2	Bovine: Kidney	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
3.2	IE adult	3.2	Sheep: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
2.3	WHO Cluster diet F	2.2	Bovine: Kidney	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
1.8	DK child	1.8	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.8	UK Toddler	1.5	Bovine: Kidney	0.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
1.6	ES child	0.9	Swine: Liver	0.5	Swine: Kidney	0.2	Bovine: Liver	
0.8	WHO regional European diet	0.7	Bovine: Kidney	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.8	DK adult	0.8	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.8	UK Adult	0.6	Bovine: Kidney	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.6	NL general	0.4	Swine: Liver	0.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.6	ES adult	0.2	Swine: Liver	0.2	Swine: Kidney	0.2	Bovine: Liver	
0.5	LT adult	0.3	Swine: Liver	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.2	WHO cluster diet E	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:

The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I004 is unlikely to present a public health concern.

Table 3.2-36: Chronic Assessment of Metabolite M310I005 (neurotox trigger)

M310I005			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	1 9
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
9.3	UK Toddler	7.1	Sugar beet (root)	0.5	Rice	0.4	Wheat	
7.4	WHO Cluster diet B	1.0	Olives for oil production	0.9	Wine grapes	0.9	Wheat	
6.4	FR toddler	3.4	Spinach	0.5	Potatoes	0.4	Beans (with pods)	
6.0	NL child	1.8	Spinach	0.7	Leafy brassica	0.6	Lettuce and other salad plants	
5.2	UK infant	3.1	Sugar beet (root)	0.6	Rice	0.3	Potatoes	
5.0	IE adult	0.8	Leafy brassica	0.6	Wine grapes	0.6	Spinach	
4.3	WHO cluster diet D	1.4	Leafy brassica	0.7	Wheat	0.5	Rice	
4.2	SE general population 90th percentile	0.9	Leafy brassica	0.8	Lettuce and other salad plants	0.4	Potatoes	
4.0	FR infant	2.1	Spinach	0.4	Potatoes	0.3	Beans (with pods)	
3.9	PT General population	1.3	Wine grapes	0.7	Rice	0.6	Potatoes	
3.9	FR all population	2.0	Wine grapes	0.6	Lettuce and other salad plants	0.3	Wheat	
3.9	WHO cluster diet E	0.8	Wine grapes	0.4	Wheat	0.4	Lettuce and other salad plants	
3.8	DE child	1.0	Spinach	0.6	Table grapes	0.4	Wheat	
3.7	ES child	0.8	Lettuce and other salad plants	0.5	Rice	0.5	Wheat	
3.5	ES adult	1.1	Lettuce and other salad plants	0.4	Beet leaves (chard)	0.4	Spinach	
3.5	IT adult	1.1	Lettuce and other salad plants	0.5	Spinach	0.4	Wheat	
3.4	NL general	0.7	Spinach	0.5	Lettuce and other salad plants	0.4	Leafy brassica	
3.4	WHO regional European diet	0.8	Lettuce and other salad plants	0.4	Potatoes	0.3	Wheat	
3.3	UK vegetarian	1.2	Sugar beet (root)	0.4	Wine grapes	0.4	Rice	
3.3	IT kids/toddler	0.8	Lettuce and other salad plants	0.7	Wheat	0.3	Beet leaves (chard)	
3.3	WHO Cluster diet F	0.6	Lettuce and other salad plants	0.5	Leafy brassica	0.4	Wheat	
3.1	UK Adult	1.2	Sugar beet (root)	0.5	Wine grapes	0.3	Rice	
2.6	DK child	0.6	Wheat	0.5	Rye	0.3	Lettuce and other salad plants	
1.9	DK adult	0.7	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
1.3	LT adult	0.3	Potatoes	0.2	Rice	0.1	Head cabbage	
1.2	FI adult	0.2	Lettuce and other salad plants	0.2	Wine grapes	0.1	Leafy brassica	
1.1	PL general population	0.4	Potatoes	0.2	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I005 is unlikely to present a public health concern.

Table 3.2-37: Chronic Assessment of Metabolite M310I006 (neurotox trigger)

M310I006

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	1 10
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
9.6	UK Toddler	7.3	Sugar beet (root)	0.6	Rice	0.4	Wheat	
7.6	WHO Cluster diet B	1.0	Olives for oil production	0.9	Wine grapes	0.9	Wheat	
6.6	FR toddler	3.5	Spinach	0.5	Potatoes	0.4	Beans (with pods)	
6.2	NL child	1.8	Spinach	0.7	Leafy brassica	0.6	Lettuce and other salad plants	
5.3	UK Infant	3.2	Sugar beet (root)	0.6	Rice	0.3	Potatoes	
5.2	IE adult	0.8	Leafy brassica	0.7	Wine grapes	0.6	Spinach	
4.4	WHO cluster diet D	1.4	Leafy brassica	0.7	Wheat	0.5	Rice	
4.4	SE general population 90th percentile	1.0	Leafy brassica	0.8	Lettuce and other salad plants	0.4	Potatoes	
4.1	FR infant	2.2	Spinach	0.4	Potatoes	0.3	Beans (with pods)	
4.1	PT General population	1.3	Wine grapes	0.8	Rice	0.6	Potatoes	
4.0	FR all population	2.1	Wine grapes	0.7	Lettuce and other salad plants	0.4	Wheat	
4.0	WHO cluster diet E	0.8	Wine grapes	0.4	Wheat	0.4	Lettuce and other salad plants	
3.9	DE child	1.0	Spinach	0.7	Table grapes	0.4	Wheat	
3.8	ES child	0.9	Lettuce and other salad plants	0.5	Rice	0.5	Wheat	
3.6	ES adult	1.1	Lettuce and other salad plants	0.4	Beet leaves (chard)	0.4	Spinach	
3.6	IT adult	1.2	Lettuce and other salad plants	0.5	Spinach	0.4	Wheat	
3.6	NL general	0.7	Spinach	0.5	Lettuce and other salad plants	0.4	Leafy brassica	
3.5	WHO regional European diet	0.9	Lettuce and other salad plants	0.4	Potatoes	0.3	Wheat	
3.4	UK vegetarian	1.2	Sugar beet (root)	0.4	Wine grapes	0.4	Rice	
3.4	IT kids/toddler	0.9	Lettuce and other salad plants	0.7	Wheat	0.3	Beet leaves (chard)	
3.4	WHO Cluster diet F	0.6	Lettuce and other salad plants	0.5	Leafy brassica	0.4	Wheat	
3.2	UK Adult	1.3	Sugar beet (root)	0.6	Wine grapes	0.4	Rice	
2.7	DK child	0.6	Wheat	0.5	Rye	0.3	Lettuce and other salad plants	
2.0	DK adult	0.7	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
1.3	LT adult	0.3	Potatoes	0.2	Rice	0.1	Head cabbage	
1.2	FI adult	0.2	Lettuce and other salad plants	0.2	Wine grapes	0.1	Leafy brassica	
1.1	PL general population	0.4	Potatoes	0.2	Table grapes	0.2	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I006 is unlikely to present a public health concern.

Table 3.2-38: Chronic Assessment of Metabolite M310I007 (neurotox trigger)

M310I007

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
2	17
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
17.1	UK Toddler	13.0	Sugar beet (root)	1.0	Rice	0.7	Wheat	
13.6	WHO Cluster diet B	1.8	Olives for oil production	1.7	Wine grapes	1.6	Wheat	
11.8	FR toddler	6.3	Spinach	1.0	Potatoes	0.7	Beans (with pods)	
11.0	NL child	3.3	Spinach	1.3	Leafy brassica	1.1	Lettuce and other salad plants	
9.5	UK Infant	5.7	Sugar beet (root)	1.1	Rice	0.6	Potatoes	
9.2	IE adult	1.4	Leafy brassica	1.2	Wine grapes	1.1	Spinach	
7.9	WHO cluster diet D	2.6	Leafy brassica	1.2	Wheat	1.0	Rice	
7.8	SE general population 90th percentile	1.7	Leafy brassica	1.5	Lettuce and other salad plants	0.8	Potatoes	
7.4	FR infant	3.9	Spinach	0.8	Potatoes	0.5	Beans (with pods)	
7.3	PT General population	2.3	Wine grapes	1.4	Rice	1.0	Potatoes	
7.2	FR all population	3.7	Wine grapes	1.2	Lettuce and other salad plants	0.6	Wheat	
7.1	WHO cluster diet E	1.5	Wine grapes	0.7	Wheat	0.7	Lettuce and other salad plants	
7.0	DE child	1.8	Spinach	1.2	Table grapes	0.8	Wheat	
6.8	ES child	1.5	Lettuce and other salad plants	0.8	Rice	0.8	Wheat	
6.5	ES adult	2.0	Lettuce and other salad plants	0.7	Beet leaves (chard)	0.7	Spinach	
6.5	IT adult	2.1	Lettuce and other salad plants	0.6	Spinach	0.6	Wheat	
6.3	NL general	1.3	Spinach	0.9	Lettuce and other salad plants	0.8	Leafy brassica	
6.2	WHO regional European diet	1.5	Lettuce and other salad plants	0.8	Potatoes	0.6	Wheat	
6.1	UK vegetarian	2.2	Sugar beet (root)	0.8	Wine grapes	0.7	Rice	
6.0	IT kids/toddler	1.5	Lettuce and other salad plants	1.3	Wheat	0.5	Beet leaves (chard)	
6.0	WHO Cluster diet F	1.1	Lettuce and other salad plants	0.9	Leafy brassica	0.7	Wheat	
5.8	UK Adult	2.3	Sugar beet (root)	1.0	Wine grapes	0.6	Rice	
4.8	DK child	1.0	Wheat	0.8	Rye	0.5	Lettuce and other salad plants	
3.6	DK adult	1.3	Wine grapes	0.5	Lettuce and other salad plants	0.4	Wheat	
2.4	LT adult	0.6	Potatoes	0.4	Rice	0.3	Head cabbage	
2.2	FI adult	0.3	Lettuce and other salad plants	0.3	Wine grapes	0.3	Leafy brassica	
2.0	PL general population	0.7	Potatoes	0.3	Table grapes	0.3	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I007 is unlikely to present a public health concern.

Table 3.2-39: Chronic Assessment of Metabolite M310I008 (neurotox trigger)

M310I008

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
2	17
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
17.1	UK Toddler	13.0	Sugar beet (root)	1.0	Rice	0.7	Wheat	
13.6	WHO Cluster diet B	1.8	Olives for oil production	1.7	Wine grapes	1.6	Wheat	
11.8	FR toddler	6.3	Spinach	1.0	Potatoes	0.7	Beans (with pods)	
11.0	NL child	3.3	Spinach	1.3	Leafy brassica	1.1	Lettuce and other salad plants	
9.5	UK Infant	5.7	Sugar beet (root)	1.1	Rice	0.6	Potatoes	
9.2	IE adult	1.4	Leafy brassica	1.2	Wine grapes	1.1	Spinach	
7.9	WHO cluster diet D	2.6	Leafy brassica	1.2	Wheat	1.0	Rice	
7.8	SE general population 90th percentile	1.7	Leafy brassica	1.5	Lettuce and other salad plants	0.8	Potatoes	
7.4	FR infant	3.9	Spinach	0.8	Potatoes	0.5	Beans (with pods)	
7.3	PT General population	2.3	Wine grapes	1.4	Rice	1.0	Potatoes	
7.2	FR all population	3.7	Wine grapes	1.2	Lettuce and other salad plants	0.6	Wheat	
7.1	WHO cluster diet E	1.5	Wine grapes	0.7	Wheat	0.7	Lettuce and other salad plants	
7.0	DE child	1.8	Spinach	1.2	Table grapes	0.8	Wheat	
6.8	ES child	1.5	Lettuce and other salad plants	0.8	Rice	0.8	Wheat	
6.5	ES adult	2.0	Lettuce and other salad plants	0.7	Beet leaves (chard)	0.7	Spinach	
6.5	IT adult	2.1	Lettuce and other salad plants	0.6	Spinach	0.6	Wheat	
6.3	NL general	1.3	Spinach	0.9	Lettuce and other salad plants	0.8	Leafy brassica	
6.2	WHO regional European diet	1.5	Lettuce and other salad plants	0.8	Potatoes	0.6	Wheat	
6.1	UK vegetarian	2.2	Sugar beet (root)	0.8	Wine grapes	0.7	Rice	
6.0	IT kids/toddler	1.5	Lettuce and other salad plants	1.3	Wheat	0.5	Beet leaves (chard)	
6.0	WHO Cluster diet F	1.1	Lettuce and other salad plants	0.9	Leafy brassica	0.7	Wheat	
5.8	UK Adult	2.3	Sugar beet (root)	1.0	Wine grapes	0.6	Rice	
4.8	DK child	1.0	Wheat	0.8	Rye	0.5	Lettuce and other salad plants	
3.6	DK adult	1.3	Wine grapes	0.5	Lettuce and other salad plants	0.4	Wheat	
2.4	LT adult	0.6	Potatoes	0.4	Rice	0.3	Head cabbage	
2.2	FI adult	0.3	Lettuce and other salad plants	0.3	Wine grapes	0.3	Leafy brassica	
2.0	PL general population	0.7	Potatoes	0.3	Table grapes	0.3	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I008 is unlikely to present a public health concern.

Table 3.2-40: Chronic Assessment of Metabolite M310I009 (neurotox trigger)

M310I009

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
3	21
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
21.0	UK Toddler	16.0	Sugar beet (root)	1.2	Rice	0.9	Wheat	
16.7	WHO Cluster diet B	2.2	Olives for oil production	2.0	Wine grapes	2.0	Wheat	
14.5	FR toddler	7.7	Spinach	1.2	Potatoes	0.9	Beans (with pods)	
13.5	NL child	4.0	Spinach	1.6	Leafy brassica	1.4	Lettuce and other salad plants	
11.7	UK infant	7.1	Sugar beet (root)	1.4	Rice	0.8	Potatoes	
11.3	IE adult	1.7	Leafy brassica	1.4	Wine grapes	1.4	Spinach	
9.7	WHO cluster diet D	3.1	Leafy brassica	1.5	Wheat	1.2	Rice	
9.5	SE general population 90th percentile	2.1	Leafy brassica	1.8	Lettuce and other salad plants	1.0	Potatoes	
9.0	FR infant	4.8	Spinach	1.0	Potatoes	0.7	Beans (with pods)	
8.9	PT General population	2.8	Wine grapes	1.7	Rice	1.2	Potatoes	
8.8	FR all population	4.6	Wine grapes	1.5	Lettuce and other salad plants	0.8	Wheat	
8.7	WHO cluster diet E	1.8	Wine grapes	0.9	Wheat	0.9	Lettuce and other salad plants	
8.6	DE child	2.2	Spinach	1.5	Table grapes	1.0	Wheat	
8.3	ES child	1.9	Lettuce and other salad plants	1.0	Rice	1.0	Wheat	
7.9	ES adult	2.4	Lettuce and other salad plants	0.9	Beet leaves (chard)	0.8	Spinach	
7.9	IT adult	2.5	Lettuce and other salad plants	1.0	Spinach	1.0	Wheat	
7.8	NL general	1.5	Spinach	1.1	Lettuce and other salad plants	1.0	Leafy brassica	
7.6	WHO regional European diet	1.9	Lettuce and other salad plants	0.9	Potatoes	0.7	Wheat	
7.5	UK vegetarian	2.6	Sugar beet (root)	0.9	Wine grapes	0.8	Rice	
7.4	IT kids/toddler	1.9	Lettuce and other salad plants	1.6	Wheat	0.7	Beet leaves (chard)	
7.3	WHO Cluster diet F	1.4	Lettuce and other salad plants	1.1	Leafy brassica	0.8	Wheat	
7.1	UK Adult	2.8	Sugar beet (root)	1.2	Wine grapes	0.8	Rice	
5.9	DK child	1.3	Wheat	1.0	Rye	0.6	Lettuce and other salad plants	
4.4	DK adult	1.6	Wine grapes	0.6	Lettuce and other salad plants	0.5	Wheat	
2.9	LT adult	0.7	Potatoes	0.5	Rice	0.3	Head cabbage	
2.6	FI adult	0.4	Lettuce and other salad plants	0.3	Wine grapes	0.3	Leafy brassica	
2.5	PL general population	0.8	Potatoes	0.4	Table grapes	0.3	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I009 is unlikely to present a public health concern.

Table 3.2-41: Chronic Assessment of Metabolite M310I010 (neurotox trigger)

M310I010			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
0	51
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
50.9	UK Infant	47.7	Milk and cream,	1.9	Bovine: Kidney	0.7	Bovine: Liver	0.4
49.5	FR toddler	48.9	Milk and cream,	0.3	Spinach	0.1	Potatoes	0.2
38.8	NL child	36.2	Milk and cream,	0.6	Bovine: Liver	0.5	Swine: Liver	0.2
32.1	FR infant	31.7	Milk and cream,	0.2	Spinach	0.0	Potatoes	0.1
27.1	UK Toddler	25.5	Milk and cream,	0.7	Sugar beet (root)	0.6	Bovine: Kidney	0.8
18.0	DE child	17.6	Milk and cream,	0.1	Spinach	0.1	Table grapes	0.1
16.8	DK child	15.6	Milk and cream,	0.9	Bovine: Liver	0.1	Wheat	0.2
16.5	ES child	15.4	Milk and cream,	0.5	Swine: Liver	0.2	Swine: Kidney	0.1
15.7	SE general population 90th percentile	15.3	Milk and cream,	0.1	Leafy brassica	0.1	Lettuce and other salad plants	0.1
11.4	WHO Cluster diet B	6.2	Bovine: Kidney	3.9	Milk and cream,	0.5	Bovine: Liver	0.3
8.7	NL general	8.1	Milk and cream,	0.2	Swine: Liver	0.1	Bovine: Liver	0.1
8.0	WHO cluster diet D	6.2	Milk and cream,	1.3	Bovine: Kidney	0.1	Leafy brassica	0.2
7.2	DK adult	6.6	Milk and cream,	0.4	Bovine: Liver	0.1	Wine grapes	0.1
7.1	FI adult	7.0	Milk and cream,	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0
6.7	ES adult	6.1	Milk and cream,	0.1	Swine: Liver	0.1	Lettuce and other salad plants	0.1
6.6	WHO regional European diet	5.9	Milk and cream,	0.3	Bovine: Kidney	0.1	Lettuce and other salad plants	0.1
6.1	WHO Cluster diet F	4.9	Milk and cream,	0.8	Bovine: Kidney	0.1	Bovine: Liver	0.1
5.6	IE adult	3.4	Milk and cream,	1.7	Sheep: Liver	0.1	Leafy brassica	0.1
5.3	LT adult	4.9	Milk and cream,	0.2	Swine: Liver	0.1	Bovine: Liver	0.1
4.3	UK vegetarian	4.0	Milk and cream,	0.1	Sugar beet (root)	0.0	Wine grapes	0.2
4.3	UK Adult	3.7	Milk and cream,	0.2	Bovine: Kidney	0.1	Sugar beet (root)	0.2
4.2	WHO cluster diet E	3.7	Milk and cream,	0.1	Bovine: Liver	0.1	Wine grapes	0.1
3.7	FR all population	3.3	Milk and cream,	0.2	Wine grapes	0.1	Lettuce and other salad plants	0.1
0.4	PT General population	0.1	Wine grapes	0.1	Rice	0.1	Potatoes	0.1
0.3	IT adult	0.1	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	0.1
0.3	IT kids/toddler	0.1	Lettuce and other salad plants	0.1	Wheat	0.0	Beet leaves (chard)	0.1
0.1	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.1

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I010 is unlikely to present a public health concern.

Table 3.2-42: Chronic Assessment of Metabolite M310I011 (neurotox trigger)

M310I011

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
3	21
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
21.3	UK Toddler	16.0	Sugar beet (root)	1.2	Rice	0.9	Wheat	
19.1	WHO Cluster diet B	2.2	Olives for oil production	2.1	Bovine: Kidney	2.0	Wine grapes	
14.5	FR toddler	7.7	Spinach	1.2	Potatoes	0.9	Beans (with pods)	
14.3	NL child	4.0	Spinach	1.6	Leafy brassica	1.4	Lettuce and other salad plants	
12.6	UK Infant	7.1	Sugar beet (root)	1.4	Rice	0.8	Potatoes	
12.0	IE adult	1.7	Leafy brassica	1.4	Wine grapes	1.4	Spinach	
10.1	WHO cluster diet D	3.1	Leafy brassica	1.5	Wheat	1.2	Rice	
9.5	SE general population 90th percentile	2.1	Leafy brassica	1.8	Lettuce and other salad plants	1.0	Potatoes	
9.0	FR infant	4.8	Spinach	1.0	Potatoes	0.7	Beans (with pods)	
8.9	PT General population	2.8	Wine grapes	1.7	Rice	1.2	Potatoes	
8.8	FR all population	4.6	Wine grapes	1.5	Lettuce and other salad plants	0.8	Wheat	
8.8	WHO cluster diet E	1.8	Wine grapes	0.9	Wheat	0.9	Lettuce and other salad plants	
8.7	ES child	1.9	Lettuce and other salad plants	1.0	Rice	1.0	Wheat	
8.6	DE child	2.2	Spinach	1.5	Table grapes	1.0	Wheat	
8.0	ES adult	2.4	Lettuce and other salad plants	0.9	Beet leaves (chard)	0.8	Spinach	
7.9	IT adult	2.5	Lettuce and other salad plants	1.0	Spinach	1.0	Wheat	
7.9	NL general	1.5	Spinach	1.1	Lettuce and other salad plants	1.0	Leafy brassica	
7.8	WHO regional European diet	1.9	Lettuce and other salad plants	0.9	Potatoes	0.7	Wheat	
7.7	WHO Cluster diet F	1.4	Lettuce and other salad plants	1.1	Leafy brassica	0.8	Wheat	
7.5	UK vegetarian	2.6	Sugar beet (root)	0.9	Wine grapes	0.8	Rice	
7.4	IT kids/toddler	1.9	Lettuce and other salad plants	1.6	Wheat	0.7	Beet leaves (chard)	
7.2	UK Adult	2.8	Sugar beet (root)	1.2	Wine grapes	0.8	Rice	
6.3	DK child	1.3	Wheat	1.0	Rye	0.6	Lettuce and other salad plants	
4.6	DK adult	1.6	Wine grapes	0.6	Lettuce and other salad plants	0.5	Wheat	
3.0	LT adult	0.7	Potatoes	0.5	Rice	0.3	Head cabbage	
2.6	FI adult	0.4	Lettuce and other salad plants	0.3	Wine grapes	0.3	Leafy brassica	
2.5	PL general population	0.8	Potatoes	0.4	Table grapes	0.3	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I011 is unlikely to present a public health concern.

Table 3.2-43: Chronic Assessment of Metabolite M310I013 (neurotox trigger)

M310I013			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
0	2
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1.8	UK Toddler	1.4	Sugar beet (root)	0.1	Rice	0.1	Wheat	0.2
1.4	WHO Cluster diet B	0.2	Olives for oil production	0.2	Wine grapes	0.2	Wheat	0.4
1.2	FR toddler	0.7	Spinach	0.1	Potatoes	0.1	Beans (with pods)	0.3
1.2	NL child	0.3	Spinach	0.1	Leafy brassica	0.1	Lettuce and other salad plants	0.3
1.0	UK Infant	0.6	Sugar beet (root)	0.1	Rice	0.1	Potatoes	0.2
1.0	IE adult	0.1	Leafy brassica	0.1	Wine grapes	0.1	Spinach	0.2
0.8	WHO cluster diet D	0.3	Leafy brassica	0.1	Wheat	0.1	Rice	0.3
0.8	SE general population 90th percentile	0.2	Leafy brassica	0.2	Lettuce and other salad plants	0.1	Potatoes	0.2
0.8	FR infant	0.4	Spinach	0.1	Potatoes	0.1	Beans (with pods)	0.2
0.8	PT General population	0.2	Wine grapes	0.1	Rice	0.1	Potatoes	0.3
0.8	FR all population	0.4	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	0.1
0.7	WHO cluster diet E	0.2	Wine grapes	0.1	Wheat	0.1	Lettuce and other salad plants	0.2
0.7	DE child	0.2	Spinach	0.1	Table grapes	0.1	Wheat	0.2
0.7	ES child	0.2	Lettuce and other salad plants	0.1	Rice	0.1	Wheat	0.2
0.7	ES adult	0.2	Lettuce and other salad plants	0.1	Beet leaves (chard)	0.1	Spinach	0.1
0.7	IT adult	0.2	Lettuce and other salad plants	0.1	Spinach	0.1	Wheat	0.2
0.7	NL general	0.1	Spinach	0.1	Lettuce and other salad plants	0.1	Leafy brassica	0.1
0.7	WHO regional European diet	0.2	Lettuce and other salad plants	0.1	Potatoes	0.1	Wheat	0.2
0.6	UK vegetarian	0.2	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	0.1
0.6	IT kids/toddler	0.2	Lettuce and other salad plants	0.1	Wheat	0.1	Beet leaves (chard)	0.2
0.6	WHO Cluster diet F	0.1	Lettuce and other salad plants	0.1	Leafy brassica	0.1	Wheat	0.2
0.6	UK Adult	0.2	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	0.1
0.5	DK child	0.1	Wheat	0.1	Rye	0.1	Lettuce and other salad plants	0.4
0.4	DK adult	0.1	Wine grapes	0.1	Lettuce and other salad plants	0.0	Wheat	0.1
0.2	LT adult	0.1	Potatoes	0.0	Rice	0.0	Head cabbage	0.1
0.2	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	0.1
0.2	PL general population	0.1	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.1

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I013 is unlikely to present a public health concern.

Table 3.2-44: Chronic Assessment of Metabolite M310I017 (neurotox trigger)

M310I017			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
5	45
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
45.0	UK Toddler	34.3	Sugar beet (root)	2.6	Rice	2.0	Wheat	
35.8	WHO Cluster diet B	4.8	Olives for oil production	4.4	Wine grapes	4.3	Wheat	
31.0	FR toddler	16.5	Spinach	2.5	Potatoes	1.9	Beans (with pods)	
29.0	NL child	8.6	Spinach	3.3	Leafy brassica	3.0	Lettuce and other salad plants	
25.0	UK Infant	15.1	Sugar beet (root)	2.9	Rice	1.6	Potatoes	
24.3	IE adult	3.7	Leafy brassica	3.1	Wine grapes	2.9	Spinach	
20.7	WHO cluster diet D	6.7	Leafy brassica	3.3	Wheat	2.5	Rice	
20.5	SE general population 90th percentile	4.6	Leafy brassica	3.9	Lettuce and other salad plants	2.1	Potatoes	
19.4	FR infant	10.3	Spinach	2.1	Potatoes	1.4	Beans (with pods)	
19.1	PT General population	6.1	Wine grapes	3.6	Rice	2.7	Potatoes	
18.9	FR all population	9.8	Wine grapes	3.1	Lettuce and other salad plants	1.6	Wheat	
18.7	WHO cluster diet E	3.9	Wine grapes	2.0	Wheat	1.9	Lettuce and other salad plants	
18.5	DE child	4.8	Spinach	3.1	Table grapes	2.1	Wheat	
17.9	ES child	4.1	Lettuce and other salad plants	2.2	Rice	2.2	Wheat	
17.0	ES adult	5.2	Lettuce and other salad plants	1.9	Beet leaves (chard)	1.7	Spinach	
17.0	IT adult	5.4	Lettuce and other salad plants	2.2	Spinach	2.1	Wheat	
16.7	NL general	3.3	Spinach	2.3	Lettuce and other salad plants	2.1	Leafy brassica	
16.4	WHO regional European diet	4.0	Lettuce and other salad plants	2.0	Potatoes	1.5	Wheat	
16.1	UK vegetarian	5.7	Sugar beet (root)	2.0	Wine grapes	1.8	Rice	
15.8	IT kids/toddler	4.0	Lettuce and other salad plants	3.3	Wheat	1.4	Beet leaves (chard)	
15.7	WHO Cluster diet F	2.9	Lettuce and other salad plants	2.3	Leafy brassica	1.8	Wheat	
15.1	UK Adult	6.0	Sugar beet (root)	2.7	Wine grapes	1.7	Rice	
12.7	DK child	2.8	Wheat	2.2	Rye	1.4	Lettuce and other salad plants	
9.4	DK adult	3.4	Wine grapes	1.3	Lettuce and other salad plants	1.0	Wheat	
6.2	LT adult	1.6	Potatoes	1.0	Rice	0.7	Head cabbage	
5.7	FI adult	0.8	Lettuce and other salad plants	0.7	Wine grapes	0.7	Leafy brassica	
5.3	PL general population	1.7	Potatoes	0.8	Table grapes	0.7	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I017 is unlikely to present a public health concern.

Table 3.2-45: Chronic Assessment of Metabolite M310I018 (neurotox trigger)

PbAld			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
0	2
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
2.1	UK Toddler	1.6	Sugar beet (root)	0.1	Rice	0.1	Wheat	0.2
1.7	WHO Cluster diet B	0.2	Olives for oil production	0.2	Wine grapes	0.2	Wheat	0.4
1.4	FR toddler	0.8	Spinach	0.1	Potatoes	0.1	Beans (with pods)	0.3
1.4	NL child	0.4	Spinach	0.2	Leafy brassica	0.1	Lettuce and other salad plants	0.3
1.2	UK Infant	0.7	Sugar beet (root)	0.1	Rice	0.1	Potatoes	0.2
1.1	IE adult	0.2	Leafy brassica	0.1	Wine grapes	0.1	Spinach	0.3
1.0	WHO cluster diet D	0.3	Leafy brassica	0.2	Wheat	0.1	Rice	0.3
1.0	SE general population 90th percentile	0.2	Leafy brassica	0.2	Lettuce and other salad plants	0.1	Potatoes	0.2
0.9	FR infant	0.5	Spinach	0.1	Potatoes	0.1	Beans (with pods)	0.2
0.9	PT General population	0.3	Wine grapes	0.2	Rice	0.1	Potatoes	0.3
0.9	FR all population	0.5	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	0.1
0.9	WHO cluster diet E	0.2	Wine grapes	0.1	Wheat	0.1	Lettuce and other salad plants	0.3
0.9	DE child	0.2	Spinach	0.1	Table grapes	0.1	Wheat	0.2
0.8	ES child	0.2	Lettuce and other salad plants	0.1	Rice	0.1	Wheat	0.2
0.8	ES adult	0.2	Lettuce and other salad plants	0.1	Beet leaves (chard)	0.1	Spinach	0.1
0.8	IT adult	0.3	Lettuce and other salad plants	0.1	Spinach	0.1	Wheat	0.1
0.8	NL general	0.2	Spinach	0.1	Lettuce and other salad plants	0.1	Leafy brassica	0.1
0.8	WHO regional European diet	0.2	Lettuce and other salad plants	0.1	Potatoes	0.1	Wheat	0.2
0.7	UK vegetarian	0.3	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	0.1
0.7	IT kids/toddler	0.2	Lettuce and other salad plants	0.2	Wheat	0.1	Beet leaves (chard)	0.2
0.7	WHO Cluster diet F	0.1	Lettuce and other salad plants	0.1	Leafy brassica	0.1	Wheat	0.2
0.7	UK Adult	0.3	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	0.1
0.6	DK child	0.1	Wheat	0.1	Rye	0.1	Lettuce and other salad plants	0.4
0.4	DK adult	0.2	Wine grapes	0.1	Lettuce and other salad plants	0.0	Wheat	0.1
0.3	LT adult	0.1	Potatoes	0.0	Rice	0.0	Head cabbage	0.1
0.3	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	0.1
0.2	PL general population	0.1	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.1

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of PbAld is unlikely to present a public health concern.

Table 3.2-46: Chronic Assessment of Metabolite M310I019 (neurotox trigger)

M310I019

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
0.2	NL child	0.2	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	WHO regional European diet	0.2	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.0	NL general	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I019 is unlikely to present a public health concern.

Table 3.2-47: Chronic Assessment of Metabolite M310I021 (neurotox trigger)

Prepare workbook for refined calculations

Undo refined calculations

M310I021			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
10

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
9.6	IE adult	9.6	Sheep: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
6.3	NL child	3.2	Bovine: Liver	3.1	Swine: Liver		FRUIT (FRESH OR FROZEN)	
5.4	DK child	5.4	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
3.9	UK Infant	3.9	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
3.4	ES child	2.7	Swine: Liver	0.7	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
3.1	WHO Cluster diet B	3.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
2.3	DK adult	2.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.9	NL general	1.1	Swine: Liver	0.8	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
1.6	LT adult	0.9	Swine: Liver	0.7	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
1.2	ES adult	0.7	Swine: Liver	0.5	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.9	UK Toddler	0.9	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.7	WHO cluster diet E	0.7	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.6	WHO cluster diet D	0.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.6	UK Adult	0.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.4	WHO Cluster diet F	0.4	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.3	WHO regional European diet	0.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I021 is unlikely to present a public health concern.

Table 3.2-48: Chronic Assessment of Metabolite M310I024 (neurotox trigger)

PbAlc

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
3	21
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
21.0	UK Toddler	16.0	Sugar beet (root)	1.2	Rice	0.9	Wheat	
16.7	WHO Cluster diet B	2.2	Olives for oil production	2.0	Wine grapes	2.0	Wheat	
14.5	FR toddler	7.7	Spinach	1.2	Potatoes	0.9	Beans (with pods)	
13.5	NL child	4.0	Spinach	1.6	Leafy brassica	1.4	Lettuce and other salad plants	
11.7	UK infant	7.1	Sugar beet (root)	1.4	Rice	0.8	Potatoes	
11.3	IE adult	1.7	Leafy brassica	1.4	Wine grapes	1.4	Spinach	
9.7	WHO cluster diet D	3.1	Leafy brassica	1.5	Wheat	1.2	Rice	
9.5	SE general population 90th percentile	2.1	Leafy brassica	1.8	Lettuce and other salad plants	1.0	Potatoes	
9.0	FR infant	4.8	Spinach	1.0	Potatoes	0.7	Beans (with pods)	
8.9	PT General population	2.8	Wine grapes	1.7	Rice	1.2	Potatoes	
8.8	FR all population	4.6	Wine grapes	1.5	Lettuce and other salad plants	0.8	Wheat	
8.7	WHO cluster diet E	1.8	Wine grapes	0.9	Wheat	0.9	Lettuce and other salad plants	
8.6	DE child	2.2	Spinach	1.5	Table grapes	1.0	Wheat	
8.3	ES child	1.9	Lettuce and other salad plants	1.0	Rice	1.0	Wheat	
7.9	ES adult	2.4	Lettuce and other salad plants	0.9	Beet leaves (chard)	0.8	Spinach	
7.9	IT adult	2.5	Lettuce and other salad plants	1.0	Spinach	1.0	Wheat	
7.8	NL general	1.5	Spinach	1.1	Lettuce and other salad plants	1.0	Leafy brassica	
7.6	WHO regional European diet	1.9	Lettuce and other salad plants	0.9	Potatoes	0.7	Wheat	
7.5	UK vegetarian	2.6	Sugar beet (root)	0.9	Wine grapes	0.8	Rice	
7.4	IT kids/toddler	1.9	Lettuce and other salad plants	1.6	Wheat	0.7	Beet leaves (chard)	
7.3	WHO Cluster diet F	1.4	Lettuce and other salad plants	1.1	Leafy brassica	0.8	Wheat	
7.1	UK Adult	2.8	Sugar beet (root)	1.2	Wine grapes	0.8	Rice	
5.9	DK child	1.3	Wheat	1.0	Rye	0.6	Lettuce and other salad plants	
4.4	DK adult	1.6	Wine grapes	0.6	Lettuce and other salad plants	0.5	Wheat	
2.9	LT adult	0.7	Potatoes	0.5	Rice	0.3	Head cabbage	
2.6	FI adult	0.4	Lettuce and other salad plants	0.3	Wine grapes	0.3	Leafy brassica	
2.5	PL general population	0.8	Potatoes	0.4	Table grapes	0.3	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of PbAlc is unlikely to present a public health concern.

Table 3.2-49: Chronic Assessment of Metabolite M310I025 (neurotox trigger)

M310I025			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
3	24
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
24.0	UK Toddler	18.3	Sugar beet (root)	1.4	Rice	1.0	Wheat	
19.1	WHO Cluster diet B	2.6	Olives for oil production	2.3	Wine grapes	2.3	Wheat	
16.5	FR toddler	8.8	Spinach	1.4	Potatoes	1.0	Beans (with pods)	
15.5	NL child	4.6	Spinach	1.8	Leafy brassica	1.6	Lettuce and other salad plants	
13.3	UK Infant	8.1	Sugar beet (root)	1.6	Rice	0.9	Potatoes	
12.9	IE adult	2.0	Leafy brassica	1.6	Wine grapes	1.6	Spinach	
11.0	WHO cluster diet D	3.6	Leafy brassica	1.7	Wheat	1.4	Rice	
10.9	SE general population 90th percentile	2.4	Leafy brassica	2.1	Lettuce and other salad plants	1.1	Potatoes	
10.3	FR infant	5.5	Spinach	1.1	Potatoes	0.8	Beans (with pods)	
10.2	PT General population	3.3	Wine grapes	1.9	Rice	1.4	Potatoes	
10.1	FR all population	5.2	Wine grapes	1.7	Lettuce and other salad plants	0.9	Wheat	
9.9	WHO cluster diet E	2.1	Wine grapes	1.1	Wheat	1.0	Lettuce and other salad plants	
9.9	DE child	2.5	Spinach	1.7	Table grapes	1.1	Wheat	
9.5	ES child	2.2	Lettuce and other salad plants	1.2	Rice	1.2	Wheat	
9.1	ES adult	2.8	Lettuce and other salad plants	1.0	Beet leaves (chard)	0.9	Spinach	
9.1	IT adult	2.9	Lettuce and other salad plants	1.2	Spinach	1.1	Wheat	
8.9	NL general	1.8	Spinach	1.2	Lettuce and other salad plants	1.1	Leafy brassica	
8.7	WHO regional European diet	2.1	Lettuce and other salad plants	1.1	Potatoes	0.8	Wheat	
8.6	UK vegetarian	3.0	Sugar beet (root)	1.1	Wine grapes	0.9	Rice	
8.4	IT kids/toddler	2.1	Lettuce and other salad plants	1.8	Wheat	0.7	Beet leaves (chard)	
8.4	WHO Cluster diet F	1.6	Lettuce and other salad plants	1.2	Leafy brassica	1.0	Wheat	
8.1	UK Adult	3.2	Sugar beet (root)	1.4	Wine grapes	0.9	Rice	
6.8	DK child	1.5	Wheat	1.2	Rye	0.7	Lettuce and other salad plants	
5.0	DK adult	1.8	Wine grapes	0.7	Lettuce and other salad plants	0.5	Wheat	
3.3	LT adult	0.8	Potatoes	0.5	Rice	0.4	Head cabbage	
3.0	FI adult	0.4	Lettuce and other salad plants	0.4	Wine grapes	0.4	Leafy brassica	
2.8	PL general population	0.9	Potatoes	0.4	Table grapes	0.4	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I025 is unlikely to present a public health concern.

Table 3.2-50: Chronic Assessment of Metabolite M310I026 (neurotox trigger)

M310I026			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0003	ARfD (mg/kg bw):	0.005
Source of ADI:	Neurotox	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	0 1
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
0.9	UK Toddler	0.7	Sugar beet (root)	0.1	Rice	0.0	Wheat	0.8
0.7	WHO Cluster diet B	0.1	Olives for oil production	0.1	Wine grapes	0.1	Wheat	0.3
0.6	FR toddler	0.3	Spinach	0.1	Potatoes	0.0	Beans (with pods)	0.2
0.6	NL child	0.2	Spinach	0.1	Leafy brassica	0.1	Lettuce and other salad plants	0.2
0.5	UK Infant	0.3	Sugar beet (root)	0.1	Rice	0.0	Potatoes	0.4
0.5	IE adult	0.1	Leafy brassica	0.1	Wine grapes	0.1	Spinach	0.1
0.4	WHO cluster diet D	0.1	Leafy brassica	0.1	Wheat	0.1	Rice	0.2
0.4	SE general population 90th percentile	0.1	Leafy brassica	0.1	Lettuce and other salad plants	0.0	Potatoes	0.1
0.4	FR infant	0.2	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.1
0.4	PT General population	0.1	Wine grapes	0.1	Rice	0.1	Potatoes	0.1
0.4	FR all population	0.2	Wine grapes	0.1	Lettuce and other salad plants	0.0	Wheat	0.1
0.4	WHO cluster diet E	0.1	Wine grapes	0.0	Wheat	0.0	Lettuce and other salad plants	0.1
0.4	DE child	0.1	Spinach	0.1	Table grapes	0.0	Wheat	0.1
0.4	ES child	0.1	Lettuce and other salad plants	0.0	Rice	0.0	Wheat	0.1
0.3	ES adult	0.1	Lettuce and other salad plants	0.0	Beet leaves (chard)	0.0	Spinach	0.1
0.3	IT adult	0.1	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	0.1
0.3	NL general	0.1	Spinach	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.1
0.3	WHO regional European diet	0.1	Lettuce and other salad plants	0.0	Potatoes	0.0	Wheat	0.1
0.3	UK vegetarian	0.1	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.2
0.3	IT kids/toddler	0.1	Lettuce and other salad plants	0.1	Wheat	0.0	Beet leaves (chard)	0.1
0.3	WHO Cluster diet F	0.1	Lettuce and other salad plants	0.0	Leafy brassica	0.0	Wheat	0.1
0.3	UK Adult	0.1	Sugar beet (root)	0.1	Wine grapes	0.0	Rice	0.2
0.3	DK child	0.1	Wheat	0.0	Rye	0.0	Lettuce and other salad plants	0.2
0.2	DK adult	0.1	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.1
0.1	LT adult	0.0	Potatoes	0.0	Rice	0.0	Head cabbage	0.1
0.1	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	0.0
0.1	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.1

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I026 is unlikely to present a public health concern.

Table 3.2-51: Chronic Assessment of Metabolite M310I001 (Cramer class III trigger)

M310I001

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 0 – 13

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
13.1	ES child	11.3	Poultry: Meat	0.9	Birds' eggs	0.2	Swine: Liver	
11.3	WHO Cluster diet B	8.6	Poultry: Meat	1.0	Bovine: Kidney	0.6	Birds' eggs	
10.3	WHO cluster diet E	8.9	Poultry: Meat	0.8	Birds' eggs	0.1	Wine grapes	
9.4	FR toddler	7.4	Poultry: Meat	1.2	Birds' eggs	0.4	Spinach	
9.2	WHO regional European diet	7.7	Poultry: Meat	0.8	Birds' eggs	0.1	Swine: Meat	
8.4	NL child	6.2	Poultry: Meat	0.7	Birds' eggs	0.2	Bovine: Liver	
7.0	DE child	5.2	Poultry: Meat	1.4	Birds' eggs	0.1	Spinach	
6.7	ES adult	5.6	Poultry: Meat	0.6	Birds' eggs	0.1	Lettuce and other salad plants	
6.1	FR all population	5.3	Poultry: Meat	0.4	Birds' eggs	0.2	Wine grapes	
6.0	FR infant	5.0	Poultry: Meat	0.5	Birds' eggs	0.2	Spinach	
5.3	WHO Cluster diet F	4.0	Poultry: Meat	0.6	Birds' eggs	0.1	Bovine: Kidney	
4.8	WHO cluster diet D	3.5	Poultry: Meat	0.5	Birds' eggs	0.2	Bovine: Kidney	
4.7	IE adult	3.1	Poultry: Meat	0.6	Sheep: Liver	0.3	Birds' eggs	
4.0	NL general	3.0	Poultry: Meat	0.4	Birds' eggs	0.1	Swine: Meat	
3.5	LT adult	2.8	Poultry: Meat	0.4	Birds' eggs	0.1	Swine: Meat	
2.7	UK Infant	1.6	Birds' eggs	0.3	Sugar beet (root)	0.3	Bovine: Kidney	
2.2	UK Toddler	1.1	Birds' eggs	0.7	Sugar beet (root)	0.1	Bovine: Kidney	
1.7	DK child	1.1	Birds' eggs	0.3	Bovine: Liver	0.1	Wheat	
1.5	SE general population 90th percentile	1.1	Birds' eggs	0.1	Leafy brassica	0.1	Lettuce and other salad plants	
1.1	UK vegetarian	0.4	Birds' eggs	0.3	Poultry: Meat	0.1	Sugar beet (root)	
0.8	DK adult	0.4	Birds' eggs	0.1	Bovine: Liver	0.1	Wine grapes	
0.8	UK Adult	0.4	Birds' eggs	0.1	Sugar beet (root)	0.1	Wine grapes	
0.4	PT General population	0.1	Wine grapes	0.1	Rice	0.1	Potatoes	
0.4	FI adult	0.3	Birds' eggs	0.0	Lettuce and other salad plants	0.0	Wine grapes	
0.4	IT adult	0.1	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	
0.3	IT kids/toddler	0.1	Lettuce and other salad plants	0.1	Wheat	0.0	Beet leaves (chard)	
0.1	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I001 is unlikely to present a public health concern.

Table 3.2-52: Chronic Assessment of Metabolite M310I003 (Cramer class III trigger)

M310I003			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	1
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1.0	UK Infant	0.8	Birds' eggs	0.1	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
0.7	WHO Cluster diet B	0.4	Bovine: Kidney	0.3	Birds' eggs		FRUIT (FRESH OR FROZEN)	
0.7	DE child	0.7	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.6	FR toddler	0.6	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.6	UK Toddler	0.6	Birds' eggs	0.0	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
0.5	SE general population 90th percentile	0.5	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.5	DK child	0.5	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.5	ES child	0.4	Birds' eggs	0.0	Swine: Kidney		FRUIT (FRESH OR FROZEN)	
0.4	NL child	0.4	Birds' eggs	0.0	Swine: Kidney	0.0	Swine: Kidney	
0.4	WHO regional European diet	0.4	Birds' eggs	0.0	Bovine: Kidney	0.0	Poultry: Liver	
0.4	WHO cluster diet E	0.4	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.3	WHO Cluster diet F	0.3	Birds' eggs	0.1	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
0.3	WHO cluster diet D	0.3	Birds' eggs	0.1	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
0.3	ES adult	0.3	Birds' eggs	0.0	Swine: Kidney		FRUIT (FRESH OR FROZEN)	
0.3	FR infant	0.3	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	DK adult	0.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	UK vegetarian	0.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	UK Adult	0.2	Birds' eggs	0.0	Bovine: Kidney		FRUIT (FRESH OR FROZEN)	
0.2	LT adult	0.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	FR all population	0.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	NL general	0.2	Birds' eggs	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)	
0.2	IE adult	0.2	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.1	FI adult	0.1	Birds' eggs		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	IT adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I003 is unlikely to present a public health concern.

Table 3.2-53: Chronic Assessment of Metabolite M310I004 (Cramer class III trigger)

M310I004

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
3

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
3.4	WHO Cluster diet B	3.2	Bovine: Kidney	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
1.3	UK Infant	1.0	Bovine: Kidney	0.3	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.9	NL child	0.3	Swine: Kidney	0.3	Swine: Kidney	0.2	Bovine: Liver	
0.7	WHO cluster diet D	0.6	Bovine: Kidney	0.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.6	IE adult	0.6	Sheep: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.5	WHO Cluster diet F	0.4	Bovine: Kidney	0.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.4	DK child	0.4	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.4	UK Toddler	0.3	Bovine: Kidney	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.3	ES child	0.2	Swine: Liver	0.1	Swine: Kidney	0.0	Bovine: Liver	
0.2	WHO regional European diet	0.1	Bovine: Kidney	0.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.2	DK adult	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.2	UK Adult	0.1	Bovine: Kidney	0.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.1	NL general	0.1	Swine: Liver	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.1	ES adult	0.0	Swine: Liver	0.0	Swine: Kidney	0.0	Bovine: Liver	
0.1	LT adult	0.1	Swine: Liver	0.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.0	WHO cluster diet E	0.0	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I004 is unlikely to present a public health concern.

Table 3.2-54: Chronic Assessment of Metabolite M310I005 (Cramer class III trigger)

M310I005

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	0 2
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1.9	UK Toddler	1.4	Sugar beet (root)	0.1	Rice	0.1	Wheat	
1.5	WHO Cluster diet B	0.2	Olives for oil production	0.2	Wine grapes	0.2	Wheat	
1.3	FR toddler	0.7	Spinach	0.1	Potatoes	0.1	Beans (with pods)	
1.2	NL child	0.4	Spinach	0.1	Leafy brassica	0.1	Lettuce and other salad plants	
1.0	UK Infant	0.6	Sugar beet (root)	0.1	Rice	0.1	Potatoes	
1.0	IE adult	0.2	Leafy brassica	0.1	Wine grapes	0.1	Spinach	
0.9	WHO cluster diet D	0.3	Leafy brassica	0.1	Wheat	0.1	Rice	
0.8	SE general population 90th percentile	0.2	Leafy brassica	0.2	Lettuce and other salad plants	0.1	Potatoes	
0.8	FR infant	0.4	Spinach	0.1	Potatoes	0.1	Beans (with pods)	
0.8	PT General population	0.3	Wine grapes	0.1	Rice	0.1	Potatoes	
0.8	FR all population	0.4	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.8	WHO cluster diet E	0.2	Wine grapes	0.1	Wheat	0.1	Lettuce and other salad plants	
0.8	DE child	0.2	Spinach	0.1	Table grapes	0.1	Wheat	
0.7	ES child	0.2	Lettuce and other salad plants	0.1	Rice	0.1	Wheat	
0.7	ES adult	0.2	Lettuce and other salad plants	0.1	Beet leaves (chard)	0.1	Spinach	
0.7	IT adult	0.2	Lettuce and other salad plants	0.1	Spinach	0.1	Wheat	
0.7	NL general	0.1	Spinach	0.1	Lettuce and other salad plants	0.1	Leafy brassica	
0.7	WHO regional European diet	0.2	Lettuce and other salad plants	0.1	Potatoes	0.1	Wheat	
0.7	UK vegetarian	0.2	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	
0.7	IT kids/toddler	0.2	Lettuce and other salad plants	0.1	Wheat	0.1	Beet leaves (chard)	
0.7	WHO Cluster diet F	0.1	Lettuce and other salad plants	0.1	Leafy brassica	0.1	Wheat	
0.6	UK Adult	0.2	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	
0.5	DK child	0.1	Wheat	0.1	Rye	0.1	Lettuce and other salad plants	
0.4	DK adult	0.1	Wine grapes	0.1	Lettuce and other salad plants	0.0	Wheat	
0.3	LT adult	0.1	Potatoes	0.0	Rice	0.0	Head cabbage	
0.2	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	
0.2	PL general population	0.1	Potatoes	0.0	Table grapes	0.0	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I005 is unlikely to present a public health concern.

Table 3.2-55: Chronic Assessment of Metabolite M310I006 (Cramer class III trigger)

M310I006

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	0 2
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1.9	UK Toddler	1.5	Sugar beet (root)	0.1	Rice	0.1	Wheat	
1.5	WHO Cluster diet B	0.2	Olives for oil production	0.2	Wine grapes	0.2	Wheat	
1.3	FR toddler	0.7	Spinach	0.1	Potatoes	0.1	Beans (with pods)	
1.2	NL child	0.4	Spinach	0.1	Leafy brassica	0.1	Lettuce and other salad plants	
1.1	UK Infant	0.6	Sugar beet (root)	0.1	Rice	0.1	Potatoes	
1.0	IE adult	0.2	Leafy brassica	0.1	Wine grapes	0.1	Spinach	
0.9	WHO cluster diet D	0.3	Leafy brassica	0.1	Wheat	0.1	Rice	
0.9	SE general population 90th percentile	0.2	Leafy brassica	0.2	Lettuce and other salad plants	0.1	Potatoes	
0.8	FR infant	0.4	Spinach	0.1	Potatoes	0.1	Beans (with pods)	
0.8	PT General population	0.3	Wine grapes	0.2	Rice	0.1	Potatoes	
0.8	FR all population	0.4	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.8	WHO cluster diet E	0.2	Wine grapes	0.1	Wheat	0.1	Lettuce and other salad plants	
0.8	DE child	0.2	Spinach	0.1	Table grapes	0.1	Wheat	
0.8	ES child	0.2	Lettuce and other salad plants	0.1	Rice	0.1	Wheat	
0.7	ES adult	0.2	Lettuce and other salad plants	0.1	Beet leaves (chard)	0.1	Spinach	
0.7	IT adult	0.2	Lettuce and other salad plants	0.1	Spinach	0.1	Wheat	
0.7	NL general	0.1	Spinach	0.1	Lettuce and other salad plants	0.1	Leafy brassica	
0.7	WHO regional European diet	0.2	Lettuce and other salad plants	0.1	Potatoes	0.1	Wheat	
0.7	UK vegetarian	0.2	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	
0.7	IT kids/toddler	0.2	Lettuce and other salad plants	0.1	Wheat	0.1	Beet leaves (chard)	
0.7	WHO Cluster diet F	0.1	Lettuce and other salad plants	0.1	Leafy brassica	0.1	Wheat	
0.6	UK Adult	0.3	Sugar beet (root)	0.1	Wine grapes	0.1	Rice	
0.5	DK child	0.1	Wheat	0.1	Rye	0.1	Lettuce and other salad plants	
0.4	DK adult	0.1	Wine grapes	0.1	Lettuce and other salad plants	0.0	Wheat	
0.3	LT adult	0.1	Potatoes	0.0	Rice	0.0	Head cabbage	
0.2	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	
0.2	PL general population	0.1	Potatoes	0.0	Table grapes	0.0	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I006 is unlikely to present a public health concern.

Table 3.2-56: Chronic Assessment of Metabolite M310I007 (Cramer class III trigger)

M310I007

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
0	3
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
3.4	UK Toddler	2.6	Sugar beet (root)	0.2	Rice	0.1	Wheat	
2.7	WHO Cluster diet B	0.4	Olives for oil production	0.3	Wine grapes	0.3	Wheat	
2.4	FR toddler	1.3	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
2.2	NL child	0.7	Spinach	0.3	Leafy brassica	0.2	Lettuce and other salad plants	
1.9	UK Infant	1.1	Sugar beet (root)	0.2	Rice	0.1	Potatoes	
1.8	IE adult	0.3	Leafy brassica	0.2	Wine grapes	0.2	Spinach	
1.6	WHO cluster diet D	0.5	Leafy brassica	0.2	Wheat	0.2	Rice	
1.6	SE general population 90th percentile	0.3	Leafy brassica	0.3	Lettuce and other salad plants	0.2	Potatoes	
1.5	FR infant	0.8	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
1.5	PT General population	0.5	Wine grapes	0.3	Rice	0.2	Potatoes	
1.4	FR all population	0.7	Wine grapes	0.2	Lettuce and other salad plants	0.1	Wheat	
1.4	WHO cluster diet E	0.3	Wine grapes	0.1	Wheat	0.1	Lettuce and other salad plants	
1.4	DE child	0.4	Spinach	0.2	Table grapes	0.2	Wheat	
1.4	ES child	0.3	Lettuce and other salad plants	0.2	Rice	0.2	Wheat	
1.3	ES adult	0.4	Lettuce and other salad plants	0.1	Beet leaves (chard)	0.1	Spinach	
1.3	IT adult	0.4	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.3	NL general	0.3	Spinach	0.2	Lettuce and other salad plants	0.2	Leafy brassica	
1.2	WHO regional European diet	0.3	Lettuce and other salad plants	0.2	Potatoes	0.1	Wheat	
1.2	UK vegetarian	0.4	Sugar beet (root)	0.2	Wine grapes	0.1	Rice	
1.2	IT kids/toddler	0.3	Lettuce and other salad plants	0.3	Wheat	0.1	Beet leaves (chard)	
1.2	WHO Cluster diet F	0.2	Lettuce and other salad plants	0.2	Leafy brassica	0.1	Wheat	
1.2	UK Adult	0.5	Sugar beet (root)	0.2	Wine grapes	0.1	Rice	
1.0	DK child	0.2	Wheat	0.2	Rye	0.1	Lettuce and other salad plants	
0.7	DK adult	0.3	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.5	LT adult	0.1	Potatoes	0.1	Rice	0.1	Head cabbage	
0.4	FI adult	0.1	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
0.4	PL general population	0.1	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:

The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I007 is unlikely to present a public health concern.

Table 3.2-57: Chronic Assessment of Metabolite M310I008 (Cramer class III trigger)

M310I008

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	
0	3

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
3.4	UK Toddler	2.6	Sugar beet (root)	0.2	Rice	0.1	Wheat	
2.7	WHO Cluster diet B	0.4	Olives for oil production	0.3	Wine grapes	0.3	Wheat	
2.4	FR toddler	1.3	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
2.2	NL child	0.7	Spinach	0.3	Leafy brassica	0.2	Lettuce and other salad plants	
1.9	UK infant	1.1	Sugar beet (root)	0.2	Rice	0.1	Potatoes	
1.8	IE adult	0.3	Leafy brassica	0.2	Wine grapes	0.2	Spinach	
1.6	WHO cluster diet D	0.5	Leafy brassica	0.2	Wheat	0.2	Rice	
1.6	SE general population 90th percentile	0.3	Leafy brassica	0.3	Lettuce and other salad plants	0.2	Potatoes	
1.5	FR infant	0.8	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
1.5	PT General population	0.5	Wine grapes	0.3	Rice	0.2	Potatoes	
1.4	FR all population	0.7	Wine grapes	0.2	Lettuce and other salad plants	0.1	Wheat	
1.4	WHO cluster diet E	0.3	Wine grapes	0.1	Wheat	0.1	Lettuce and other salad plants	
1.4	DE child	0.4	Spinach	0.2	Table grapes	0.2	Wheat	
1.4	ES child	0.3	Lettuce and other salad plants	0.2	Rice	0.2	Wheat	
1.3	ES adult	0.4	Lettuce and other salad plants	0.1	Beet leaves (chard)	0.1	Spinach	
1.3	IT adult	0.4	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.3	NL general	0.3	Spinach	0.2	Lettuce and other salad plants	0.2	Leafy brassica	
1.2	WHO regional European diet	0.3	Lettuce and other salad plants	0.2	Potatoes	0.1	Wheat	
1.2	UK vegetarian	0.4	Sugar beet (root)	0.2	Wine grapes	0.1	Rice	
1.2	IT kids/toddler	0.3	Lettuce and other salad plants	0.3	Wheat	0.1	Beet leaves (chard)	
1.2	WHO Cluster diet F	0.2	Lettuce and other salad plants	0.2	Leafy brassica	0.1	Wheat	
1.2	UK Adult	0.5	Sugar beet (root)	0.2	Wine grapes	0.1	Rice	
1.0	DK child	0.2	Wheat	0.2	Rye	0.1	Lettuce and other salad plants	
0.7	DK adult	0.3	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.5	LT adult	0.1	Potatoes	0.1	Rice	0.1	Head cabbage	
0.4	FI adult	0.1	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
0.4	PL general population	0.1	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I008 is unlikely to present a public health concern.

Table 3.2-58: Chronic Assessment of Metabolite M310I009 (Cramer class III trigger)

M310I009

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 1 - 4

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
4.2	UK Toddler	3.2	Sugar beet (root)	0.2	Rice	0.2	Wheat	
3.3	WHO Cluster diet B	0.4	Olives for oil production	0.4	Wine grapes	0.4	Wheat	
2.9	FR toddler	1.5	Spinach	0.2	Potatoes	0.2	Beans (with pods)	
2.7	NL child	0.8	Spinach	0.3	Leafy brassica	0.3	Lettuce and other salad plants	
2.3	UK Infant	1.4	Sugar beet (root)	0.3	Rice	0.2	Potatoes	
2.3	IE adult	0.3	Leafy brassica	0.3	Wine grapes	0.3	Spinach	
1.9	WHO cluster diet D	0.6	Leafy brassica	0.3	Wheat	0.2	Rice	
1.9	SE general population 90th percentile	0.4	Leafy brassica	0.4	Lettuce and other salad plants	0.2	Potatoes	
1.8	FR infant	1.0	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
1.8	PT General population	0.6	Wine grapes	0.3	Rice	0.2	Potatoes	
1.8	FR all population	0.9	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
1.7	WHO cluster diet E	0.4	Wine grapes	0.2	Wheat	0.2	Lettuce and other salad plants	
1.7	DE child	0.4	Spinach	0.3	Table grapes	0.2	Wheat	
1.7	ES child	0.4	Lettuce and other salad plants	0.2	Rice	0.2	Wheat	
1.6	ES adult	0.5	Lettuce and other salad plants	0.2	Beet leaves (chard)	0.2	Spinach	
1.6	IT adult	0.5	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.6	NL general	0.3	Spinach	0.2	Lettuce and other salad plants	0.2	Leafy brassica	
1.5	WHO regional European diet	0.4	Lettuce and other salad plants	0.2	Potatoes	0.1	Wheat	
1.5	UK vegetarian	0.5	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.5	IT kids/toddler	0.4	Lettuce and other salad plants	0.3	Wheat	0.1	Beet leaves (chard)	
1.5	WHO Cluster diet F	0.3	Lettuce and other salad plants	0.2	Leafy brassica	0.2	Wheat	
1.4	UK Adult	0.6	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.2	DK child	0.3	Wheat	0.2	Rye	0.1	Lettuce and other salad plants	
0.9	DK adult	0.3	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.6	LT adult	0.1	Potatoes	0.1	Rice	0.1	Head cabbage	
0.5	FI adult	0.1	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
0.5	PL general population	0.2	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I009 is unlikely to present a public health concern.

Table 3.2-59: Chronic Assessment of Metabolite M310I010 (Cramer class III trigger)

M310I010

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 10

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
10.2	UK Infant	9.5	Milk and cream,	0.4	Bovine: Kidney	0.1	Bovine: Liver	0.1
9.9	FR toddler	9.8	Milk and cream,	0.1	Spinach	0.0	Potatoes	0.0
7.8	NL child	7.2	Milk and cream,	0.1	Bovine: Liver	0.1	Swine: Liver	0.0
6.4	FR infant	6.3	Milk and cream,	0.0	Spinach	0.0	Potatoes	0.0
5.4	UK Toddler	5.1	Milk and cream,	0.1	Sugar beet (root)	0.1	Bovine: Kidney	0.2
3.6	DE child	3.5	Milk and cream,	0.0	Spinach	0.0	Table grapes	0.0
3.4	DK child	3.1	Milk and cream,	0.2	Bovine: Liver	0.0	Wheat	0.0
3.3	ES child	3.1	Milk and cream,	0.1	Swine: Liver	0.0	Swine: Kidney	0.0
3.1	SE general population 90th percentile	3.1	Milk and cream,	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.0
2.3	WHO Cluster diet B	1.2	Bovine: Kidney	0.8	Milk and cream,	0.1	Bovine: Liver	0.1
1.7	NL general	1.6	Milk and cream,	0.0	Swine: Liver	0.0	Bovine: Liver	0.0
1.6	WHO cluster diet D	1.2	Milk and cream,	0.3	Bovine: Kidney	0.0	Leafy brassica	0.0
1.4	DK adult	1.3	Milk and cream,	0.1	Bovine: Liver	0.0	Wine grapes	0.0
1.4	FI adult	1.4	Milk and cream,	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0
1.3	ES adult	1.2	Milk and cream,	0.0	Swine: Liver	0.0	Lettuce and other salad plants	0.0
1.3	WHO regional European diet	1.2	Milk and cream,	0.1	Bovine: Kidney	0.0	Lettuce and other salad plants	0.0
1.2	WHO Cluster diet F	1.0	Milk and cream,	0.2	Bovine: Kidney	0.0	Bovine: Liver	0.0
1.1	IE adult	0.7	Milk and cream,	0.3	Sheep: Liver	0.0	Leafy brassica	0.0
1.1	LT adult	1.0	Milk and cream,	0.0	Swine: Liver	0.0	Bovine: Liver	0.0
0.9	UK vegetarian	0.8	Milk and cream,	0.0	Sugar beet (root)	0.0	Wine grapes	0.0
0.9	UK Adult	0.7	Milk and cream,	0.0	Bovine: Kidney	0.0	Sugar beet (root)	0.0
0.8	WHO cluster diet E	0.7	Milk and cream,	0.0	Bovine: Liver	0.0	Wine grapes	0.0
0.7	FR all population	0.7	Milk and cream,	0.0	Wine grapes	0.0	Lettuce and other salad plants	0.0
0.1	PT General population	0.0	Wine grapes	0.0	Rice	0.0	Potatoes	0.0
0.1	IT adult	0.0	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	0.0
0.1	IT kids/toddler	0.0	Lettuce and other salad plants	0.0	Wheat	0.0	Beet leaves (chard)	0.0
0.0	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.0

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I010 is unlikely to present a public health concern.

Table 3.2-60: Chronic Assessment of Metabolite M310I011 (Cramer class III trigger)

M310I011

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum: 1 - 4

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
4.3	UK Toddler	3.2	Sugar beet (root)	0.2	Rice	0.2	Wheat	
3.8	WHO Cluster diet B	0.4	Olives for oil production	0.4	Bovine: Kidney	0.4	Wine grapes	
2.9	FR toddler	1.5	Spinach	0.2	Potatoes	0.2	Beans (with pods)	
2.9	NL child	0.8	Spinach	0.3	Leafy brassica	0.3	Lettuce and other salad plants	
2.5	UK Infant	1.4	Sugar beet (root)	0.3	Rice	0.2	Potatoes	
2.4	IE adult	0.3	Leafy brassica	0.3	Wine grapes	0.3	Spinach	
2.0	WHO cluster diet D	0.6	Leafy brassica	0.3	Wheat	0.2	Rice	
1.9	SE general population 90th percentile	0.4	Leafy brassica	0.4	Lettuce and other salad plants	0.2	Potatoes	
1.8	FR infant	1.0	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
1.8	PT General population	0.6	Wine grapes	0.3	Rice	0.2	Potatoes	
1.8	FR all population	0.9	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
1.8	WHO cluster diet E	0.4	Wine grapes	0.2	Wheat	0.2	Lettuce and other salad plants	
1.7	ES child	0.4	Lettuce and other salad plants	0.2	Rice	0.2	Wheat	
1.7	DE child	0.4	Spinach	0.3	Table grapes	0.2	Wheat	
1.6	ES adult	0.5	Lettuce and other salad plants	0.2	Beet leaves (chard)	0.2	Spinach	
1.6	IT adult	0.5	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.6	NL general	0.3	Spinach	0.2	Lettuce and other salad plants	0.2	Leafy brassica	
1.6	WHO regional European diet	0.4	Lettuce and other salad plants	0.2	Potatoes	0.1	Wheat	
1.5	WHO Cluster diet F	0.3	Lettuce and other salad plants	0.2	Leafy brassica	0.2	Wheat	
1.5	UK vegetarian	0.5	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.5	IT kids/toddler	0.4	Lettuce and other salad plants	0.3	Wheat	0.1	Beet leaves (chard)	
1.4	UK Adult	0.6	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.3	DK child	0.3	Wheat	0.2	Rye	0.1	Lettuce and other salad plants	
0.9	DK adult	0.3	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.6	LT adult	0.1	Potatoes	0.1	Rice	0.1	Head cabbage	
0.5	FI adult	0.1	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
0.5	PL general population	0.2	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I011 is unlikely to present a public health concern.

Table 3.2-61: Chronic Assessment of Metabolite M310I013 (Cramer class III trigger)

M310I013

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
0.4	UK Toddler	0.3	Sugar beet (root)	0.0	Rice	0.0	Wheat	0.0
0.3	WHO Cluster diet B	0.0	Olives for oil production	0.0	Wine grapes	0.0	Wheat	0.1
0.2	FR toddler	0.1	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.1
0.2	NL child	0.1	Spinach	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.1
0.2	UK Infant	0.1	Sugar beet (root)	0.0	Rice	0.0	Potatoes	0.0
0.2	IE adult	0.0	Leafy brassica	0.0	Wine grapes	0.0	Spinach	0.0
0.2	WHO cluster diet D	0.1	Leafy brassica	0.0	Wheat	0.0	Rice	0.1
0.2	SE general population 90th percentile	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.0	Potatoes	0.0
0.2	FR infant	0.1	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.0
0.2	PT General population	0.0	Wine grapes	0.0	Rice	0.0	Potatoes	0.1
0.2	FR all population	0.1	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.0
0.1	WHO cluster diet E	0.0	Wine grapes	0.0	Wheat	0.0	Lettuce and other salad plants	0.0
0.1	DE child	0.0	Spinach	0.0	Table grapes	0.0	Wheat	0.0
0.1	ES child	0.0	Lettuce and other salad plants	0.0	Rice	0.0	Wheat	0.0
0.1	ES adult	0.0	Lettuce and other salad plants	0.0	Beet leaves (chard)	0.0	Spinach	0.0
0.1	IT adult	0.0	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	0.0
0.1	NL general	0.0	Spinach	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.0
0.1	WHO regional European diet	0.0	Lettuce and other salad plants	0.0	Potatoes	0.0	Wheat	0.0
0.1	UK vegetarian	0.0	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.0
0.1	IT kids/toddler	0.0	Lettuce and other salad plants	0.0	Wheat	0.0	Beet leaves (chard)	0.0
0.1	WHO Cluster diet F	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.0	Wheat	0.0
0.1	UK Adult	0.0	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.0
0.1	DK child	0.0	Wheat	0.0	Rye	0.0	Lettuce and other salad plants	0.1
0.1	DK adult	0.0	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.0
0.0	LT adult	0.0	Potatoes	0.0	Rice	0.0	Head cabbage	0.0
0.0	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	0.0
0.0	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.0

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I013 is unlikely to present a public health concern.

Table 3.2-62: Chronic Assessment of Metabolite M310I017 (Cramer class III trigger)

M310I017

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum - maximum	1 – 9
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
9.0	UK Toddler	6.9	Sugar beet (root)	0.5	Rice	0.4	Wheat	
7.2	WHO Cluster diet B	1.0	Olives for oil production	0.9	Wine grapes	0.9	Wheat	
6.2	FR toddler	3.3	Spinach	0.5	Potatoes	0.4	Beans (with pods)	
5.8	NL child	1.7	Spinach	0.7	Leafy brassica	0.6	Lettuce and other salad plants	
5.0	UK infant	3.0	Sugar beet (root)	0.6	Rice	0.3	Potatoes	
4.9	IE adult	0.7	Leafy brassica	0.6	Wine grapes	0.6	Spinach	
4.1	WHO cluster diet D	1.3	Leafy brassica	0.7	Wheat	0.5	Rice	
4.1	SE general population 90th percentile	0.9	Leafy brassica	0.8	Lettuce and other salad plants	0.4	Potatoes	
3.9	FR infant	2.1	Spinach	0.4	Potatoes	0.3	Beans (with pods)	
3.8	PT General population	1.2	Wine grapes	0.7	Rice	0.5	Potatoes	
3.8	FR all population	2.0	Wine grapes	0.6	Lettuce and other salad plants	0.3	Wheat	
3.7	WHO cluster diet E	0.8	Wine grapes	0.4	Wheat	0.4	Lettuce and other salad plants	
3.7	DE child	1.0	Spinach	0.6	Table grapes	0.4	Wheat	
3.6	ES child	0.8	Lettuce and other salad plants	0.4	Rice	0.4	Wheat	
3.4	ES adult	1.0	Lettuce and other salad plants	0.4	Beet leaves (chard)	0.3	Spinach	
3.4	IT adult	1.1	Lettuce and other salad plants	0.4	Spinach	0.4	Wheat	
3.3	NL general	0.7	Spinach	0.5	Lettuce and other salad plants	0.4	Leafy brassica	
3.3	WHO regional European diet	0.8	Lettuce and other salad plants	0.4	Potatoes	0.3	Wheat	
3.2	UK vegetarian	1.1	Sugar beet (root)	0.4	Wine grapes	0.4	Rice	
3.2	IT kids/toddler	0.8	Lettuce and other salad plants	0.7	Wheat	0.3	Beet leaves (chard)	
3.1	WHO Cluster diet F	0.6	Lettuce and other salad plants	0.5	Leafy brassica	0.4	Wheat	
3.0	UK Adult	1.2	Sugar beet (root)	0.5	Wine grapes	0.3	Rice	
2.5	DK child	0.6	Wheat	0.4	Rye	0.3	Lettuce and other salad plants	
1.9	DK adult	0.7	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
1.2	LT adult	0.3	Potatoes	0.2	Rice	0.1	Head cabbage	
1.1	FI adult	0.2	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
1.1	PL general population	0.3	Potatoes	0.2	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I017 is unlikely to present a public health concern.

Table 3.2-63: Chronic Assessment of Metabolite M310I018 (Cramer class III trigger)

PbAld

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum

No of diets exceeding ADI: —

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
0.4	UK Toddler	0.3	Sugar beet (root)	0.0	Rice	0.0	Wheat	0.0
0.3	WHO Cluster diet B	0.0	Olives for oil production	0.0	Wine grapes	0.0	Wheat	0.1
0.3	FR toddler	0.2	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.1
0.3	NL child	0.1	Spinach	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.1
0.2	UK Infant	0.1	Sugar beet (root)	0.0	Rice	0.0	Potatoes	0.0
0.2	IE adult	0.0	Leafy brassica	0.0	Wine grapes	0.0	Spinach	0.1
0.2	WHO cluster diet D	0.1	Leafy brassica	0.0	Wheat	0.0	Rice	0.1
0.2	SE general population 90th percentile	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.0	Potatoes	0.0
0.2	FR infant	0.1	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.0
0.2	PT General population	0.1	Wine grapes	0.0	Rice	0.0	Potatoes	0.1
0.2	FR all population	0.1	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.0
0.2	WHO cluster diet E	0.0	Wine grapes	0.0	Wheat	0.0	Lettuce and other salad plants	0.1
0.2	DE child	0.0	Spinach	0.0	Table grapes	0.0	Wheat	0.0
0.2	ES child	0.0	Lettuce and other salad plants	0.0	Rice	0.0	Wheat	0.0
0.2	ES adult	0.0	Lettuce and other salad plants	0.0	Beet leaves (chard)	0.0	Spinach	0.0
0.2	IT adult	0.1	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	0.0
0.2	NL general	0.0	Spinach	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.0
0.2	WHO regional European diet	0.0	Lettuce and other salad plants	0.0	Potatoes	0.0	Wheat	0.0
0.1	UK vegetarian	0.1	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.0
0.1	IT kids/toddler	0.0	Lettuce and other salad plants	0.0	Wheat	0.0	Beet leaves (chard)	0.0
0.1	WHO Cluster diet F	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.0	Wheat	0.0
0.1	UK Adult	0.1	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.0
0.1	DK child	0.0	Wheat	0.0	Rye	0.0	Lettuce and other salad plants	0.1
0.1	DK adult	0.0	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.0
0.1	LT adult	0.0	Potatoes	0.0	Rice	0.0	Head cabbage	0.0
0.1	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	0.0
0.0	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.0

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of PbAld is unlikely to present a public health concern.

Table 3.2-64: Chronic Assessment of Metabolite M310I019 (Cramer class III trigger)

M310I019

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
0.0	NL child	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.0	WHO regional European diet	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.0	NL general	0.0	Poultry: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	DK adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I019 is unlikely to present a public health concern.

Table 3.2-65: Chronic Assessment of Metabolite M310I021 (Cramer class III trigger)

M310I021

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
2

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
1.9	IE adult	1.9	Sheep: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
1.3	NL child	0.6	Bovine: Liver	0.6	Swine: Liver		FRUIT (FRESH OR FROZEN)	
1.1	DK child	1.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.8	UK Infant	0.8	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.7	ES child	0.5	Swine: Liver	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.6	WHO Cluster diet B	0.6	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.5	DK adult	0.5	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.4	NL general	0.2	Swine: Liver	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.3	LT adult	0.2	Swine: Liver	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.2	ES adult	0.1	Swine: Liver	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)	
0.2	UK Toddler	0.2	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.1	WHO cluster diet E	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.1	WHO cluster diet D	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.1	UK Adult	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.1	WHO Cluster diet F	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
0.1	WHO regional European diet	0.1	Bovine: Liver		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	
	FI adult		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I021 is unlikely to present a public health concern.

Table 3.2-66: Chronic Assessment of Metabolite M310I024 (Cramer class III trigger)

PbAlc

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

	TMDI (range) in % of ADI minimum - maximum
	1 — 4
No of diets exceeding ADI:	—

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
4.2	UK Toddler	3.2	Sugar beet (root)	0.2	Rice	0.2	Wheat	
3.3	WHO Cluster diet B	0.4	Olives for oil production	0.4	Wine grapes	0.4	Wheat	
2.9	FR toddler	1.5	Spinach	0.2	Potatoes	0.2	Beans (with pods)	
2.7	NL child	0.8	Spinach	0.3	Leafy brassica	0.3	Lettuce and other salad plants	
2.3	UK infant	1.4	Sugar beet (root)	0.3	Rice	0.2	Potatoes	
2.3	IE adult	0.3	Leafy brassica	0.3	Wine grapes	0.3	Spinach	
1.9	WHO cluster diet D	0.6	Leafy brassica	0.3	Wheat	0.2	Rice	
1.9	SE general population 90th percentile	0.4	Leafy brassica	0.4	Lettuce and other salad plants	0.2	Potatoes	
1.8	FR infant	1.0	Spinach	0.2	Potatoes	0.1	Beans (with pods)	
1.8	PT General population	0.6	Wine grapes	0.3	Rice	0.2	Potatoes	
1.8	FR all population	0.9	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
1.7	WHO cluster diet E	0.4	Wine grapes	0.2	Wheat	0.2	Lettuce and other salad plants	
1.7	DE child	0.4	Spinach	0.3	Table grapes	0.2	Wheat	
1.7	ES child	0.4	Lettuce and other salad plants	0.2	Rice	0.2	Wheat	
1.6	ES adult	0.5	Lettuce and other salad plants	0.2	Beet leaves (chard)	0.2	Spinach	
1.6	IT adult	0.5	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.6	NL general	0.3	Spinach	0.2	Lettuce and other salad plants	0.2	Leafy brassica	
1.5	WHO regional European diet	0.4	Lettuce and other salad plants	0.2	Potatoes	0.1	Wheat	
1.5	UK vegetarian	0.5	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.5	IT kids/toddler	0.4	Lettuce and other salad plants	0.3	Wheat	0.1	Beet leaves (chard)	
1.5	WHO Cluster diet F	0.3	Lettuce and other salad plants	0.2	Leafy brassica	0.2	Wheat	
1.4	UK Adult	0.6	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.2	DK child	0.3	Wheat	0.2	Rye	0.1	Lettuce and other salad plants	
0.9	DK adult	0.3	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.6	LT adult	0.1	Potatoes	0.1	Rice	0.1	Head cabbage	
0.5	FI adult	0.1	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
0.5	PL general population	0.2	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of PbAlc is unlikely to present a public health concern.

Table 3.2-67: Chronic Assessment of Metabolite M310I025 (Cramer class III trigger)

M310I025

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum
1 5

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
4.8	UK Toddler	3.7	Sugar beet (root)	0.3	Rice	0.2	Wheat	
3.8	WHO Cluster diet B	0.5	Olives for oil production	0.5	Wine grapes	0.5	Wheat	
3.3	FR toddler	1.8	Spinach	0.3	Potatoes	0.2	Beans (with pods)	
3.1	NL child	0.9	Spinach	0.4	Leafy brassica	0.3	Lettuce and other salad plants	
2.7	UK Infant	1.6	Sugar beet (root)	0.3	Rice	0.2	Potatoes	
2.6	IE adult	0.4	Leafy brassica	0.3	Wine grapes	0.3	Spinach	
2.2	WHO cluster diet D	0.7	Leafy brassica	0.3	Wheat	0.3	Rice	
2.2	SE general population 90th percentile	0.5	Leafy brassica	0.4	Lettuce and other salad plants	0.2	Potatoes	
2.1	FR infant	1.1	Spinach	0.2	Potatoes	0.2	Beans (with pods)	
2.0	PT General population	0.7	Wine grapes	0.4	Rice	0.3	Potatoes	
2.0	FR all population	1.0	Wine grapes	0.3	Lettuce and other salad plants	0.2	Wheat	
2.0	WHO cluster diet E	0.4	Wine grapes	0.2	Wheat	0.2	Lettuce and other salad plants	
2.0	DE child	0.5	Spinach	0.3	Table grapes	0.2	Wheat	
1.9	ES child	0.4	Lettuce and other salad plants	0.2	Rice	0.2	Wheat	
1.8	ES adult	0.6	Lettuce and other salad plants	0.2	Beet leaves (chard)	0.2	Spinach	
1.8	IT adult	0.6	Lettuce and other salad plants	0.2	Spinach	0.2	Wheat	
1.8	NL general	0.4	Spinach	0.2	Lettuce and other salad plants	0.2	Leafy brassica	
1.7	WHO regional European diet	0.4	Lettuce and other salad plants	0.2	Potatoes	0.2	Wheat	
1.7	UK vegetarian	0.6	Sugar beet (root)	0.2	Wine grapes	0.2	Rice	
1.7	IT kids/toddler	0.4	Lettuce and other salad plants	0.4	Wheat	0.1	Beet leaves (chard)	
1.7	WHO Cluster diet F	0.3	Lettuce and other salad plants	0.2	Leafy brassica	0.2	Wheat	
1.6	UK Adult	0.6	Sugar beet (root)	0.3	Wine grapes	0.2	Rice	
1.4	DK child	0.3	Wheat	0.2	Rye	0.1	Lettuce and other salad plants	
1.0	DK adult	0.4	Wine grapes	0.1	Lettuce and other salad plants	0.1	Wheat	
0.7	LT adult	0.2	Potatoes	0.1	Rice	0.1	Head cabbage	
0.6	FI adult	0.1	Lettuce and other salad plants	0.1	Wine grapes	0.1	Leafy brassica	
0.6	PL general population	0.2	Potatoes	0.1	Table grapes	0.1	Tomatoes	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I025 is unlikely to present a public health concern.

Table 3.2-68: Chronic Assessment of Metabolite M310I026 (Cramer class III trigger)

M310I026			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):		proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	0.0015	ARfD (mg/kg bw):	0.005
Source of ADI:	CramerIII	Source of ARfD:	CramerIII
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum - maximum

No of diets exceeding ADI: –

Highest calculated TMDI values in % of ADI	MS Diet	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
0.2	UK Toddler	0.1	Sugar beet (root)	0.0	Rice	0.0	Wheat	0.2
0.1	WHO Cluster diet B	0.0	Olives for oil production	0.0	Wine grapes	0.0	Wheat	0.1
0.1	FR toddler	0.1	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.0
0.1	NL child	0.0	Spinach	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.0
0.1	UK Infant	0.1	Sugar beet (root)	0.0	Rice	0.0	Potatoes	0.1
0.1	IE adult	0.0	Leafy brassica	0.0	Wine grapes	0.0	Spinach	0.0
0.1	WHO cluster diet D	0.0	Leafy brassica	0.0	Wheat	0.0	Rice	0.0
0.1	SE general population 90th percentile	0.0	Leafy brassica	0.0	Lettuce and other salad plants	0.0	Potatoes	0.0
0.1	FR infant	0.0	Spinach	0.0	Potatoes	0.0	Beans (with pods)	0.0
0.1	PT General population	0.0	Wine grapes	0.0	Rice	0.0	Potatoes	0.0
0.1	FR all population	0.0	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.0
0.1	WHO cluster diet E	0.0	Wine grapes	0.0	Wheat	0.0	Lettuce and other salad plants	0.0
0.1	DE child	0.0	Spinach	0.0	Table grapes	0.0	Wheat	0.0
0.1	ES child	0.0	Lettuce and other salad plants	0.0	Rice	0.0	Wheat	0.0
0.1	ES adult	0.0	Lettuce and other salad plants	0.0	Beet leaves (chard)	0.0	Spinach	0.0
0.1	IT adult	0.0	Lettuce and other salad plants	0.0	Spinach	0.0	Wheat	0.0
0.1	NL general	0.0	Spinach	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.0
0.1	WHO regional European diet	0.0	Lettuce and other salad plants	0.0	Potatoes	0.0	Wheat	0.0
0.1	UK vegetarian	0.0	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.0
0.1	IT kids/toddler	0.0	Lettuce and other salad plants	0.0	Wheat	0.0	Beet leaves (chard)	0.0
0.1	WHO Cluster diet F	0.0	Lettuce and other salad plants	0.0	Leafy brassica	0.0	Wheat	0.0
0.1	UK Adult	0.0	Sugar beet (root)	0.0	Wine grapes	0.0	Rice	0.0
0.1	DK child	0.0	Wheat	0.0	Rye	0.0	Lettuce and other salad plants	0.0
0.0	DK adult	0.0	Wine grapes	0.0	Lettuce and other salad plants	0.0	Wheat	0.0
0.0	LT adult	0.0	Potatoes	0.0	Rice	0.0	Head cabbage	0.0
0.0	FI adult	0.0	Lettuce and other salad plants	0.0	Wine grapes	0.0	Leafy brassica	0.0
0.0	PL general population	0.0	Potatoes	0.0	Table grapes	0.0	Tomatoes	0.0

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of M310I026 is unlikely to present a public health concern.

3.2.2 Acute Assessments

The acute assessments were conducted using the EFSA PRIMo model (version 2). Acute assessments were run against the trigger value of 0.005 mg/kg (Cramer class III).

The input values (parent/metabolite ratios) of Table 3.2-13 were multiplied with the HRs of Table 3.2-14 and run against the trigger value 0.005 mg/kg bw using the EFSA PRIMo model (version 2).

In Table 3.2-69 to Table 3.2-86 the results of the EFSA PRIMo calculations are shown.

Table 3.2-69: Acute Assessment of Metabolite M310I001 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	29.5	Poultry: Meat	0.1315 / -	29.5	Poultry: Meat	0.1315 / -	30.9	Poultry: Meat	0.1315 / -	30.9	Poultry: Meat	0.1315 / -
	20.2	Scarole (broad-leaf	0.011552 / -	15.2	Bovine: Kidney	0.2015 / -	6.9	Bovine: Kidney	0.2015 / -	6.9	Bovine: Kidney	0.2015 / -
	15.2	Bovine: Kidney	0.2015 / -	12.1	Scarole (broad-leaf	0.011552 / -	6.7	Chinese cabbage	0.00944 / -	6.7	Chinese cabbage	0.00944 / -
	12.8	Kale	0.00944 / -	9.1	Kale	0.00944 / -	5.7	Swine: Kidney	0.2015 / -	5.7	Swine: Kidney	0.2015 / -
	8.0	Spinach	0.01776 / -	8.0	Spinach	0.01776 / -	3.8	Kale	0.00944 / -	3.3	Purslane	0.01776 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-70: Acute Assessment of Metabolite M310I003 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
No of commodities for which ARfD/ADI is exceeded (IESTI 1):		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—
IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
5.9	Bovine: Kidney	0.079 / -	5.9	Bovine: Kidney	0.079 / -	2.7	Bovine: Kidney	0.079 / -	2.7	Bovine: Kidney	0.079 / -
2.3	Birds' eggs	0.0093 / -	2.3	Birds' eggs	0.0093 / -	2.3	Swine: Kidney	0.079 / -	2.3	Swine: Kidney	0.079 / -
2.0	Swine: Kidney	0.079 / -	2.0	Swine: Kidney	0.079 / -	0.7	Birds' eggs	0.0093 / -	0.7	Birds' eggs	0.0093 / -
						0.0	Poultry: Liver	0.00054 / -	0.0	Poultry: Liver	0.00054 / -
No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-71: Acute Assessment of Metabolite M310I004 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	48.8	Bovine: Kidney	0.6475 / -	48.8	Bovine: Kidney	0.6475 / -	22.0	Bovine: Kidney	0.6475 / -	22.0	Bovine: Kidney	0.6475 / -
	16.4	Swine: Kidney	0.6475 / -	16.4	Swine: Kidney	0.6475 / -	18.4	Swine: Kidney	0.6475 / -	18.4	Swine: Kidney	0.6475 / -
	6.9	Bovine: Liver	0.04285 / -	6.9	Bovine: Liver	0.04285 / -	2.3	Bovine: Liver	0.04285 / -	2.3	Bovine: Liver	0.04285 / -
	1.0	Swine: Liver	0.04285 / -	1.0	Swine: Liver	0.04285 / -	0.6	Sheep: Liver	0.04285 / -	0.6	Sheep: Liver	0.04285 / -
							0.6	Swine: Liver	0.04285 / -	0.6	Swine: Liver	0.04285 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-72: Acute Assessment of Metabolite M310I005 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	39.1	Scarole (broad-leaf	0.022382 / -	23.5	Scarole (broad-leaf	0.022382 / -	13.1	Chinese cabbage	0.01829 / -	13.1	Chinese cabbage	0.01829 / -
	24.7	Kale	0.01829 / -	17.7	Kale	0.01829 / -	7.5	Kale	0.01829 / -	6.5	Purslane	0.03441 / -
	15.6	Spinach	0.03441 / -	15.6	Spinach	0.03441 / -	7.1	Purslane	0.03441 / -	6.2	Spinach	0.03441 / -
	13.6	Chinese cabbage	0.01829 / -	13.6	Chinese cabbage	0.01829 / -	6.2	Spinach	0.03441 / -	5.5	Kale	0.01829 / -
	12.1	Beet leaves (chard)	0.03441 / -	9.2	Beet leaves	0.03441 / -	5.1	Beet leaves (chard)	0.03441 / -	4.3	Beet leaves (chard)	0.03441 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-73: Acute Assessment of Metabolite M310I006 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	40.4	Scarole (broad-leaf	0.023104 / -	24.2	Scarole (broad-leaf	0.023104 / -	13.5	Chinese cabbage	0.01888 / -	13.5	Chinese cabbage	0.01888 / -
	25.5	Kale	0.01888 / -	18.2	Kale	0.01888 / -	7.7	Kale	0.01888 / -	6.7	Purslane	0.03552 / -
	16.1	Spinach	0.03552 / -	16.1	Spinach	0.03552 / -	7.3	Purslane	0.03552 / -	6.3	Spinach	0.03552 / -
	14.0	Chinese cabbage	0.01888 / -	14.0	Chinese cabbage	0.01888 / -	6.3	Spinach	0.03552 / -	5.7	Kale	0.01888 / -
	12.5	Beet leaves (chard)	0.03552 / -	9.5	Beet leaves	0.03552 / -	5.3	Beet leaves (chard)	0.03552 / -	4.5	Beet leaves (chard)	0.03552 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-74: Acute Assessment of Metabolite M310I007 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	72.0	Scarole (broad-leaf	0.041154 / -	43.2	Scarole (broad-leaf	0.041154 / -	24.0	Chinese cabbage	0.03363 / -	24.0	Chinese cabbage	0.03363 / -
	45.5	Kale	0.03363 / -	32.5	Kale	0.03363 / -	13.7	Kale	0.03363 / -	11.9	Purslane	0.06327 / -
	28.6	Spinach	0.06327 / -	28.6	Spinach	0.06327 / -	13.1	Purslane	0.06327 / -	11.3	Spinach	0.06327 / -
	25.0	Chinese cabbage	0.03363 / -	25.0	Chinese cabbage	0.03363 / -	11.3	Spinach	0.06327 / -	10.2	Kale	0.03363 / -
	22.2	Beet leaves (chard)	0.06327 / -	16.9	Beet leaves	0.06327 / -	9.4	Beet leaves (chard)	0.06327 / -	7.9	Beet leaves (chard)	0.06327 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-75: Acute Assessment of Metabolite M310I008 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
No of commodities for which ARfD/ADI is exceeded (IESTI 1):		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—
IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
72.0	Scarole (broad-leaf	0.041154 / -	43.2	Scarole (broad-leaf	0.041154 / -	24.0	Chinese cabbage	0.03363 / -	24.0	Chinese cabbage	0.03363 / -
45.5	Kale	0.03363 / -	32.5	Kale	0.03363 / -	13.7	Kale	0.03363 / -	11.9	Purslane	0.06327 / -
28.6	Spinach	0.06327 / -	28.6	Spinach	0.06327 / -	13.1	Purslane	0.06327 / -	11.3	Spinach	0.06327 / -
25.0	Chinese cabbage	0.03363 / -	25.0	Chinese cabbage	0.03363 / -	11.3	Spinach	0.06327 / -	10.2	Kale	0.03363 / -
22.2	Beet leaves (chard)	0.06327 / -	16.9	Beet leaves	0.06327 / -	9.4	Beet leaves (chard)	0.06327 / -	7.9	Beet leaves (chard)	0.06327 / -
No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-76: Acute Assessment of Metabolite M310I009 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
No of commodities for which ARfD/ADI is exceeded (IESTI 1):		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—
IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
88.4	Scarole (broad-leaf	0.05054 / -	53.0	Scarole (broad-leaf	0.05054 / -	29.5	Chinese cabbage	0.0413 / -	29.5	Chinese cabbage	0.0413 / -
55.8	Kale	0.0413 / -	39.9	Kale	0.0413 / -	16.8	Kale	0.0413 / -	14.6	Purslane	0.0777 / -
35.1	Spinach	0.0777 / -	35.1	Spinach	0.0777 / -	16.1	Purslane	0.0777 / -	13.9	Spinach	0.0777 / -
30.7	Chinese cabbage	0.0413 / -	30.7	Chinese cabbage	0.0413 / -	13.9	Spinach	0.0777 / -	12.5	Kale	0.0413 / -
27.3	Beet leaves (chard)	0.0777 / -	20.7	Beet leaves	0.0777 / -	11.5	Beet leaves (chard)	0.0777 / -	9.7	Beet leaves (chard)	0.0777 / -
No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-77: Acute Assessment of Metabolite M310I010 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
No of commodities for which ARfD/ADI is exceeded (IESTI 1):		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—
IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
19.1	Bovine: Kidney	0.253 / -	19.1	Bovine: Kidney	0.253 / -	8.6	Bovine: Kidney	0.253 / -	8.6	Bovine: Kidney	0.253 / -
9.2	Milk and milk	0.0037 / -	9.2	Milk and milk	0.0037 / -	7.2	Swine: Kidney	0.253 / -	7.2	Swine: Kidney	0.253 / -
6.4	Swine: Kidney	0.253 / -	6.4	Swine: Kidney	0.253 / -	1.3	Milk and milk	0.0037 / -	1.3	Milk and milk products: Cattle	0.0037 / -
3.8	Scarole (broad-leaf	0.002166 / -	3.6	Bovine: Liver	0.022 / -	1.3	Chinese cabbage	0.00177 / -	1.3	Chinese cabbage	0.00177 / -
3.6	Bovine: Liver	0.022 / -	2.3	Scarole (broad-leaf	0.002166 / -	1.2	Bovine: Liver	0.022 / -	1.2	Bovine: Liver	0.022 / -
No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-78: Acute Assessment of Metabolite M310I011 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
No of commodities for which ARfD/ADI is exceeded (IESTI 1):		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—
IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
88.4	Scarole (broad-leaf	0.05054 / -	53.0	Scarole (broad-leaf	0.05054 / -	29.5	Chinese cabbage	0.0413 / -	29.5	Chinese cabbage	0.0413 / -
55.8	Kale	0.0413 / -	39.9	Kale	0.0413 / -	16.8	Kale	0.0413 / -	14.6	Purslane	0.0777 / -
35.1	Spinach	0.0777 / -	35.1	Spinach	0.0777 / -	16.1	Purslane	0.0777 / -	13.9	Spinach	0.0777 / -
30.7	Chinese cabbage	0.0413 / -	30.7	Chinese cabbage	0.0413 / -	13.9	Spinach	0.0777 / -	12.5	Kale	0.0413 / -
27.3	Beet leaves (chard)	0.0777 / -	26.4	Bovine: Kidney	0.35 / -	11.9	Bovine: Kidney	0.35 / -	11.9	Bovine: Kidney	0.35 / -
No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-79: Acute Assessment of Metabolite M310I013 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
No of commodities for which ARfD/ADI is exceeded (IESTI 1):		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**		—
IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
7.6	Scarole (broad-leaf	0.004332 / -	4.5	Scarole (broad-leaf	0.004332 / -	2.5	Chinese cabbage	0.00354 / -	2.5	Chinese cabbage	0.00354 / -
4.8	Kale	0.00354 / -	3.4	Kale	0.00354 / -	1.4	Kale	0.00354 / -	1.3	Purslane	0.00666 / -
3.0	Spinach	0.00666 / -	3.0	Spinach	0.00666 / -	1.4	Purslane	0.00666 / -	1.2	Spinach	0.00666 / -
2.6	Chinese cabbage	0.00354 / -	2.6	Chinese cabbage	0.00354 / -	1.2	Spinach	0.00666 / -	1.1	Kale	0.00354 / -
2.3	Beet leaves (chard)	0.00666 / -	1.8	Beet leaves	0.00666 / -	1.0	Beet leaves (chard)	0.00666 / -	0.8	Beet leaves (chard)	0.00666 / -
No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-80: Acute Assessment of Metabolite M310I017 (Cramer class III trigger)

	Acute risk assessment /children						Acute risk assessment / adults / general population					
	The acute risk assessment is based on the ARfD.											
	For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
	In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
	Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		2	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		1	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		---	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		---
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	189.4	Scarole (broad-leaf	0.1083 / 0.05	113.6	Scarole (broad-leaf	0.1083 / 0.09	63.2	Chinese cabbage	0.0885 / -	63.2	Chinese cabbage	0 0885 / -
	119.7	Kale	0.0885 / 0.07	85.5	Kale	0.0885 / -	36.1	Kale	0.0885 / -	31.3	Purslane	0.1665 / -
	75.3	Spinach	0.1665 / -	75.3	Spinach	0.1665 / -	34.4	Purslane	0.1665 / -	29.8	Spinach	0.1665 / -
	65.7	Chinese cabbage	0.0885 / -	65.7	Chinese cabbage	0.0885 / -	29.8	Spinach	0.1665 / -	26.8	Kale	0.0885 / -
	58.5	Beet leaves (chard)	0.1665 / -	44.4	Beet leaves	0.1665 / -	24.7	Beet leaves (chard)	0.1665 / -	20.9	Beet leaves (chard)	0.1665 / -
	No of critical MRLs (IESTI 1)		2				**No of critical MRLs (IESTI 2)**		1			

Table 3.2-81: Acute Assessment of Metabolite M310I018 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	8.8	Scarole (broad-leaf	0.005054 / -	5.3	Scarole (broad-leaf	0.005054 / -	3.0	Chinese cabbage	0.00413 / -	3.0	Chinese cabbage	0.00413 / -
	5.6	Kale	0.00413 / -	4.0	Kale	0.00413 / -	1.7	Kale	0.00413 / -	1.5	Purslane	0.00777 / -
	3.5	Spinach	0.00777 / -	3.5	Spinach	0.00777 / -	1.6	Purslane	0.00777 / -	1.4	Spinach	0.00777 / -
	3.1	Chinese cabbage	0.00413 / -	3.1	Chinese cabbage	0.00413 / -	1.4	Spinach	0.00777 / -	1.3	Kale	0.00413 / -
	2.7	Beet leaves (chard)	0.00777 / -	2.1	Beet leaves	0.00777 / -	1.2	Beet leaves (chard)	0.00777 / -	1.0	Beet leaves (chard)	0.00777 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-82: Acute Assessment of Metabolite M310I019 (Cramer class III trigger)

	Acute risk assessment /children						Acute risk assessment / adults / general population					
	The acute risk assessment is based on the ARfD.											
	For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
	In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
	Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
							9.1	Poultry: Liver	0.1025 / -	9.1	Poultry: Liver	0.1025 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-83: Acute Assessment of Metabolite M310I021 (Cramer class III trigger)

	Acute risk assessment /children						Acute risk assessment / adults / general population					
	The acute risk assessment is based on the ARfD.											
	For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
	In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
	Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	20.6	Bovine: Liver	0.1275 / -	20.6	Bovine: Liver	0.1275 / -	6.9	Bovine: Liver	0.1275 / -	6.9	Bovine: Liver	0.1275 / -
	2.8	Swine: Liver	0.1275 / -	2.8	Swine: Liver	0.1275 / -	1.7	Sheep: Liver	0.1275 / -	1.7	Sheep: Liver	0.1275 / -
							1.7	Swine: Liver	0.1275 / -	1.7	Swine: Liver	0.1275 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-84: Acute Assessment of Metabolite M310I024 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	88.4	Scarole (broad-leaf	0.05054 / -	53.0	Scarole (broad-leaf	0.05054 / -	29.5	Chinese cabbage	0.0413 / -	29.5	Chinese cabbage	0.0413 / -
	55.8	Kale	0.0413 / -	39.9	Kale	0.0413 / -	16.8	Kale	0.0413 / -	14.6	Purslane	0.0777 / -
	35.1	Spinach	0.0777 / -	35.1	Spinach	0.0777 / -	16.1	Purslane	0.0777 / -	13.9	Spinach	0.0777 / -
	30.7	Chinese cabbage	0.0413 / -	30.7	Chinese cabbage	0.0413 / -	13.9	Spinach	0.0777 / -	12.5	Kale	0.0413 / -
	27.3	Beet leaves (chard)	0.0777 / -	20.7	Beet leaves	0.0777 / -	11.5	Beet leaves (chard)	0.0777 / -	9.7	Beet leaves (chard)	0.0777 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

Table 3.2-85: Acute Assessment of Metabolite M310I025 (Cramer class III trigger)

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		1	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	101 0	Scarole (broad-leaf	0.05776 / 0.05	60.6	Scarole (broad-leaf	0.05776 / -	33.7	Chinese cabbage	0.0472 / -	33.7	Chinese cabbage	0 0472 / -
	63.8	Kale	0.0472 / -	45.6	Kale	0.0472 / -	19.2	Kale	0.0472 / -	16.7	Purslane	0.0888 / -
	40.1	Spinach	0.0888 / -	40.1	Spinach	0.0888 / -	18.4	Purslane	0.0888 / -	15.9	Spinach	0.0888 / -
	35.1	Chinese cabbage	0.0472 / -	35.1	Chinese cabbage	0.0472 / -	15.9	Spinach	0.0888 / -	14.3	Kale	0.0472 / -
	31.2	Beet leaves (chard)	0.0888 / -	23.7	Beet leaves	0.0888 / -	13.2	Beet leaves (chard)	0.0888 / -	11.1	Beet leaves (chard)	0.0888 / -
	No of critical MRLs (IESTI 1)		1				**No of critical MRLs (IESTI 2)**			—		

Table 3.2-86: Acute Assessment of Metabolite M310I026 (Cramer class III trigger)

	Acute risk assessment /children						Acute risk assessment / adults / general population					
	The acute risk assessment is based on the ARfD.											
	For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the ESTI calculation.											
	In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
	Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 1)**		—	**No of commodities for which ARfD/ADI is exceeded (IESTI 2)**		—
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	3.8	Scarole (broad-leaf	0.002166 / -	2.3	Scarole (broad-leaf	0.002166 / -	1.3	Chinese cabbage	0.00177 / -	1.3	Chinese cabbage	0.00177 / -
	2.4	Kale	0.00177 / -	1.7	Kale	0.00177 / -	0.7	Kale	0.00177 / -	0.6	Purslane	0.00333 / -
	1.5	Spinach	0.00333 / -	1.5	Spinach	0.00333 / -	0.7	Purslane	0.00333 / -	0.6	Spinach	0.00333 / -
	1.3	Chinese cabbage	0.00177 / -	1.3	Chinese cabbage	0.00177 / -	0.6	Spinach	0.00333 / -	0.5	Kale	0.00177 / -
	1.2	Beet leaves (chard)	0.00333 / -	0.9	Beet leaves	0.00333 / -	0.5	Beet leaves (chard)	0.00333 / -	0.4	Beet leaves (chard)	0.00333 / -
	No of critical MRLs (IESTI 1)			—			**No of critical MRLs (IESTI 2)**			—		

4 Conclusion

The summarized results of the TTC calculations (input values see Table 3.2-14) for the metabolites on concern are given in the table below.

Table 4-1: Results of TTC calculations applying the genotoxic, neurotoxic and chronic (Cramer class III) TTC values (0.0000025, 0.0003 and 0.0015 mg/kg bw/d) as ADI and the ARfD of 0.005 mg/kg bw/d [Input values used are based on STMRs of alpha-Cypermethrin (chronic) and on HRs of alpha-CYP (acute) representing a refinement scenario]

Metabolite	Chronic assessment						Acute assessment	Occurrence			
	TTC genotox		TTC neurotox		TTC (Cramer class III)		ARfD exceeded?	Livestock		Rat	Plant
	max. ADI utilization	exceeded?	max. ADI utilization	exceeded?	max. ADI utilization	exceeded?		Hen	Goat		
M310I001	7835%	**yes**	65.3%	no	13.1%	no	no (max. 30.9%)	x	x	x	x
M310I003	573%	**yes**	4.8%	no	1.0%	no	no (max. 5.9%)	x	x		
M310I004	2025%	**yes**	16.9%	no	3.4%	no	no (max. 48.8%)		x	x	
M310I005	1117%	**yes**	9.3%	no	1.9%	no	no (max. 39.1%)				x
M310I006	1153%	**yes**	9.6%	no	1.9%	no	no (max. 40.4%)				x
M310I007	2053%	**yes**	17.1%	no	3.4%	no	no (max. 72.0%)				x
M310I008	2053%	**yes**	17.1%	no	3.4%	no	no (max. 72.0%)				x
M310I009	2521%	**yes**	21.0%	no	4.2%	no	no (max. 88.4%)				x
M310I010	6103%	**yes**	50.9%	no	10.2%	no	no (max. 19.1%)		x	x	x
M310I011	2553%	**yes**	21.3%	no	4.3%	no	no (max. 88.4%)	x	x	x	x
M310I013	216%	**yes**	1.8%	no	0.4%	no	no (max. 7.6%)				
M310I017	5402%	**yes**	45.0%	no	9.0%	no	**yes (max. 189%)**		x	x	x
M310I018	252%	**yes**	2.1%	no	0.4%	no	no (max. 8.8%)				x
M310I019	24.0%	no	0.2%	no	0.0%	no	no (max. 9.1%)	x			
M310I021	1153%	**yes**	9.6%	no	1.9%	no	no (max. 20.6%)		x		
M310I024	2521%	**yes**	21.0%	no	4.2%	no	no (max. 88.4%)				x
M310I025	2881%	**yes**	24.0%	no	4.8%	no	**yes (max. 101%)**			x	x
M310I026	108%	**yes**	0.9%	no	0.2%	no	no (max. 3.8%)				x

A summary of the Toxicological Threshroshold of Concern (TTC) evaluation is presented in dossier section M-CA 6.9 and the evaluation of the toxicological relevance of the plant and animal metabolites of alpha-cypermethrin based on these results are further discussed in dossier section KCA 5.8.1

5 Appendix

All trials from the table below were used for further evaluation, together with the HRs and STMRs derived from these residue data and used for risk assessment.

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Grape	2005/1006474	FAN/19/03	N	<0.05	7	0.064	0.032	0.069	0.041
Grape	2005/1006474	FAN/20/03	N	<0.05	7				
Grape	2005/1006474	FBM/14/03	N	<0.05	7				
Grape	2005/1006474	FBM/15/03	N	<0.05	7				
Grape	2005/1007591	DU2/06/04	N	0.03	7				
Grape	2005/1007591	DU4/06/04	N	0.031	7				
Grape	2005/1007591	FAN/12/04	N	0.018	7				
Grape	2005/1007591	FBM/06/04	N	0.020	14				
Grape	2006/1026853	AF/8830/BA/1	N	0.064	7				
Grape	2006/1026853	AF/8830/BA/2	N	0.033	7				
Grape	2006/1026853	AF/8830/BA/3	N	0.037	14				
Grape	2006/1026853	AF/8830/BA/4	N	0.043	7				
Grape	2007/1008492	A/NF/I/06/127	N	0.012	6				
Grape	2007/1008492	A/NF/I/06/128	N	0.019	8				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Grape	2007/1008492	A/GE/I/06/129	N	0.021	14				
Grape	2007/1008492	A/GE/I/06/130	N	0.028	14				
Grape	2005/1004977	ALO/15/03	S	<0.05	7	0.069	0.049		
Grape	2005/1004977	ALO/24/03	S	<0.05	7				
Grape	2005/1004977	ITA/10/03	S	<0.05	6				
Grape	2005/1004977	ITA/11/03	S	<0.05	6				
Grape	2005/1007589	FBD/01/04	S	0.069	14				
Grape	2005/1007589	FTL/01/04	S	0.061	14				
Grape	2005/1007589	GRE/03/04	S	0.048	7				
Grape	2005/1007589	GRE/04/04	S	0.051	7				
Grape	2006/1026853	AF/8830/BA/5	S	0.051	14				
Grape	2006/1026853	AF/8830/BA/6	S	<0.01	7				
Grape	2006/1026853	AF/8830/BA/7	S	0.045	7				
Grape	2006/1026853	AF/8830/BA/8	S	<0.01	7				
Grape	2007/1008492	A/SF/I/06/131	S	0.015	7				
Grape	2007/1008492	A/GR/I/06/132	S	0.012	7				
Grape	2007/1008492	A/SP/I/06/133	S	0.039	13				
Grape	2007/1008492	A/IT/I/06/134	S	0.030	6				
Strawberry	2007/1008489	A/BE/I/05/88	N	<0.01	3	0.031	0.01	0.054	0.01
Strawberry	2007/1008489	A/GE/I/05/81	N	<0.01	3				
Strawberry	2007/1008489	A/NF/I/05/80	N	0.031	3				
Strawberry	2007/1008489	A/UK/I/05/83	N	<0.01	3				
Strawberry	2007/1008493	A/NF/I/06/88	N	<0.01	3				
Strawberry	2007/1008493	A/GE/I/06/89	N	<0.01	2				
Strawberry	2007/1008493	A/NL/I/06/90	N	<0.01	4				
Strawberry	2007/1008493	A/UK/I/06/91	N	0.023	7				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Strawberry	2007/1008489	A/IT/I/05/86	S	<0.01	3	0.028	0.01		
Strawberry	2007/1008489	A/IT/I/05/87	S	0.028	1				
Strawberry	2007/1008489	A/SF/I/05/84	S	<0.01	3				
Strawberry	2007/1008489	A/SP/I/05/85	S	<0.01	3				
Strawberry	2007/1008493	A/SF/I/06/92	S	0.018	3				
Strawberry	2007/1008493	A/SP/I/06/93	S	0.017	3				
Strawberry	2007/1008493	A/GR/I/06/95	S	<0.01	3				
Strawberry	2007/1008493	A/IT/I/06/94	S	<0.01	3				
Strawberry	2007/1007935	A/BE/I/05/78	Indoor (N)	0.040	3	0.054	0.02		
Strawberry	2007/1007935	A/GE/I/05/72	Indoor (N)	<0.01	3				
Strawberry	2007/1007935	A/GE/I/05/73	Indoor (N)	<0.01	3				
Strawberry	2007/1007935	A/GR/I/05/77	Indoor (S)	0.048	3				
Strawberry	2007/1007935	A/IT/I/05/76	Indoor (S)	0.029	3				
Strawberry	2007/1007935	A/NF/I/05/70	Indoor (N)	0.054	3				
Strawberry	2007/1007935	A/SF/I/05/71	Indoor (S)	<0.01	3				
Strawberry	2007/1007935	A/SP/I/05/75	Indoor (S)	<0.01	3				
Olive	2012/1157548	L110427	S	0.14	7	0.26	0.05		
Olive	2012/1157548	L110428	S	0.26	7				
Olive	2005/1004975	ALO/16/03	S	<0.05	6				
Olive	2005/1004975	ALO/17/03	S	<0.05	6				
Olive	2005/1004975	GRE/11/03	S	<0.05	7				
Olive	2005/1004975	GRE/12/03	S	<0.05	8				
Olive	2005/1007582	ALO/23/04	S	0.04	7				
Olive	2005/1007582	ALO/24/04	S	0.09	7				
Olive	2005/1007582	GRE/14/04	S	0.04	7				
Olive	2005/1007582	GRE/15/04	S	<0.01	7				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Potato	2006/1026846	AF/8831/BA/1	N	<0.01	7	0.01	0.01	0.01	0.01
Potato	2006/1026846	AF/8831/BA/2	N	<0.01	7				
Potato	2006/1026846	AF/8831/BA/3	N	<0.01	7				
Potato	2006/1026846	AF/8831/BA/4	N	<0.01	7				
Potato	2007/1007945	AF/10497/BA/1	N	<0.01	7				
Potato	2007/1007945	AF/10497/BA/2	N	<0.01	7				
Potato	2007/1007945	AF/10497/BA/3	N	<0.01	7				
Potato	2007/1007945	AF/10497/BA/9	N	<0.01	7				
Potato	2008/1002704	L070446	N	<0.01	6				
Potato	2008/1002704	L070447	N	<0.01	8				
Potato	2012/1157550	L110423	N	<0.01	7				
Potato	2012/1157550	L110424	N	<0.01	7				
Potato	2007/1007945	AF/10497/BA/5	S	<0.01	14	0.01	0.01		
Potato	2007/1007945	AF/10497/BA/6	S	<0.01	14				
Potato	2007/1007945	AF/10497/BA/7	S	<0.01	14				
Potato	2007/1007945	AF/10497/BA/8	S	<0.01	14				
Potato	2006/1026846	AF/8831/BA/5	S	<0.01	14				
Potato	2006/1026846	AF/8831/BA/6	S	<0.01	14				
Potato	2006/1026846	AF/8831/BA/7	S	<0.01	14				
Potato	2006/1026846	AF/8831/BA/8	S	<0.01	14				
Potato	2005/1007592	GRE/02/04	S	<0.01	14				
Potato	2005/1007592	ITA/01/04	S	<0.01	14				
Potato	2008/1002704	L070448	S	<0.01	15				
Potato	2008/1002704	L070449	S	<0.01	14				
Potato	2012/1157550	L110425	S	<0.01	14				
Potato	2012/1157550	L110426	S	<0.01	14				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Carrot	2005/1035294	RU-I-13 04 RP NW 2/1	N	<0.01	3	0.01	0.01		
Carrot	2005/1035294	RU-I-13 04 RP NW 2/2	N	<0.01	3				
Carrot	2005/1035294	RU-I-13 04 RP NW 2/3	N	<0.01	3				
Carrot	2006/1029533	RU-I-06 05 NW BN 2/1	N	<0.01	3				
Carrot	2006/1029533	RU-I-06 05 NW BN 221	N	<0.01	7				
Carrot	2006/1029533	RU-I-06 05 SH KI 2/1	N	<0.01	7				
Carrot	2006/1029533	RU-I-06 05 SH KI 2/2	N	<0.01	7				
Carrot	2006/1029533	RU-I-06 05 BY FS 2/1	N	<0.01	7				
Carrot	2006/1029533	RU-I-06 05 BY FS 2/2	N	<0.01	7				
Onion	2006/1026854	AF/8827/BA/1	N	0.01	7	0.01	0.01		
Onion	2006/1026854	AF/8827/BA/2	N	0.01	7				
Onion	2006/1026854	AF/8827/BA/3	N	<0.01	7				
Onion	2006/1026854	AF/8827/BA/4	N	<0.01	7				
Onion	2007/1008499	A/NF/I/06/150	N	<0.01	8				
Onion	2007/1008499	A/GE/I/06/151	N	<0.01	7				
Onion	2007/1008499	A/GE/I/06/152	N	<0.01	7				
Onion	2007/1008499	A/NL/I/06/153	N	<0.01	7				
Tomato (F)	2007/1008488	A/NF/I/05/60	N	<0.01	3	0.01	0.03	0.037	0.01
Tomato (F)	2007/1008488	A/NF/I/05/61	N	<0.01	3				
Tomato (F)	2007/1008488	A/GE/I/05/62	N	<0.01	3				
Tomato (F)	2007/1008488	A/BE/I/05/63	N	<0.01	3				
Tomato (F)	2007/1008494	A/NF/I/06/97	N	<0.01	4				
Tomato (F)	2007/1008494	A/GE/I/06/98	N	0.01	3				
Tomato (F)	2007/1008494	A/GE/I/06/99	N	<0.01	3				
Tomato (F)	2007/1008494	A/GE/I/06/100	N	<0.01	3				
Tomato (F)	2007/1007937	AF/10504/BA/1	N	<0.01	3				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Tomato (F)	2007/1007937	AF/10504/BA/2	N	<0.01	3				
Tomato (F)	2009/1090704	L080173	N	0.03	2				
Tomato (F)	2009/1090704	L080174	N	0.01	8				
Tomato (F)	2007/1008488	A/SF/I/05/64	S	0.013	7	0.021	0.01		
Tomato (F)	2007/1008488	A/SF/I/05/65	S	<0.01	3				
Tomato (F)	2007/1008488	A/SP/I/05/66	S	<0.01	3				
Tomato (F)	2007/1008488	A/IT/I/05/67	S	0.021	3				
Tomato (F)	2007/1008494	A/SF/I/06/101	S	<0.01	3				
Tomato (F)	2007/1008494	A/GR/I/06/102	S	<0.01	4				
Tomato (F)	2007/1008494	A/SP/I/06/103	S	<0.01	3				
Tomato (F)	2007/1008494	A/IT/I/06/104	S	0.019	2				
Tomato (F)	2007/1007937	AF/10504/BA/3	S	0.013	3				
Tomato (F)	2007/1007937	AF/10504/BA/4	S	<0.01	3				
Tomato (F)	2009/1090704	L080175	S	<0.01	3				
Tomato (F)	2009/1090704	L080176	S	<0.01	3				
Tomato (I)	2007/1007934	A/NF/I/05/50	Indoor (N)	0.016	14	0.037	0.016		
Tomato (I)	2007/1007934	A/SF/I/05/51	Indoor (S)	<0.01	3				
Tomato (I)	2007/1007934	A/BE/I/05/54	Indoor (N)	0.015	3				
Tomato (I)	2007/1007934	A/SP/I/05/55	Indoor (S)	0.024	3				
Tomato (I)	2007/1007934	A/GR/I/05/57	Indoor (S)	0.014	7				
Tomato (I)	2007/1007934	A/GE/I/05/52	Indoor (N)	0.025	2				
Tomato (I)	2007/1007934	A/GE/I/05/53	Indoor (N)	0.016	3				
Tomato (I)	2007/1007934	A/IT/I/05/56	Indoor (S)	0.037	3				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Pepper, sweet (F)	2006/1026860	AF/8820/BA/1	S	<0.01	7	0.28	0.013	0.033	0.017
Pepper, sweet (F)	2006/1026860	AF/8820/BA/2	S	<0.01	7				
Pepper, sweet (F)	2006/1026860	AF/8820/BA/3	S	0.028	6				
Pepper, sweet (F)	2006/1026860	AF/8820/BA/4	S	0.015	14				
Pepper, sweet (F)	2007/1008497	A/SF/I/06/162	S	<0.01	8				
Pepper, sweet (F)	2007/1008497	A/IT/I/06/163	S	<0.01	8				
Pepper, sweet (F)	2007/1008497	A/SP/I/06/164	S	0.024	8				
Pepper, sweet (F)	2007/1008497	A/GR/I/06/165	S	0.017	7				
Pepper, sweet (I)	2006/1036933	05I CL FR P35	Indoor (S)	0.016	3	0.033	0.018		
Pepper, sweet (I)	2006/1036933	05I CL FR P39	Indoor (S)	0.017	3				
Pepper, sweet (I)	2006/1036933	05ES086R	Indoor (S)	0.029	7				
Pepper, sweet (I)	2006/1036933	05RF047	Indoor (S)	0.016	3				
Pepper, sweet (I)	2006/1036933	G023-05 I	Indoor (N)	0.033	14				
Pepper, sweet (I)	2006/1036933	IR05BASL51PL01	Indoor (S)	0.018	7				
Cucumber (I)	2004/5000720	ACK/08/04	Indoor (N)	<0.01	3	0.012	0.010	0.037	0.010
Cucumber (I)	2004/5000720	AGR/10/04	Indoor (N)	<0.01	3				
Cucumber (I)	2004/5000720	ALB/07/04	Indoor (N)	0.012	4				
Cucumber (I)	2004/5000720	ALO/17/04	Indoor (S)	<0.01	4				
Cucumber (I)	2004/5000720	FAN/10/04	Indoor (N)	<0.01	2				
Cucumber (I)	2004/5000720	FBD/11/04	Indoor (S)	<0.01	3				
Cucumber (I)	2004/5000720	GRE/12/04	Indoor (S)	<0.01	4				
Cucumber (I)	2004/5000720	ITA/09/04	Indoor (S)	<0.01	3				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Cucumber (I)	2006/1036934	05 I CL FR P32	Indoor (S)	0.012	3	0.37	0.014		
Cucumber (I)	2006/1036934	AT-05/007-1	Indoor (N)	<0.01	3				
Cucumber (I)	2006/1036934	G022-05 I-B	Indoor (N)	0.014	3				
Cucumber (I)	2006/1036934	ALB/190507-01	Indoor (N)	0.020	3				
Cucumber (I)	2006/1036934	05 I CL FR P36	Indoor (S)	0.031	3				
Cucumber (I)	2006/1036934	IR05BASL11LG01	Indoor (S)	0.037	3				
Cucumber (I)	2006/1036934	05ES087R	Indoor (S)	<0.01	3				
Cucumber (I)	2006/1036934	05RF044	Indoor (S)	0.014	3				
Melon (F)	2006/1024607 2007/1011007	AF/8816/BA/1	S	<0.01	3	0.010	0.014	0.010	0.048
Melon (F)	2006/1024607 2007/1011007	AF/8816/BA/2	S	<0.01	3				
Melon (F)	2006/1024607 2007/1011007	AF/8816/BA/3	S	<0.01	3				
Melon (F)	2006/1024607 2007/1011007	AF/8816/BA/4	S	<0.01	2				
Melon (F)	2007/1007940	AF/10490/BA/1	S	<0.01	2				
Melon (F)	2007/1007940	AF/10490/BA/2	S	0.014	3				
Melon (F)	2007/1007940	AF/10490/BA/3	S	<0.01	3				
Melon (F)	2007/1007940	AF/10490/BA/4	S	<0.01	3				
Melon (I)	2006/1037507	05I CL FR P34	I (S)	<0.01	7	0.01	0.048		
Melon (I)	2006/1037507	AGR/54/05	I (N)	0.022	7				
Melon (I)	2006/1037507	AGR/53/05	I (N)	0.029	8				
Melon (I)	2006/1037507	ALB/190508-01	I (N)	<0.01	7				
Melon (I)	2006/1037507	05 I CL FR P38	I (S)	<0.01	7				
Melon (I)	2006/1037507	IR05BASG61LG01	I (S)	<0.01	7				
Melon (I)	2006/1037507	05ES/085R	I (S)	0.048	7				
Melon (I)	2006/1037507	05RF046	I (S)	<0.01	7				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Broccoli (F)	2006/1026863	AF/8814/BA/1	N	<0.01	7	0.029	0.016	0.047	0.018
Broccoli (F)	2006/1026863	AF/8814/BA/2	N	0.015	7				
Broccoli (F)	2006/1026863	AF/8814/BA/3	N	<0.01	7				
Broccoli (F)	2006/1026863	AF/8814/BA/4	N	0.018	7				
Broccoli (F)	2007/1013274	A/NF/I/06/120	N	0.011	7				
Broccoli (F)	2007/1013274	A/GE/I/06/121	N	0.029	6				
Broccoli (F)	2007/1013274	A/DK/I/06/122	N	0.017	7				
Broccoli (F)	2007/1013274	A/UK/I/06/123	N	0.018	7				
Broccoli (F)	2006/1026863	AF/8814/BA/5	S	0.028	3	0.047	0.03		
Broccoli (F)	2006/1026863	AF/8814/BA/6	S	0.047	3				
Broccoli (F)	2007/1013274	A/GR/I/06/124	S	0.032	3				
Broccoli (F)	2007/1013274	A/SP/I/06/125	S	0.026	4				
Cauliflower (F)	2006/1026864	AF/8813/BA/1	N	<0.01	7	0.085	0.01	0.085	0.01
Cauliflower (F)	2006/1026864	AF/8813/BA/2	N	<0.01	7				
Cauliflower (F)	2006/1026864	AF/8813/BA/3	N	0.085	7				
Cauliflower (F)	2006/1026864	AF/8813/BA/4	N	<0.01	7				
Cauliflower (F)	2007/1008495	A/NF/I/06/136	N	<0.01	6				
Cauliflower (F)	2007/1008495	A/UK/I/06/137	N	<0.01	7				
Cauliflower (F)	2007/1008495	A/NL/I/06/138	N	0.010	7				
Cauliflower (F)	2007/1008495	A/DK/I/06/139	N	0.011	8				
Cauliflower (F)	2007/1007936	AF/10502/BA/1	N	<0.01	7				
Cauliflower (F)	2007/1007936	AF/10502/BA/2	N	<0.01	6				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Cauliflower (F)	2006/1026864	AF/8813/BA/5	S	<0.01	3	0.083	0.01		
Cauliflower (F)	2006/1026864	AF/8813/BA/6	S	<0.01	3				
Cauliflower (F)	2007/1008495	A/SP/I/06/140	S	<0.01	4				
Cauliflower (F)	2007/1008495	A/GR/I/06/141	S	<0.01	3				
Cauliflower (F)	2007/1007936	AF/10502/BA/3	S	0.083	2				
Cauliflower (F)	2007/1007936	AF/10502/BA/4	S	<0.01	3				
Brussels sprouts (F)	2006/1026859	AF/8818/BA/1	N	0.014	7	0.046	0.018	0.046	0.016
Brussels sprouts (F)	2006/1026859	AF/8818/BA/2	N	0.017	7				
Brussels sprouts (F)	2006/1026859	AF/8818/BA/3	N	0.046	7				
Brussels sprouts (F)	2006/1026859	AF/8818/BA/4	N	<0.01	7				
Brussels sprouts (F)	2007/1007943	AF/10499/BA/1	N	0.025	7				
Brussels sprouts (F)	2007/1007943	AF/10499/BA/2	N	0.018	7				
Brussels sprouts (F)	2007/1007943	AF/10499/BA/3	N	0.020	7				
Brussels sprouts (F)	2007/1007943	AF/10499/BA/4	N	0.010	7				
Brussels sprouts (F)	2006/1026859	AF/8818/BA/5	S	<0.01	3	0.029	0.012		
Brussels sprouts (F)	2006/1026859	AF/8818/BA/6	S	0.013	3				
Brussels sprouts (F)	2007/1007943	AF/10499/BA/5	S	0.029	4				
Brussels sprouts (F)	2007/1007943	AF/10499/BA/6	S	<0.01	3				
Cabbage, head (F)	2004/1006470	ACK/14/03	N	<0.050	7	0.105	0.034	0.105	0.0.014
Cabbage, head (F)	2004/1006470	ALB/12/03	N	<0.050	7				
Cabbage, head (F)	2004/1006470	FAN/21/03	N	<0.050	7				
Cabbage, head (F)	2004/1006470	OAT/18/03	N	<0.050	6				
Cabbage, head (F)	2006/1026852	AF/8829/BA/1	N	0.018	7				
Cabbage, head (F)	2006/1026852	AF/8829/BA/2	N	<0.010	7				
Cabbage, head (F)	2006/1026852	AF/8829/BA/4	N	0.104	7				
Cabbage, head (F)	2006/1026852	AF/8829/BA/7	N	0.014	7				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Cabbage, head (F)	2007/1013148	A/NF/I/06/143	N	<0.010	8				
Cabbage, head (F)	2007/1013148	A/BE/I/06/144	N	<0.010	7				
Cabbage, head (F)	2007/1013148	A/UK/I/06/145	N	0.105	7				
Cabbage, head (F)	2007/1013148	A/GE/I/06/146	N	<0.010	7				
Cabbage, head (F)	2006/1026852	AF/8829/BA/5	S	<0.010	3				
Cabbage, head (F)	2006/1026852	AF/8829/BA/6	S	0.013	3	0.013	0.012		
Cabbage, head (F)	2007/1013148	A/SF/I/06/147	S	0.010	3				
Cabbage, head (F)	2007/1013148	A/SP/I/06/148	S	0.013	6				
Leafy brassica (F)	2007/1013342	A/NF/I/06/115	N	0.590	3				
Leafy brassica (F)	2007/1013342	A/NL/I/06/116	N	0.350	4	0.59	0.367		
Leafy brassica (F)	2006/1026862	AF/8815/BA/1	N	0.170	3				
Leafy brassica (F)	2006/1026862	AF/8815/BA/5	N	0.383	7				
Leafy brassica (F)	2006/1026862	AF/8815/BA/3	S	0.436	3				
Leafy brassica (F)	2006/1026862	AF/8815/BA/4	S	0 278	3				
Leafy brassica (F)	2007/1013342	A/SF/I/06/117	S	0.046	3			0.59	0.14
Leafy brassica (F)	2007/1013342	A/SP/I/06/118	S	0.056	3				
Chinese cabbage (F)	2013/1416285	S13-00427-01 / L130046	S	0.086	3	0.436	0.075		
Chinese cabbage (F)	2013/1416285	S13-00427-02 / L130047	S	0.110	3				
Chinese cabbage (F)	2013/1416285	S13-00427-03 / L130048	S	0.064	3				
Chinese cabbage (F)	2013/1416285	S13-00427-04 / L130049	S	0.019	2				
Lettuce, head (F)	2006/1026855	AF/8817/BA/1	N	0.722	2				
Lettuce, head (F)	2006/1026855	AF/8817/BA/2	N	0.244	3				
Lettuce, head (F)	2006/1026855	AF/8817/BA/3	N	0.125	3	0.722	0.123	0.722	0.125
Lettuce, head (F)	2006/1026855	AF/8817/BA/4	N	0.221	3				
Lettuce, head (F)	2007/1007938	AF/10503/BA/1	N	0.072	3				
Lettuce, head (F)	2007/1007938	AF/10503/BA/2	N	<0.01	3				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Lettuce, head (F)	2007/1008496	A/NF/I/06/106	N	0.27	3				
Lettuce, head (F)	2007/1008496	A/GE/I/06/107	N	0.021	2				
Lettuce, head (F)	2007/1008496	A/DK/I/06/108	N	0.12	3				
Lettuce, head (F)	2007/1008496	A/UK/I/06/109	N	0.055	3				
Lettuce (F)	AL-726-004	Trial No. W/FR/E 81/446	S	0.28	3	0.585	0.195		
Lettuce, head (F)	2006/1026855	AF/8817/BA/5	S	0.103	3				
Lettuce, head (F)	2006/1026855	AF/8817/BA/6	S	0.305	3				
Lettuce, head (F)	2006/1026855	AF/8817/BA/7	S	0.210	3				
Lettuce, head (F)	2006/1026855	AF/8817/BA/8	S	0.585	3				
Lettuce, head (F)	2007/1008496	A/SF/I/06/110	S	0.32	3				
Lettuce, head (F)	2007/1008496	A/SP/I/06/111	S	0.11	3				
Lettuce, head (F)	2007/1008496	A/IT/I/06/112	S	0.41	3				
Lettuce, head (F)	2007/1008496	A/GR/I/06/113	S	<0.01	4				
Lettuce, head (F)	2007/1007938	AF/10503/BA/3	S	0.195	3				
Lettuce, head (F)	2007/1007938	AF/10503/BA/4	S	0.387	2				
Lettuce, leaf (F)	2014/1140312	L130050	S	0.24	3				
Lettuce, leaf (F)	2014/1140312	L130051	S	0.075	2				
Lettuce, leaf (F)	2014/1140312	L130052	S	0.032	3				
Lettuce, leaf (F)	2014/1140312	L130053	S	0.21	3				
Lettuce, leaf (F)	2014/1140312	L130054	S	0.049	3				
Lettuce, leaf (F)	2014/1140312	L130055	S	0.11	3				
Lettuce, leaf (F)	2014/1140312	L130056	S	0.037	2				
Lettuce, leaf (F)	2014/1140312	L130057	S	0.091	3				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Spinach (F)	2006/1026849	AF/8819/BA/1	N	0.135	7	1.113	0.465		
Spinach (F)	2007/1007944	AF/10498/BA/1	N	0.891	7				
Spinach (F)	2007/1007944	AF/10498/BA/2	N	0.185	7				
Spinach (F)	2007/1007944	AF/10498/BA/6	N	1.113	7				
Spinach (F)	2007/1035745	RU-I-18 06 SN DD 1/1	N	0.46	7				
Spinach (F)	2007/1035745	RU-I-18 06 BB FO 1/1	N	0.47	7				
Spinach (F)	2007/1035745	RU-I-18 06 NW BN 1/1	N	0.36	7				
Spinach (F)	2007/1035745	RU-I-18 06 NW BN 1/2	N	0.48	7				
Green beans with pods (F)	2006/1026857	AF/8825/BA/1	N	0.026	7	0.026	0.016	0.063	0.020
Green beans with pods (F)	2006/1026857	AF/8825/BA/2	N	0.011	7				
Green beans with pods (F)	2006/1026857	AF/8825/BA/3	N	0.014	14				
Green beans with pods (F)	2006/1026857	AF/8825/BA/4	N	0.015	7				
Green beans with pods (F)	2007/1007950	AF/10492/BA/1	N	<0.01	7				
Green beans with pods (F)	2007/1007950	AF/10492/BA/2	N	0.017	7				
Green beans with pods (F)	2007/1007950	AF/10492/BA/3	N	0.019	7				
Green beans with pods (F)	2007/1007950	AF/10492/BA/4	N	0.018	8				
Green beans with pods (F)	2006/1026857	AF/8825/BA/5	S	0.028	3	0.063	0.034		
Green beans with pods (F)	2006/1026857	AF/8825/BA/6	S	0.063	3				
Green beans with pods (F)	2006/1026857	AF/8825/BA/7	S	0.020	3				
Green beans with pods (F)	2006/1026857	AF/8825/BA/8	S	0.018	7				
Green beans with pods (F)	2007/1007950	AF/10492/BA/5	S	0.045	3				
Green beans with pods (F)	2007/1007950	AF/10492/BA/6	S	0.028	3				
Green beans with pods (F)	2007/1007950	AF/10492/BA/7	S	0.050	3				
Green beans with pods (F)	2007/1007950	AF/10492/BA/8	S	0.039	3				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Green peas without pods (F)	2006/1026856	AF/8826/BA/2	N	<0.01	3	0.01	0.01	0.01	0.01
Green peas without pods (F)	2006/1026856	AF/8826/BA/3	N	<0.01	3				
Green peas without pods (F)	2006/1026856	AF/8826/BA/4	N	<0.01	3				
Green peas without pods (F)	2006/1026856	AF/8826/BA/7	N	<0.01	3				
Green peas without pods (F)	2007/1007951	AF/10491/BA/1	N	<0.01	3				
Green peas without pods (F)	2007/1007951	AF/10491/BA/2	N	<0.01	2				
Green peas without pods (F)	2007/1007951	AF/10491/BA/3	N	<0.01	3				
Green peas without pods (F)	2007/1007951	AF/10491/BA/4	N	<0.01	4				
Green peas without pods (F)	2006/1026856	AF/8826/BA/5	S	<0.01	3	0.01	0.01		
Green peas without pods (F)	2006/1026856	AF/8826/BA/6	S	<0.01	3				
Green peas without pods (F)	2007/1007951	AF/10491/BA/5	S	<0.01	2				
Green peas without pods (F)	2007/1007951	AF/10491/BA/6	S	<0.01	3				
Artichoke (F)	2006/1026845	AF/8828/BA/1	S	0.021	7	0.04	0.023		
Artichoke (F)	2006/1026845	AF/8828/BA/2	S	0.040	7				
Artichoke (F)	2007/1007948	AF/10494/BA/1	S	0.019	7				
Artichoke (F)	2007/1007948	AF/10494/BA/2	S	0.025	7				
Celery (F)	2006/1029533	RU-I-1605 RPNW 2/1	N	0.1443	7	0.30	0.22		
Celery (F)	2006/1029533	RU-I-1605 RPNW 2/2	N	0.1822	7				
Celery (F)	2007/1035745	RU-I-1706 RPNW 1/1	N	0.30	7				
Celery (F)	2007/1035745	RU-I-1706 RPNW 1/2	N	0.23	7				
Celery (F)	2007/1035745	RU-I-1706 NWBN 1/1	N	0.22	7				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Leek (F)	2006/1026850	AF/8821/BA/1	N	0.058	14	0.071	0.044	0.105	0.044
Leek (F)	2006/1026850	AF/8821/BA/2	N	0.053	7				
Leek (F)	2006/1026850	AF/8821/BA/3	N	0.071	7				
Leek (F)	2006/1026850	AF/8821/BA/4	N	0.033	7				
Leek (F)	2007/1008498	A/NF/I/06/155	N	0.018	7				
Leek (F)	2007/1008498	A/BE/I/06/157	N	0.039	7				
Leek (F)	2007/1008498	A/GE/I/06/158	N	0.032	7				
Leek (F)	2007/1008498	A/UK/I/06/156	N	0.048	7				
Leek (F)	2006/1026850	AF/8821/BA/5	S	0.062	3	0.105	0.047		
Leek (F)	2006/1026850	AF/8821/BA/6	S	0.105	3				
Leek (F)	2007/1008498	A/SP/I/06/159	S	0.017	3				
Leek (F)	2007/1008498	A/SF/I/06/160	S	0.032	3				
Dry beans	2006/1026858	AF/8824/BA/5	N	<0.01	14	0.01	0.01	0.01	0.01
Dry beans	2006/1026858	AF/8824/BA/6	N	<0.01	14				
Dry beans	2007/1007949	AF/10493/BA/6	N	<0.01	14				
Dry beans	2007/1007949	AF/10493/BA/9	N	<0.01	14				
Dry beans	2006/1026858	AF/8824/BA/7	S	<0.01	14	0.01	0.01		
Dry beans	2006/1026858	AF/8824/BA/8	S	<0.01	14				
Dry beans	2007/1007949	AF/10493/BA/8	S	<0.01	14				
Dry beans	2007/1007949	AF/10493/BA/11	S	<0.01	14				
Dry peas	2006/1026858	AF/8824/BA/1	N	<0.01	14	0.01	0.01	0.042	0.01
Dry peas	2006/1026858	AF/8824/BA/2	N	<0.01	14				
Dry peas	2007/1007949	AF/10493/BA/1	N	<0.01	14				
Dry peas	2007/1007949	AF/10493/BA/2	N	<0.01	13				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Dry peas	2006/1026858	AF/8824/BA/3	S	<0.01	14	0.042	0.01		
Dry peas	2006/1026858	AF/8824/BA/4	S	0.042	28				
Dry peas	2007/1007949	AF/10493/BA/3	S	<0.01	13				
Dry peas	2007/1007949	AF/10493/BA/4	S	<0.01	13				
Cotton seed	2006/1026847	AF/8822/BA/1	S	<0.01	7	0.018	0.01		
Cotton seed	2006/1026847	AF/8822/BA/2	S	0.018	13				
Cotton seed	2006/1026847	AF/8822/BA/3	S	<0.01	6				
Cotton seed	2006/1026847	AF/8822/BA/4	S	<0.01	7				
Cotton seed	2007/1007947	AF/10495/BA/1	S	<0.01	7				
Cotton seed	2007/1007947	AF/10495/BA/2	S	<0.01	7				
Cotton seed	2007/1007947	AF/10495/BA/3	S	<0.01	7				
Cotton seed	2007/1007947	AF/10495/BA/4	S	<0.01	7				
Rape seed	AL-750-004	Not reported	N	<0.01	61	0.01	0.01	0.06	0.01
Rape seed	AL-750-004	WROSR 3	N	<0.01	69				
Rape seed	AL-750-004	83/307	N	<0.01	73				
Rape seed	AL-750-004	83/306	N	<0.01	70				
Rape seed	AL-750-004	83/305	N	<0.01	75				
Rape seed	2008/1019999	AF/12151/BA/2	N	<0.01	28				
Rape seed	2008/1019999	AF/12151/BA/3	N	<0.01	28				
Rape seed	2013/1037957	L120460	N	<0.01	29				
Rape seed	2013/1037957	L120461	N	<0.01	29				
Rape seed	2013/1416283	S13-00443-01 / L130038	N	<0.01	29				
Rape seed	2013/1416283	S13-00443-02/ L130039	N	<0.01	28				
Rape seed	2013/1416283	S13-00443-03/ L130040	N	<0.01	28				
Rape seed	2013/1416283	S13-00443-04/ L130041	N	<0.01	27				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Rape seed	AL-750-021	W/FR/R/92/244	S	<0.01	64	0.06	0.01		
Rape seed	AL-750-021	W/FR/R/92/244	S	<0.01	64				
Rape seed	AL-750-021	W/FR/R/92/246	S	<0.01	75				
Rape seed	AL-750-021	W/FR/R/92/246	S	<0.01	75				
Rape seed	2002/1004087	ALO/10/01	S	0.06	42				
Rape seed	2002/1004087	FTL/07/01	S	<0.05	43				
Rape seed	2008/1019999	AF/12151/BA/6	S	<0.01	27				
Rape seed	2008/1019999	AF/12151/BA/7	S	<0.01	28				
Rape seed	2013/1037957	L120462	S	<0.01	28				
Rape seed	2013/1037957	L120463	S	<0.01	28				
Rape seed	2013/1037957	L120464	S	<0.01	28				
Rape seed	2013/1037957	L120465	S	<0.01	29				
Rape seed	2013/1416283	S13-00443-05/ L130042	S	<0.01	29				
Rape seed	2013/1416283	S13-00443-06/ L130043	S	0.015	29				
Rape seed	2013/1416283	S13-00443-07/ L130044	S	0.012	29				
Rape seed	2013/1416283	S13-00443-08/ L130045	S	<0.01	28				
Barley grain	2013/1388974	L120444	N	0.035	29	0.079	0.033	0.083	0.035
Barley grain	2013/1388974	L120445	N	0.053	29				
Barley grain	2013/1388974	L120446	N	0.032	28				
Barley grain	2013/1388974	L120447	N	0.031	28				
Barley grain	2013/1416284	S13-00441-01 / L130026	N	0.020	27				
Barley grain	2013/1416284	S13-00441-03 / L130028	N	0.077	22				
Barley grain	2013/1416284	S13-00441-04 / L130029	N	0.079	28				
Barley grain	2014/1173599	S14-00678 / L140092	N	0.030	28±1				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Barley grain	AL-730-029	W/FR/E/92/238	S	<0.01	48	0.083	0.035	0.01	0.01
Barley grain	2013/1388974	L120448	S	0.050	26				
Barley grain	2013/1388974	L120450	S	0.066	21				
Barley grain	2013/1388974	L120451	S	0.026	28				
Barley grain	2013/1416284	S13-00441-05 / L130030	S	0.034	20				
Barley grain	2013/1416284	S13-00441-06 / L130031	S	0.079	26				
Barley grain	2013/1416284	S13-00441-07 / L130032	S	0.035	28				
Barley grain	2013/1416284	S13-00441-08 / L130033	S	0.024	36				
Barley grain	2013/1416281	S13-00446-01 / L130003	S	0.083	28				
Maize	2009/1125196	L090227	N	<0.01	20	0.01	0.01		
Maize	2009/1125196	L090228	N	<0.01	14				
Maize	AL-730-013	W/FR/E81/883	N	<0.01	95				
Maize	AL-730-057	R32-86	N	<0.01	57				
Maize	AL-730-058	R31-86	N	<0.01	61				
Maize	AL-730-059	R33-86	N	<0.01	59				
Maize	AL-730-060	R34-86	N	<0.01	59				
Maize	AL-730-062	R94/87	N	<0.01	63				
Maize	AL-730-063	R95/87	N	<0.01	56				
Maize	AL-730-064	R96/87	N	<0.01	60				
Maize	AL-730-065	R97/87	N	<0.01	69				
Maize	2009/1125196	L090229	S	<0.01	21	0.01	0.01		
Maize	AL-730-013	W/FR/E81/221	S	<0.01	100				
Maize	AL-730-019	S/FR/E89/161	S	<0.01	50				
Maize	AL-730-019	S/FR/E89/416	S	<0.01	42				
Maize	AL-730-027	S/FR/E90/160	S	<0.01	13				
Maize	AL-730-027	S/FR/E90/424	S	<0.01	40				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Maize	AL-730-027	S/FR/E90/447	S	<0.01	22				
Maize	AL-730-014	S/SA/E81/443	South Africa	<0.01	14				
Maize	AL-730-015	I/RE/MI-01/82	Brazil	<0.01	22				
Rice	2006/1026848	AF/8823/BA/1	S	0.148	14	0.206	0.092		
Rice	2006/1026848	AF/8823/BA/2	S	0.011	21				
Rice	2006/1026848	AF/8823/BA/3	S	0.206	28				
Rice	2006/1026848	AF/8823/BA/4	S	0.039	21				
Rice	2007/1007946	AF/10496/BA/1	S	0.066	21				
Rice	2007/1007946	AF/10496/BA/2	S	0 13	20				
Rice	2007/1007946	AF/10496/BA/3	S	0.110	22				
Rice	2007/1007946	AF/10496/BA/4	S	0.073	21				
Wheat grain	AL-730-001	S/FR/E83/878	N	<0.01	46	0.01	0.01	0.01	0.01
Wheat grain	AL-730-003	S/FR/E84/890	N	<0.01	28				
Wheat grain	AL-730-003	S/FR/E84/891	N	<0.01	20				
Wheat grain	AL-730-003	S/FR/E84/892	N	<0.01	28				
Wheat grain	AL-730-003	S/FR/E84/893	N	<0.01	27				
Wheat grain	AL-730-031	BE61	N	<0.01	34				
Wheat grain	2008/1002701	L070422	N	<0.01	35				
Wheat grain	2008/1002701	L070423	N	<0.01	36				
Wheat grain	2008/1002701	L070424	N	<0.01	27				
Wheat grain	2008/1002701	L070425	N	0.010	34				
Wheat grain	2014/1028112	L120452	N	<0.01	28				
Wheat grain	2014/1028112	L120453	N	<0.01	29				
Wheat grain	2014/1028112	L120454	N	<0.01	65				
Wheat grain	2014/1028112	L120455	N	<0.01	28				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Wheat grain	2013/1416282	S13-00445-01 / L130034	N	<0.01	27				
Wheat grain	2013/1416282	S13-00445-02 / L130035	N	<0.01	28				
Wheat grain	AL-730-001	S/FR/E83/220	S	<0.01	38				
Wheat grain	AL-730-001	S/FR/E83/419	S	<0.01	43				
Wheat grain	AL-730-003	S/FE/E84/217	S	<0.01	28				
Wheat grain	AL-730-003	S/FR/E84/218	S	<0.01	28				
Wheat grain	AL-730-003	S/FR/E84/347	S	<0.01	28				
Wheat grain	AL-730-003	S/FR/E84/348	S	<0.01	28				
Wheat grain	AL-730-029	W/FR/E/92/239	S	<0.01	48				
Wheat grain	AL-730-030	W/FR/E/92/209	S	<0.01	50				
Wheat grain	AL-730-030	W/FR/E/92/869	S	<0.01	38				
Wheat grain	2008/1002701	L070426	S	<0.01	35	0.01	0.01		
Wheat grain	2008/1002701	L070427	S	<0.01	27				
Wheat grain	2008/1002701	L070428	S	<0.01	28				
Wheat grain	2008/1002701	L070429	S	<0.01	36				
Wheat grain	2014/1028112	L120456	S	<0.01	28				
Wheat grain	2014/1028112	L120457	S	<0.01	28				
Wheat grain	2014/1028112	L120458	S	<0.01	36				
Wheat grain	2014/1028112	L120459	S	<0.01	28				
Wheat grain	2013/1416282	S13-00445-03 / L130036	S	<0.01	27				
Wheat grain	2013/1416282	S13-00445-04 / L130037	S	<0.01	28				

Table 5-1: Residue levels of alpha-cypermethrin, BAS 310 I (Raw Data)

Crop	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		N+S	
						HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Sugar beet root	2003/1021716	AF/6792/BA/1	N	<0.010	92	0.228	0.03	0.228	0.01
Sugar beet root	2003/1021716	AF/6792/BA/2	N	<0.010	92				
Sugar beet root	2003/1021716	AF/6792/BA/3	N	<0.010	88				
Sugar beet root	2003/1021716	AF/6792/BA/4	N	<0.010	84				
Sugar beet root	2004/1006468	ACK/02/03	N	<0.050	25				
Sugar beet root	2004/1006468	AGR/02/03	N	<0.050	14				
Sugar beet root	2004/1006468	DU4/02/03	N	<0.050	30				
Sugar beet root	2004/1006468	DU2/02/03	N	0.228	21				
Sugar beet root	2006/1026851	AF/8832/BA/1	S	<0.010	7	0.01	0.01		
Sugar beet root	2006/1026851	AF/8832/BA/2	S	<0.010	7				
Sugar beet root	2006/1026851	AF/8832/BA/3	S	<0.010	7				
Sugar beet root	2006/1026851	AF/8832/BA/4	S	<0.010	7				
Sugar beet root	2007/1007941	AF/10501/BA/1	S	<0.010	7				
Sugar beet root	2007/1007941	AF/10501/BA/2	S	<0.010	7				
Sugar beet root	2007/1007941	AF/10501/BA/3	S	<0.010	7				
Sugar beet root	2007/1007941	AF/10501/BA/4	S	<0.010	7				

All trials from the table below were used for further evaluation. In the last column of the table it is shown which trials were used to derive the MRLs, HRs and STMRs used for evaluation.

Table 5-2: Residue data used for the calculation of the livestock dietary burden (Raw Data)

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Potato	Tuber	2006/1026846	AF/8831/BA/1	N	<0.01	7	0.01	0.01	0.01	0.01
Potato	Tuber	2006/1026846	AF/8831/BA/2	N	<0.01	7				
Potato	Tuber	2006/1026846	AF/8831/BA/3	N	<0.01	7				
Potato	Tuber	2006/1026846	AF/8831/BA/4	N	<0.01	7				
Potato	Tuber	2007/1007945	AF/10497/BA/1	N	<0.01	7				
Potato	Tuber	2007/1007945	AF/10497/BA/2	N	<0.01	7				
Potato	Tuber	2007/1007945	AF/10497/BA/3	N	<0.01	7				
Potato	Tuber	2007/1007945	AF/10497/BA/9	N	<0.01	7				
Potato	Tuber	2008/1002704	L070446	N	<0.01	6				
Potato	Tuber	2008/1002704	L070447	N	<0.01	8				
Potato	Tuber	2012/1157550	L110423	N	<0.01	7				
Potato	Tuber	2012/1157550	L110424	N	<0.01	7				
Potato	Tuber	2007/1007945	AF/10497/BA/5	S	<0.01	14	0.01	0.01		
Potato	Tuber	2007/1007945	AF/10497/BA/6	S	<0.01	14				
Potato	Tuber	2007/1007945	AF/10497/BA/7	S	<0.01	14				
Potato	Tuber	2007/1007945	AF/10497/BA/8	S	<0.01	14				
Potato	Tuber	2006/1026846	AF/8831/BA/5	S	<0.01	14				
Potato	Tuber	2006/1026846	AF/8831/BA/6	S	<0.01	14				
Potato	Tuber	2006/1026846	AF/8831/BA/7	S	<0.01	14				
Potato	Tuber	2006/1026846	AF/8831/BA/8	S	<0.01	14				
Potato	Tuber	2005/1007592	GRE/02/04	S	<0.01	14				
Potato	Tuber	2005/1007592	ITA/01/04	S	<0.01	14				
Potato	Tuber	2008/1002704	L070448	S	<0.01	15				
Potato	Tuber	2008/1002704	L070449	S	<0.01	14				
Potato	Tuber	2012/1157550	L110425	S	<0.01	14				
Potato	Tuber	2012/1157550	L110426	S	<0.01	14				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Carrot	Root	2005/1035294	RU-I-13 04 RP NW 2/1	N	<0.01	3	0.01	0.01		
Carrot	Root	2005/1035294	RU-I-13 04 RP NW 2/2	N	<0.01	3				
Carrot	Root	2005/1035294	RU-I-13 04 RP NW 2/3	N	<0.01	3				
Carrot	Root	2006/1029533	RU-I-06 05 NW BN 2/1	N	<0.01	3				
Carrot	Root	2006/1029533	RU-I-06 05 NW BN 221	N	<0.01	7				
Carrot	Root	2006/1029533	RU-I-06 05 SH KI 2/1	N	<0.01	7				
Carrot	Root	2006/1029533	RU-I-06 05 SH KI 2/2	N	<0.01	7				
Carrot	Root	2006/1029533	RU-I-06 05 BY FS 2/1	N	<0.01	7				
Carrot	Root	2006/1029533	RU-I-06 05 BY FS 2/2	N	<0.01	7				
Cabbage	Head	2004/1006470	ACK/14/03	N	<0.050	7	0.105	0.034	0.105	0.014
Cabbage	Head	2004/1006470	ALB/12/03	N	<0.050	7				
Cabbage	Head	2004/1006470	FAN/21/03	N	<0.050	7				
Cabbage	Head	2004/1006470	OAT/18/03	N	<0.050	6				
Cabbage	Head	2006/1026852	AF/8829/BA/1	N	0.018	7				
Cabbage	Head	2006/1026852	AF/8829/BA/2	N	<0.010	7				
Cabbage	Head	2006/1026852	AF/8829/BA/4	N	0.104	4				
Cabbage	Head	2006/1026852	AF/8829/BA/7	N	0.014	7				
Cabbage	Head	2007/1013148	A/NF/I/06/143	N	<0.010	8				
Cabbage	Head	2007/1013148	A/BE/I/06/144	N	<0.010	7				
Cabbage	Head	2007/1013148	A/UK/I/06/145	N	0.105	7				
Cabbage	Head	2007/1013148	A/GE/I/06/146	N	<0.010	7				
Cabbage	Head	2006/1026852	AF/8829/BA/5	S	<0.010	3	0.013	0.012		
Cabbage	Head	2006/1026852	AF/8829/BA/6	S	0.013	3				
Cabbage	Head	2007/1013148	A/SF/I/06/147	S	0.010	3				
Cabbage	Head	2007/1013148	A/SP/I/06/148	S	0.013	6				
Leafy brassica	Leaves	2007/1013342	A/NF/I/06/115	N	0.590	3	0.59	0.367	0.59	0.14
Leafy brassica	Leaves	2007/1013342	A/NL/I/06/116	N	0.350	4				
Leafy brassica	Leaves	2006/1026862	AF/8815/BA/1	N	0.170	3				
Leafy brassica	Leaves	2006/1026862	AF/8815/BA/5	N	0.383	7				
Leafy brassica	Leaves	2006/1026862	AF/8815/BA/3	S	0.436	3	0.436	0.075		
Leafy brassica	Leaves	2006/1026862	AF/8815/BA/4	S	0.278	3				
Leafy brassica	Leaves	2007/1013342	A/SF/I/06/117	S	0.046	3				
Leafy brassica	Leaves	2007/1013342	A/SP/I/06/118	S	0.056	3				
Chinese cabbage	Leaves	2013/1140312	S13-00427-01 / L130046	S	0.086	3				
Chinese cabbage	Leaves	2013/1140312	S13-00427-02 / L130047	S	0.110	3				
Chinese cabbage	Leaves	2013/1140312	S13-00427-03 / L130048	S	0.064	3				
Chinese cabbage	Leaves	2013/1140312	S13-00427-04 / L130049	S	0.019	2				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Dry beans	Seed	2006/1026858	AF/8824/BA/5	N	<0.01	14	0.01	0.01	0.01	0.01
Dry beans	Seed	2006/1026858	AF/8824/BA/6	N	<0.01	14				
Dry beans	Seed	2007/1007949	AF/10493/BA/6	N	<0.01	14				
Dry beans	Seed	2007/1007949	AF/10493/BA/9	N	<0.01	14				
Dry beans	Seed	2006/1026858	AF/8824/BA/7	S	<0.01	14	0.01	0.01		
Dry beans	Seed	2006/1026858	AF/8824/BA/8	S	<0.01	14				
Dry beans	Seed	2007/1007949	AF/10493/BA/8	S	<0.01	14				
Dry beans	Seed	2007/1007949	AF/10493/BA/11	S	<0.01	14				
Dry peas	Seed	2006/1026858	AF/8824/BA/1	N	<0.01	14	0.01	0.01	0.042	0.01
Dry peas	Seed	2006/1026858	AF/8824/BA/2	N	<0.01	14				
Dry peas	Seed	2007/1007949	AF/10493/BA/1	N	<0.01	14				
Dry peas	Seed	2007/1007949	AF/10493/BA/2	N	<0.01	13				
Dry peas	Seed	2006/1026858	AF/8824/BA/3	S	<0.01	14	0.042	0.01		
Dry peas	Seed	2006/1026858	AF/8824/BA/4	S	0.042	28				
Dry peas	Seed	2007/1007949	AF/10493/BA/3	S	<0.01	13				
Dry peas	Seed	2007/1007949	AF/10493/BA/4	S	<0.01	13				
Bean	straw	2006/1026858	AF/8824/BA/5	N	0.396	21	0.593	0.300	0.593	0.246
Bean	straw	2006/1026858	AF/8824/BA/6	N	0.593	28				
Bean	straw	2007/1007949	AF/10493/BA/6	N	0.167	14				
Bean	straw	2007/1007949	AF/10493/BA/9	N	0.203	28				
Bean	straw	2006/1026858	AF/8824/BA/7	S	0.095	14	0.424	0.198		
Bean	straw	2006/1026858	AF/8824/BA/8	S	0.289	14				
Bean	straw	2007/1007949	AF/10493/BA/8	S	0.424	14				
Bean	straw	2007/1007949	AF/10493/BA/11	S	0.107	14				
Pea	straw	2006/1026858	AF/8824/BA/1	N	0.524	21	1.008	0.450	1.008	0.515
Pea	straw	2006/1026858	AF/8824/BA/2	N	0.365	21				
Pea	straw	2007/1007949	AF/10493/BA/1	N	1.008	21				
Pea	straw	2007/1007949	AF/10493/BA/2	N	0.376	13				
Pea	straw	2006/1026858	AF/8824/BA/3	S	0.673	21	0.997	0.589		
Pea	straw	2006/1026858	AF/8824/BA/4	S	0.477	21				
Pea	straw	2007/1007949	AF/10493/BA/3	S	0.997	21				
Pea	straw	2007/1007949	AF/10493/BA/4	S	0.505	27				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Bean	Vines	2006/1026857	AF/8825/BA/1	N	0.087	7	0.177	0.125	1.013	0.188
Bean	Vines	2006/1026857	AF/8825/BA/2	N	0.144	7				
Bean	Vines	2006/1026857	AF/8825/BA/3	N	0.092	14				
Bean	Vines	2006/1026857	AF/8825/BA/4	N	0.177	7				
Bean	Vines	2007/1007950	AF/10492/BA/1	N	0.089	7				
Bean	Vines	2007/1007950	AF/10492/BA/2	N	0.172	7				
Bean	Vines	2007/1007950	AF/10492/BA/3	N	0.127	7				
Bean	Vines	2007/1007950	AF/10492/BA/4	N	0.123	8				
Bean	Vines	2006/1026857	AF/8825/BA/5	S	0.256	3	1.013	0.614		
Bean	Vines	2006/1026857	AF/8825/BA/6	S	0.796	3				
Bean	Vines	2006/1026857	AF/8825/BA/7	S	0.455	3				
Bean	Vines	2006/1026857	AF/8825/BA/8	S	0.594	3				
Bean	Vines	2007/1007950	AF/10492/BA/5	S	1.013	3				
Bean	Vines	2007/1007950	AF/10492/BA/6	S	0.634	3				
Bean	Vines	2007/1007950	AF/10492/BA/7	S	0.704	3				
Bean	Vines	2007/1007950	AF/10492/BA/8	S	0.198	3				
Pea	Vines	2006/1026856	AF/8826/BA/2	N	0.076	7	0.430	0.264	0.431	0.275
Pea	Vines	2006/1026856	AF/8826/BA/3	N	0.057	3				
Pea	Vines	2006/1026856	AF/8826/BA/4	N	0.354	3				
Pea	Vines	2006/1026856	AF/8826/BA/7	N	0.301	7				
Pea	Vines	2007/1007951	AF/10491/BA/1	N	0.104	7				
Pea	Vines	2007/1007951	AF/10491/BA/2	N	0.430	2				
Pea	Vines	2007/1007951	AF/10491/BA/3	N	0.226	14				
Pea	Vines	2007/1007951	AF/10491/BA/4	N	0.346	7				
Pea	Vines	2006/1026856	AF/8826/BA/5	S	0.267	3	0.431	0.275		
Pea	Vines	2006/1026856	AF/8826/BA/6	S	0.282	7				
Pea	Vines	2007/1007951	AF/10491/BA/5	S	0.165	2				
Pea	Vines	2007/1007951	AF/10491/BA/6	S	0.431	3				
Cotton	Seed	2006/1026847	AF/8822/BA/1	S	<0.01	7	0.018	0.01		
Cotton	Seed	2006/1026847	AF/8822/BA/2	S	0.018	13				
Cotton	Seed	2006/1026847	AF/8822/BA/3	S	<0.01	6				
Cotton	Seed	2006/1026847	AF/8822/BA/4	S	<0.01	7				
Cotton	Seed	2007/1007947	AF/10495/BA/1	S	<0.01	7				
Cotton	Seed	2007/1007947	AF/10495/BA/2	S	<0.01	7				
Cotton	Seed	2007/1007947	AF/10495/BA/3	S	<0.01	7				
Cotton	Seed	2007/1007947	AF/10495/BA/4	S	<0.01	7				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Rape	Seed	AL-750-004	Not reported	N	<0.01	61	0.01	0.01	0.06	0.01
Rape	Seed	AL-750-004	WROSR 3	N	<0.01	69				
Rape	Seed	AL-750-004	83/307	N	<0.01	73				
Rape	Seed	AL-750-004	83/306	N	<0.01	70				
Rape	Seed	AL-750-004	83/305	N	<0.01	75				
Rape	Seed	2008/1019999	AF/12151/BA/2	N	<0.01	28				
Rape	Seed	2008/1019999	AF/12151/BA/3	N	<0.01	28				
Rape	Seed	2013/1037957	L120460	N	<0.01	29				
Rape	Seed	2013/1037957	L120461	N	<0.01	29				
Rape	Seed	2013/1416283	S13-00443-01 / L130038	N	<0.01	29				
Rape	Seed	2013/1416283	S13-00443-02/ L130039	N	<0.01	28				
Rape	Seed	2013/1416283	S13-00443-03/ L130040	N	<0.01	28				
Rape	Seed	2013/1416283	S13-00443-04/ L130041	N	<0.01	27				
Rape	Seed	AL-750-021	W/FR/R/92/244	S	<0.01	64	0.06	0.01		
Rape	Seed	AL-750-021	W/FR/R/92/244	S	<0.01	64				
Rape	Seed	AL-750-021	W/FR/R/92/246	S	<0.01	75				
Rape	Seed	AL-750-021	W/FR/R/92/246	S	<0.01	75				
Rape	Seed	2002/1004087	ALO/10/01	S	0.06	42				
Rape	Seed	2002/1004087	FTL/07/01	S	<0.05	43				
Rape	Seed	2008/1019999	AF/12151/BA/6	S	<0.01	27				
Rape	Seed	2008/1019999	AF/12151/BA/7	S	<0.01	28				
Rape	Seed	2013/1037957	L120462	S	<0.01	28				
Rape	Seed	2013/1037957	L120463	S	<0.01	28				
Rape	Seed	2013/1037957	L120464	S	<0.01	28				
Rape	Seed	2013/1037957	L120465	S	<0.01	29				
Rape	Seed	2013/1416283	S13-00443-05/ L130042	S	<0.01	29				
Rape	Seed	2013/1416283	S13-00443-06/ L130043	S	0.015	29				
Rape	Seed	2013/1416283	S13-00443-07/ L130044	S	0.012	29				
Rape	Seed	2013/1416283	S13-00443-08/ L130045	S	<0.01	28				
Rape	Forage	2013/1037957	L120460	N	0.160	29	0.27	0.135	0.270	0.101
Rape	Forage	2013/1037957	L120461	N	0.110	29				
Rape	Forage	2013/1416283	S13-00443-01 / L130038	N	0.140	29				
Rape	Forage	2013/1416283	S13-00443-02/ L130039	N	0.066	28				
Rape	Forage	2013/1416283	S13-00443-03/ L130040	N	0.270	28				
Rape	Forage	2013/1416283	S13-00443-04/ L130041	N	0.130	27				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Rape	Forage	2002/1004087	ALO/10/01	S	0.24	29	0.24	0.076		
Rape	Forage	2002/1004087	FTL/07/01	S	<0.05	29				
Rape	Forage	2013/1037957	L120462	S	0.067	28				
Rape	Forage	2013/1037957	L120463	S	0.018	28				
Rape	Forage	2013/1037957	L120464	S	0.190	28				
Rape	Forage	2013/1037957	L120465	S	0.130	29				
Rape	Forage	2013/1416283	S13-00443-05/ L130042	S	0.040	29				
Rape	Forage	2013/1416283	S13-00443-06/ L130043	S	0.092	29				
Rape	Forage	2013/1416283	S13-00443-07/ L130044	S	0.084	29				
Rape	Forage	2013/1416283	S13-00443-08/ L130045	S	0.060	28				
Barley	Grain	2013/1388974	L120444	N	0.035	29	0.079	0.033	0.083	0.035
Barley	Grain	2013/1388974	L120445	N	0.053	29				
Barley	Grain	2013/1388974	L120446	N	0.032	28				
Barley	Grain	2013/1388974	L120447	N	0.031	28				
Barley	Grain	2013/1416284	S13-00441-01 / L130026	N	0.020	27				
Barley	Grain	2013/1416284	S13-00441-03 / L130028	N	0.077	22				
Barley	Grain	2013/1416284	S13-00441-04 / L130029	N	0.079	28				
Barley	Grain	2014/1173599	S14-00678 / L140092	N	0.030	28±1				
Barley	Grain	AL-730-029	W/FR/E/92/238	S	<0.01	48	0.083	0.035		
Barley	Grain	2013/1388974	L120448	S	0.050	26				
Barley	Grain	2013/1388974	L120450	S	0.066	21				
Barley	Grain	2013/1388974	L120451	S	0.026	28				
Barley	Grain	2013/1416284	S13-00441-05 / L130030	S	0.034	20				
Barley	Grain	2013/1416284	S13-00441-06 / L130031	S	0.079	26				
Barley	Grain	2013/1416284	S13-00441-07 / L130032	S	0.035	28				
Barley	Grain	2013/1416284	S13-00441-08 / L130033	S	0.024	36				
Barley	Grain	2013/1416281	S13-00446-01 / L130003	S	0.083	28				
Barley	Forage	2013/1416284	S13-00441-08 / L130033	S	0.38	28				
Barley	Straw	2013/1388974	L120444	N	0.250	29	0.49	0.38	0.92	0.400
Barley	Straw	2013/1388974	L120445	N	0.490	29				
Barley	Straw	2013/1388974	L120446	N	0.260	28				
Barley	Straw	2013/1388974	L120447	N	0.470	28				
Barley	Straw	2013/1416284	S13-00441-01 / L130026	N	0.350	27				
Barley	Straw	2013/1416284	S13-00441-03 / L130028	N	0.480	22				
Barley	Straw	2013/1416284	S13-00441-04 / L130029	N	0.410	28				
Barley	Straw	2014/1173599	S14-00678 / L140092	N	0.200	28±1				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Barley	Straw	AL-730-029	W/FR/E/92/238	S	0.08	48				
Barley	Straw	2013/1388974	L120448	S	0.400	26				
Barley	Straw	2013/1388974	L120450	S	0.240	21				
Barley	Straw	2013/1388974	L120451	S	0.210	28				
Barley	Straw	2013/1416284	S13-00441-05 / L130030	S	0.920	20	0.92	0.40		
Barley	Straw	2013/1416284	S13-00441-06 / L130031	S	0.470	26				
Barley	Straw	2013/1416284	S13-00441-07 / L130032	S	0.680	28				
Barley	Straw	2013/1416284	S13-00441-08 / L130033	S	0.400	36				
Barley	Straw	2013/1416281	S13-00446-01 / L130003	S	0.430	28				
Maize	Grain	2009/1125196	L090227	N	<0.01	20				
Maize	Grain	2009/1125196	L090228	N	<0.01	14				
Maize	Grain	AL-730-013	W/FR/E81/883	N	<0.01	95				
Maize	Grain	AL-730-057	R32-86	N	<0.01	57				
Maize	Grain	AL-730-058	R31-86	N	<0.01	61				
Maize	Grain	AL-730-059	R33-86	N	<0.01	59	0.01	0.01		
Maize	Grain	AL-730-060	R34-86	N	<0.01	59				
Maize	Grain	AL-730-062	R94/87	N	<0.01	63				
Maize	Grain	AL-730-063	R95/87	N	<0.01	56				
Maize	Grain	AL-730-064	R96/87	N	<0.01	60			0.01	0.01
Maize	Grain	AL-730-065	R97/87	N	<0.01	69				
Maize	Grain	2009/1125196	L090229	S	<0.01	21				
Maize	Grain	AL-730-013	W/FR/E81/221	S	<0.01	100				
Maize	Grain	AL-730-019	S/FR/E89/161	S	<0.01	50				
Maize	Grain	AL-730-019	S/FR/E89/416	S	<0.01	42	0.01	0.01		
Maize	Grain	AL-730-027	S/FR/E90/160	S	<0.01	13				
Maize	Grain	AL-730-027	S/FR/E90/424	S	<0.01	40				
Maize	Grain	AL-730-027	S/FR/E90/447	S	<0.01	22				
Maize	Grain	AL-730-014	S/SA/E81/443	South Africa	<0.01	14				
Maize	Grain	AL-730-015	I/RE/MI-01/82	Brazil	<0.01	22				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Maize	Silage	AL-730-013	W/FR/E81/883	N	<0.010	95	0.46	0.18	0.460	0.180
Maize	Silage	AL-730-062	R94/87	N	0.020	21				
Maize	Silage	AL-730-058	R31-86	N	0.12	14				
Maize	Silage	2009/1125196	L090227	N	0.134	20				
Maize	Silage	AL-730-059	R33-86	N	0.18	14				
Maize	Silage	AL-730-064	R96/87	N	0.18	21				
Maize	Silage	2009/1125196	L090228	N	0.214	14				
Maize	Silage	AL-730-063	R95/87	N	0.290	14				
Maize	Silage	AL-730-060	R34-86	N	0.320	14				
Maize	Silage	AL-730-057	R32-86	N	0.330	14				
Maize	Silage	AL-730-065	R97/87	N	0.46	14				
Maize	Silage	AL-730-013	W/FR/E81/221	S	<0.010	65	0.168	0.089		
Maize	Silage	2009/1125196	L090229	S	0.168	21				
Rice	Grain	2006/1026848	AF/8823/BA/1	S	0.148	14	0.206	0.092		
Rice	Grain	2006/1026848	AF/8823/BA/2	S	0.011	21				
Rice	Grain	2006/1026848	AF/8823/BA/3	S	0.206	28				
Rice	Grain	2006/1026848	AF/8823/BA/4	S	0.039	21				
Rice	Grain	2007/1007946	AF/10496/BA/1	S	0.066	21				
Rice	Grain	2007/1007946	AF/10496/BA/2	S	0.13	20				
Rice	Grain	2007/1007946	AF/10496/BA/3	S	0.110	22				
Rice	Grain	2007/1007946	AF/10496/BA/4	S	0.073	21				
Rice	Straw	2006/1026848	AF/8823/BA/1	S	0.131	21	0.354	0.136		
Rice	Straw	2006/1026848	AF/8823/BA/2	S	0.194	21				
Rice	Straw	2006/1026848	AF/8823/BA/3	S	0.354	21				
Rice	Straw	2006/1026848	AF/8823/BA/4	S	0.141	21				
Rice	Straw	2007/1007946	AF/10496/BA/1	S	0.11	21				
Rice	Straw	2007/1007946	AF/10496/BA/2	S	0.091	20				
Rice	Straw	2007/1007946	AF/10496/BA/3	S	0.190	22				
Rice	Straw	2007/1007946	AF/10496/BA/4	S	0.13	27				
Wheat	Grain	AL-730-001	S/FR/E83/878	N	<0.01	46	0.01	0.01	0.01	0.01
Wheat	Grain	AL-730-003	S/FR/E84/890	N	<0.01	28				
Wheat	Grain	AL-730-003	S/FR/E84/891	N	<0.01	20				
Wheat	Grain	AL-730-003	S/FR/E84/892	N	<0.01	28				
Wheat	Grain	AL-730-003	S/FR/E84/893	N	<0.01	27				
Wheat	Grain	AL-730-031	BE61	N	<0.01	34				
Wheat	Grain	2008/1002701	L070422	N	<0.01	35				
Wheat	Grain	2008/1002701	L070423	N	<0.01	36				
Wheat	Grain	2008/1002701	L070424	N	<0.01	27				
Wheat	Grain	2008/1002701	L070425	N	0.010	34				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Wheat	Grain	2014/1028112	L120452	N	<0.01	28				
Wheat	Grain	2014/1028112	L120453	N	<0.01	29				
Wheat	Grain	2014/1028112	L120454	N	<0.01	65				
Wheat	Grain	2014/1028112	L120455	N	<0.01	28				
Wheat	Grain	2013/1416282	S13-00445-01 / L130034	N	<0.01	27				
Wheat	Grain	2013/1416282	S13-00445-02 / L130035	N	<0.01	28				
Wheat	Grain	AL-730-001	S/FR/E83/220	S	<0.01	38				
Wheat	Grain	AL-730-001	S/FR/E83/419	S	<0.01	43				
Wheat	Grain	AL-730-003	S/FE/E84/217	S	<0.01	28				
Wheat	Grain	AL-730-003	S/FR/E84/218	S	<0.01	28				
Wheat	Grain	AL-730-003	S/FR/E84/347	S	<0.01	28				
Wheat	Grain	AL-730-003	S/FR/E84/348	S	<0.01	28				
Wheat	Grain	AL-730-029	W/FR/E/92/239	S	<0.01	48				
Wheat	Grain	AL-730-030	W/FR/E/92/209	S	<0.01	50				
Wheat	Grain	AL-730-030	W/FR/E/92/869	S	<0.01	38				
Wheat	Grain	2008/1002701	L070426	S	<0.01	35	0.01	0.01		
Wheat	Grain	2008/1002701	L070427	S	<0.01	27				
Wheat	Grain	2008/1002701	L070428	S	<0.01	28				
Wheat	Grain	2008/1002701	L070429	S	<0.01	36				
Wheat	Grain	2014/1028112	L120456	S	<0.01	28				
Wheat	Grain	2014/1028112	L120457	S	<0.01	28				
Wheat	Grain	2014/1028112	L120458	S	<0.01	36				
Wheat	Grain	2014/1028112	L120459	S	<0.01	28				
Wheat	Grain	2013/1416282	S13-00445-03 / L130036	S	<0.01	27				
Wheat	Grain	2013/1416282	S13-00445-04 / L130037	S	<0.01	28				
Wheat	Forage	2008/1002701	L070422	N	0.340	28				
Wheat	Forage	2008/1002701	L070423	N	0.730	28	0.730	0.335		
Wheat	Forage	2014/1028112	L120454	N	0.130	29				
Wheat	Forage	2014/1028112	L120455	N	0.330	28				
Wheat	Forage	2008/1002701	L070426	S	0.990	28			0.99	0.265
Wheat	Forage	2008/1002701	L070429	S	0.150	27	0.99	0.175		
Wheat	Forage	2014/1028112	L120458	S	0.110	28				
Wheat	Forage	2014/1028112	L120459	S	0.200	28				
Wheat	Straw	AL-730-001	S/FR/E83/878	N	0.17	46				
Wheat	Straw	AL-730-003	S/FR/E84/890	N	0.07	41				
Wheat	Straw	AL-730-003	S/FR/E84/891	N	0.29	20	0.82	0.325	1.040	0.33
Wheat	Straw	AL-730-003	S/FR/E84/892	N	0.19	28				
Wheat	Straw	AL-730-003	S/FR/E84/893	N	0.12	27				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Wheat	Straw	AL-730-031	BE61	N	0.03	34				
Wheat	Straw	2014/1028112	L120452	N	0.530	28				
Wheat	Straw	2014/1028112	L120453	N	0.290	29				
Wheat	Straw	2014/1028112	L120454	N	0.181	65				
Wheat	Straw	2014/1028112	L120455	N	0.400	28				
Wheat	Straw	2013/1416282	S13-00445-01 / L130034	N	0.360	27				
Wheat	Straw	2013/1416282	S13-00445-02 / L130035	N	0.400	28				
Wheat	Straw	2008/1002701	L070422	N	0.360	35				
Wheat	Straw	2008/1002701	L070423	N	0.730	36				
Wheat	Straw	2008/1002701	L070424	N	0.510	27				
Wheat	Straw	2008/1002701	L070425	N	0.820	42				
Wheat	Straw	AL-730-001	S/FR/E83/220	S	0.44	38				
Wheat	Straw	AL-730-001	S/FR/E83/419	S	0.29	43				
Wheat	Straw	AL-730-003	S/FE/E84/217	S	0.12	42				
Wheat	Straw	AL-730-003	S/FR/E84/218	S	0.13	62				
Wheat	Straw	AL-730-003	S/FR/E84/347	S	0.16	28				
Wheat	Straw	AL-730-003	S/FR/E84/348	S	0.06	42				
Wheat	Straw	AL-730-029	W/FR/E/92/239	S	0.05	48				
Wheat	Straw	AL-730-030	W/FR/E/92/209	S	0.09	50				
Wheat	Straw	AL-730-030	W/FR/E/92/869	S	0.75	38				
Wheat	Straw	2013/1416282	S13-00445-03 / L130036	S	0.370	27	1.040	0.33		
Wheat	Straw	2013/1416282	S13-00445-04 / L130037	S	0.470	28				
Wheat	Straw	2008/1002701	L070426	S	0.800	35				
Wheat	Straw	2008/1002701	L070427	S	0.820	42				
Wheat	Straw	2008/1002701	L070428	S	1.040	28				
Wheat	Straw	2008/1002701	L070429	S	0.200	36				
Wheat	Straw	2014/1028112	L120456	S	0.540	28				
Wheat	Straw	2014/1028112	L120457	S	0.330	28				
Wheat	Straw	2014/1028112	L120458	S	0.180	36				
Wheat	Straw	2014/1028112	L120459	S	0.350	28				
Sugar beet	Root	2003/1021716	AF/6792/BA/1	N	<0.010	92				
Sugar beet	Root	2003/1021716	AF/6792/BA/2	N	<0.010	92				
Sugar beet	Root	2003/1021716	AF/6792/BA/3	N	<0.010	88				
Sugar beet	Root	2003/1021716	AF/6792/BA/4	N	<0.010	84				
Sugar beet	Root	2004/1006468	ACK/02/03	N	<0.050	25	0.228	0.010	0.228	0.010
Sugar beet	Root	2004/1006468	AGR/02/03	N	<0.050	14				
Sugar beet	Root	2004/1006468	DU4/02/03	N	<0.050	30				
Sugar beet	Root	2004/1006468	DU2/02/03	N	<0.228	21				

Crop	Commodity	DocID	Trial No.	EU Region	Residues BAS 310 I [mg/kg]	PHI [days]	Per EU Region		Overall	
							HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]
Sugar beet	Root	2006/1026851	AF/8832/BA/1	S	<0.010	7	0.01	0.01		
Sugar beet	Root	2006/1026851	AF/8832/BA/2	S	<0.010	7				
Sugar beet	Root	2006/1026851	AF/8832/BA/3	S	<0.010	7				
Sugar beet	Root	2006/1026851	AF/8832/BA/4	S	<0.010	7				
Sugar beet	Root	2007/1007941	AF/10501/BA/1	S	<0.010	7				
Sugar beet	Root	2007/1007941	AF/10501/BA/2	S	<0.010	7				
Sugar beet	Root	2007/1007941	AF/10501/BA/3	S	<0.010	7				
Sugar beet	Root	2007/1007941	AF/10501/BA/4	S	<0.010	7				
Sugar beet	Tops	2003/1021716	AF/6792/BA/1	N	<0.010	92	0.292	0.030	0.292	0.073
Sugar beet	Tops	2003/1021716	AF/6792/BA/2	N	<0.010	92				
Sugar beet	Tops	2003/1021716	AF/6792/BA/3	N	<0.010	88				
Sugar beet	Tops	2003/1021716	AF/6792/BA/4	N	<0.010	84				
Sugar beet	Tops	2004/1006468	ACK/02/03	N	<0.050	25				
Sugar beet	Tops	2004/1006468	AGR/02/03	N	0.07	14				
Sugar beet	Tops	2004/1006468	DU4/02/03	N	<0.050	30				
Sugar beet	Tops	2004/1006468	DU2/02/03	N	0.292	21				
Sugar beet	Tops	2006/1026851	AF/8832/BA/3	S	0.048	14	0.236	0.128		
Sugar beet	Tops	2007/1007941	AF/10501/BA/4	S	0.075	7				
Sugar beet	Tops	2006/1026851	AF/8832/BA/2	S	0.079	7				
Sugar beet	Tops	2007/1007941	AF/10501/BA/1	S	0.103	7				
Sugar beet	Tops	2006/1026851	AF/8832/BA/4	S	0.152	7				
Sugar beet	Tops	2007/1007941	AF/10501/BA/2	S	0.173	7				
Sugar beet	Tops	2006/1026851	AF/8832/BA/1	S	0.227	7				
Sugar beet	Tops	2007/1007941	AF/10501/BA/3	S	0.236	7				

BASF
The Chemical Company

Alpha-Cypermethrin

Document M-CA, Section 6

RESIDUES IN OR ON TREATED PRODUCTS, FOOD AND FEED AND PLANT METABOLISM

Compiled by:

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
10/Jul/2017		BASF DocID 2017/1134415

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 6 RESIDUES IN OR ON TREATED PRODUCTS, FOOD AND FEED AND PLANT METABOLISM

An overview of metabolites identified during consumer safety studies is given below.

Table 6-1: Notations of parent and metabolites of alpha-cypermethrin

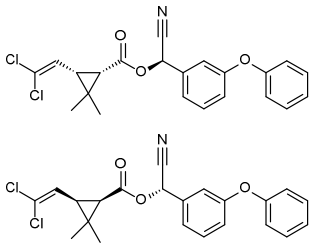
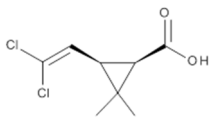
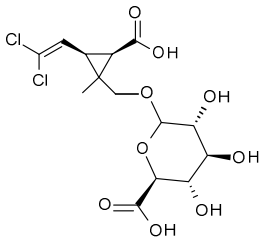
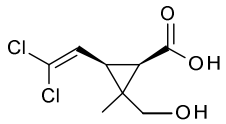
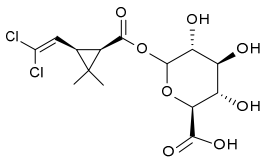
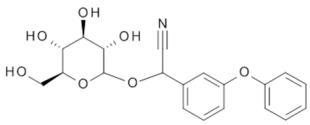
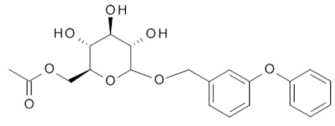
Substance/ Metabolite Code	Reg. No	Synonyms	CAS-No	Compound found in	Structure/Name
BAS 310 I Alpha-cypermethrin	4078193	WL 85871 CL 900049 FMC 63318 FMC 39391	67375-30-8	Plant (wheat, cabbage, lettuce) Livestock (goat, hen) Rat	
M310I001	4080830	DCVA DCVC acid Cis-DVA CL 912554 CL 194198 (cis)	59042-49-8	Rat Livestock (goat, hen) Crop (wheat)	
M310I002					
M310I003				Rat Livestock (goat, hen)	
M310I004				Rat Livestock (goat)	
M310I005				Crop (lettuce)	
M310I006				Crop (lettuce)	

Table 6-1: Notations of parent and metabolites of alpha-cypermethrin

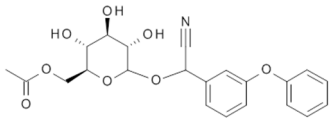
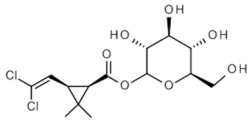
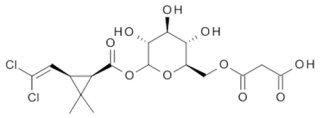
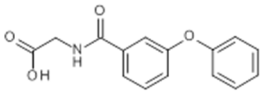
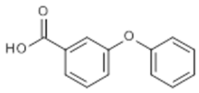
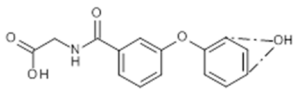
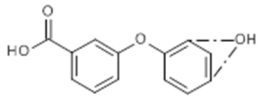
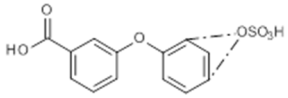
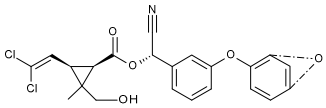
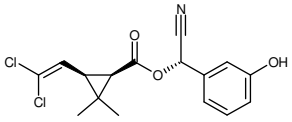
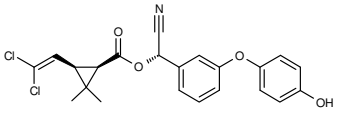
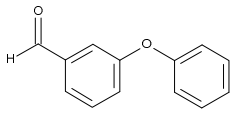
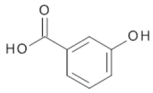
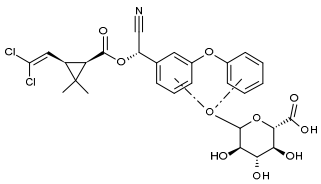
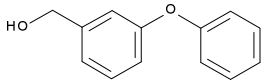
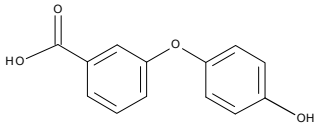
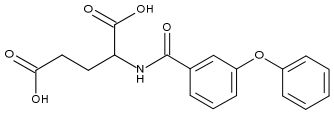
Substance/ Metabolite Code	Reg. No	Synonyms	CAS-No	Compound found in	Structure/Name
M310I007				Crop (lettuce)	
M310I008				Crop (lettuce)	
M310I009				Crop (lettuce)	
M310I010 N-(3-phenoxybenzoyl)glycine	4108084	3PBA glycine WL 46194 CL 117585		Rat Livestock (goat) Crop (wheat)	
M310I011 3-phenoxybenzoic acid	130213	PBA m-PB acid 3-PB acid WL 44607 CL 206128	3739-38-6	Rat Livestock (goat, hen) Crop (wheat)	
M310I012				Rat	
M310I013		WL 46114 CL 213336		Rat Crop (wheat, cabbage)	
M310I014				Rat	
M310I015				Rat Livestock (goat)	
M310I016	6004475			Rat	
M310I017 4'hydroxy-alpha-cypermethrin	6002320	CL 194198 (cis-form CL1010235)		Rat Livestock (goat) Crop (wheat, cabbage)	

Table 6-1: Notations of parent and metabolites of alpha-cypermethrin

Substance/ Metabolite Code	Reg. No	Synonyms	CAS-No	Compound found in	Structure/Name
M310I018 3-phenoxy-benzaldehyd	4080665	WL 42049 CL 206969 3-PBAD 3-PBAD III m-PBAD CPBT	39515-51-0	Crop (wheat)	
M310I019 3-hydroxy-benzoic acid			CAS-No. 99-06-9	Livestock (hen)	
M310I021				Livestock (goat)	
M310I024 3-phenoxy-benzyl-alcohol	207323	PBAlc WL 40673 CL 206138		Crop (wheat, cabbage)	
M310I025	4110493	4-hydroxy-3-phenoxy-benzaldehyd WL46114		Crop (wheat, cabbage)	
M310I026 3-phenoxy-benzoyl glutamic acid	4110960			Crop (wheat)	

CA 6.1 Storage stability of residues

In the framework of the original inclusion into Annex I according to Directive 91/414/EEC, storage stability in crop commodities of high water (lettuce), starch (cereal grain) and oil (oilseed rape) content have been evaluated, as well as data on cattle tissues and milk.

Additional storage stability data in crop commodities of high water (tomato), acid (pineapple), starch (barley grain), protein (bean seed), and oil content (oilseed rape) are available in order to support the representative uses cucumber/courgette, leafy brassica, lettuce, barley, wheat and oil seed rape. These new studies will be summarized in the following.
Storage stability for animal matrices is not provided as no new feeding studies have been conducted.

Storage stability data supporting the terrestrial field dissipation are provided and are addressed under paragraph 7.1.2.2.1 in the application. Storage stability for water as matrix is not required.

An overview on the storage stability studies is presented in Table 6.1-1.

Table 6.1-1: Overview on storage stability studies

Matrix	Study code	DocID	EU reviewed
Soybean, wheat, apple, soil	SBGR.85.044	AL-326-006	Yes
Cereals	CFS 1998-097	AL-730-066 1998/7001651	Yes
Lettuce	CFS 1998-026	AL-726-015 1998/7001641	No[1]
Oilseed rape	4807	AL-750-040	Yes
Oilseed rape	4832 (interim) 00602F	AL-730-068 ~~2003/1009950~~ 2003/1009948	Yes No
Cattle tissues and milk	RES 01-002	AL-326-035	Yes
Tomatoes	CFS 1998-098 DER98	AL-723-029 1998/7001639	No
Oilseed rape	00602F	2003/1009948	No
Oilseed rape	98402F	2003/1009950	No
Pineapple (fruit), barley (grain), bean (seed)	331420	2014/3006687	No
Soil[2]	699256	2014/1152598	No

1 study was submitted in the context of the peer review under Directive 91/414/EEC, but it was not assessed
2 mentioned for completeness; will be addressed under 7.1.2.2.1

During the peer review under Directive 91/414/EEC, a storage stability study in lettuce (AL-726-015) was submitted. This study was not assessed in the 3rd Addendum to the Monograph: Addendum Chapter B-7 (Residue data), January 2003). This study was not included in the application as it had been submitted already with the alpha-cypermethrin addendum dossier. For the sake of completeness, it is summarized again below under point 6.1/1. All subsequent study summaries appear in the same sequence as indicated in the application, starting with 6.1/2.

Report:	CA 6.1/1 Mueller U., 1998a Alphacypermethrin (CL 900049): Storage stability of CL 900049 residues at < -18 C in lettuce (Germany, 1997) AL-726-015
Guidelines:	BBA IV 3-3, IVA Guideline Residue Chemistry Part II Storage Stability (1990), EEC 91/414 Annex II (Part A Section 6.2), EEC 91/414 Annex III (Part A Section 8.1)
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

In 1997 a study was initiated in order to investigate the stability of alpha-cypermethrin (CL 900049) residues in lettuce during storage at ≤ -18° C. This storage stability study was conducted using sample material with incurred residues of alpha-cypermethrin from a 1996 residue study. Aliquots of this sample material were analyzed for residues of alpha-cypermethrin after 0, 3, 6, 9 and 12 months storage at ≤ -18°C according to residue method FAMS 045-02 by means of capillary gas chromatography with nitrogen/phosphorus detection (GC/NPD).
No residues of alpha-cypermethrin were detected in any of the control samples, which had been stored at ≤ -18°C. Mean residue values obtained after 0, 3, 6, 9 and 12 month storage at ≤ -18°C from treated lettuce sample material with incurred alpha-cypermethrin residues ranged from 0.13 to 0.14 mg/kg. Concurrent recoveries for alpha-cypermethrin were performed at a level of 0.10 mg/kg and ranged from 73 to 98 %.
These results show that alpha-cypermethrin residues are stable in lettuce samples for at least 12 months, when residue samples are stored at ≤ -18°C.

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:** Alpha-cypermethrin CF 05898
Description: CF 05898 (EC)
Lot/Batch #: B011: 100 g/L
Purity: not reported
CAS#: 67375-30-8 alpha-cypermethrin
Development code: not reported
Spiking levels: 0.1 mg/kg

2. **Test Commodity:**
Crop: Lettuce
Type: Leafy vegetables
Variety: not reported
Botanical name: *Lactuca sativa*
Crop parts(s) or processed commodity: Leaves
Sample size: 15 g (aliquots)

B. STUDY DESIGN AND METHODS

1. Test procedure

An untreated and a treated sample from a lettuce residue study were used as sample material. The deep frozen lettuce samples were ground using a mincing machine.
At the start of the study (0 month storage), five aliquots of the treated sample material were analyzed together with one aliquot of the untreated sample for residues of alpha-cypermethrin in order to prove the homogeneity of the residue sample according to residue method FAMS 045-02 by means of capillary gas chromatography with nitrogen/phosphorus detection (GC/NPD). The limit of quantitation of the method (LOQ) was 0.01 mg/kg.
After 3, 6, 9 and 12 months storage at ≤ - 18°C, three aliquots of the treated sample together with one aliquot of the untreated sample were analyzed for residues of alpha-cypermethrin.
With each set of samples, two concurrent recoveries for alpha-cypermethrin were conducted in the range of the expected residue level (0.10 mg/kg). These concurrent recoveries were performed using the same untreated lettuce sample.

2. Description of analytical procedures

The method used for the analysis was FAMS 045-02 (Cyanamid Forschung GmbH) of March 31, 1995. A 15 g aliquot of sample material was extracted with 120 mL of hexane/acetone (8/2 v/v) using an Ultra-Turrax. The mixture was then centrifuged for 5 min at 3800 rpm (=2900 g). The extract solution was partitioned three times with 100 mL of water. The combined organic phases were reduced in volume almost to dryness by means of a rotary evaporator and the residue redissolved in 7.5 mL of acidic methanol. Further cleanup was carried out by gel permeation chromatography (GPC) by injecting an aliquot of this solution into the GPC system. The residues of alpha-cypermethrin were determined by capillary gas chromatography with nitrogen/phosphorus detection (GC/NPD). The limit of quantitation (LOQ) of the method was 0.01 mg/kg.

Method FAMS 045-02 is basically the same as method FAMS 066-01 which was employed in the storage stability study on alpha-cypermethrin in cereals AL-730-066, reviewed in the framework of the original inclusion into Annex I according to Directive 91/414/EEC. It only describes a solid phase extraction step as an alternative to gel permeation clean-up. The final extract is redissolved in cyclohexanone in method FAMS 045-02 instead of ethyl acetate as in method FAMS 066-01.
In the lettuce storage stability study described here, gel permeation clean-up was applied instead of a solid-phase extraction. Thus, the work-up procedure used was essentially the same as in the peer-reviewed cereal storage stability study AL-730-066. The gas chromatographic conditions applied in both studies were identical with the only exception of the purge valve off-on time (1.3 min for lettuce instead of 1.0 min for cereals).

Table 6.1-2: Summary of concurrent recoveries in lettuce

Matrix	Fortification Level (mg/kg)	Summary Recoveries				
		Recoveries (%)	n	Mean (%)	SD (+/-)	RSD (%)
Method FAMS 045-02		alpha-cypermethrin				
Lettuce	0.1	93, 96, 98, 96, 95, 91, 97, 95, 74, 73	10	91	9.3	10.3

II. RESULTS AND DISCUSSION

A summary of the frozen storage stability of alpha-cypermethrin residues in lettuce is given in Table 6.1-3.

No residues of alpha-cypermethrin were detected in any of the aliquots of the control sample, which had been stored at ≤ -18°C. Mean residue values obtained from the lettuce sample with incurred alpha-cypermetrin residues after 0, 3, 6, 9 and 12 month storage ranged from 0.14 to 0.13 mg/kg. Mean residue values, which were corrected for the average of concurrent recoveries obtained from each measuring date, ranged from 0.14 to 0.18 mg/kg.
Concurrent recoveries for alpha-cypermetrin were performed at a level of 0.10 mg/kg and ranged from 73 to 98 %. The average of all concurrent recoveries obtained during the study was 91 % (RSD = 10.3 %).
These results show that alpha-cypermethrin residues are stable in lettuce samples for at least 12 months, when residue samples are stored at ≤ -18°C.

Table 6.1-3: Summary of the storage stability of alpha-cypermethrin in lettuce

Storage period (months)	Mean recovery (%)		
	BAS 310 I found (mg/kg)	Concurrent BAS 310 I recovery (%)	BAS 310 I in stored samples, corrected for concurrent recovery (mg/kg)
0	0.14	95	0.15
3	0.14	97	0.14
6	0.14	93	0.15
9	0.14	96	0.15
12	0.13	74	0.18

III. CONCLUSION

The results of the storage stability study in lettuce show that alpha-cypermethrin residues are stable in lettuce samples for at least 12 months, when residue samples are stored at < -18°C.

Report:	CA 6.1/2 Mueller U., 1998b Alphacypermethrin (CL 900049): Storage stability of CL 900049 residues at < -18 degree C in tomatoes (Germany 1997) AL-723-029
Guidelines:	BBA IV 3-3, IVA Guideline Residue Chemistry Part II Storage Stability 1992, EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III 8.1
GLP:	yes (certified by Landesanstalt fuer Pflanzenbau und Pflanzenschutz Rheinland-Pfalz, Mainz)

Executive Summary

A freezer storage stability study was performed in order to investigate the stability of alpha-cypermethrin (BAS 310 I; former code: CL 900049) residues in tomatoes during storage at ≤ -18° C. In the course of the study 15 g aliquots of commercially purchased sample material were fortified with alpha-cypermethrin at a level of 0.10 mg/kg and stored in the deep freezer room. Immediately after the fortification, as well as after 3, 6, 9, and 12 months storage, fortified samples were analyzed together with one control sample for residues of alpha-cypermethrin according to residue method FAMS 045-02 by means of capillary gas chromatography with nitrogen/phosphorus detection (GC/NPD).

No residues of alpha-cypermethrin were detected in any of the control samples, which had been stored at ≤ -18° C. The mean residue value of alpha-cypermethrin obtained after 0 month (initial value) and 12 months storage (end value) at ≤ -18°C was 0.09 mg/kg, each. The mean residue values for alpha-cypermethrin obtained after 0, 3, 6, 9 and 12 months ranged from 0.08 to 0.10 mg/kg. Mean residue values from fortified tomato samples, which were corrected for the average of concurrent recoveries obtained from each set of samples, ranged from 0.09 to 0.10 mg/kg. Concurrent recoveries for alpha-cypermethrin were performed at a level of 0.10 mg/kg and ranged from 88 to 100 %.

These results show that alpha-cypermethrin residues in tomatoes are stable in frozen storage (≤ -18°C) for at least one year.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin (BAS 310 I; former code: CL 900049)
Description:
Lot/Batch #: AC 9575-006
Purity: 99.8%
CAS#: 67375-30-8 alpha-cypermethrin
Development code: not reported
Spiking levels: 0.10 mg/kg

2. Test Commodity:
Crop: Tomato
Type: Fruiting vegetables
Variety: not reported, commercially available sample material
Botanical name: *Lycopersicon esculentum*
Crop parts(s) or processed commodity: Fruit
Sample size: 15 g (aliquots)

B. STUDY DESIGN AND METHODS

1. Test procedure

In 1997 a study was initiated in order to investigate the stability of alpha-cypermethrin (BAS 310 I) residues in tomatoes during storage at ≤ - 18° C. In the course of the study 15 g aliquots of commercially purchased sample material were fortified with alpha-cypermethrin (BAS 310 I) at a level of 0.10 mg/kg and stored in the deep freezer room. Immediately after the fortification, as well as after 3, 6, 9, and 12 months storage, three fortified and one control sample, which was stored under the same conditions, were analyzed for residues of alpha-cypermethrin according to residue method FAMS 045-02 by means of capillary gas chromatography with nitrogen/phosphorus detection (GC/NPD). With each set of samples, two concurrent recoveries for alpha-cypermethrin were conducted in the range of the expected residue level (0.10 mg/kg).

2. Description of analytical procedures

The method used for the analysis was FAMS 045-02 (Cyanamid Forschung GmbH) of March 31, 1995. A 15 g aliquot of sample material was extracted with 120 mL of hexane/acetone (8/2 v/v) using an Ultra-Turrax. The mixture was then centrifuged for 5 min at 3800 rpm (=2900 g). The extract solution was partitioned three times with 100 mL of water. The combined organic phases were reduced in volume almost to dryness by means of a rotary evaporator and the residue redissolved in 7.5 mL of acidic methanol. Further cleanup was carried out by gel permeation chromatography (GPC) according to the method by injecting an aliquot of this solution into the GPC system. The residues of alpha-cypermethrin were determined by capillary gas chromatography with nitrogen/phosphorus detection (GC/NPD). The limit of quantitation (LOQ) of the method was 0.01 mg/kg.

Method FAMS 045-02 is basically the same as method FAMS 066-01 which was employed in the storage stability study on alpha-cypermethrin in cereals AL-730-066, reviewed in the framework of the original inclusion into Annex I according to Directive 91/414/EEC. It only describes a solid phase extraction step as an alternative to gel permeation clean-up. The final extract is redissolved in cyclohexanone in method FAMS 045-02 instead of ethyl acetate as in method FAMS 066-01.

In the tomato storage stability study described here, gel permeation clean-up was applied instead of a solid-phase extraction. Thus, the work-up procedure used was essentially the same as in the peer-reviewed cereal storage stability study AL-730-066. The gas chromatographic conditions applied in both studies were identical.

Method recoveries were analyzed with each set of stored samples. The results are summarized in Table 6.1-4.

Table 6.1-4: Summary of concurrent recoveries in tomato fruit

Matrix	Fortification Level (mg/kg)	Summary Recoveries				
		Recoveries (%)	n	Mean (%)	SD (+/-)	RSD (%)
Method FAMS 045-02		alpha-cypermethrin				
Tomato fruit	0.10	97, 94, 91, 91, 92, 93, 99, 100, 95, 88	10	94	3.8	4.0

II. RESULTS AND DISCUSSION

A summary of the frozen storage stability of alpha-cypermethrin residues in tomatoes is given in Table 6.1-5.

No residues of alpha-cypermethrin were detected in any of the control samples, which had been stored at ≤ -18° C.
The mean residue value of alpha-cypermethrin obtained after 0 months (initial value) and 12 months storage (end value) at ≤ -18°C was 0.09 mg/kg, each. The mean residue values for alpha-cypermethrin obtained after 0, 3, 6, 9 and 12 months ranged from 0.08 to 0.10 mg/kg. Mean residue values from fortified tomato samples, which were corrected for the average of concurrent recoveries obtained from each set of samples, ranged from 0.09 to 0.10 mg/kg. Concurrent recoveries for alpha-cypermethrin were performed at a level of 0.10 mg/kg and ranged from 88 to 100%. The average of all concurrent recoveries obtained during the study was 94 % (RSD = 4.0 %).

Table 6.1-5: Summary of the storage stability of alpha-cypermethrin in tomatoes

Storage period (months)	Mean recovery (%)		
	BAS 310 I found (mg/kg)	Concurrent BAS 310 I recovery (%)	BAS 310 I in stored samples, corrected for concurrent recovery (mg/kg))
0	0.09	96	0.09
3	0.08	91	0.09
6	0.09	93	0.10
9	0.10	100	0.10
12	0.09	92	0.10

III. CONCLUSION

These results show that alpha-cypermethrin residues in tomatoes are stable in frozen storage (≤ -18°C) for at least one year.

Report: CA 6.1/3
Dale T., 2003a
Alphacypermethrin - Freezer storage stability in oil seed rape (whole plant, whole pod and seed)
2003/1009948
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Executive Summary

A freezer storage stability study was performed investigating the stability of alpha-cypermethrin in oilseed rape whole plant, whole pod and seed samples. The study was performed over a period of 12 months.
Specimens of oil seed rape whole plant, whole pod and seed which were fortified with BAS 310 I (alpha-cypermethrin) at 0.5 mg/kg, were analysed in duplicate after approximately 1.5, 3, 6 and 12 months. No initial data was obtained the first specimens were assayed after a nominal 1.5 months.
When corrected for concurrently performed recoveries none of the individual results obtained at the nominal 12 month timepoint (52 weeks) falls below 70.9%.
The results indicate that alpha-cypermethrin residues are stable in oilseed rape whole plant, whole pods and seeds under frozen conditions for at least 52-53 weeks.
An interim report was issued detailing the results up to and including the nominal 3 month timepoint. This study has already been peer-reviewed in the framework of the original inclusion into Annex I according to Directive 91/414/EEC (BASF RDI-No. AL-730-068; DocID 2002/1011586).
This final report includes all the data from the completed study.

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:** Alpha-Cypermethrin (BAS 310 I, former code: AC 900049)
Description:
Lot/Batch #: AC 12395-18
Purity: 95.1%
CAS#: 67375-30-8 alpha-cypermethrin
Development code: not reported
Spiking levels: 0.5 mg/kg

2. **Test Commodity:**
Crop: Oilseed rape
Type: Oilseeds
Variety: not reported
Botanical name: *Brassica napus*
Crop parts(s) or processed commodity: whole plant, whole pods, seeds
Sample size: 10 g (aliquots)

B. STUDY DESIGN AND METHODS

1. Test procedure

Aliquots of 10 g of each specimen matrix were weighed into screw top glass bottles. For every assay timepoint a set of 5 bottles were filled with specimen material. Two of these were fortified with BAS 310 I to give a level of 0.5 mg/kg. One was used as control and the other two used for concurrent procedural recoveries. Spare aliquots were also prepared in case of analytical problems or breakages. One group of specimens for each matrix were to be used to show that the control specimen was free of any contamination. This group consisted of two controls to assay.
The glass containers were tightly closed and transferred to a freezer. No initial specimens were taken for assay because the method could not be established within the analytical laboratory at this time. For the same reason no control group assays were done prior to fortification. The specimens were kept at a temperature of approximately minus 20°C in the dark over the entire period of the study until needed for an assay timepoint.
Specimen assay timepoints were at about 1.5, 3, 6 and 12 months.

An interim report provided results from the first two sampling intervals (6-weeks and 12 weeks) and was already evaluated (BASF RDI-No. AL-730-068; DocID 2002/1011586).

2. Description of analytical procedures

The samples were analyzed using the GC/NPD method, BASF Agro Research method RLA 12594V. The limit of quantification of this method was 0.05 mg/kg. This method was validated in study AL-244-007 (2000/7000960) which had been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414.

Method recoveries were analyzed with each set of stored samples. The results are summarized in Table 6.1-6.

Table 6.1-6: Summary of concurrent recoveries in oilseed rape matrices

Matrix	Fortification level (mg/kg)	Summary recoveries			
		n	Mean (%)	SD (%)	RSD (%)
BASF method No. RLA 12594V		**BAS 310 I**			
Whole plant	0.5	8	88.6	6.4	7.2
Whole pods	0.5	8	89.6	5.3	6.0
Seeds	0.5	8	97.1	5.0	5.2
Overall	0.5	24	91.8	6.6	7.2

II. RESULTS AND DISCUSSION

A summary of the frozen storage stability of alpha-cypermethrin residues in whole OSR plants, OSR pods, and OSR seeds is given in Table 6.1-7 - Table 6.1-9. In order to account for possible variations over the time investigated, the procedural recovery results are given in addition.The stability results are expressed as average percentage of the nominal fortification and are reported both uncorrected and as relative recovery after correction for procedural recovery. The stored recoveries were the actual percent of the residues recovered in the stored samples based on the amount of alpha-cypermethrin added to control samples. These recoveries give insight into the validity of the uncorrected residues reported in RAC and processing studies when stored under similar conditions. Corrected recoveries in stored samples equate to actual loss of residues during storage. Corrections were made for the experimental variability observed in the method spikes that were analyzed with the stored samples. Corrected recovery values above 70% were considered to indicate that the residues were stable.
Both the stored and corrected recoveries in the tested seed samples were above 70% when stored under frozen conditions for 12 months. In whole plant, a stored recovery value of 53.2% (corrected 60.5%) was found in one individual sample after 13 weeks, while the duplicate sample stored for the same time and analyzed at the same time point had a stored recovery of 72.2% (corrected 82.0%). Moreover, all whole plant samples analysed at later time points showed stored recoveries between 79.2-86.0% (corrected 87.3-99.3%); after 53 weeks, the mean stored uncorrected recovery lay at 85.15%. The isolated finding at 13 weeks storage should therefore be regarded as an outlier and not indicative of residue degradation during deep-frozen storage of rape seed whole plant specimen.

In oilseed rape whole pods, a trend to lower stored recoveries was observed beginning after 25 weeks of storage (25 weeks: 61.4/69.1%, corrected 73.1/82.3%; 53 weeks: 62.7/66.2%, corrected 70.9/74.8%). However, when corrected for concurrently performed recoveries none of the individual results obtained at the nominal 12 month timepoint falls below 70.9%.

In seeds, alpha-cypermethrin was recovered at more than 70 % during the whole time course of the study.
These results indicate that alpha-cypermethrin residues are stable in oilseed rape whole plant, whole pods and seeds under frozen conditions for at least 52-53 weeks.

Table 6.1-7: Summary of the storage stability of alpha-cypermethrin in oilseed rape whole plant

Weeks of storage	BAS 310 I		
	Procedural recovery (%)[1]	Stored recovery mg/kg (%)[2]	Relative recovery mg/kg (%)[3]
7 weeks	90 90	0.3926 (78.5)	0.4362 (87.2)
	(90.0)	0.4298 (86.0)	0.4776 (95.5)
13 weeks	86 90	0.2660 (53.2)	0.3023 (60.5)
	(88.0)	0.3610 (72.2)	0.4102 (82.0)
25 weeks	79 81	0.3970 (79.4)	0.4963 (99.3)
	(80.0)	0.3958 (79.2)	0.4948 (99.0)
53 weeks	97 96	0.4300 (86.0)	0.4456 (89.1)
	(96.5)	0.4214 (84.3)	0.4367 (87.3)

1 concurrent procedural recoveries were performed in duplicate at each assay timepoint. The average procedural recovery value is shown **in bold in brackets**.
2 uncorrected
3 relative recovery= stored recovery/average procedural recovery x 100

Table 6.1-8: Summary of the storage stability of alpha-cypermethrin in oilseed rape whole pods

Weeks of storage	BAS 310 I		
	Procedural recovery (%)[1]	Stored recovery mg/ kg (%)[2]	Relative recovery mg/kg (%)[3]
8 weeks	96 99	0.3958 (79.2)	0.4060 (81.2)
	(97.5)	0.4342 (86.8)	0.4453 (89.1)
13 weeks	88 89	0.3920 (78.4)	0.4429 (88.6)
	(88.5)	0.4272 (85.4)	0.4827 (96.5)
25 weeks	85 83	0.3456 (69.1)	0.4114 (82.3)
	(84.0)	0.3072 (61.4)	0.3657 (73.1)
53 weeks	89 88	0.3308 (66.2)	0.3738 (74.8)
	(88.5)	0.3136 (62.7)	0.3544 (70.9)

1 concurrent procedural recoveries were performed in duplicate at each assay timepoint. The average procedural recovery value is shown **in bold in brackets**.
2 uncorrected
3 relative recovery= stored recovery/average procedural recovery x 100

Table 6.1-9: Summary of the storage stability of alpha-cypermethrin in oilseed rape seeds

Weeks of storage	BAS 310 I		
	Procedural recovery (%)[1]	**Stored recovery mg/kg (%)[2]**	**Relative recovery mg/kg (%)[3]**
6 weeks	91 89	0.4334 (86.7)	0.4816 (96.3)
	(90.0)	0.4526 (90.5)	0.5029 (100.6)
12 weeks	99 105	0.5000 (100.0)	0.4902 (98.0)
	(102.0)	0.4282 (85.6)	0.4198 (84.0)
24 weeks	99 97	0.3940 (78.8)	0.4020 (80.4)
	(98.0)	0.3616 (72.3)	0.3690 (73.8)
52 weeks	99 98	0.3680 (73.6)	0.3736 (74.7)
	(98.5)	0.3722 (74.4)	0.3779 (75.6)

1 concurrent procedural recoveries were performed in duplicate at each assay timepoint. The average procedural recovery value is shown in bold in brackets.
2 uncorrected
3 relative recovery= stored recovery/average procedural recovery x 100

III. CONCLUSION

The results indicate that alpha-cypermethrin residues are stable in OSR whole plant, whole pods and seeds under frozen conditions for at least 52-53 weeks.

Report: CA 6.1/4
Dale T., 2003b
Alphacypermethrin - Freezer storage stability in oil seed rape (whole plant, whole pod and seed)
2003/1009950
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Executive Summary

A freezer storage stability study was performed investigating the storage stability of alpha-cypermethrin (BAS 310 I) in oilseed rape whole plant, whole pod and seed samples spiked at 0.5 mg/kg. The samples were analysed in duplicate after approximately 0, 1, 3, 6 and 11 months. When corrected for concurrently performed recoveries none of the individual results obtained at the nominal 11 month timepoint (48 weeks) falls below 70.7%.
The results indicate that alpha-cypermethrin residues are stable in oilseed rape whole plant, whole pods and seeds under frozen conditions for at least 48 weeks.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-cypermethrin (BAS 310 I, former code: AC 900049)
Description:
Lot/Batch #: AC 12395-18
Purity: 95.1%
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.5 mg/kg

2. Test Commodity:
Crop: Oilseed rape
Type: Oilseed
Variety: not reported
Botanical name: *Brassica napus*
Crop parts(s) or processed commodity: whole plant, whole pods, seeds
Sample size: 10 g (aliquots)

B. STUDY DESIGN AND METHODS

1. Test procedure

The freezer storage stability of alpha-cypermethrin residues over a period of 11 months was investigated using untreated control whole oil seed rape (OSR) plant, OSR pod, and OSR seed specimens.
Aliquots of 10 g of each specimen matrix were weighed into screw top glass bottles. For every assay timepoint a set of 5 bottles were filled with specimen material. Two of these were fortified with BAS 310 I to give a level of 0.5 mg/kg. One was used as control and the other two used for concurrent procedural recoveries. Spare aliquots were also prepared in case of analytical problems or breakages.
One group of specimens for each matrix were to be used to show that the control specimen was free of any contamination. This group consisted of two controls to assay and another two to act as concurrent procedural recoveries.
The glass containers were tightly closed and transferred to a freezer. Specimens analysed at the 0 timepoint were not taken to the freezer but were analysed immediately.
The specimens were kept at a temperature of approximately minus 20°C in the dark over the entire period of the study until needed for an assay timepoint. .
Specimen assay timepoints were at about 0, 1, 3, 6 and 11 months.

2. Description of analytical procedures

The samples were analyzed using the BASF Agro Research method RLA 12594V. The limit of quantitation of this method was 0.05 mg/kg.

Method recoveries were analyzed with each set of stored samples. The results are summarized in Table 6.1-10.

Table 6.1-10: Summary of concurrent recoveries in oilseed rape matrices

Matrix	Fortification level (mg/kg)	Summary recoveries			
		n	Mean (%)	SD (%)	RSD (%)
BASF method No. RLA 12594V		**BAS 310 I**			
Whole plant	0.5	10	92.7	4.0	4.3
Whole pods	0.5	12	88.5	3.6	4.0
Seeds	0.5	12	95.7	6.9	7.2
Overall	0.5	34	92.3	5.8	6.3

II. RESULTS AND DISCUSSION

The results of the analysis of the control samples used for spiking revealed no residues above the limit of quantitation for rape whole pods and seeds. No results for controls are available for whole plant specimens because of analysis failure. The results are shown in Table 6.1-11.

A summary of the frozen storage stability of alpha-cypermethrin residues in whole oilseed rape plants, pods, and seeds is given in Table 6.1-12–Table 6.1-14. In order to account for possible variations over the time investigated, the procedural recovery results are given in addition.The stability results are expressed as average percentage of the nominal fortification and are reported both uncorrected and as relative recovery after correction for procedural recovery. The stored recoveries were the actual percent of the residues recovered in the stored samples based on the amount of alpha-cypermethrin added to control samples. These recoveries give insight into the validity of the uncorrected residues reported in RAC and processing studies when stored under similar conditions. Corrected recoveries in stored samples equate to actual loss of residues during storage. Corrections were made for the experimental variability observed in the method spikes that were analyzed with the stored samples. Corrected recovery values above 70% were considered to indicate that the residues were stable.
Both the stored and corrected recoveries in the tested oilseed rape whole plant and seed samples were above 70% when stored under frozen conditions for approximately 49 and 48 weeks, respectively.
In oilseed rape whole pods, stored and corrected recoveries were higher than 70% up to and including a storage interval of 26 weeks. At the final time point of 48 weeks, the stored recoveries were 64.0/68.0%; however, the corrected recoveries still exceeded 70% (70.7/75.2%).

These results indicate that alpha-cypermethrin residues are stable in oilseed rape whole plant, whole pods and seeds under frozen conditions for at least 48 weeks.

Table 6.1-11: Analytical results for control matrices used

Matrix	BAS 310 I			
	Results of control (mg/kg)		Procedural recovery (%)	
Whole plant*	N/A	N/A	N/A	
Whole pod	<0.05	<0.05	84	80
Seeds	<0.05	<0.05	85	88

* Analysis failed – no result available

Table 6.1-12: Summary of the storage stability of alpha-cypermethrin in oilseed rape whole plant

Weeks of storage	BAS 310 I		
	Procedural recovery (%)[1]	Stored recovery mg/kg (%)[2]	Relative recovery mg/kg (%)[3]
0 weeks	90 94 **(92.0)**	0.4382 (87.6)	0.4763 (95.3)
		0.4334 (86.7)	0.4711 (94.2)
5 weeks	96 90 **(93.0)**	0.4226 (84.5)	0.4544 (90.9)
		0.3882 (77.6)	0.4174 (83.5)
14 weeks	88 86 **(87.0)**	0.4184 (83.7)	0.4809 (96.2)
		0.3978 (79.6)	0.4572 (91.4)
27 weeks	98 96 **(97.0)**	0.3786 (75.7)	0.3903 (78.1)
		0.3814 (76.3)	0.3932 (78.6)
49 weeks	93 96 **(94.5)**	0.4170 (83.4)	0.4413 (88.3)
		0.3936 (78.7)	0.4165 (83.3)

1 concurrent procedural recoveries were performed in duplicate at each assay timepoint. The average procedural recovery value is shown **in bold in brackets**.
2 uncorrected
3 relative recovery= stored recovery/average procedural recovery x 100

Table 6.1-13: Summary of the storage stability of alpha-cypermethrin in oilseed rape whole pods

Weeks of storage	BAS 310 I		
	Procedural recovery (%)[1]	Stored recovery mg/ kg (%)[2]	Relative recovery mg/kg (%)[3]
0 weeks	88 90 **(89.0)**	0.4296 (85.9)	0.4827 (96.5)
		0.4530 (90.6)	0.5090 (101.8)
4 weeks	89 93 **(91.0)**	0.4252 (85.0)	0.4673 (93.5)
		0.3752 (75.0)	0.4123 (82.5)
12 weeks	87 89 **(88.0)**	0.4020 (80.4)	0.4568 (91.4)
		0.3544 (70.9)	0.4027 (80.5)
26 weeks	89 92 **(90.5)**	0.3972 (79.4)	0.4389 (87.8)
		0.3912 (78.2)	0.4323 (86.5)
48 weeks	90 91 **(90.5)**	0.3402 (68.0)	0.3759 (75.2)
		0.3198 (64.0)	0.3534 (70.7)

1 concurrent procedural recoveries were performed in duplicate at each assay timepoint. The average procedural recovery value is shown **in bold in brackets**.
2 uncorrected
3 relative recovery= stored recovery/average procedural recovery x 100

Table 6.1-14: Summary of the storage stability of alpha-cypermethrin in oilseed rape seeds

Weeks of storage	BAS 310 I		
	Procedural recovery (%)[1]	**Stored recovery mg/kg (%)[2]**	**Relative recovery mg/kg (%)[3]**
0 weeks	86 87 **(86.5)**	0.3828 (76.6)	0.4425 (88.5)
		0.4084 (81.7)	0.4721 (94.4)
5 weeks	99 99 **(99.0)**	0.4684 (93.7)	0.4731 (94.6)
		0.4720 (94.4)	0.4768 (95.4)
15 weeks	102 100 **(101)**	0.4832 (96.6)	0.4784 (95.7)
		0.4884 (97.7)	0.4836 (96.7)
26 weeks	103 100 **(101.5)**	0.4522 (90.4)	0.4455 (89.1)
		0.4352 (87.0)	0.4288 (85.8)
48 weeks	101 98 **(99.5)**	0.3672 (73.4)	0.3690 (73.8)
		0.3742 (74.8)	0.3761 (75.2)

1 concurrent procedural recoveries were performed in duplicate at each assay timepoint. The average procedural recovery value is shown **in bold in brackets**.
2 uncorrected
3 relative recovery= stored recovery/average procedural recovery x 100

III. CONCLUSION

The results indicate that alpha-cypermethrin residues are stable in oilseed rape whole plant, whole pods and seeds under frozen conditions for at least 48 weeks.

Report: CA 6.1/5
Porto F., 2014a
Investigation of the storage stability of Alphacypermethrin residues in pineapple (fruit), barley (grain) and bean (seed) at -20°C
2014/3006687

Guidelines: OECD 506 (Oct. 2007), FAO manual on submission and evaluation of pesticide residues data for the estimation of maximum residue levels in food and feed - Roma 2009 (Second edition), EEC 396/2005, EEC 7032/VI/95 rev. 5, EPA 860.1380

GLP: yes
(certified by Instituto Nacional de Metrologia, Normalizacao e Qualidade Industrial - INMETRO, Rio de Janeiro, Brazil)

Executive Summary

The stability of alpha-cypermethrin in pineapple (fruit), barley (grain) and bean (seed) at -20° C was investigated over a time period of at least 728 days.
Pineapple, barley and bean samples were spiked with the test item at the level of 0.1 mg/kg (10 x LOQ). After that, they were stored in a cold chamber under the usual storage conditions for laboratory samples, -20°C or lower, in the dark. Untreated control samples were fortified and analysed along with the five stored samples within each analytical series in order to prove the validity and applicability of the analytical method.
Residues found in the stored samples were corrected by the mean value of the fortified samples and were evaluated by means of an exponential function.
The samples were analysed by means of LC/MS/MS with an LOQ of 0.01 mg/kg (BASF method 567/0). The efficiency of the method was determined at each analysis time period by fortifying control samples with alpha-cypermethrin. The average recoveries for alpha-cypermethrin during the current study were 91.1 ± 10 (n=32), 88.5 ± 11 (n=19) and 90.1 ± 12 (n=19), pineapple, barley and bean respectively.
The results obtained from the stored fortified samples indicate that alpha-cypermethrin is stable at -20°C in pineapple, barley and bean samples for at least 728 days.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-cypermethrin (BAS 310 I, CL-No. 900049)
Description:
Lot/Batch #: L80-24
Purity: 99.4%
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.1 mg/kg

2. Test Commodity:

Crop:	1) Pineapple 2) Barley 3) Bean
Type:	1) Miscellaneous fruit, inedible peel, large 2) Oilseeds 3) Pulses
Variety:	not reported
Botanical name:	1) *Ananas comosus* 2) *Brassica napus* 3) not reported
Crop parts(s) or processed commodity:	1) Pineapple fruit 2) Barley grain 3) Bean seed
Sample size:	5 g (aliquots)

B. STUDY DESIGN AND METHODS

1. Test procedure

The stability of alpha-cypermethrin in pineapple (fruit), barley (grain) and bean (seed) at -20° C was investigated over a time period of at least 728 days. For each sampling interval, and each matrix, 11 samples of 5 g were prepared, stored at -20ºC or lower temperature and analyzed, as described below:
- 5 samples, which were spiked at the concentration of 10 x LOQ (0.1 mg/kg)
- 1 sample not spiked (control sample)
- 5 further controls for procedural recoveries were stored together with the samples. These samples were spiked just before analysis with a fresh solution which contained the active ingredients.

For each sampling interval, there was a set of backup samples.
The sampling of 0 day is an exception (analysis directly after samples spiking), where 6 instead of 11 samples were prepared (1 control and 5 samples spiked with the concentration of 10 x LOQ = 0.1 mg/kg, for each matrix).
The extraction containers were closed with caps and transferred into a cold chamber. The samples analyzed at day 0 (starting values) were not frozen, but were analyzed immediately after their preparation. The containers were kept at a temperature of about – 20oC or lower, in the dark, over the entire period of the experiment.
Pineapple, barley and bean samples containing alpha-cypermethrin were analyzed at 0 day, 29, 153, 202, 365, 544, 735 days (pineapple), 162, 365, 545, 729 days (barley) and 155, 365, 545, 728 days (bean).

2. Description of analytical procedures

The samples were analysed by means of LC/MS/MS with an LOQ of 0.01 mg/kg (BASF method 567/0).

Aliquots were extracted with 70/25/5 methanol/ water/HCl 2 mol/L (v/v/v), centrifuged, extracted with water / cyclohexane 5/25 (v/v) and centrifuged again. An aliquot was evaporated to dryness. The residue was dissolved in methanol/water (80/20), filtered if necessary, and finally the residues were determined by means of LC/MS/MS.

Method recoveries were analyzed with each set of stored samples. The results are summarized in Table 6.1-15.

Table 6.1-15: Summary of concurrent recoveries in pineapple fruit, barley grain and bean seed

Matrix	Fortification level (mg/kg)	Summary recoveries				
		Range	n	Mean (%)	SD (%)	RSD (%)[1)]
BASF method No. 567/0			**BAS 310 I**			
Pineapple fruit	0.1	70.1-124	33	91.1	10	11
Barley grain	0.1	73.2-107	19	88.5	11	12
Bean seed	0.1	55.6-110	19	90.1	12	13

[1)] derived by dividing SD by Mean (values not reported)

II. RESULTS AND DISCUSSION

A summary of the frozen storage stability of alpha-cypermethrin residues in pineapple fruit, barley grain and bean seed for up to 728 days at -20°C or lower is given in Table 6.1-16 - Table 6.1-18. Recoveries from the stored fortified samples were corrected by procedural recoveries to account for experimental variability observed in the method spikes that were analyzed with the stored samples. The results obtained for barley: 0, 29 and 35 sampling days and for bean: 0, 29, 35 and 51 sampling days were not be reported in this final report due to recovery problems with the analytical method. Inclusion of new samples for these timepoints in the study was deemed not necessary since the analytical results from later time points clearly demonstrated storage stability of alpha-cypermethrin in barley grain and bean seeds.

All uncorrected and corrected average recoveries determined in the stored samples are clearly above 70 % for pineapple fruit, barley grain and bean seed, respectively, up to the end of the storage period.
The storage stability study demonstrates that residues of alpha-cypermethrin are stable for at least 728 days for beans, 729 days for barley and 735 days for pineapple, based on all the results described.

Table 6.1-16: Summary of the storage stability of alpha-cypermethrin in pineapple fruit

Storage period (months)	Average recovery (%)		
	BAS 310 I found (%)[1]	Procedural BAS 310 I recovery (%)	BAS 310 I in stored samples, corrected for procedural recovery
0	100	95.1	105
29	88.6	93.5	94.8
153	86.2	92.5	93.2
202	78.6	90.7	86.7
365	84.1	97.3	86.4
544	75.2	85.2	88.3
735	75.0	85.6	87.6

1 of initially fortified

Table 6.1-17: Summary of the storage stability of alpha-cypermethrin in barley grain

Storage period (months)	Average recovery (%)		
	BAS 310 I found (%)[1]	Procedural BAS 310 I recovery (%)	BAS 310 I in stored samples, corrected for procedural recovery
162	91.9	92.6	99.3
365	79.1	91.0	86.9
545	80.2	89.0	90.0
729	74.3	79.7	93.2

1 of initially fortified

Table 6.1-18: Summary of the storage stability of alpha-cypermethrin in bean seed

Storage period (months)	Average recovery (%)		
	BAS 310 I found (%)[1]	Procedural BAS 310 I recovery (%)	BAS 310 I in stored samples, corrected for procedural recovery
155	95.7	89.7	107
365	93.9	97.6	96.3
545	76.8	86.2	89.1
728	86.5	86.8	100

1 of initially fortified

III. CONCLUSION

The storage stability study demonstrates that residues of alpha-cypermethrin are stable for at least 728 days for beans, 729 days for barley and 735 days for pineapple.

CA 6.2 Metabolism, distribution and expression of residues

CA 6.2.1 Metabolism, distribution and expression of residues in plants

In the framework of the original inclusion into Annex I according to Directive 91/414/EEC the following studies investigating of the metabolism of the active substance alpha-cypermethrin in plant were peer-reviewed: on vegetables (cabbage) and cereals (wheat). Studies were done using ^{14}C-benzyl-labelled alpha-cypermethrin or cis-cypermethrin, respectively (cabbage; AL-640-001), vinyl- and benzyl-^{14}C labeled alpha-cypermethrin (wheat, AL-640-002 and AL-640-003), or benzyl-(ring-U-^{14}C/7-^{13}C] and [cyclopropane-1-(^{14}C/^{13}C)] (wheat; AL-640-004). The characteristics of these studies are summarized in Table 6.2.1-1.

Table 6.2.1-1: Summary of metabolism studies in plants previously available

Group	Crop	Test substance	Label position	Method, F or G (a)	Rate (kg a.s./ha)	No	Sampling (DAT)	Year	DocID
Brassica vegetables	Cabbage	Alpha-cypermethrin (WL85871)	^{14}C-benzyl	Foliar, F	0.05 1)	3	43	1983	AL-640-001
		Cis-cypermethrin (WL43481)	^{14}C-benzyl						
Cereals	Winter wheat	Alpha-cypermethrin (WL85871)	^{14}C-vinyl	Foliar, F	0.01 2) 0.1 2)	1	57	1994	AL-640-002
	Winter wheat	Alpha-cypermethrin (WL85871)	^{14}C-benzyl	Foliar, F	0.01 2) 0.1 2)	1	62	1994	AL-640-003
	Spring wheat	Alpha-cypermethrin (CL 900049)	benzyl-(ring-U-^{14}C/7-^{13}C	Foliar, F	0.08 3)	2	0 DAT-1 7 DAT-1 0 DAT -2 21 DAT -2 42 DAT -2	2001	AL-640-004
			cyclopropane-1-^{14}C, -^{13}C	Foliar, F	0.08 3)	2			

(a): Outdoor/field application (F) or glasshouse/protected/indoor application (G)
DAT: Days after (last) treatment; DAT-n denoting day(s) after nth treatment)
1) as EC (100 g/L, EF05835)
2) in formulation based on recipe CF05898
3) formulated as FASTAC®

After three foliar applications at a rate equivalent to 50 g a.s./ha to **cabbage**, the level of total radioactive residues was higher in the old outer leaves than in the new ones indicating a lack of translocation. The principal constituant of the residue at harvest was the parent insecticide in the outer leaves and stalk of cabbage. *Cis-trans* isomerization had occurred in the leaves. The identification of the different metabolites and their quantification were tentatively investigated by TLC but not confirmed: *cis*- and *trans*-isomers of 4-hydroxy-cypermethrin (WL48393, WL48394), the amide analogue of cypermethrin (WL47133), 3-phenoxybenzoic acid (SD36750), 4-HO-3-PBA (WL46114) and 3-phenoxybenzyl alcohol (WL40673).

After a single foliar application at a rate equivalent either to 0.01 or 0.1 kg a.s./ha to **wheat**, alpha-cypermethrin was found to be a major constituent of the residue in wheat straw (~~24% to 44%~~ 45% to 53% TRR). 3-Phenoxybenzoic acid was also present at low level (3.3% of TRR)

After two foliar applications at a rate equivalent to 0.08 kg a.s./ha to **wheat**, major component of the residue in wheat forage, hay, straw and grain was found to be the parent alpha-cypermethrin which had undergone isomerization from cis- to trans-isomer in forage, hay and straw. In grain samples from the plot treated with cyclopropane label, the parent compound accounted for 22.5% of the TRR and was confirmed as being the unchanged cis-2 isomer. The unidentified fractions appeared to be a mixture of polar material and lignin-enriched radioactivity.

The incurred total radioactive residues in wheat forage, hay and straw included low levels of mono-ring-hydroxylated parent (4'-OH parent, max. 4.8 % TRR in straw 42 DALT), the ester hydrolysis products dichlororvinylcyclopropane acid (DCVA; max 2.3 and 1.5% of TRR for cis-DCVA and trans-DCVA, respectively, in straw 42 DALT) and the various phenoxybenozates (3-PBAld, 3-PBA, 3-PBAlc, 4'-OH-3-PBA and 3-PBA-amino acids; 0.2 % TRR - 1.2% TRR in forage, hay and straw). In grain 42 DALT, residues of 4'-OH parent, the phenoxybenzoates (3-PBAld, 3-PBA, 3-PBAlc, 4'-OH-3-PBA and 3-PBA-amino acids) and *cis*- and *trans*-DCVA were not detected (below limit of quantification).
Most of the identified metabolites in this wheat metabolism study are recovered in the rat metabolism and are therefore out of any toxicological relevance. The metabolites 3-PBAld and 3-PBAlc were not detected in the rat metabolism but were not present in wheat grain and were present at very low levels in the other matrices.

The metabolism studies performed between 1983 and 2001 show essential similarities: In all cases unchanged parent alpha-cypermethrin was reported to be the major compound. *Cis-trans* isomerization was observed. The metabolite 3-phenoxybenzoic acid (3-PBA) was found in all studies. While in the earlier wheat metabolism study only 3-PBA was identified as a metabolite, the later wheat and the cabbage study both demonstrated formation of 4-hydroxycypermethrin, 4'-OH-3-PBA and 3-phenoxybenzyl alcohol. *Cis*-DCVA and *trans*-DCVA were identified in the later wheat study after treatment with cyclopropane-labelled alpha-cypermethrin.

The degradation of alpha-cypermethrin in wheat and cabbage proceeded primarily via two independent pathways:

-oxidative ring hydroxylation of the phenyl ring
-hydrolysis at the ester linkage with the formation of 2 moieties:
a) the phenoxybenzoyl portion which leads to a multiplicity of metabolites as acids, aldehydes, alcohols and converted into amino acid conjugate;
b) the dichlorovinyl propane acid portion of the molecule (cis and trans-DCVA isomers)

In the cabbage metabolism study considerable amounts of the total radioactive residue could not be identified. The reason for incomplete identification is certainly due to the relative simplicity of the analytical equipment in the early 1980s. The thin layer chromatography used in this time was not potent and sensitive enough to detect minor components in very low absolute residue amounts.

Against this background it was intended to perform one more metabolism study. Considering the crop groups covered by metabolism studies as well as the intended uses it was decided to conduct a new metabolism study in lettuce as representative of the leafy vegetables.

Report:	CA 6.2.1/1 Schreiner D., Possienke M. 2013a Metabolism of ^{14}C-alpha-cypermethrin in lettuce. 2012/1084223
Guidelines:	EPA 860.1300: Nature of the Residue in Plants Livestock,EPA 860.1000,PMRA Residue Chemistry Guidelines Section 97.2 Nature of the Residue - Plants - Livestock (Canada),JMAFF 59 NohSan No 4200,Lundehn III: 7028/VI/95 rev. 3 Appendix A (EU) Metabolism and distribution in plants (draft),OECD 501
GLP:	Yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

The objective of the present study was to investigate the metabolism of alpha-cypermethrin (BAS 310 I) in lettuce after foliar spray application. The test item was either labeled in the benzyl moiety or in the cyclopropane moiety. Lettuce was treated twice with BAS 310 I at a nominal application rate of 50 g a.s./ha at each application (approximately 0.045 lb/A). The lettuce heads were sampled 3 and 7 days after the last application (DALA). By enantiomer-specific analysis it was investigated whether an enantiomer of alpha-cypermethrin (BAS 310 I) is preferentially metabolized in lettuce.

The levels of total radioactive residues (TRR) at 3 DALA accounted for 0.873 mg/kg and 1.587 mg/kg for the benzyl label and the cyclopropane label, respectively. At 7 DALA, 1.318 mg/kg for the benzyl label and 1.372 mg/kg for the cyclopropane label were calculated.

For the investigations of metabolic patterns, subsamples of lettuce heads from both labels (benzyl and cyclopropane label) and both sampling time points (3 DALA and 7 DALA) were extracted with acetonitrile and water. The extractability of the radioactive residues from lettuce with acetonitrile and water was very high for both labels and both sampling time points (>97% TRR). The predominant part of the radioactive residues was extracted with acetonitrile, and only small portions were extracted with water. Due to low levels of radioactivity (up to approximately 2% TRR) in the residual radioactive residues after solvent extraction no further solubilization steps were performed.

Structural elucidation of metabolites was based on HPLC-MS investigations of isolated fractions from acetonitrile extracts. Peak assignment for BAS 310 I and its metabolites was based on comparison of the retention times of the isolated components with the ^{14}C-signals of the HPLC analyses. In the lettuce heads, BAS 310 I was the main component identified for both labels at 3 DALA and at 7 DALA (88.8 to 98.0% TRR). For the benzyl label, three metabolites (M310I005, M310I006 and M310I007) were identified, which lost the cyclopropane moiety and were therefore benzyl label-specific. For the cyclopropane label two label-specific metabolites (M310I008 and M310I009) were identified.

The metabolite M310I005 is a glucose conjugate of a proposed intermediate. M310I005 is subsequently metabolized via an acetylation reaction at the glucose moiety into M310I007, which was identified in the benzyl label at levels up to 4.9% TRR. The loss of the nitrile group of M310I007, which occurred presumably via hydrolysis and subsequent decarboxylation, leads to the formation of metabolite M310I006. The peak assigned to M310I005 and M310I006 accounted for up to 3.1% TRR. A second cleavage product of the ester bond hydrolysis is presumably 3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecarboxylic acid (DCVA), of which the glucose conjugate M310I008 was identified as metabolite in lettuce. This glycosylated compound is by malonylation on the glucose moiety metabolized into M310I009. The peak assigned to M310I008 and M310I009 accounted for up to 5.9% TRR.
The enantiomer-specific analyses showed that the ratio of enantiomer 1 to enantiomer 2 of BAS 310 I calculated from the HPLC analyses of the application solution was 1:1. The ratio found for fractions from lettuce extracts was similar to the enantiomer ratio in the application solution.
All samples were stored at approximately -18°C during the course of the study. All extractions were performed within less than three months after sampling. HPLC analyses were carried out within two months after extraction. Therefore, no storage stability investigations were necessary.
The extractability with three alternative extraction procedures according to residue methods for samples from both labels at 3 DALA was high (BASF residue analytical method 567/1: ≥92% TRR, multimethod QuEChERS: >75% TRR, multimethod DFG S19: >88% TRR), but the alternative extractions yielded slightly lower amounts of extracted radioactive residues than the acetonitrile/water extraction used in the metabolism study.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description:	a racemic mixture of two of the four cis isomers ((+)-*1R-cis-αS* and (-)-*1S-cis-αR*) comprising cypermethrin ^{14}C-alpha-cypermethrin (benzylring-U-^{14}C label) ^{13}C-alpha-cypermethrin (phenoxy-1,2,3,4,5,6-^{13}C label) ^{14}C-alpha-cypermethrin (cyclopropane-1-^{14}C label)
Lot/Batch #:	^{14}Calpha-cypermethrin (benzylring-U-^{14}C label): 775-0401 ^{13}Calpha-cypermethrin (phenoxy-1,2,3,4,5,6-^{13}C label): 1025-1034 ^{14}C-alpha-cypermethrin (cyclopropane-1-^{14}C label): 986-1046 ^{13}C-alpha-cypermethrin (cyclopropane-1-^{13}C label): 990-1024
Purity:	Radiochemical purity: ^{14}C-alpha-cypermethrin (benzylring-U-^{14}C label): 96.1% ^{14}C-alpha-cypermethrin (cyclopropane-1-^{14}C label): 98.2% Chemical purity: ^{14}C-alpha-cypermethrin (benzylring-U-^{14}C label): 93.7% ^{13}C-alpha-cypermethrin (phenoxy-1,2,3,4,5,6-^{13}C label): 95.1% ^{14}C-alpha-cypermethrin (cyclopropane-1-^{14}C label): 93.6% ^{13}C-alpha-cypermethrin (cyclopropane-1-^{13}C label): 97.7% Specific activity: ^{14}C-alpha-cypermethrin (benzylring-U-^{14}C label): 4.94 MBq/mg ^{14}C-alpha-cypermethrin (cyclopropane-1-^{14}C label): 4.9 MBq/mg
CAS#:	67375-30-8

Stability of test compound: All samples were stored at approximately -18°C during the course of the study. All extractions were performed within less than three months after sampling. HPLC analyses were carried out within two months after extraction. Therefore, no storage stability investigations were necessary.

2. Test Commodity:

Crop: Lettuce
Type: Leaf
Variety: Matilda
Botanical name: *Lactuca sativa*
Crop parts(s) or processed commodity: lettuce heads
Sample size: Not relevant

3. Soil: A sandy loam was used. The soil physicochemical properties are described below (see Table 6.2.1-2).

Table 6.2.1-2: Soil physicochemical properties

Soil series	Soil type	pH	OM %	Sand %	Silt %	Clay %	Maximal water holding capacity[1]	CEC[2] cmol/kg
Bruch West	*Sandy Loam	**7.3	***2.8	*63.7	*25.1	*11.3	29.2	11.9

* USDA scheme ** ($CaCl_2$) ***Organic matter calculated as 1.72 x percent organic carbon
1 in g/100 g dry soil
2 Cation exchange capacity

B. STUDY DESIGN

The study was carried out at the Agricultural Research Centre of BASF in Limburgerhof, Germany. The plant uptake part of the study was conducted in plastic containers located in climatic chambers and in the greenhouse, respectively.

1. Test procedure

Lettuce plants were planted into containers filled with sandy loam soil. The maintenance of the crop was performed in accordance with normal agricultural practice; fertilizers and additional pesticides were applied to achieve an adequate plant growth. According to the study protocol, the crop was treated twice with radiolabeled test items at a nominal rate of 50 g a.s./ha (approximately 0.045 lb/A) at each application. The first application was carried out 17 days before the last sampling date; the second was performed 7 days before the last sampling date. Samples of lettuce were taken 3 and 7 days after last application (DALA).

The in-life part was performed with two ^{14}C-labeled test substances, one radiolabeled in the benzyl ring (benzyl label) and the other in the cyclopropane ring (cyclopropane label). The ^{14}C-labeled test items were mixed with a ^{13}C-labeled test item each in a ratio of 2:1. For the preparation of the application formulation with the radiolabeled benzyl label and cyclopropane label, calculated amounts of ^{14}C-benzyl labeled and ^{13}C-phenoxy-1,2,3,4,5,6 labeled test items and calculated amounts of ^{14}C-cyclopropane labeled and ^{13}C-cyclopropane labeled test items were weighed, respectively.
The test item mixtures were dissolved in the blank formulation BAS 310 DZ I and water. Thereafter, the mixture was sonicated to generate a homogenous emulsion. The purity of the application solutions was confirmed using HPLC method and the isotope pattern were analysed by GC-CI-MS analysis.
The application formulations were applied with an automatic spray track at actual total application rates of 99.7 g a.s./ha for the benzyl label and of 101.9 g a.s./ha for the cyclopropane label (corresponding to a spray volume of 221.9 L/ha (benzyl label) and of 218.4 L/ha (cyclopropane label). Considering the loss recovered in the washing liquids of the spray track system, 99.28% (benzyl label) and 99.19% (cyclopropane label) of the application solution were actually applied as well as 99.05% (benzyl label) and 99.20% (cyclopropane label) at the second application.
Samples of lettuce were taken 3 and 7 days after last application (DALA) homogenized and frozen at -18°C or below. Extracts were stored in a refrigerator or, for longer periods in a freezer.

2. Description of analytical procedures

Combustion: Homogenized solid plant samples were weighed, pressed and combusted by means of an automatic sample oxidizer. The $^{14}CO_2$ was trapped by an absorption and scintillation liquid, and the collected radioactivity was measured by liquid scintillation counting. ^{14}C standards were combusted to determine the recovered radioactivity and the measurements were corrected accordingly. In order to determine the background radioactivity, aliquots of untreated lettuce samples were combusted under the same conditions. This resulted in a measured background activity of 0 dpm/g. Therefore, the limit of quantitation in mg/kg was not calculated from the background radioactivity level.
Homogenization / solvent extraction: All samples were homogenized with dry ice. After sublimation of the dry ice, the samples were weighed and radioassayed. Lettuce homogenized plant material was extracted with acetonitrile and with water. After centrifugation, the supernatant was filtered and the corresponding acetonitrile extracts and water extracts were pooled. Extracts were concentrated, analysed using HPLC and radioassayed (LSC). The residue after solvent extraction was dried, homogenized and radioassayed
Metabolite isolation for MS and enantiomer-specific analysis: Radioassayed, concentrated lettuce extracts (7 DALA) were redissolved in acetonitrile / water (1/9), loaded on a preconditioned SPE column and cleaned-up.
The eluates were analyzed by HPLC and HPLC-MS. In addition, enantiomer-specific analysis (both labels) was performed using HPLC.
HPLC: Extracts were concentrated, assayed by LSC (liquid scintillation counter for the quantitation of radioactivity in liquid samples) and characterized by HPLC. Metabolite patterns were confirmed using further HPLC methods.

LSC measurement of liquid samples: For the quantitation of radioactivity in liquid samples a liquid scintillation counter (LSC) was used. Aliquots of liquid samples were mixed with a sufficient volume of a suitable scintillator prior to measurement. All data were corrected using appropriate quench curves and are expressed in decays per minute (dpm).
Extraction procedures according to Residue Methods:
In addition to the solvent extractions, samples from 3 DALA of both labels were extracted following alternative extraction protocols according to residue methods. According to BASF residue analytical method 567/1 extraction was performed using a solvent mixture of methanol, water and HCl. The supernatants were radioassayed and analyzed using HPLC. Further extraction procedures according to multimethod QuEChERS and multimethod DFG S19 were performed and supernatants were concentrated and subsequently analyzed using HPLC.
Storage Stability:
Since storage intervals for frozen plant samples prior to extraction were below three months and for the extracts prior to HPLC analysis were below two months, respectively, no storage stability investigations were performed.
Calculations:
All calculations were performed with the complete numerical data exported from the LIMS. The numerical data were generally shown as rounded values (smaller degree of precision) to increase readability.

3. Identification of metabolites

The structure elucidation of metabolites was based on HPLC-MS investigations of isolated fractions from acetonitrile extracts 7 DALA of both labels. The identification (peak assignment) was based on comparison of the retention times of the isolated components with the ^{14}C-signals of the quantitative and confirmatory HPLC analyses.

II. RESULTS AND DISCUSSION

A. TOTAL RADIOACTIVE RESIDUES (TRRs)

The TRR was calculated by summarizing the extractable radioactive residues (ERR) and the residual radioactive residues (RRR) after solvent extraction. The calculated TRR for the benzyl label sampled 3 DALA was 0.873 mg/kg and 1.318 mg/kg at 7 DALA (see Table 6.2.1-3). For the cyclopropane label, the TRR for lettuce sampled 3 DALA and 7 DALA was 1.587 mg/kg and 1.372 mg/kg, respectively. Additionally, the TRR was measured by direct combustion analysis followed by LSC. The measured TRR of both labels of 3 DALA showed no major differences to the calculated TRR values. For 7 DALA, the calculated and measured concentrations differed to some extend (benzyl label: 2.433 mg/kg for TRR measured and 1.318 mg/kg for TRR calculated, cyclopropane label: 1.977 mg/kg for TRR measured and 1.372 mg/kg for TRR calculated). The differences were most probably due to slight inhomogeneity.
No background radioactivity was measured (0 dpm); therefore no calculation of the limit of quantitation could be performed.

Table 6.2.1-3: Total radioactive residues (TRRs) in lettuce samples

Matrix	DALA[1]	TRR determined by direct combustion [mg/kg]	TRR calculated [mg/kg]*
Lettuce head		Benzyl label	
	3	0.814	0.873
	7	2.433	1.318
		Cyclopropane label	
	3	1.538	1.587
	7	1.977	1.372

1 DALA = Days after last application
* Sum of ERR (extraction with acetonitrile and water) and RRR (extraction residue)

B. EXTRACTION, CHARACTERIZATION AND IDENTIFICATION OF RESIDUES

The extractability of lettuce for the two labels and two time points with acetonitrile and water is summarized in Table 6.2.1-4.

Table 6.2.1-4: Extraction efficiency of radioactive residues in lettuce samples

Matrix	DALA[1]	TRR calculated*	Distribution of radioactive residues				ERR[2]		RRR[3]	
			Acetonitrile extract		Aqueous extract					
		[mg/kg]	[mg/kg]	[%TRR]	[mg/kg]	[%TRR]	[mg/kg]	[%TRR]	[mg/kg]	[%TRR]
Lettuce head		Benzyl label								
	3	0.873	0.851	97.5	0.005	0.5	0.856	98.0	0.017	2.0
	7	1.318	1.277	96.9	0.011	0.8	1.288	97.7	0.030	2.3
		Cyclopropane label								
	3	1.587	1.559	98.2	0.010	0.6	1.569	98.8	0.019	1.2
	7	1.372	1.340	97.6	0.012	0.9	1.352	98.5	0.021	1.5

1 DALA = Days after last application
* TRR was calculated as the sum of ERR + RRR
2 ERR = extractable radioactive residue, calculated as the sum of acetonitrile extract and water extract
3 RRR = residual radioactive residue

1. Extraction and characterization of residues in lettuce

The extractability of lettuce with acetonitrile and water was very high and accounted for 97.7 to 98.8% of the TRR. For the benzyl label 97.5% TRR (3 DALA) and 96.9% TRR (7 DALA) were extracted with acetonitrile, whereas for the cyclopropane label 98.2% TRR (3 DALA) and 97.6% TRR (7 DALA) were extracted. Thus, virtually the complete radioactivity was extracted with acetonitrile and the subsequent water extraction released only minor amounts of 0.5 to 0.9% TRR.

Table 6.2.1-5: Metabolites detected in lettuce

Components	Acetonitrile extract (3 DALA)				Sum of extracts* (7 DALA)			
	Benzyl label		Cyclopropane label		Benzyl label		Cyclopropane label	
	[mg/kg]	[%TRR[2]]	[mg/kg]	[%TRR[2]]	[mg/kg]	[%TRR[2]]	[mg/kg]	[%TRR[2]]
Alpha-cypermethrin (BAS 310 I)	0.814	93.3	1.556	98.0	1.171	88.8	1.271	92.6
M310I005 / M310I006	0.027	3.1	n.d.	n.d.	0.030	2.3	n.d.	n.d.
M310I007	0.021	2.4	n.d.	n.d.	0.065	4.9	n.d.	n.d.
M310I008 / M310I009	n.d.	n.d.	0.049	3.1	n.d.	n.d.	0.081	5.9
Total identified from ERR[1]	0.862	98.7	1.605	101.1	1.266	96.1	1.352.	98.5.
Pooled water extract	0.005	0.5	0.010	0.6	n.d.	n.d.	n.d.	n.d.
Total characterized from ERR[1]	0.005	0.5	0.010	0.6	0.002	0.1	0.007	0.5
Total identified and characterized from ERR[1]	0.867	99.2	1.615	101.7	1.268	96.2	1.360	99.1
Residue After Solvent Extraction	0.017	2.0	0.019	1.2	0.030	2.3	0.021	1.5
Total Identified and Characterized + Residue	0.884	101.2	1.633	102.9	1.298	98.5	1.380	100.6

* The extracts (acetonitrile + aqueous) were combined and analyzed
1 ERR = extractable radioactive residue
2 TRR = total radioactive residue

2. Identification and quantitation of extractable residues in lettuce

Benzyl label, 3 DALA

Analysis of the acetonitrile extract of lettuce using HPLC method resulted in a pattern of three peaks, which were all identified. The main peak was found to be the parent compound BAS 310 I and accounted for 0.814 mg/kg or 93.3% TRR. The second peak was assigned to M310I005 and M310I006 and accounted for 0.027 mg/kg or 3.1% TRR, while the third peak, metabolite M310I007 was identified at a level of 0.021 mg/kg or 2.4% TRR.

In the water extract only minor amounts of radioactive residues were measured (0.005 mg/kg or 0.5% TRR) in the radioassay. Due to low levels of radioactivity (0.017 mg/kg or 2.0% TRR), the RRR after solvent extraction was not further solubilized. The results of the analysis of lettuce sampled 3 DALA (benzyl label) are compiled in Table 6.2.1-5. In summary, 98.7% of the total radioactive residues were identified in the ERR.

Benzyl label, 7 DALA

In the acetonitrile extract the same components as in the acetonitrile extract from lettuce sampled after 3 DALA were identified with HPLC. Likewise, the parent compound BAS 310 I was the main constituent at 1.171 mg/kg or 88.8% TRR. The second peak, metabolites M310I005 and M310I006 accounted for 0.026 mg/kg or 2.0% TRR, while the third peak, metabolite M310I007 was identified at a level of 0.061 mg/kg or 4.6% TRR.

Analysis of the water extract using HPLC resulted in a pattern of three peaks, of which two were identified. These are the more polar metabolites M310I005 and M310I006 as well as the metabolite M310I007 which were identified at levels of up to 0.3% TRR.

In the ERR, 96.1% of the total radioactive residues were identified and 0.1% TRR were characterized by HPLC. The RRR after solvent extraction was, due to low levels of radioactivity (0.030 mg/kg or 2.3% TRR), not further solubilized. Results of the sum of acetonitrile and water extracts of lettuce sampled 7 DALA (benzyl label) are summarized in Table 6.2.1-5.

Cyclopropane label, 3 DALA

Analysis of the acetonitrile extract of lettuce using HPLC resulted in a pattern of two peaks, which were both identified. The main peak was found to be the parent compound BAS 310 I and accounted for 1.556 mg/kg or 98.0% TRR. The second peak (assigned to M310I008 and M310I009) accounted for 0.049 mg/kg or 3.1% TRR.

In the water extract only minor amounts of radioactive residues were measured (0.010 mg/kg or 0.6% TRR) in the radioassay. Due to low levels of radioactivity (0.019 mg/kg or 1.2% TRR), the RRR after solvent extraction was not further solubilized. In the ERR 101.1% of the total radioactive residues were identified. The results of the analysis of lettuce sampled 3 DALA (cyclopropane label) are summarized in Table 6.2.1-5.

Cyclopropane label, 7 DALA

In the acetonitrile extract the same components as in the acetonitrile extract from lettuce sampled after 3 DALA were identified with HPLC. Likewise, the parent compound BAS 310 I was the main constituent at 1.271 mg/kg or 92.6% TRR. The remaining peak, which consists of M310I008 and M310I009, accounted for 0.076 mg/kg or 5.6% TRR. Analysis of the water extract using HPLC resulted in a pattern of three peaks, of which one was identified. M310I008 and M310I009 (assigned to the peak) accounted for 0.005 mg/kg or 0.4% TRR. In the ERR, 98.5% of the total radioactive residues were identified and 0.5% TRR were characterized by HPLC. The RRR after solvent extraction was, due to low levels of radioactivity (0.021 mg/kg or 1.5% TRR), not further solubilized. Results of the sum of acetonitrile and water extracts of lettuce sampled 7 DALA (cyclopropane label) are summarized in Table 6.2.1-5.

Enantiomer Ratio of BAS 310 I

In order to analyze if one enantiomer of BAS 310 I was preferably metabolized in lettuce, enantiomer-specific analyses were performed.

For the application solution the ratio of enantiomer 1 to enantiomer 2 was found to be approximately 1:1 for both labels (benzyl and cyclopropane label). For the determination of the enantiomer ratio in lettuce samples, the parent compound was isolated from fractions of the acetonitrile extract of both labels at 7 DALA and analyzed using HPLC. Thereby, a similar ratio of enantiomer 1 to enantiomer 2 was calculated compared to the application solution. The origin of a slight shoulder in front of the second peak could not be addressed as cypermethrin isomers, neither by HPLC analysis of the acetonitrile extracts of both labels nor by analysis of the diluted application solutions using UV chromatograms. The amounts of the parent substance were too low for detection.

3. Metabolic pathway

The proposed metabolic pathway of BAS 310 I (alpha-cypermethrin) in lettuce is shown in Figure 6.2.1-1.

A first step of the metabolism of BAS 310 I in lettuce after foliar application is the hydrolysis of the ester bond which releases two proposed intermediates. One of these intermediates is in a further step conjugated to a glucose resulting in the detected metabolite M310I005. Subsequent to the glucose conjugation reaction, M310I005 is acetylated at the glucose moiety resulting in the formation of the metabolite M310I007. The loss of the nitrile group of M310I007 (presumably via hydrolysis and subsequent decarboxylation) leads to the formation of metabolite M310I006.

The second cleavage product of the ester bond hydrolysis is presumably 3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecarboxylic acid (DCVA), which was not detected in this study. Nevertheless, was the glucose conjugate of DCVA, which was code-named M310I008 identified as a metabolite in lettuce. This conjugate is by malonylation on the glucose moiety metabolized to M310I009, the corresponding DCVA malonylglucose.

4. Extraction procedures according to Residue Methods

Additional to the acetonitrile and water extractions of the metabolism investigations samples of both labels from 3 DALA were also extracted with alternative extraction procedures according to residue methods.

The extraction procedure according to BASF residue analytical method 567/1, the method adapted from the multimethod QuEChERS and the extraction procedure according to multimethod DFG S19 resulted in slightly lower amounts of extracted radioactive residues than the acetonitrile/water extraction used in the metabolism study, except for the multimethod DFG S19 where for the benzyl label 3 DALA one slightly higher extractability was detected (Table 6.2.1-6). The concentrated solvent supernatants of the alternative extraction methods were analyzed using HPLC. The results of the HPLC analyses are summarized in Table 6.2.1-7.

Table 6.2.1-6: Extraction efficiency of radioactive residues in lettuce samples obtained with extraction procedures according to residue methods

Matrix	DALA[1]	Metabolism investigations (3 x acetonitrile + 2 x water)[3]		BASF residue analytical method 567/1		Multimethod QuEChERS		Multimethod DFG S19	
		[mg/kg]	[%TRR][2]	[mg/kg]	[%TRR][2]	[mg/kg]	[%TRR][2]	[mg/kg]	[%TRR][2]
Lettuce head		Benzyl label							
	3	0.856	98.0	0.803	92.0	0.781	89.5	0.882	101.0
		Cyclopropane label							
	3	1.569	98.8	1.465	92.3	1.204	75.9	1.409	88.8

1 DALA = Days after last application
2 TRR was calculated as the sum of ERR + RRR
3 Calculated as sum of the pooled extracts, method used for quantitation of residues

Table 6.2.1-7: Total identified radioactive residues extracted from lettuce samples 3 DALA with extraction procedures according to residue methods

Extraction method	Metabolism investigations	BASF residue analytical method 567/1	Multimethod QuEChERS	Multimethod DFG S19
Metabolite	[% TRR]			
	Benzyl label			
BAS 310 I	93.3	76.1	85.4	83.5
M310I005 / M310I006	3.1	4.4	2.8	1.9
M310I007	2.4	1.4	1.8	0.7
	Cyclopropane label			
BAS 310 I	98.0	79.8	71.3	75.0
M310I008 / M310I009	3.1	1.6	2.7	2.1

5. Storage stability

All samples were stored in a freezer (–18°C or below) directly after sampling until used. Subsamples of lettuce sampled 3 DALA and 7 DALA of both labels were extracted 2 to 18 days after sampling. Aliquots of extracts were analyzed for quantitation and confirmation by HPLC within 33 days after extraction. Enantiomer-specific analysis was performed 35 days after extraction, while fractionation of extracts was performed 17 to 59 days after extraction. Subsamples used for alternative solvent extraction procedures were extracted 70 to 74 days after sampling and analyzed by HPLC within the next 2 days.
Since the storage intervals for the frozen plant samples prior to extraction were below three months and the storage intervals for the extracts prior to the HPLC analysis were below two months, no storage stability investigations were necessary.

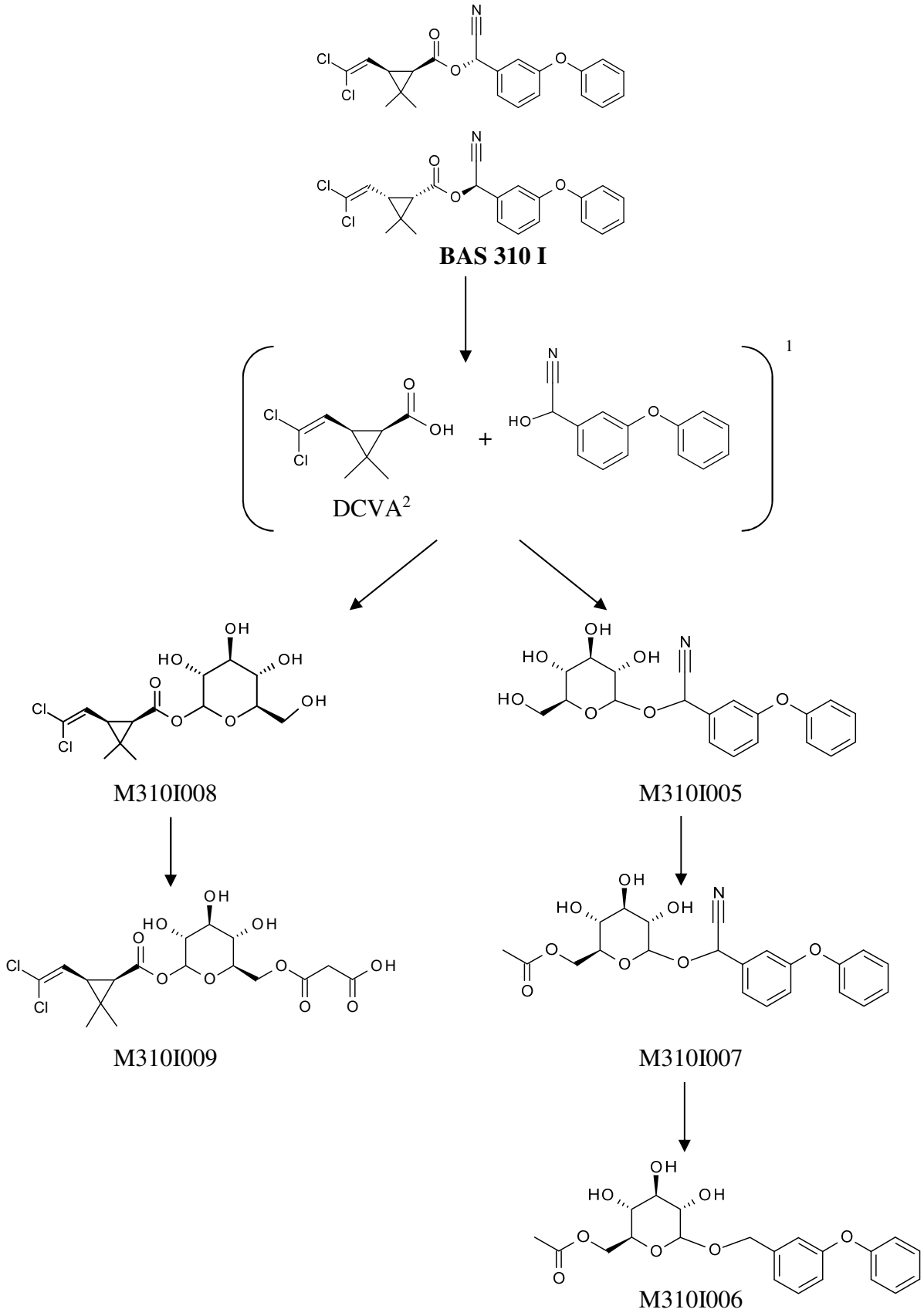
III. CONCLUSION

The insecticide alpha-cypermethrin (BAS 310 I), which was radioactive labeled in the benzyl or in the cyclopropane moiety, was applied to lettuce by two foliar applications at a nominal rate of 2 x 50 g a.s./ha. The lettuce heads were sampled 3 days and 7 days after the last application.
The levels of total radioactive residues (TRR) at 3 DALA accounted for 0.873 mg/kg and 1.587 mg/kg for the benzyl label and the cyclopropane label, respectively. At 7 DALA, 1.318 mg/kg for the benzyl label and 1.372 mg/kg for the cyclopropane label were calculated.
The extractability of the radioactive residues from lettuce with acetonitrile and water was very high for both labels and both sampling time points (>97% TRR). The predominant part of the radioactive residues was extracted with acetonitrile, and only small portions were extracted with water. Due to low levels of radioactivity (up to approximately 2% TRR) in the residual radioactive residues after solvent extraction no further solubilization steps were performed.

Structural elucidation of metabolites was based on HPLC-MS investigations of isolated fractions from acetonitrile extract of the benzyl label at 7 DALA. In these fractions, additional to the parent compound the metabolites M310I005, M310I006 and M310I007 were identified. HPLC-MS analyses of an isolated fraction from acetonitrile extract of the cyclopropane label at 7 DALA identified M310I008 and M310I009 as label-specific metabolites. The peak assignment for BAS 310 I and its metabolites was based on comparison of the retention times of the isolated components with the ^{14}C-signals of the HPLC analyses.

In the lettuce heads, BAS 310 I was the main component identified for both labels at 3 DALA and at 7 DALA (>88% TRR). For the benzyl label, three metabolites (M310I005, M310I006 and M310I007) were identified, which lost the cyclopropane moiety and were therefore benzyl label-specific. For the cyclopropane label two label-specific metabolites (M310I008 and M310I009) were identified.
An ester bond cleavage of the parent compound leads presumably to two cleavage products. Those intermediates were not detected in this study, but due to a low level of metabolized parent compound, low amounts of cleavage products are expected.
The metabolite M310I005 is a glucose conjugate of one of the proposed intermediates. M310I005 is subsequently metabolized via an acetylation reaction at the glucose moiety into M310I007, which was identified in the benzyl label at levels up to 5% TRR. The loss of the nitrile group of M310I007, which occurred presumably via hydrolysis and subsequent decarboxylation, leads to the formation of metabolite M310I006. The peak assigned to M310I005 and M310I006 accounted for up to approximately 3% TRR.
The second cleavage product of the ester bond hydrolysis is presumably 3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecarboxylic acid (DCVA), of which the glucose conjugate M310I008 was identified as metabolite in lettuce. This glycosylated compound is by malonylation on the glucose moiety metabolized to M310I009. The peak assigned to M310I008 and M310I009 accounted for up to approximately 6% TRR.
The enantiomer-specific analyses showed that the ratio of enantiomer 1 to enantiomer 2 of BAS 310 I calculated from the HPLC analyses of the application solution was 1:1. The ratio of fractions from lettuce extracts was similar to the enantiomer ratio in the application solution.
The extractability with three alternative extraction procedures for samples from both labels at 3 DALA was high (BASF residue analytical method 567/1: ≥92% TRR, multimethod QuEChERS: >75% TRR, multimethod DFG S19: >88% TRR), but the alternative extractions yielded slightly lower amounts of extracted radioactive residues than the acetonitrile/water extraction used in the metabolism study. All methods allow extracting and determining the parent compound with high recoveries (>75%).

Figure 6.2.1-1: Proposed metabolic pathway of alpha-cypermethrin (BAS 310 I) in lettuce



[1] putative intermediates not found
[2] DCVA: 3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecarboxylic acid

Overall Summary of Plant Metabolism

The present Annex I renewal dossier provides a new metabolism study in lettuce as representative for leafy vegetables. The new study mainly confirms the findings and degradation reactions known so far and complements additional details due to more sensitive technology.

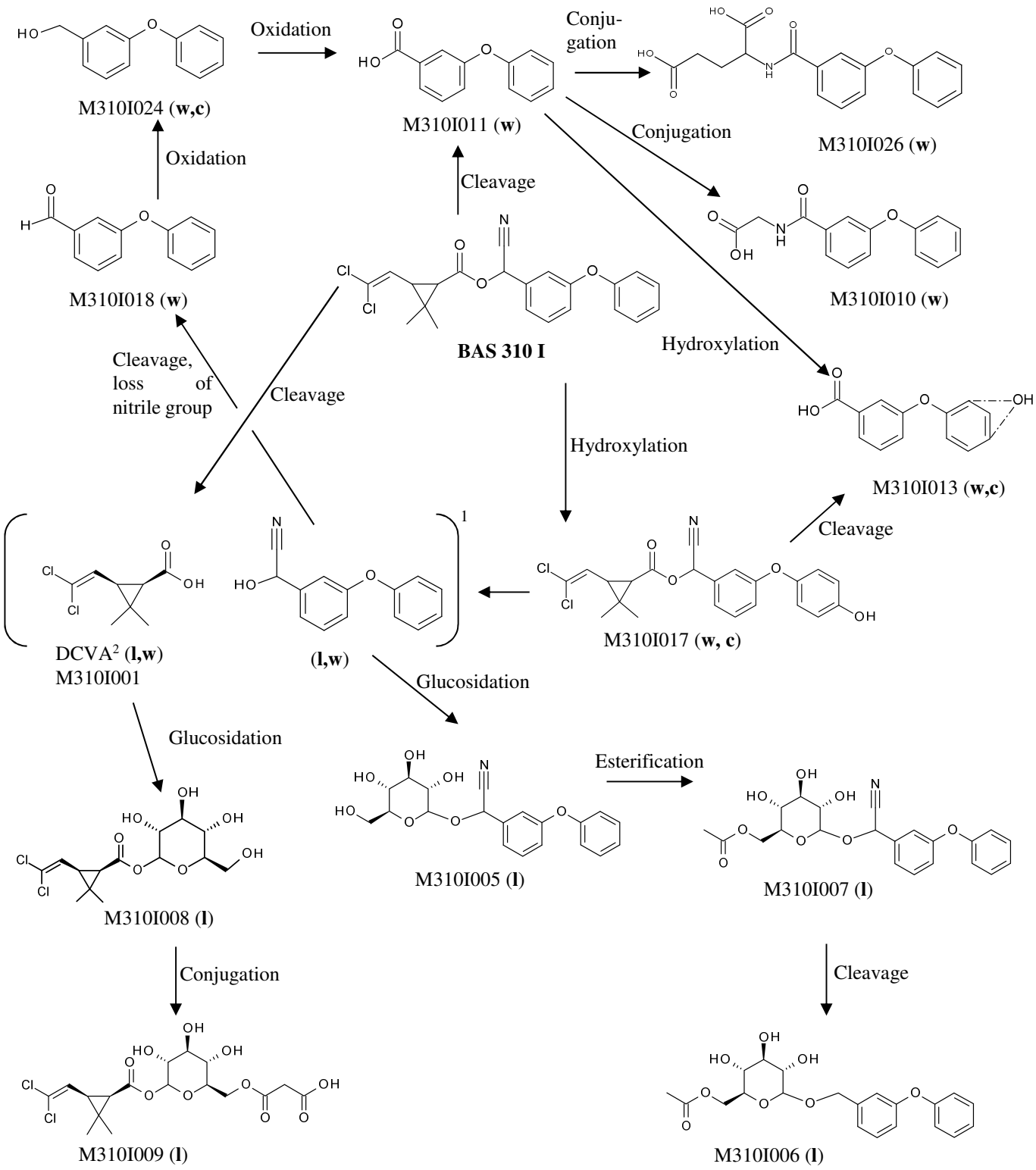
Combining the findings of the older metabolism studies and the new one, a system of degradation reactions results which can not only explain most of the facts gathered so far but also lead to a more complete picture.

In all studies, the parent is the major residue constituent in the RAC. *Cis*/*trans* isomerization of BAS 310 I was observed in wheat and cabbage which can be attributed to sunlight exposure. In lettuce (indoor study), no difference in the BAS 310 I isomer ratio was found compared to the application solution, which could probably be attributed to the shorter PHI and as such to the shorter exposure to artificial light.

Common in all metabolism studies is the hydrolysis at the ester linkage with the formation of two moieties, the phenoxybenzoyl portion and the dichlorovinyl cyclopropane acid portion of the molecule. Oxidative ring hydroxylation of the phenyl ring was observed only in wheat and cabbage.
In **lettuce**, the alpha-cypermethrin cleavage products (M310I001 (DCVA) and the phenoxybenzoyl moiety) were not detected, but due to a low level of metabolized parent compound, low amounts were to be expected. From the proposed intermediate DCVA a glucose conjugate is formed (M310I008), which is further metabolized to M310I009 by malonylation on the glucose moiety. The phenoxybenzoyl portion is conjugated with glucose to form M310I005. M310I005 is subsequently metabolized via an acetylation reaction at the glucose moiety into M310I007. The loss of the nitrile group of M310I007, which occurred presumably via hydrolysis, leads to the formation of metabolite M310I006.
In **wheat**, low levels of the cleavage product DCVA were detected. The phenoxybenzoyl portion was metabolized to the various phenoxybenozates (3-PBAld, 3-PBA, 3-PBAlc, 4'-OH-3-PBA and 3-PBA-amino acids). 3-phenoxybenzoic acid, 4-HO-3-PBA and 3-phenoxybenzyl alcohol (WL40673) were also detected in **cabbage**.
The difference in the metabolites found resulting from cleavage of alpha-cypermethrin in lettuce as compared to cabbage and wheat can be mainly attributed to the considerably shorter PHI/sampling intervals in the lettuce study.

A complete picture on plant metabolism is shown in Figure 6.2.1-2.

Figure 6.2.1-2: Proposed metabolic pathway of alpha-cypermethrin (BAS 310 I) in plant (lettuce (l), cabbage (c) and wheat (w))



[1] putative intermediates not found

[2] DCVA: 3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecarboxylic acid

This entry is taken from public literature and was not included in the study list of the application.

Report: CA 6.2.1/2
Fujisawa T. et al., 2009a
Application of separated leaf cell suspension to Xenobiotic - Metabolism in plant
2009/1131382
Guidelines: none
GLP: no

Executive Summary

Metabolic profiles of ^{14}C-labeled primary metabolites from several pesticides, 4-cyanophenol **(1)**, 3-phenoxybenzoic acid **(2)**, 3-phenoxybenzyl alcohol **(3)**, 3,5-dichloroaniline **(4)**, and (1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropanecarboxylic acid **(5)**, were examined by using enzymatically separated leaf cell suspension from seedlings of cabbage (*Brassica oleracea*) and tomato (*Lycopersicon esculentum*). After 1 day of incubation, the metabolites were extensively transformed in cabbage, whereas they were scarcely metabolized in tomato. The major metabolic pathways were the phase II reactions leading to a number of conjugates such as glucoside/malonylglucoside of **1-5**, malate of **2**, and glutamate of **4**. The oxidation of **1** and **2** was observed as a minor reaction to produce 4-hydroxybezoic acid and 3-(4-hydroxyphenoxy)benzoic acid. The chemical identities of the secondary metabolites were determined by various spectrometric analyses (LC-MS, LC-MS/MS, and NMR) and/or HPLC cochromatography with the synthetic reference standards. As a result, this separated leaf cell suspension system was found to well reproduce the in vivo plant metabolism.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: ^{14}C labelled 4-cyanophenol **(1)**,
^{14}C labelled 3-phenoxybenzoic acid **(2)**,
^{14}C labelled 3-phenoxybenzyl alcohol **(3)**,
^{14}C labelled 3,5-dichloroaniline **(4),**
^{14}C labelled (1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropanecarboxylic acid **(5)**

Description: ^{14}C phenyl ring labelled (0.82 GBq/mmol) 4-cyanophenol **(1)**,
^{14}C phenoxyphenyl ring labelled (2.45 GBq/mmol) phenoxybenzoic acid **(2)**,
^{14}C phenoxyphenyl ring labelled (2.25 GBq/mmol) phenoxybenzyl alcohol **(3)**,
^{14}C phenyl ring labelled (2.22 GBq/mmol) 3,5-dichloroaniline **(4)**
^{14}C cyclopropyl-1 ring labelled (0.98 GBq/mmol) (1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropanecarboxylic acid **(5)**

Lot/Batch #: not reported
Purity: Radiochemical purity for compound **1-5** >97%
CAS#: not applicable
Development code: not applicable
Stability of test Compound: not reported

2. Test Commodity: Brassica vegetables, Fruiting vegetables
Crop: Head cabbages, Tomatoes
Type: not reported
Variety: Kinkei 201, Patio
Botanical name: *Brassica oleracea, Lycopersicon esculentum*
Crop parts(s) or processed
Commodity: leaves
Sample size: Cabbage and tomato: 5 g

3. Soil: moist compost (Kureha Chemical Co., Ltd.)

B. STUDY DESIGN

1. Test procedure

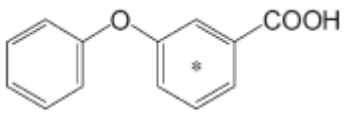
Metabolism studies were conducted using the separated leaf cell suspension of cabbage (*Brassica oleracea*) and tomato (*Lycopersicon esculentum*) with phenolic, alcoholic, anilinic, and carboxylic compounds, which are the primary metabolites from various kinds of agrochemicals. The ^{14}C-labeled primary metabolites 4-cyanophenol **(1)**, 3-phenoxybenzoic acid **(2)**, 3-phenoxybenzyl alcohol **(3)**, 3,5-dichloroaniline **(4)** and (1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropanecarboxylic acid **(5)** are shown from Figure 6.2.1-3 to Figure 6.2.1-7.
Seeds of cabbage (*B. oleracea*) and tomato (*L. esculentum*) were sown in 50 mL plastic pots containing a moist compost and grown until third- to fourth-leaf stage in a temperature controlled cabinet at 25 °C for day temperature and 22 °C for night temperature. Light with an intensity of 19500 lx at the level of primary leaves was supplied by a combination of fluorescent and incandescent lights with a 16 h light and 8 h dark sequence. Irrigation was appropriately conducted until sampling for approximately a month. Five grams of third and fourth-leaves in fresh weight were used for the experiments.
Sampled leaves were immersed in 80% ethanol (v/v) for 30 s, sterilized by dipping into a 0.5% sodium hypochlorite solution for 10 min, and rinsed three times with sterilized deionized water. The leaves were then transferred into a homogenizer cup filled with 120 mL of Murashige-Skoog medium, cut into small pieces and homogenized. Prior to the experiment, the osmotic pressure and pH of the MS medium were adjusted to 0.3-0.4 M and 6.5, respectively, and the prepared medium was sterilized by autoclaving. 20 mL portions of the leaf homogenate were divided into 100 mL Erlenmeyer flasks, and 1% of Macerozyme R-10 was added to each. The methanol dose solutions of ^{14}C-**1-5** were individually prepared by isotopically diluting them with corresponding non-radiolabeled reference standards to give the specific activity of approximately
4.3 kBq/µg. Each of the dose solution was spiked into the leaf suspension medium. Sampling was conducted at 1, 2, and 4 days of exposure.

Figure 6.2.1-3: Structural formula of ^{14}C phenyl ring labelled 4-cyanophenol (1)

1

* : labelled position

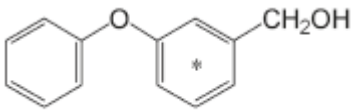
Figure 6.2.1-4: Structural formula of ^{14}C phenoxyphenyl ring labelled phenoxybenzoic acid (2)



2

* : labelled position

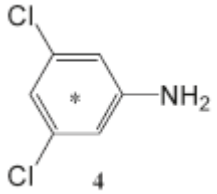
Figure 6.2.1-5: Structural formula of ^{14}C phenoxyphenyl ring labelled phenoxybenzyl alcohol (3)



3

* : labelled position

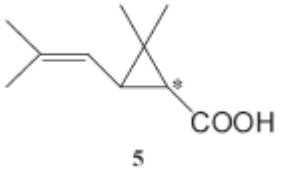
Figure 6.2.1-6: Structural formula of ^{14}C phenyl ring labelled 3,5-dichloroaniline (4)



4

* : labelled position

Figure 6.2.1-7: Structural formula of ^{14}C cyclopropyl-1 ring labelled (1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropanecarboxylic acid (5)



5

* : labelled position

2. Description of analytical procedures

The incubated samples were first divided into leaf cells and culture medium by filtration and were stored in a freezer (<-20 °C) before analyses. The leaf cells were extracted with acetone/water (4:1, v/v). The homogenate was filtered and the residue on the filter was extracted. The culture medium was partitioned three times with a 3-fold volume of ethyl acetate. Aliquots of the plant extracts as well as the organic and water layers from culture medium were radioassayed by LSC. The plant residues were air-dried in open vessels at room temperature for 1 week, and subsamples of the dried residues were subjected to combustion analysis to determine the remaining radioactivity.
For the purpose of isolating unknown metabolites for spectroscopic analyses, the plant extracts and the organic layer of the culture medium were individually subjected to the preparative purification by a solid-phase absorbent cartridge. The chemical identities of the secondary metabolites were determined by various spectrometric analyses (LC-MS, LC-MS/MS, and NMR) and/or HPLC cochromatography with the synthetic reference standards.

II. RESULTS AND DISCUSSION

A. TOTAL RADIOACTIVE RESIDUES (TRRs) and EXTRACTION

The recoveries of the ^{14}C labelled compounds **1-5** from the test system after 1 day of incubation are shown in Table 6.2.1-8.

Table 6.2.1-8: Recoveries of the ^{14}C labelled compounds 1-5 from the test system after 1 day of incubation

Compound	Recovery of applied radioactivity (^{14}C) [%]
1	78.8
2	82.3
3	79.8
4	80.6
5	78.6

The unrecovered ^{14}C increased as the exposure period was extended, possibly due to azeotropic vaporization (data not shown). There were insignificant differences in the loss of ^{14}C from the test system among the test substances, although the Henry's law constants of **1-5** greatly varied by 3 orders. The distribution of ^{14}C within the test system after 1 day of exposure was examined in duplicate, and the result is shown in Table 6.2.1-9. The radioactivity taken up by cabbage leaf cell was estimated to be 36.2% TRR **(1)**, 46.3% TRR **(2)**, 35.9% TRR **(3)**, 50.8% TRR **(4)** and 47.2% TRR **(5)** by combining extractable and unextractable ^{14}C. For tomato leaf cell, an increase in rate of the ^{14}C taken up was observed compared to the one in cabbage, as 80.8% TRR **(1)**, 79.7% TRR **(2)**, 91.4% TRR **(3)**, 84.3% TRR **(4)**, and 86.6% TRR **(5)**. The unextractable ^{14}C for compound **1-5** amounted to 3.0-5.4% TRR in cabbage and 2.4-6.9% TRR in tomato.

Table 6.2.1-9: ^{14}C distribution of compound 1-5 in the cell suspension system

Total radioactive residues (TRR) [%]					
Compound	**1**	**2**	**3**	**4**	**5**
Cabbage					
Leaf cell*	36.2	46.3	35.9	50.8	47.2
extractable	30.8	42.4	32.3	47.8	43.3
unextravtable	5.4	3.9	3.6	3.0	3.9
Medium ^{14}C	63.8	53.7	64.1	49.2	52.8
Tomato					
Leaf cell*	80.8	79.7	91.4	84.3	86.6
extractable	78.4	74.5	84.5	79.0	81.9
unextravtable	2.4	5.2	6.9	5.3	4.7
Medium ^{14}C	18.9	20.3	8.7	15.7	13.4

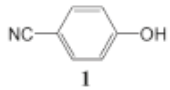
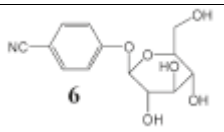
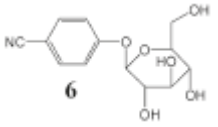
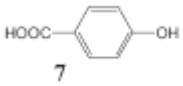
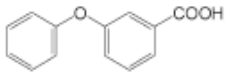
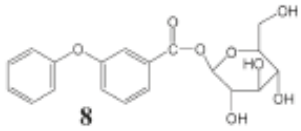
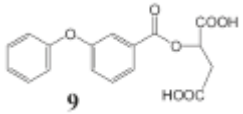
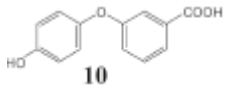
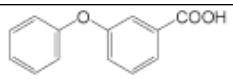
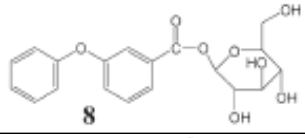
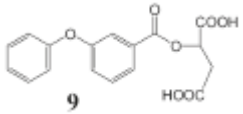
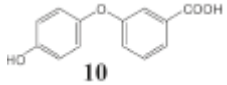
* sum of extractable and unextractable TRRs

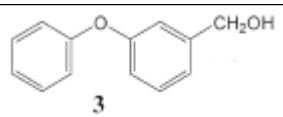
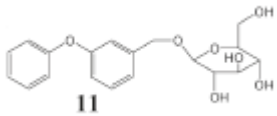
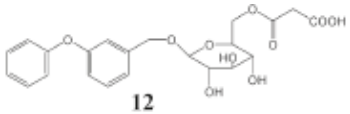
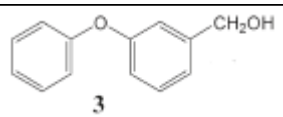
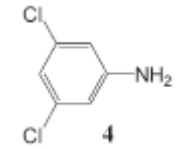
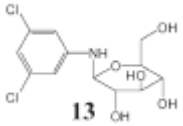
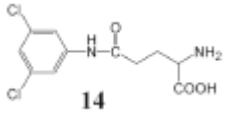
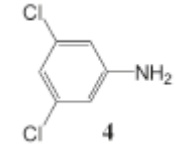
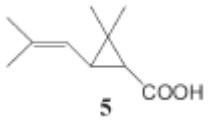
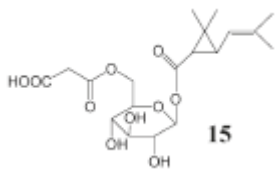
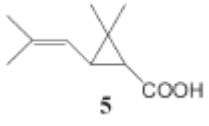
B. CHARACTERIZATION AND IDENTIFICATION OF RESIDUES

1. Identification, characterization and quantitation of extractable residues

The formation and decline of metabolites were examined in detail for the compound [^{14}C]-5 using the cabbage leaf cell suspension with exposure periods of 1, 2, and 4 days. The metabolic distribution reached a plateau within 1 day, and approximately 100% of the radioactivity detected in the cabbage leaf cell was the malonylglucose conjugate **(15)** of compound **5**. On the basis of these data, the incubation period in the study examining metabolic profiles of the compounds [^{14}C]-**1-5** was determined to be 1 day after treatment. The metabolic distribution in the extract of leaf cells is shown in Table 6.2.1-10. In general, the test compounds were extensively metabolized in the leaf cells of cabbage as compared with tomato cells. In cabbage, the glucose conjugate **(6)** was the only metabolite from compound **(1)** and amounted to 27.6% TRR with the remaining radioactivity (3.2% TRR) being unchanged compound **(1)**. Both compounds **(3)** and **(4)** were fully transformed within 1 day of exposure as none of the corresponding test compounds remained in the extract of the leaf cells. The metabolites of compound **(3)** were glucoside **(11)** and malonylglucoside **(12)** conjugates, each at 4.6% and 27.7% TRR, and compound **(4)** was metabolized to glucoside **(13)** and glutamate **(14)** conjugates at 23.7% and 16.3% TRR, respectively. In the case of compounds **(2)** and **(5)**, differences were observed in the metabolic profiles, such as compound **(2)** being transformed to three major metabolites, glucoside **(8)** and malate **(9)** conjugates and a hydroxylated free form **(10)** at 6.6%, 18.4% and 2.4% TRR, respectively, whereas compound **(5)** was solely metabolized to its malonylglucoside conjugate **(15)** at 43.3% TRR. Some of the metabolites derived from compounds **(1)** and **(2)** were detected in the culture medium. The metabolic distribution of compound **(1)** in the medium of cabbage was 9.4% TRR for metabolite **(6)** and 7.4% TRR for metabolite **(7)**. In the case of compound **(2)**, metabolites **(8)**, **(9)**, and **(10)** were detected in the cabbage culture medium at 34.3%, 7.6% and 0.8% TRR, respectively. With respect to tomato plant, compounds **(1)**, **(2)** and **(4)** were transformed to their corresponding glucose conjugates at the levels of 9.1% TRR **(6)**, 2.2% TRR **(8)**, and 15.1% TRR **(13)**, respectively, within the total test system. No transformation products were observed for compounds **(3)** or **(5)**.

Table 6.2.1-10: Metabolic distribution of compound 1-5 in the cell suspension system

Compound	Matrix	Metabolite identity		TRR [%]	
		Designation	Chemical structur	Cabbage	Tomato
4-cyanophenol (**1**)	leaf extract	4-cyanophenol (**1**)	 **1**	3.2	72.3
		glucose conjugate	 **6**	27.6	6.1
	medium	4-cyanophenol (**1**)	 **1**	24.4	15.9
		glucose conjugate	 **6**	9.4	3.0
		4-hydroxybenzoic acid	 **7**	7.4	nd
phenoxybenzoic acid (**2**)	leaf extract	phenoxybenzoic acid (**2**)	 **2**	13.4	72.8
		glucose conjugate	 **8**	6.6	1.7
		malate conjugate	 **9**	18.4	nd
		3-(4-hydroxy phenoxy)benzoic acid	 **10**	2.4	nd
	medium	phenoxybenzoic acid (**2**)	 **2**	10.7	19.8
		glucose conjugate (**8**)	 **8**	34.3	0.5
		malate conjugate (**9**)	 **9**	7.6	nd
		3-(4-hydroxy phenoxy)benzoic acid (**10**)	 **10**	0.8	nd

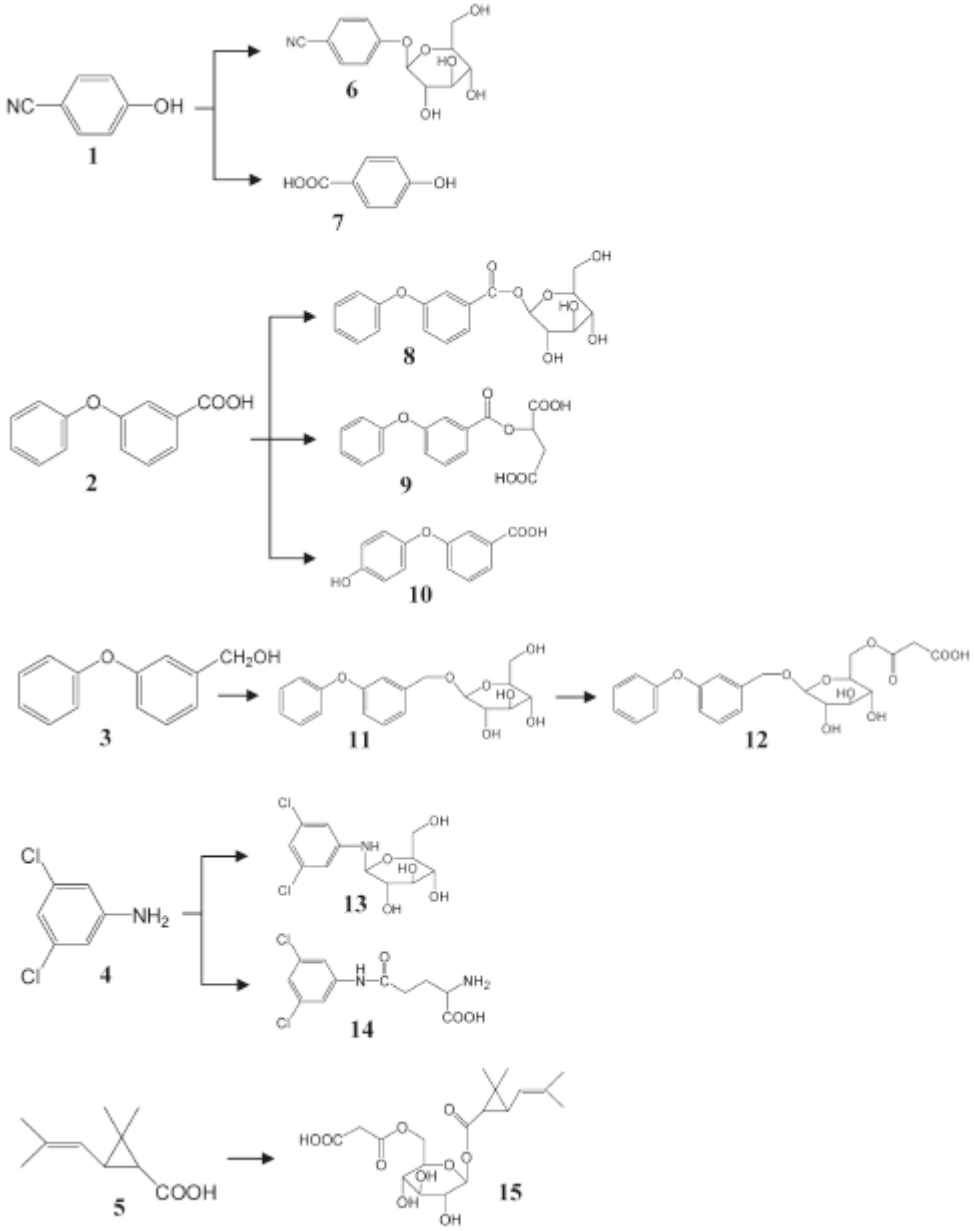
Compound	Matrix	Metabolite identity		TRR [%]	
		Designation	Chemical structur	Cabbage	Tomato
phenoxybenzyl alcohol **(3)**	leaf extract	phenoxybenzyl alcohol **(3)**	 **3**	nd	84.5
		O-glucoside conjugate **(11)**	 **11**	4.6	nd
		malonylglucoside conjugate **(12)**	 **12**	27.7	nd
	medium	phenoxybenzyl alcohol **(3)**	 **3**	64.1	8.7
3,5-dichloroaniline **(4)**	leaf extract	3,5-dichloroaniline **(4)**	 **4**	nd	63.9
		N-glucoside conjugate **(13)**	 **13**	23.7	15.1
		glutamate conjugate **(14)**	 **14**	16.3	nd
	medium	3,5-dichloroaniline **(4)**	 **4**	49.2	15.7
(1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropane carboxylic acid **(5)**	leaf extract	(1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropane carboxylic acid **(5)**	 **5**	nd	81.9
		malonylglucose conjugate **(15)**	 **15**	43.3	nd
	medium	(1RS)-trans-2,2-dimethyl-3-(2-methylprop-1-enyl)cyclopropane carboxylic acid **(5)**	 **5**	52.8	13.4

nd = not detectable

2. Proposed metabolic pathway

The chemical identities of the secondary metabolites were determined by various spectrometric analyses (LC-MS, LC-MS/MS, and NMR) and/or HPLC cochromatography with the synthetic reference standards. On the basis of these identifications, the metabolic pathways of the compounds 1-5 are summarized in Figure 6.2.1-8.

Figure 6.2.1-8: Proposed metabolic pathways of compounds 1-5



III. CONCLUSION

After 1 day of incubation, the metabolites were extensively transformed in cabbage, whereas they were scarcely metabolized in tomato. The major metabolic pathways were the phase II reactions leading to a number of conjugates such as glucoside/malonylglucoside of compounds (1)-(5), malate of compound (2) and glutamate of compound (4). The oxidation of compounds (1) and (2) was observed as a minor reaction to produce 4-hydroxybezoic acid and 3-(4-hydroxyphenoxy)benzoic acid.

In conclusion, the results presented in the study substantiate the view that xenobiotic metabolism conducted using the separated leaf cell suspension is a useful adjunct to the conventional whole plant approach. The study has shown that the oxidative reactions and/or conjugation reactions observed in vivo were also found in the separated leaf cell suspension for compounds 1-5. Furthermore, the species difference detected in plants was considered to be reflected in this test system, with respect to both the nature of phase I and phase II metabolites formed and the amount of parent compounds remained. Especially, the cabbage leaf cell suspension clearly demonstrated the high efficiency of xenobiotic catabolism showing quantitative transformation of the substrates within a day. From this fact, it can be assumed that this system will be an effective tool for the research on metabolic features in whole plant.

CA 6.2.2 Poultry

In the framework of the original inclusion into Annex I according to Directive 91/414/EEC a metabolism study in laying hens (AL-440-021) was peer-reviewed. The characteristics of this study are summarized in Table 6.2.2-1.

Table 6.2.2-1: Metabolism study in laying hen previously available

Group	Species	Label position	No of animal	Application details		Sample details		Year	DocID
				Rate (mg/kg bw per d)	Duration (days)	Commodity	Time		
Laying poultry	Hens	Benzyl ring-U-^{14}C Cycloprop ane-l-^{14}C	32 (4 groups of 8 hens)	7.5-7.75 (low dose) 17.13-18.01 (high dose)	14	Eggs	Daily during the dosing period	2001	AL-440-021
						Excreta			
						Tissues	22 h after last administration		

In laying hens, the major part of the radioactivity was excreted with urine (up to 93% of the total administered dose). The highest residue levels were found in liver and fat (skin+abdominal) both for the 2 dosing groups. Extractability of the residues with various solvents and after enzymatic digestion exceeded 90% of the TRR in the edible tissues and eggs.
Residue levels in eggs sampled at the high dose levels reached a plateau after 6 days (0.092 mg/kg) and 8 days (0.053 mg/kg) respectively for cyclopropane and benzyl labelling forms at the high dose. The major part of the extractable residues could be identified (at least 75% of the TRR except in liver for which the identification rate was rather low and ranged between 13% and 43% of the TRR). Characterization and identification of the radioactive residues in the different matrices revealed that parent compound was the predominant component in fat (skin+abdominal) and in eggs (at least 70% of the TRR). In liver, the metabolite cis-DCVA was recovered as the major metabolite for the cyclopropane labelling form (30% of the TRR) while a major amount of the radioactive residues remained unidentified (35 to 55% of the TRR).
Interconversion of cis and trans isomers didn't occur neither in the eggs nor tissues.

In order to confirm the results as shown above and to further clarify the remaining unidentified amounts with state-of-the-art analytic technology, it was decided to conduct a new hen metabolism study according to current guideline requirements.

Report:	CA 6.2.2/1 [redacted] 2014a The Metabolism of [14C]-Reg. No. 4078193 (BAS 310 I) in Laying Hens 2014/1140293
Guidelines:	EPA 860.1300: EPA Residue Chemistry Test Guidelines,EPA 860.1300: Nature of the Residue in Plants Livestock,EPA 860.1000: EPA Residue Chemistry Test Guidelines,EPA 860.1000: Background - PMRA Section 97.2 (Canada): Residue Chemistry Guidelines: Plants and Lifestock (June 1997), JMAFF 59 NohSan No 4200,BBA IV 3-2,EEC 7028/VI/95 rev. 3 Appendix A (EU): Metabolism and distribution in plants (draft of 22 July 1997)
GLP:	Yes (laboratory certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Description:	Alpha-cypermethrin (BAS 310 I)
Lot/Batch #:	775-0401 (benzyl-U-^{14}C) 1025-1034 (phenoxy-1,2,3,4,5,6-^{13}C) (for benzyl blend) 986-1046 (cyclopropane-1-^{14}C) 990-1024 (cyclopropane-1-^{13}C) L80-24 (unlabeled)
Purity:	96.1% (radiochemical), 4.94 MBq/mg (benzyl-U-^{14}C) 95.1% (phenoxy-1,2,3,4,5,6-^{13}C) 93.6% (radiochemical), 4.9 MBq/mg (cyclopropane-1-^{14}C) 97.7% (cyclopropane-1-^{13}C) 99.4% (unlabeled)
CAS#:	67375-30-8
Development code:	4078193
Stability of test compound:	The test item was stable for the test period

2. Test Animals

Species:	Hen
Variety:	Hy-line Brown
Gender:	Female
Age:	67-68 weeks
Weight at dosing:	1.714-2.317 kg
Number of animals:	20 (10 per label)
Acclimation period:	Min. 11 days
Diet:	Non-medicated feed ("Layer Mash"), *ad libitum*
Water:	Water, *ad libitum*
Housing:	Individually in metabolic cages

Environmental conditions -
Temperature: 17-24°C
Humidity: 12-60% (The low humidity results recorded are of no welfare concern to the birds as they had ad libitum access to water)
Photoperiod: 16 h light / 8 h dark

B. STUDY DESIGN AND METHODS

1. Dosing regime

Oral:
Amount of dose: 12 mg/kg feed/day (nominal)
Food consumption: 25-186 g food (dry weight)/animal/day
Vehicle: Gelatin capsule by oral gavage
Timing: Once daily
Duration: 14 days

2. Sample collection

Egg collection: Twice daily
Excreta collection: Daily
Interval from last dose to sacrifice: 6-7 h
Tissues harvested & analyzed: Eggs, liver, breast / thigh muscle, fat

3. Test system

The metabolism and distribution of alpha-cypermethrin was investigated in ten laying hens per label following a repeated oral administration of benzyl-U-^{14}C- or cyclopropane-1-^{14}C-BAS 310 I at a dose level of 12 mg/kg feed for 14 consecutive days. The test item was prepared in gelatin capsules and administered orally by gavage. The mean daily dose administered was 14.8 (benzyl label) and 14.0 mg/kg food consumed (dry weight equivalent) (cyclopropane label). The values exclude the final dose as it is not representative of a full 24 h cycle. Details of the study outline are summarized in Table 6.2.2-2.

Table 6.2.2-2: Dosing of laying hens with ^{14}C-alpha-cypermethrin

Animal	Treatment days	Nominal daily dose	Actual daily dose (mean)	
		mg/kg feed intake	mg/kg feed intake[1]	mg/kg bw[2]
Benzyl-U-^{14}C label				
11-20	14	12	13.0-17.8	0.81-1.04
Cyclopropane-1-^{14}C label				
1-10	14	12	12.4-16.5	0.74-1.00

1 Mean excludes day 14
2 Based on mean body weight on study days 2, 4 and 14 for animals 1-10 and on study days 1 and 14 for animals 11-20 as well as on mean total test item in capsule (1.82 mg for animals 11-20 and 1.73 mg for animals 1-10)

4. Sampling and Storage

During the dosing period eggs were collected at least twice daily (am and pm). Eggs collected in the afternoon of each day during the dosing period were stored in a fridge set to maintain a temperature of 4°C prior to analysis, eggs collected in the morning were immediately analyzed.
Excreta was collected prior to the first dose administration and at 24 h intervals thereafter until the sacrifice time. After processing and analysis, subsamples of excreta were taken and frozen at -20°C. For each label samples were pooled for further analysis on a daily basis.
Approximately 6-7 h after administration of the final dose, the hens were killed. The carcasses were plucked and the following samples removed: liver, omental fat, renal fat, subcutaneous fat with skin attached, breast muscle, thigh muscle, whole blood, partially formed eggs, GI tract and carcass. Any whole eggs still in the oviduct at sacrifice were collected. The eggs with shell collected from the oviduct were combined with the Day 14 eggs. All other partially formed eggs present in the carcass were retained separately for each hen and analyzed only for determination of the total radioactive residue. The carcasses were retained but analysis was not required.

5. Description of analytical methods

For the determination of the measured TRR, homogenized material (tissues) was combusted by means of a sample oxidizer before LSC measurement. For samples not requiring combustion (eggs), radioactivity was directly determined by mixing with a suitable scintillation fluid and LSC analysis.
The homogenized samples of both labels (egg yolk, egg white (cyclopropane label only), liver, breast muscle and thigh muscle) were extracted with acetonitrile and water. The acetonitrile extract was thereafter extracted with isohexane (egg yolk, liver, breast muscle and thigh muscle), which resulted in an acetonitrile phase and an isohexane phase. Fat of both labels was extracted similarly, but with a mixture of acetonitrile and isohexane. The corresponding phases were separated after extraction.

The extracts and / or processed samples of the extracts were analyzed with HPLC in order to characterize, identify and quantify radioactive residues. Identification of metabolites was mainly based on co-chromatography experiments. An additional metabolite was identified via HPLC-MS of a purified water extract of liver (cyclopropane label). Further components were isolated and characterized by incubation with enzymes (e.g. pancreatin).

II. RESULTS AND DISCUSSION

A. TOTAL RADIOACTIVE RESIDUES (TRRs)

In the analytical phase of the study, the TRR was calculated by summarizing the extractable radioactive residue (ERR) and the residual radioactive residue after solvent extraction (RRR). The calculated TRR was then set to 100%. The amounts of radioactivity which were detected in matrices relevant for the metabolism investigations are summarized in Table 6.2.2-3. The highest concentrations were detected in liver (benzyl label: 0.279 mg/kg and cyclopropane label: 0.383 mg/kg). The concentrations in egg yolk (benzyl label: 0.141 mg/kg and cyclopropane label: 0.149 mg/kg) and fat (benzyl label: 0.150 mg/kg and cyclopropane label: 0.084 mg/kg) were somewhat lower compared to liver. The concentrations in egg white, breast and thigh muscle were significantly lower compared to the other matrices (from 0.008 mg/kg to 0.022 mg/kg; for egg white of the benzyl label the TRR calculated was not determined, the TRR measured was 0.004 mg/kg). Overall, the amount of radioactive residues in corresponding matrices of the benzyl label and the cyclopropane label was comparable.
The TRR of all matrices was also measured by combustion analysis. For all matrices, the TRR measured was similar to the TRR calculated.

Table 6.2.2-3: Total radioactive residues in edible matrices after dosing of laying hens with [benzyl-U-^{14}C]- or [cyclopropane-1-^{14}C]-alpha-cypermethrin

Matrix	TRR [mg/kg]			
	measured	calculated	measured	calculated
	Benzyl-U-^{14}C label		Cyclopropane-1-^{14}C label	
Egg yolk	0.152	0.141	0.163	0.149
Egg white	0.004	n.r.	0.009	0.008
Liver	0.288	0.279	0.369	0.383
Breast muscle	0.007	0.009	0.010	0.011
Thigh muscle	0.021	0.022	0.017	0.019
Fat	0.149	0.150	0.082	0.084

TRR: Total radioactive residue
Calculated: Sum of ERR + RRR
n.r. Not reported

B. EXTRACTION OF RESIDUES

Egg white of the benzyl label was not extracted, due to the low residue level. For all other matrices the major part of radioactivity was recovered for both labels in the acetonitrile extract and the acetonitrile phase, respectively. Thereby, the residues ranged from 38.7% (liver, benzyl label) to 84.5% of the TRR (thigh muscle, benzyl label). The amount of radioactivity in acetonitrile extracts / phases of corresponding matrices of the benzyl label and the cyclopropane label was largely comparable.

For egg yolk significant amounts of radioactivity were also recovered in the isohexane phase (benzyl label: 18.5% TRR and cyclopropane label: 23.3% TRR), which originated from the acetonitrile extract. For fat, which was extracted simultaneously with acetonitrile and isohexane, the residues in the isohexane phase (benzyl label: 41.4% TRR and cyclopropane label: 48.0% TRR) were comparable to the residues in the acetonitrile phase (benzyl label: 55.5% TRR and cyclopropane label: 50.3% TRR).

In the water extracts of egg yolk (both labels), egg white (cyclopropane label) and liver (both labels) the residues ranged from 13.1% to 25.9% of the TRR. The amount of radioactivity in the water extracts of breast and thigh muscle of both labels was lower (from 6.0% to 12.0% TRR). The amount of radioactivity in water extracts of corresponding matrices of the benzyl label and the cyclopropane label was comparable. Overall, the ERR of both labels ranged from 57.6% (egg white, benzyl label) to 99.1% TRR (fat, benzyl label).

Table 6.2.2-4: Extractability of edible matrices with solvents (acetonitrile, isohexane, water) after dosing of laying hens with [benzyl-U-^{14}C]- or [cyclopropane-1-^{14}C]-alpha-cypermethrin

Matrix	TRR (calculated) mg/kg	Solvent extract (ERR)		RRR	
		mg/kg	% TRR	mg/kg	% TRR
Benzyl-U-^{14}C label					
Egg yolk	0.141	0.120	85.0	0.021	15.0
Liver	0.279	0.181	64.7	0.099	35.3
Breast muscle	0.009	0.007	85.8	0.001	14.2
Thigh muscle	0.022	0.020	90.5	0.002	9.5
Fat	0.150	0.148	99.1	0.001	0.9
Cyclopropane-1-^{14}C label					
Egg yolk	0.149	0.118	78.8	0.032	21.2
Egg white	0.008	0.005	57.6	0.004	42.4
Liver	0.383	0.320	83.4	0.063	16.6
Breast muscle	0.011	0.006	59.6	0.004	40.4
Thigh muscle	0.019	0.013	69.0	0.006	31.0
Fat	0.084	0.083	98.9	0.001	1.1

TRR: Total radioactive residue (sum of ERR + RRR)
ERR Extractable radioactive residue (solvents: acetonitrile, isohexane, water)
RRR: Residual radioactive residue after solvent extraction (solvents: acetonitrile, isohexane, water)

C. IDENTIFICATION AND CHARACTERIZATION OF RESIDUES

A summary of all identified metabolites and their distribution in egg, muscle, fat and liver is given in Table 6.2.2-5 and Table 6.2.2-6.

The unchanged parent compound **BAS 310 I** was identified in all examined matrices (egg yolk and egg white, liver, breast muscle, thigh muscle and fat) of both labels (aside from liver of the cyclopropane label). Thereby, BAS 310 I was the most abundant component in egg yolk (benzyl: 0.059 mg/kg or 41.6% TRR, cyclopropane: 0.051 mg/kg or 34.3% TRR), thigh muscle (benzyl: 0.013 mg/kg or 60.5% TRR, cyclopropane: 0.004 mg/kg or 20.9% TRR) and fat (benzyl: 0.094 mg/kg or 62.6% TRR, cyclopropane: 0.038 mg/kg or 45.4% TRR) of both labels. Moreover, BAS 310 I was identified in egg white, liver and breast muscle.
Additionally metabolites M310I019, M310I001 and M310I003 were identified, which were all label-specific. **M310I019** (3-hydroxybenzoic acid) was identified in liver of the benzyl label (0.043 mg/kg or 15.2% TRR). **M310I001** corresponds to the cyclopropane moiety of BAS 310 I after hydrolysis of the ester bond. M310I001 was identified in all matrices of the cyclopropane label (aside from fat) and was the most abundant component in egg white (0.002 mg/kg or 18.1% TRR), liver (0.221 mg/kg or 57.7% TRR) and breast muscle (0.002 mg/kg or 23.7% TRR). In thigh muscle of the cyclopropane label the concentration of M310I001 was similar to that of BAS 310 I. In egg yolk of the cyclopropane label only small amounts of M310I001 were present. Metabolite **M310I003** results from hydroxylation of a methyl group of M310I001. M310I003 was identified in the cyclopropane label of egg white (0.001 mg/kg or 9.2% TRR) and liver (0.011 mg/kg or 2.9% TRR).
Aside from the identified compounds, three components were characterized in some matrices of both labels. One peak corresponds probably to a component with the molecular formula $C_{21}H_{18}Cl_2O_4$. The other two compounds are probably conjugates of phase I metabolites of BAS 310 I. Overall, from 0.006 mg/kg or 53.4% TRR (breast muscle, cyclopropane label) up to 0.143 mg/kg or 95.7% TRR (fat, benzyl label) of the ERR were identified and characterized. If applicable, the RRR was further analyzed via solubilization with enzymes, HPLC, partition experiments and / or SPE fractionation. Thereby, from 0.001 mg/kg or 5.7% TRR (thigh muscle, benzyl label) up to 0.004 mg/kg or 35.7% TRR (breast muscle, cyclopropane label) were characterized.

Table 6.2.2-5: Summary of identified and characterized residues in edible matrices of laying hens after dosing with [benzyl-U-^{14}C]-BAS 310 I

Components	Egg yolk		Liver		Breast muscle		Thigh muscle		Fat	
	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]
BAS 310 I	0.059	41.6	0.021	7.4	0.004	45.2	0.013	60.5	0.094	62.6
M310I019	-	-	0.043	15.2	-	-	-	-	-	-
Total identified	**0.059**	**41.6**	**0.063**	**22.6**	**0.004**	**45.2**	**0.013**	**60.5**	**0.094**	**62.6**
Total characterized	0.077	54.6	0.191	68.2	0.004	50.9	0.007	33.4	0.050	33.1
Total identified and/or characterized	**0.136**	**96.1**	**0.254**	**90.9**	**0.008**	**96.1**	**0.021**	**93.9**	**0.143**	**95.7**
Final residue	0.008	5.9	0.017	5.9	0.001	6.2	0.003	14.5	0.001	0.9
Grand total	**0.144**	**102.0**	**0.270**	**96.8**	**0.009**	**102.3**	**0.024**	**108.4**	**0.145**	**96.6**

Table 6.2.2-6: Summary of identified and characterized residues in edible matrices of laying hens after dosing with [cyclopropane-1-^{14}C]-BAS 310 I

Components	Egg yolk		Egg white		Liver		Breast muscle		Thigh muscle		Fat	
	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]
BAS 310 I	0.051	34.3	0.001	9.9	-	-	0.001	9.0	0.004	20.9	0.038	45.4
M310I001	0.005	3.2	0.002	18.1	0.221	57.7	0.002	23.7	0.003	16.8	-	-
M310I003	-	-	0.001	9.2	0.011	2.9	-	-	-	-	-	-
Total identified	**0.056**	**37.5**	**0.003**	**37.2**	**0.232**	**60.6**	**0.003**	**32.7**	**0.007**	**37.7**	**0.038**	**45.4**
Total characterized	0.076	50.7	0.005	55.7	0.119	30.9	0.006	56.4	0.009	45.9	0.038	44.8
Total identified and/or characterized	**0.132**	**88.2**	**0.008**	**93.0**	**0.351**	**91.5**	**0.009**	**89.1**	**0.016**	**83.6**	**0.076**	**90.3**
Final residue	0.005	3.0	<0.001	5.4	0.017	4.4	0.001	7.1	0.002	9.5	0.001	1.1
Grand total	**0.136**	**91.2**	**0.008**	**98.4**	**0.367**	**95.9**	**0.010**	**96.2**	**0.018**	**93.1**	**0.077**	**91.4**

1. Metabolic pathway

The proposed metabolic pathway of BAS 310 I in hen is shown in Table 6.2.2-1. Hydrolysis of the ester bond of BAS 310 I results in a cyclopropane and a phenoxybenzyl moiety of the molecule. The cyclopropane moiety corresponds to metabolite M310I001. Hydroxylation of a methyl group of M310I001 yields metabolite M310I003. Cleavage of the nitrile group of the phenoxybenzyl moiety and oxidation yields the postulated intermediate M310I011, which was identified in the rat metabolism study. Cleavage of M310I011 yields metabolite M310I019 (3-Hydroxybenzoic acid).

2. Enantiomer ratio

The parent compound BAS 310 I was isolated from egg yolk and fat extracts in order to determine the enantiomer ratio. For both labels, the enantiomer ratio of BAS 310 I in egg yolk and fat was approximately 1:1, similar to the ratio of the application solutions.

3. Storage stability

All matrices were extracted within a maximum of 120 days after sampling. Quantitative analyses of the acetonitrile phases / extracts and the acetone supernatants of the water extracts were carried out within a maximum of 147 days after sampling, aside from the liver acetonitrile phase of the benzyl label (504 days). However, an early analysis of the same liver acetonitrile phase led to a similar metabolite pattern as the late analysis. Hence, due to timely extraction and analysis of the matrices no additional storage stability investigations were necessary.

III. CONCLUSION

The highest total radioactive residue (TRR) was detected in liver (benzyl: 0.279 mg/kg and cyclopropane: 0.383 mg/kg). The concentrations in egg yolk and fat were somewhat lower compared to liver. The concentrations in egg white, breast and thigh muscle were significantly lower compared to the other matrices (below or equal to 0.022 mg/kg). Overall, the amount of radioactive residues in corresponding matrices of the benzyl label and the cyclopropane label was comparable.

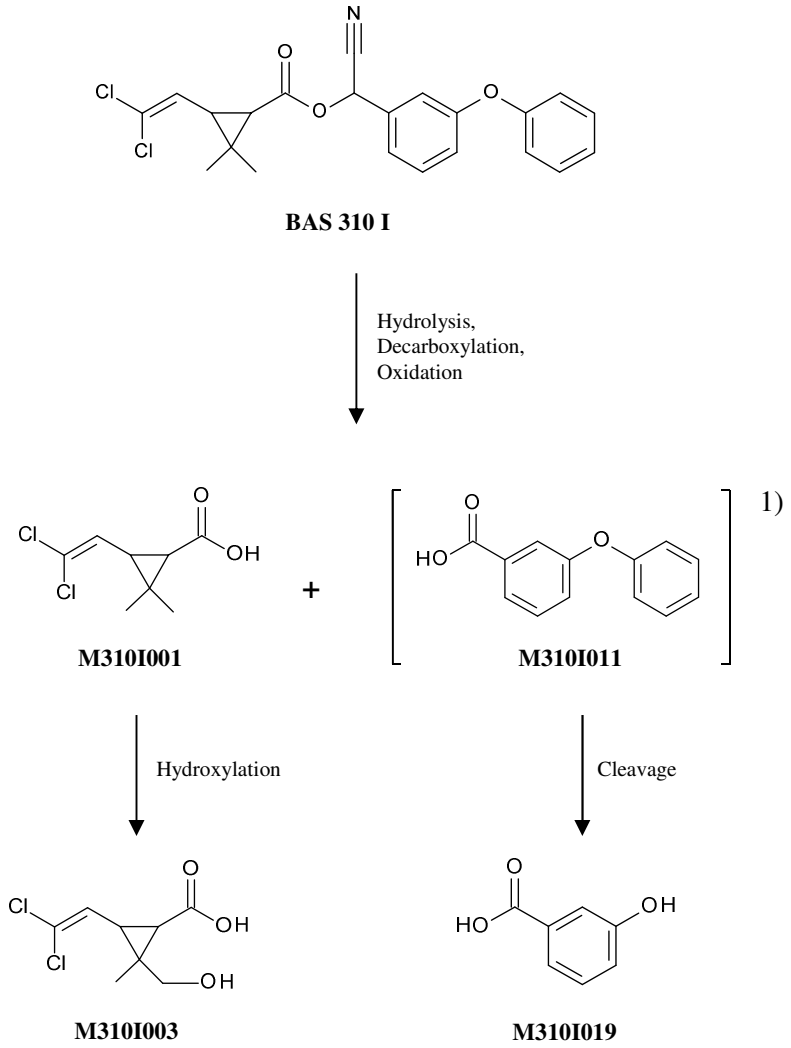
For all examined matrices, the major part of radioactivity was recovered for both labels in the acetonitrile extract and the acetonitrile phase, respectively. Thereby, the acetonitrile residues ranged from 38.7% (Liver, benzyl label) to 84.5% of the TRR (thigh muscle, benzyl label). For fat, which was extracted simultaneously with acetonitrile and isohexane, the residues in the isohexane phase were comparable to the residues in the acetonitrile phase. The amount of radioactivity in isohexane, acetonitrile and water extracts / phases of corresponding matrices of the benzyl label and the cyclopropane label was largely comparable. Overall, the ERR of both labels ranged from 57.6% (egg white, benzyl label) to 99.1% TRR (fat, benzyl label).

The unchanged parent compound BAS 310 I was identified in all examined matrices (egg yolk and egg white, liver, breast muscle, thigh muscle and fat) of both labels (aside from liver of the cyclopropane label). Thereby, BAS 310 I was the most abundant component in egg yolk (benzyl: 0.059 mg/kg or 41.6% TRR, cyclopropane: 0.051 mg/kg or 34.3% TRR), thigh muscle (benzyl: 0.013 mg/kg or 60.5% TRR, cyclopropane: 0.004 mg/kg or 20.9% TRR) and fat (benzyl: 0.094 mg/kg or 62.6% TRR, cyclopropane: 0.038 mg/kg or 45.4%TRR) of both labels. Moreover, BAS 310 I was identified in egg white, liver and breast muscle. Additionally metabolites M310I019, M310I001 and M310I003 were identified, which were all label-specific. M310I019 (3-hydroxybenzoic acid) was identified in liver of the benzyl label (0.043 mg/kg or 15.2% TRR). M310I001 corresponds to the cyclopropane moiety of BAS 310 I after hydrolysis of the ester bond. M310I001 was identified in all matrices of the cyclopropane label (aside from fat) and was the most abundant component in egg white (0.002 mg/kg or 18.1% TRR), liver (0.221 mg/kg or 57.7% TRR) and breast muscle (0.002 mg/kg or 23.7% TRR). In thigh muscle of the cyclopropane label the concentration of M310I001 was similar to that of BAS 310 I. In egg yolk of the cyclopropane label only small amounts of M310I001 were present. Metabolite M310I003 results from hydroxylation of a methyl group of M310I001. M310I003 was identified in the cyclopropane label of egg white (0.001 mg/kg or 9.2% TRR) and liver (0.011 mg/kg or 2.9 % TRR).

The parent compound BAS 310 I was also isolated from egg yolk and fat extracts in order to determine the enantiomer ratio. For both labels, the enantiomer ratio of BAS 310 I in egg yolk and fat was approximately 1:1, similar to the ratio of the application solutions.

Aside from the identified compounds, three components were characterized in some matrices of both labels. One peak corresponds probably to a component with the molecular formula $C_{21}H_{18}Cl_2O_4$. The other two compounds are probably conjugates of phase I metabolites of BAS 310 I. Overall, from 0.006 mg/kg or 53.4% TRR (breast muscle, cyclopropane label) up to 0.143 mg/kg or 95.7% TRR (fat, benzyl label) of the ERR were identified and characterized. If applicable, the RRR was further analyzed via solubilization with enzymes, HPLC, partition experiments and / or SPE fractionation. Thereby, from 0.001 mg/kg or 5.7% TRR (thigh muscle, benzyl label) up to 0.004 mg/kg or 35.7% TRR (breast muscle, cyclopropane label) were characterized.

Figure 6.2.2-1: Proposed metabolic pathway of BAS 310 I in hen



1) postulated intermediate (not detected)

CA 6.2.3 Lactating ruminants

In the framework of the original inclusion into Annex I according to Directive 91/414/EEC a metabolism study in lactating cow was peer-reviewed. The characteristics of this study are summarized in Table 6.2.3-1.

Table 6.2.3-1: Metabolism study in lactating cow previously available

Group	Species	Label position	No of animal	Application details		Sample details		Year	DocID
				Rate (mg/kg bw per d)	Dura-tion (days)	Commo-dity	Time		
Lactating ruminants	Cow	Benzyl-U-^{14}C	1 treated 1 control	0.49; overall calculated dietary concentration 19 mg/kg	4 days	Urine (excreted)	at 24 h intervals	1994	AL-440-014
						Faeces	at 24 h intervals		
						Milk	daily at ca. 09:00 and 15:00		
						Whole blood Plasma Liver Kidney Bile Fat Muscle Urine (bladder)	6 h after administration of last dose		

1 based on 250 mg/day and bw 510 kg

^{14}C-labelled alpha-cypermethrin was administered orally to a cow (overall calculated dietary concentration 19 mg/kg) via twice daily doses added to the diet. Whereas muscle, fat and milk mainly contained a single compound with similar chromatographic properties to alpha-cypermethrin (muscle 85%, fat 91%, milk 97%), the liver extract contained at least 8 metabolites with a broad range of polarities, one component (±16%) had similar chromatographic properties to those of alpha-cypermethrin. In urine, the 2 major components had identical chromatographic properties to N-(3-phenoxybenzoyl)glutamic acid and N-(3-phenoxybenzoyl)glycine, respectively. A minor component had identical chromatographic properties (HPLC) to 3-phenoxybenzoic acid (3%).

No metabolic pathway could be established as only the parent compound and some conjugates were identified. No further investigation was carried out to identify and quantify the other metabolites.

In order to further clarify the metabolic pathway in lactating ruminants with state-of-the-art analytic technology, it was decided to conduct a new goat metabolism study according to current guideline requirements.

Report:	CA 6.2.3/1 [redacted] 2014a The Metabolism of ^{14}C Reg. No. 4078193 (BAS 310 I) in Lactating Goats 2014/1083330
Guidelines:	EPA 860.1300: Nature of the Residue in Plants Livestock,EPA 860.1000,OECD Test Guideline 503 - Metabolism in livestock,EEC 91/414 (7030(VI/95 Rev. 3)
GLP:	Yes (laboratory certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Description:	Alpha-cypermethrin (BAS 310 I)
Lot/Batch #:	775-0401 (benzyl-U-^{14}C) 1025-1034 (phenoxy-1,2,3,4,5,6-^{13}C) (for benzyl blend) 986-2015 (cyclopropane-1-^{14}C) 990-1025 (cyclopropane-1-^{13}C) L80-24 (unlabeled)
Purity:	96.1% (radiochemical), 4.94 MBq/mg (benzyl-U-^{14}C) 95.1% (phenoxy-1,2,3,4,5,6-^{13}C) 98.9% (radiochemical), 4.7 MBq/mg (cyclopropane-1-^{14}C) 97.7% (cyclopropane-1-^{13}C) 99.4% (unlabeled)
CAS#:	67375-30-8
Development code:	4078193
Stability of test compound:	The test item was stable for the test period

2. Test Animals

Species:	Goat
Variety:	“Saanen cross Toggenburg” or pure "Saanan
Gender:	Female
Age:	Not reported
Weight at dosing:	40.0-68.5 kg
Number of animals:	4 (2 per label)
Acclimation period:	20 days
Diet:	2 x 0.5 kg non-medicated supplementary concentrate + hay *ad libitum*
Water:	Tap water, *ad libitum*
Housing:	Metabolic cages
Environmental conditions	
Temperature:	17-23°C
Humidity:	17-89%
Photoperiod:	12 h light / 12 h dark

B. STUDY DESIGN AND METHODS

1. Dosing regime

Oral:

Amount of dose:	12 mg/kg feed/day
Food consumption:	1.087-2.117 kg/animal/day
Vehicle:	Gelatin capsule by oral gavage
Timing:	Once daily
Duration:	7 days

2. Sample collection

Milk collection:	Twice daily
Urine and feces collection:	Daily
Interval from last dose to sacrifice:	5-8 h
Tissues harvested & analyzed:	Urine, feces, milk, bile, liver, kidney, muscle, fat

3. Test system

The metabolism and distribution of alpha-cypermethrin was investigated in two lactating goats per label following a repeated oral administration of benzyl-U-^{14}C-BAS 310 I or cyclopropane-1-^{14}C-BAS 310 I at a dose level of 12 mg/kg feed for seven consecutive days. By mistake within the preparation of the application solution of the cyclopropane label for the ^{13}C-labeled BAS 310 I, instead of alpha-cypermethrin (comprising the isomers (S)-cyano-(1R,3R) and (R)-cyano-(1S,3S)) a different pair of cis isomers of cypermethrin (comprising the isomers (S)-cyano-(1S,3S) and (R)-cyano-(1R,3R)) was used. Therefore, for the cyclopropane label, the actual dose of the applied alpha-cypermethrin was less than the target dose, and amounted to 8.55 mg/kg feed. This has no relevant impact on the results of the study. The test item was prepared in gelatin capsules and administered orally by gavage. Details of the study outline are summarized in Table 6.2.3-2.

Table 6.2.3-2: Dosing of lactating goats with ^{14}C-alpha-cypermethrin

Animal	Treatment days	Nominal daily dose	Actual daily dose		Sacrifice time after last dose (hours)
		mg/kg feed intake	mg/kg feed intake	mg/kg bw	
Benzyl-U-^{14}C label					
1+2	7	12	13.29-14.73	0.36-0.42	5
Cyclopropane-1-^{14}C label					
3+4	7	12	8.35-8.75	0.26-0.29	8

4. Sampling and storage

During the application period, aliquots of milk, urine and feces were taken for the determination of the radioactive residues twice daily for milk and daily for urine and feces. Tissues and organs (liver, kidney, muscle and fat samples) and bile were collected after animal sacrifice. The animals were killed approximately 5 h and 8 h after administration of the last dose at the time of the maximum residue concentration in plasma (t_{max}). All samples were stored at –18°C or below. Representative pool samples of urine and feces were combined for 24-144 h (benzyl label) and 24-156 h (cyclopropane label), respectively. A pool sample of milk was made for 55-96 h (cyclopropane label) and 103-127 h (benzyl and cyclopropane label). The pool samples of urine, feces and milk and tissue homogenates of liver, kidney, bile, muscle and fat were also analyzed for total radioactive residues.

5. Description of analytical methods

Aliquots of liquid samples (milk, urine) were directly analyzed after mixing with scintillation cocktail without additional treatment. Fat samples were treated with tissue solubilizer and bleached before mixing with scintillator and LSC measurement. The total radioactivity in solid samples (feces and tissues) was determined by combustion.
The homogenized feces and tissue samples of both labels were extracted with acetonitrile and water. Milk samples (103-127 h) were extracted with a mixture of acetonitrile and isohexane. Milk pool sample (55-96 h) of the cyclopropane label was extracted with acetonitrile. Fat samples were extracted with a mixture of acetonitrile and isohexane followed by extraction with water. The extracts were subsequently pooled and aliquots were taken for LSC measurement. The residues were combusted for determination of the radioactive residues.
The residues after solvent extraction of liver (both labels) and kidney (cyclopropane label) were further solubilized with pronase.
HPLC analysis was carried out for samples with a sufficient level of radioactivity. In some cases, residues after solvent extraction were further solubilized and investigated by HPLC. Identification and assignment of metabolites was based on HPLC-MS investigations of urine and a cleaned up bile sample (benzyl label), on co-chromatography experiments, comparison of the retention times and on comparison of metabolite patterns of the extracts /solubilizates investigated with those of reference items. The quantitation of the parent compound and its metabolites is generally based on the HPLC analyses. Another HPLC method was used for the determination of the isotope pattern.
For metabolite investigations, homogenized subsamples of feces, liver, kidney and muscle were extracted with acetonitrile and water. Subsamples of pooled milk were extracted with acetonitrile (55-96 h, cyclopropane label) or with a mixture of acetonitrile and isohexane (103-127 h, both labels). Fat samples were extracted with a mixture of acetonitrile and isohexane and subsequently with water.

II. RESULTS AND DISCUSSION

A. TOTAL RADIOACTIVE RESIDUES (TRRs)

The total radioactive residues (TRR) in all matrices are summarized in Table 6.2.3-3. Rapid excretion of the radioactive residues was observed. Until sacrifice, the radioactive residues excreted via urine and feces amounted to 72.9% (benzyl label) and 74.3% (cyclopropane label) of the total radioactivity administered.

The total radioactive residues (TRR) determined in pooled urine samples (24-144 h, benzyl label and 24-156 h, cyclopropane label) of goats treated with BAS 310 I were 1.055 mg/kg (benzyl label) and 10.903 mg/kg (cyclopropane label). In feces, 7.635 mg/kg and 10.779 mg/kg were found for the benzyl and the cyclopropane label, respectively.

The total radioactive residues measured in edible tissues and organs of the benzyl and cyclopropane label were 0.086 mg/kg and 0.489 mg/kg for liver and 0.058 mg/kg and 0.345 mg/kg for kidney. For the benzyl label, samples of omental, renal and subcutaneous fat were analyzed separately and radioactive residues amounted to 0.024 mg/kg, 0.031 mg/kg and 0.011 mg/kg, respectively. For the cyclopropane label composite fat was analyzed for both of the goats separately and radioactive residues amounted to 0.049 mg/kg and 0.141 mg/kg, respectively. Residue levels detected in muscle were 0.004 mg/kg for the benzyl label and 0.029 mg/kg for the cyclopropane label. In the pooled samples of milk, residue levels of 0.010 mg/kg (103-127 h, benzyl label), 0.053 mg/kg (55-96 h, cyclopropane label) and 0.044 mg/kg (103-127 h, cyclopropane label) were found. Bile contained 0.606 mg/kg (benzyl label) and 2.290 mg/kg (cyclopropane label). Overall milk, muscle and fat showed low residue levels.

Table 6.2.3-3: Total radioactive residues in edible matrices and excreta after dosing of lactating goats with [benzyl-U-^{14}C]- or [cyclopropane-1-^{14}C]-alpha-cypermethrin

Matrix	TRR [mg/kg]	
	measured	calculated
Benzyl-U-^{14}C label		
Milk[1]	0.010	0.012
Bile	0.606	-
Liver	0.086	0.083
Kidney	0.058	0.059
Fat (omental)	0.024	0.025
Fat (renal)	0.031	0.033
Fat (subcutaneous)	0.011	0.013
Muscle (loin and flank)	0.004	0.005
Urine[2]	1.055	-
Feces[2]	7.635	7.205
Cyclopropane-1-^{14}C label		
Milk[3]	0.053	0.060
Milk[1]	0.044	0.049
Bile (goat 4)	2.290	-
Liver	0.489	0.524
Kidney	0.345	0.340
Fat (composite; goat 3)	0.049	0.050
Fat (composite; goat 4)	0.141	0.144
Muscle (composite)	0.029	0.032
Urine[2]	10.903	-
Feces[2]	14.455	10.779

TRR: Total radioactive residue (sum of ERR + RRR)
1 Milk pool sample 103-127 h
2 Total of urine and feces pool samples 24-156 h
3 Milk pool sample 55-96 h

B. EXTRACTION OF RESIDUES

The extractability was generally high and the portions of extractable radioactive residues (ERR) were equal to or above 89.9% TRR. Only for liver (both labels), kidney (cyclopropane label), and muscle (benzyl label), somewhat lower portions of radioactive residues were extracted by solvent extraction (61.1-85.4% TRR).

For most extractions, the major part of radioactive residues was found in the acetonitrile extract, while lower amounts of radioactive residues were detected in the isohexane phase after partition (4.5-11.9% TRR). Only for subcutaneous fat of the benzyl label a somewhat higher portion was in the isohexane phase (23.6% TRR). Extraction of liver and kidney yielded also some amounts of radioactive residues in the water extracts (18.4-36.3% TRR).

Due to the very low amounts of residual radioactive residues (RRR) in milk, muscle and fat (≤0.005 mg/kg), the residues after solvent extraction were not further investigated. Additional solubilization was performed the case of liver (both labels) and kidney (cyclopropane label). The respective residues after solvent extraction (RRR) were incubated with pronase, which solubilized additional 20.3% TRR from liver of the benzyl label, 38.9% TRR from liver of the cyclopropane label and 14.0% TRR from kidney of the cyclopropane label.

Table 6.2.3-4: Extractability of edible matrices and feces with solvents (acetonitrile, isohexane, water) after dosing of lactating goats with [benzyl-U-^{14}C]- or [cyclopropane-1-^{14}C]-alpha-cypermethrin

Matrix	TRR (calculated) mg/kg	Solvent extract (ERR)		RRR	
		mg/kg	% TRR	mg/kg	% TRR
Benzyl-U-^{14}C label					
Milk (103-127 h)	0.012	0.012	111.1	0.001	7.8
Liver	0.083	0.056	65.4	0.027	31.4
Kidney	0.059	0.052	90.0	0.007	12.9
Fat (omental)	0.025	0.025	102.9	<0.001	1.5
Fat (renal)	0.033	0.033	105.9	0.001	2.2
Fat (subcutaneous)	0.013	0.013	112.2	<0.001	0.8
Muscle	0.005	0.004	85.4	0.002	37.2
Feces	7.205	6.866	89.9	0.339	4.4
Cyclopropane-1-^{14}C label					
Milk (55-96 h)	0.060	0.049	92.3	0.011	19.8
Milk (103-127 h)	0.049	0.045	101.2	0.004	8.8
Liver	0.524	0.299	61.1	0.226	46.2
Kidney	0.340	0.287	83.1	0.053	15.3
Composite fat (goat 3)	0.050	0.049	100.4	<0.001	1.0
Composite fat (goat 4)	0.144	0.143	101.4	0.001	0.9
Muscle	0.032	0.028	97.5	0.003	11.7
Feces	10.779	10.475	97.2	0.305	2.8

TRR: Total radioactive residue (sum of ERR + RRR)
ERR: Extractable radioactive residue (solvents: acetonitrile, isohexane, water)
RRR: Residual radioactive residue after solvent extraction

C. IDENTIFICATION AND CHARACTERIZATION OF RESIDUES

A summary of all identified metabolites and their distribution in excreta, bile, milk, muscle, fat, liver, and kidney is given in Table 6.2.3-5 to Table 6.2.3-8.

The unchanged parent compound was the main constituent in extracts from feces, milk, fat and muscle, representing 51.9-92.6% TRR. In addition BAS 310 I was found in liver and kidney of both labels at <5.0% TRR.

In detail, **BAS 310 I** was detected in extracts of the benzyl label at 7.069 mg/kg (92.6% TRR) in feces and at 0.005 mg/kg (51.9% TRR) in milk pooled from 103-127 h. The parent compound amounted to 0.019 mg/kg (77.3% TRR) in omental fat, 0.026 mg/kg (83.8% TRR) in renal fat and 0.009 mg/kg (74.5% TRR) in subcutaneous fat.
For the cyclopropane label BAS 310 I was likewise detected at 7.284 mg/kg (67.6% TRR) in feces, 0.043 mg/kg (80.5% TRR) in milk (55-96 h) and 0.033 mg/kg (74.0% TRR) in milk (103-127 h). In composite fat it was detected at 0.035 mg/kg (71.2% TRR) for goat 3 and at 0.127 mg/kg (89.8% TRR) for goat 4. BAS 310 I was furthermore detected at 0.019 mg/kg (65.6% TRR) in composite muscle.

In urine and kidney, high abundances were found for the label-specific metabolites **M310I010**, **M310I011** (benzyl label) and **M310I001**, **M310I003** and **M310I004** (cyclopropane label).
For the benzyl label M310I010 was found at amounts of 0.729 mg/kg (69.1% TRR) in urine and 0.022 mg/kg (38.0% TRR) in kidney. M310I011 accounted for 0.176 mg/kg (16.7% TRR) in urine and 0.010 mg/kg (16.6% TRR) in kidney. M310I010 was also detected in milk (0.003 mg/kg or 29.6% TRR). Both label-specific metabolites M310I010 and M310I011 were detected at low amounts in the liver acetonitrile extract as well as in the liver pronase solubilizate (together 5.3% and 4.6% TRR, respectively).
For the cyclopropane label M310I001, M310I003 and M310I004 accounted for 2.099 mg/kg (19.3% TRR), 1.974 mg/kg (18.1% TRR) and 4.584 mg/kg (42.1% TRR) in urine and for 0.033 mg/kg (9.6% TRR), 0.012 mg/kg (3.5% TRR) and 0.058 mg/kg (16.8% TRR) in kidney. Metabolite M310I001 was also detected at lower amounts in feces, liver, fat and muscle (all <4.3% TRR). M310I004 was also found in bile (0.226 mg/kg, 9.9% TRR) and liver at 0.012 mg/kg (2.5% TRR).

The metabolite **M310I021** was the most prominent component detected in bile of both labels, accounting for 0.156 mg/kg (25.7% TRR) for the benzyl label and 1.121 mg/kg (48.9% TRR) for the cyclopropane label. M310I021 was also detected in liver of both labels, where it was one of the most abundant components (4.7% and 7.4% TRR).

Additionally, the metabolites **M310I015** and **M310I017** were detected in feces of the cyclopropane label at minor concentrations (3.3% TRR and 4.1% TRR, respectively).

A number of additional HPLC peaks in the extracts was not identified, but characterized based on their chromatographic properties. For milk and the edible tissues of both labels the characterized components were always below 0.010 mg/kg or 10% of the TRR.

Table 6.2.3-5: Summary of identified and characterized residues in excreta and bile of lactating goats after dosing with [benzyl-U-^{14}C]-BAS 310 I

Components	Urine		Feces		Bile	
	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]
BAS 310 I	-	-	7.069	92.6	-	-
M310I010	0.729	69.1	-	-	-	-
M310I011	0.176	16.7	-	-	-	-
M310I021	-	-	-	-	0.156	25.7
Total identified	**0.905**	**85.8**	**7.069**	**92.6**	**0.156**	**25.7**
Total characterized	0.150	14.2	0.459	6.0	0.450	74.3
Total identified and/or characterized	**1.055**	**100.0**	**7.528**	**98.6**	**0.606**	**100.0**
Final residue	-	-	0.339	4.4	-	-
Grand total	**1.055**	**100.0**	**7.866**	**103.0**	**0.606**	**100.0**

Table 6.2.3-6: Summary of identified and characterized residues in edible matrices of lactating goats after dosing with [benzyl-U-^{14}C]-BAS 310 I

Components	Milk (103-127 h)		Liver		Kidney		Omental fat		Renal fat		Subcutaneous fat	
	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]
BAS 310 I	0.005	51.9	0.004	4.8	0.003	4.9	0.019	77.3	0.026	83.8	0.009	74.5
M310I010	0.003	29.6	0.003	3.2	0.022	38.0	-	-	-	-	-	-
M310I011	-	-	0.002	1.8	0.010	16.6	-	-	-	-	-	-
M310I021	-	-	0.006	7.4	-	-	-	-	-	-	-	-
Total identified	**0.009**	**81.5**	**0.015**	**17.2**	**0.034**	**59.6**	**0.019**	**77.3**	**0.026**	**83.8**	**0.009**	**74.5**
Total characterized	0.002	19.8	0.054	62.8	0.014	23.9	0.006	24.5	0.007	22.6	0.004	36.0
Total identified and/or characterized	**0.011**	**101.3**	**0.069**	**80.0**	**0.048**	**83.5**	**0.025**	**101.9**	**0.033**	**106.4**	**0.013**	**110.5**
Final residue	0.001	7.8	0.007	8.4	0.007	12.9	<0.001	1.5	0.001	2.2	<0.001	0.8
Grand total	**0.011**	**109.1**	**0.076**	**88.4**	**0.056**	**96.4**	**0.025**	**103.3**	**0.034**	**108.6**	**0.013**	**111.3**

Table 6.2.3-7: Summary of identified and characterized residues in excreta and bile of lactating goats after dosing with [cyclopropane-1-^{14}C]-BAS 310 I

Components	Urine		Feces		Bile	
	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]
BAS 310 I	-	-	7.284	67.6	-	-
M310I001	2.099	19.3	0.090	0.8	-	-
M310I003	1.974	18.1	-	-	-	-
M310I004	4.585	42.1	-	-	0.226	9.9
M310I015	-	-	0.353	3.3	-	-
M310I017	-	-	0.444	4.1	-	-
M310I021					1.121	48.9
Total identified	**8.658**	**79.4**	**8.171**	**75.8**	**1.347**	**58.8**
Total characterized	2.245	20.6	2.304	21.4	0.943	41.2
Total identified and/or characterized	**10.903**	**100.0**	**10.475**	**97.2**	**2.290**	**100.0**
Final residue	-	-	0.305	2.8	-	-
Grand total	**10.903**	**100.0**	**10.779**	**100.0**	**2.290**	**100.0**

Table 6.2.3-8: Summary of identified and characterized residues in edible matrices of lactating goats after dosing with [cyclopropane-1-^{14}C]-BAS 310 I

Components	Milk (55-96 h)		Milk (103-127 h)		Liver		Kidney		Composite fat (Goat 3)		Composite fat (Goat 4)		Composite muscle	
	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]	[mg/kg]	[% TRR]
BAS 310 I	0.043	80.5	0.033	74.0	0.013	2.7	0.004	1.2	0.035	71.2	0.127	89.8	0.019	65.6
M310I001	-	-	-	-	0.021	4.2	0.033	9.6	0.001	2.9	0.003	1.9	0.001	4.1
M310I003	-	-	-	-	-	-	0.012	3.5	-	-	-	-	-	-
M310I004	-	-	-	-	0.012	2.5	0.058	16.8	-	-	-	-	-	-
M310I021	-	-	-	-	0.023	4.7	-	-	-	-	-	-	-	-
Total identified	**0.043**	**80.5**	**0.033**	**74.0**	**0.069**	**14.1**	**0.108**	**31.1**	**0.036**	**74.0**	**0.129**	**91.8**	**0.020**	**69.7**
Total characterized	0.006	10.9	0.004	8.8	0.333	68.1	0.208	60.1	0.012	25.0	0.009	6.6	0.005	16.5
Total identified and/or characterized	**0.049**	**91.4**	**0.037**	**82.7**	**0.402**	**82.2**	**0.316**	**91.2**	**0.049**	**99.0**	**0.139**	**98.4**	**0.025**	**86.2**
Final residue	0.005	9.0	0.004	8.8	0.042	8.7	0.009	2.7	<0.001	1.0	0.001	0.9	0.003	11.7
Grand total	**0.054**	**100.4**	**0.041**	**91.6**	**0.444**	**90.9**	**0.326**	**94.4**	**0.049**	**100.0**	**0.140**	**99.3**	**0.029**	**97.9**

1. Metabolic pathway

The proposed metabolic pathway of BAS 310 I in goat is shown in Figure 6.2.3-1. Metabolism of ^{14}C-BAS 310 I in the lactating goat proceeds via hydroxylation and conjugation of the phenoxybenzyl and / or cyclopropyl moiety of the molecule (metabolites M310I015, M310I017 and M310I021). A further metabolic route is the cleavage of the ester bond and loss of the nitrile group leading to the label specific metabolites M310I001 and M310I011. Hydroxylation or conjugation of the latter cleavage products with glycine or glucuronic acid leads to the metabolites M310I003, M310I004 and M310I010.

2. Enantiomer ratio

In order to analyze whether one isomer of alpha-cypermethrin was preferably metabolized in goats, enantiomer-specific analysis of the parent compound BAS 310 I, isolated from selected matrices, was performed. For both labels, matrix-specific differences were observed for the isomer ratio. While in feces the ratio of both isomers of BAS 310 I was found to be approximately 1:1 for both labels, the relative amount of the (S)-cyano-(1R,3R) (isomer 1) was lower compared to the (R)-cyano-(1S,3S) (isomer 2) in the other investigated matrices (milk and renal fat in the case of the benzyl label, milk, composite fat and composite muscle in the case of the cyclopropane label). The relative amounts of isomer 1 : isomer 2 ranged from values of 16.0% : 84.0% to 22.2% : 77.8%.

3. Storage stability

Investigations of storage stability were performed in selected goat matrices at the beginning and at the end of the study. For this purpose, initial analyses of the extracts were carried out within six months after sampling. Re-analyses of stored extracts and re-extractions were performed, namely for milk (benzyl label, investigated due to a further metabolite in comparison to the cyclopropane label), liver and kidney (both labels, due to different metabolite patterns) and muscle (cyclopropane label, only investigated for this label).
For milk (benzyl label), liver, kidney (both labels) and composite muscle (cyclopropane label), the chromatograms obtained from stored extracts and re-extracted samples were in good accordance with the initial analysis. For liver of the cyclopropane label the chromatograms from stored extracts and re-extracted samples agreed likewise well with the peak pattern of the initial extraction with the exception of the metabolite M310I021, a glucuronic acid conjugate, which was less prominent in the re-extracted as well as re-analyzed sample.
Altogether, stability of residues was demonstrated in milk, liver and kidney of the benzyl label for a storage interval of at least 584 days in stored matrix and for at least 439 days in stored extracts. For liver, kidney and composite muscle of the cyclopropane label stability of residues was demonstrated for at least 368 days in stored matrix and for at least 287 days in stored extracts.

III. CONCLUSION

The metabolism of ^{14}C-BAS 310 I (benzyl and the cyclopropane labeled test substance) was investigated in lactating goats. The test substance was fed to the animals at a nominal dose level of 12 mg/kg feed for seven consecutive days. During the application period, aliquots of milk, urine and feces were taken for the determination of the radioactive residues. Tissues and organs (liver, kidney, muscle and fat samples) and bile were collected after animal sacrifice.
For the benzyl label the major route of excretion of radioactivity was via the feces, amounting to 59.9% of the total administered dose while, 13.1% of the total administered dose were excreted via urine. For the cyclopropane label 32.5% of the total administered dose were excreted via feces and 41.8% via urine.
In the pooled urine samples 1.055 mg/kg (24-144 h, benzyl label) and 10.903 mg/kg (24-156 h, cyclopropane label) were found. In the pooled feces samples radioactive residues amounted to 7.635 mg/kg (24-144 h, benzyl label) and 10.779 mg/kg (24-156 h, cyclopropane label).
The pooled milk samples represented low residue levels of 0.010 mg/kg (103-127 h, benzyl label), 0.053 mg/kg (55-96 h, cyclopropane label) and 0.044 mg/kg (103-127 h, cyclopropane label).The total radioactive residues (TRR) in organs for the benzyl and cyclopropane label amounted to 0.086 mg/kg and 0.489 mg/kg for liver, and 0.058 mg/kg and 0.345 mg/kg for kidney. Bile contained 0.606 mg/kg (benzyl label) and 2.290 mg/kg (cyclopropane label). For the benzyl label the TRR in edible tissues amounted to 0.024 mg/kg for omental fat, 0.031 mg/kg for renal fat, 0.011 mg/kg for subcutaneous fat and 0.004 mg/kg for loin and flank muscle. Considering the cyclopropane label, the TRR in edible tissues was 0.049 mg/kg for composite fat (goat 3), 0.141 mg/kg for composite fat (goat 4), and 0.029 mg/kg for composite muscle.

The extractability of the radioactive residues was high for both labels (≥89.9% TRR). Only for liver (both labels), muscle (benzyl label) and kidney (cyclopropane label) extractability was somewhat lower (between 61.1% and 85.4% TRR). Muscle extract of the benzyl label was not further investigated due to the low residue levels (0.004 mg/kg). The predominant part of the radioactive residues was extracted with acetonitrile (or a mixture of acetonitrile and isohexane) (>85% TRR), just from liver and kidney higher portions were extracted with water (between 18.4% and 36.3% TRR). The residual radioactive residues (RRR) after solvent extraction of liver and kidney were further solubilized with pronase, resulting in an additional release of 14.0-38.9% TRR.

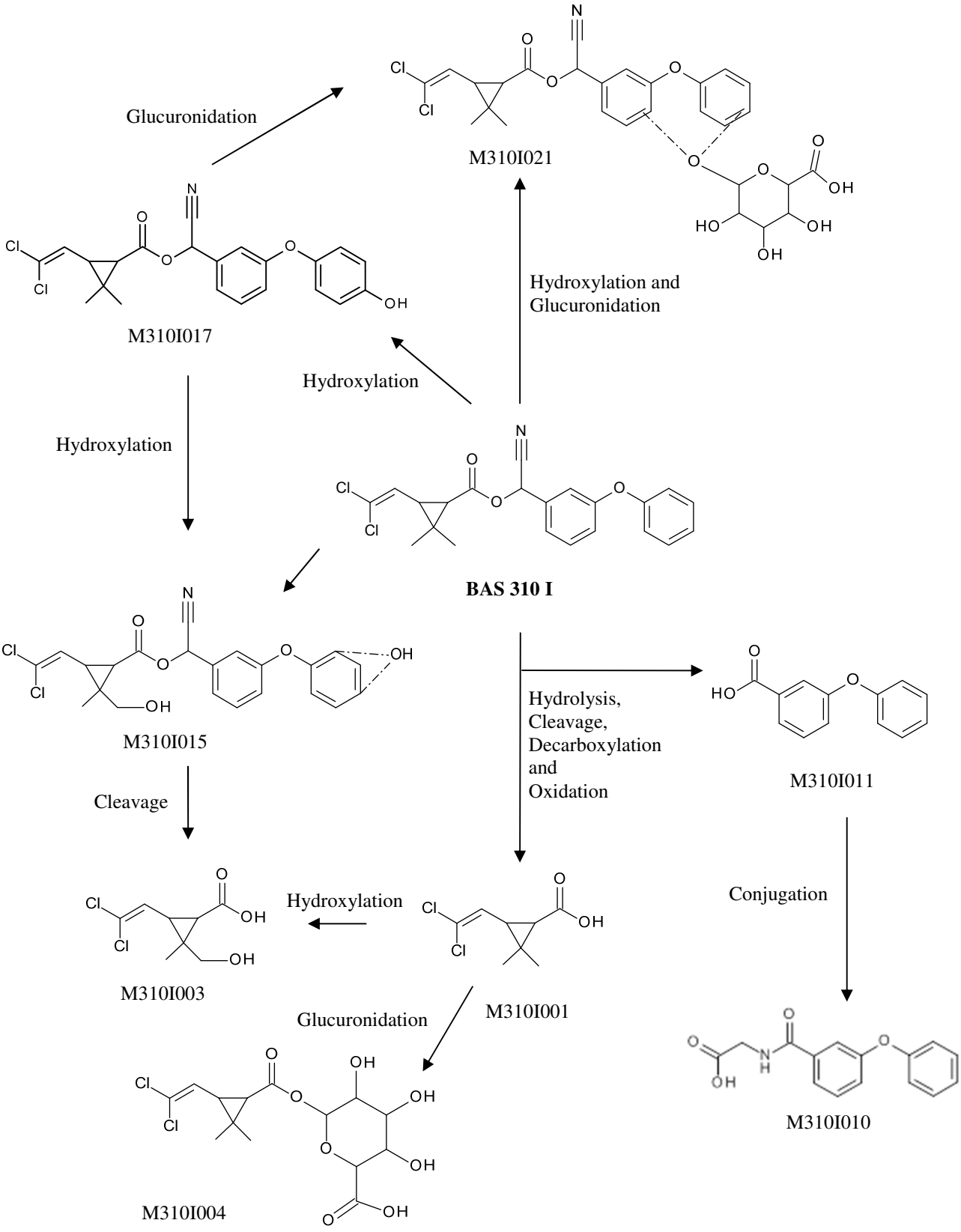
The unchanged parent compound was the main component in feces, milk, fat and muscle of both labels, representing 51.9-92.6% TRR.
High amounts of label-specific metabolites were detected in kidney and urine. For the cyclopropane label these were the metabolites M310I001 (9.6-19.3% TRR), M310I003 (3.5-18.1% TRR) and M310I004 (16.8-42.1% TRR) and for the benzyl label the metabolites M310I010 (38.0-69.1% TRR) and M310I011 (16.6-16.7% TRR).
The benzyl label specific metabolite M310I010 was also detected in milk (29.6% TRR) and liver (5.3% TRR) and label specific M310I011 at low amounts in liver (4.6% TRR).
For the cyclopropane label M310I001 was also detected in low amounts in fat and muscle (≤4.1% TRR) and M310I004 in liver and bile (2.5% and 9.9% TRR).
Further detected metabolites include M310I021 in bile (25.7-48.9% TRR) and liver (4.7-7.4% TRR) of both labels as well as M310I015 and M310I017, detected in feces of the cyclopropane label at minor concentrations (3.3% TRR and 4.1% TRR, respectively).

Several further components were characterized by their HPLC elution behavior and were below 10% TRR in all edible matrices.

Metabolism of ^{14}C-BAS 310 I in lactating goat proceeds via hydroxylation and conjugation of the phenoxybenzyl and cyclopropyl moieties of the molecule (metabolites M310I015, M310I017 and M310I021). A further metabolic route is the cleavage of the ester bond and loss of the nitrile group leading to the metabolites M310I001 and M310I011. Hydroxylation or conjugation of the latter compounds with glycine or glucuronic acid leads to the metabolites M310I003, M310I004 and M310I010. The main biotransformation reactions were also observed in rats and laying hens, so there is a consistent picture of the metabolism of ^{14}C-BAS 310 I in all animal species investigated.

In order to analyze if one enantiomer of alpha-cypermethrin was preferably metabolized in goats, enantiomer-specific analysis of the parent compound BAS 310 I, isolated from selected matrices, was performed. For feces of both labels analysis resulted in an enantiomer ratio of approximately 1:1, while the relative amount of one enantiomer was lower in the other investigated matrices (milk and renal fat in case of the benzyl label, milk, composite fat and composite muscle in case of the cyclopropane label) compared to the other enantiomer. The relative amount ranged from values from 16.0% to 22.3% TRR of the (S)-cyano-(1R,3R) enantiomer in comparison to relative amounts between 77.8% and 84.0% TRR of the (R) cyano-(1S,3S) enantiomer.

Figure 6.2.3-1: Proposed metabolic pathway of BAS 310 I in goat



CA 6.2.4 Pigs

No metabolism study was performed in pigs, since the metabolite patterns in rodents (rats) and ruminants (goats) did not differ significantly.

CA 6.2.5 Fish

According to Commission regulation 283/2013, metabolism studies in fish may be required where the plant protection product is used in crops whose parts or products, also after processing, are fed to fish and where residues in feed may occur from the intended applications.

The conditions under which such a study should be performed are further described in the Working document of the EU Commission SANCO/11187/2013, rev. 3 on the nature of pesticide residues in fish. The document specifies that the accumulation of compounds with low lipophilicity via the diet is known to be negligible and that fish metabolism studies are therefore required for active substances with a log P_{ow} equal or greater than 3 and an expected feed burden above 0.1 mg/kg DM.

In case of alpha-cypermethrin the log P_{ow} is 5.5 at 20°C without effect of pH (see Review Report, SANCO SANCO/4335/2000 final 13 February 2004). In this dossier, oilseed and cereals (barley and wheat) are submitted which are considered to serve as feed item for fish. Therefore, a metabolism study in fish was performed which is described below.

Report:	CA 6.2.5/1 [redacted] 2014a The metabolism of 14C-Alphacypermethrin ([14C]-BAS 310 I) in rainbow trout 2014/1147368
Guidelines:	SANCO/11187/2013 Rev. 3 (Working document)
GLP:	yes (certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Report:	CA 6.2.5/2 [redacted] 2017 a Report Amendment 1: The metabolism of 14C-Alphacypermethrin ([14C]-BAS 310 I) in rainbow trout 2017/1134384
Guidelines:	SANCO/11187/2013 Rev. 3 (Working document)
GLP:	yes (certified by Department of Health of the Government of the United Kingdom, United Kingdom)

Executive summary

The purpose of this study was to investigate the metabolism of alphacypermethrin (BAS 310 I) in rainbow trout following 14 consecutive daily doses of BAS 310 I fortified feed, at a target dose of 10 mg/kg.
Alphacypermethrin (BAS 310 I) metabolism in trout was investigated with [Benzyl-U-^{14}C]-BAS 310 I (radiochemical purity 99.5%) and [Cyclopropane-^{14}C]-BAS 310 I (radiochemical purity 96.9%). The in-life and analytical phases of this study were performed at Charles River Preclinical Services, Tranent, Edinburgh, EH33 2NE, UK.
The benzyl-labelled form of BAS 310 I was radiodiluted with [Phenoxy-^{13}C]-BAS 310 I in a 4:1 ratio, and the cyclopropane label was radiodiluted with non radiolabelled BAS 310 I in the same ratio. The final specific activity of the radiolabelled BAS 310 I was 3.8 MBq/mg.

Thirteen fish were treated per radiolabelled experiment (26 in total) with a nominal rate of 10 mg of test item per kg of feed. Fish were fed at a rate of ca. 2% tank biomass per day, with the rate increased on day 7 of the administration phase to take into account the increase in tissue mass.
Three whole fish were taken for each experiment 10 and 12 days following the first dose and fillets removed for total residue analysis. Daily samples included faeces which was collected prior to and following administration, and samples of tank water. On day 14, approximately 6 h post final dose all remaining fish were humanely killed and tissues (liver, fillet, skin, muscle, pyloric caeca, cleaned GI tract and remaining carcass) and the hind gut content removed post mortem.
Sub-samples of the fish samples were taken for initial TRR determination employing sample oxidation analysis or following extraction, by summing radioactivity in the extracts and unextracted residues. Fish tissue was extracted using acetonitrile and Milli-Q water. Further extractions were carried out as necessary, with protease enzyme (37ºC; 72h).

Fillet

Fish fillets were sampled from whole fish taken 10 and 12 days into treatment with BAS 310 I. The radioactive residues at day 10 were 0.070 mg/kg and 0.083 mg/kg, at day 12 residues were 0.090 and 0.053 mg/kg for fish treated with [Benzyl-U-^{14}C]-BAS 310 and [Cyclopropane-^{14}C]-BAS 310 I labels, respectively. TRR data for the fillets at day 14 were calculated from the skin and muscle tissues, in natural proportions, which were analysed separately. TRR was calculated as 0.105 and 0.083 mg/kg for [Benzyl-U-^{14}C]-BAS 310 and [Cyclopropane-^{14}C]-BAS 310 I labels, respectively. No further analysis was carried out on the 10 and 12 day fillet fractions.

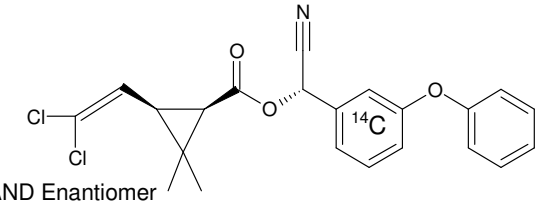
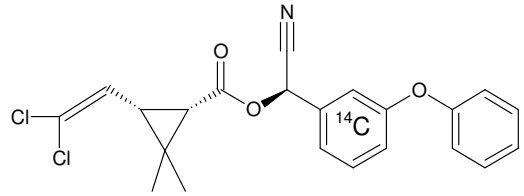
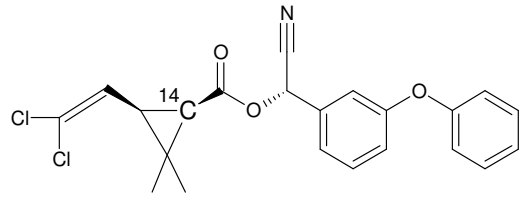
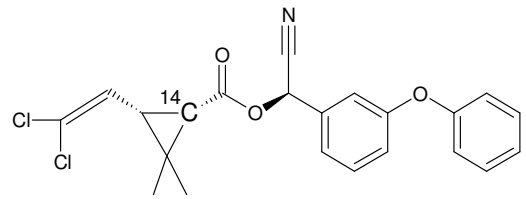
Muscle, Liver and Skin

The TRR of fish matrices taken at the final day of treatment (Day 14) with [Benzyl-U-^{14}C]-BAS 310 I were 0.094 mg/kg for muscle, 0.184 mg/kg for skin and 0.241 mg/kg for liver. Corresponding values for fish treated with [Cyclopropane-^{14}C]-BAS 310 I were 0.076 mg/kg for muscle, 0.127 mg/kg for skin and 0.299 mg/kg for liver. The majority of the residue in muscle and skin were extractable (≥90.2% TRR), further extraction of the liver with protease resulted in extractability of (≥59.1% TRR).
Parent compound BAS 310 I was detected in all fish matrices analysed at 75.9 % TRR (0.071 mg/kg) in muscle, 70.6 % TRR (0.130 mg/kg) in skin and 7.1% TRR (0.017 mg/kg) in liver for the [Benzyl-U-^{14}C]-BAS 310 I label. Corresponding values for fish treated with the [Cyclopropane-^{14}C]-BAS 310 I were 78.0 % TRR (0.060 mg/kg) in muscle, 71.7 % TRR (0.091 mg/kg) in skin and 3.8% TRR (0.011 mg/kg) in liver. Hydroxylated BAS 310 I contributed to ≤9.6% TRR (≤0.009 mg/kg) in muscle, ≤9.9% TRR (≤0.018 mg/kg) in skin and ≤5.6% TRR, (0.013 mg/kg) in liver. Multiple isomeric glucuronides of BAS 310 I were also detected as the principal residue in liver samples at ≤25.1% TRR (0.061 mg/kg). Unidentified extractable metabolites individually accounted for ≤0.006 mg/kg, and unextracted residues were ≤0.008 mg /kg in the muscle and skin and ≤0.099 mg/kg in the liver.

1. MATERIALS AND METHODS

A. MATERIALS

Test material (Analytical Standard):	BAS 310 I
Lot/Batch:	L80-24
Purity	99.4%
CAS#:	67375-30-8
Stability of test compound:	Stable under conditions of the study
Radiolabelled test material:	[^{14}C] BAS 310 I
Lot/Batch number:	[Benzyl-U-^{14}C]-BAS 310 I: Lot 775-0601 [Cyclopropane-^{14}C]-BAS 310 I: Lot 986-2101
Radiochemical Purity:	[Benzyl-U-^{14}C]-BAS 310 I: 99.5% [Cyclopropane-^{14}C]-BAS 310 I: 96.9%
Specific Activity:	[Benzyl-U-^{14}C]-BAS 310 I: 4.74 MBq/mg [Cyclopropane-^{14}C]-BAS 310 I: 4.71 MBq/mg

Chemical Structure / Positions of Radiolabels	[Benzyl-U-^{14}C]-BAS 310 I label  AND Enantiomer  [Cyclopropane-^{14}C]-BAS 310 I label  AND Enantiomer 

B. EXPERIMENTAL

The study was conducted during the period 13 February 2014 to 06 October 2014 at Charles River, Tranent, EH33 2NE, Scotland.

1. In-Life Phase

The in-life phase was conducted in round fibreglass tanks in a temperature controlled laboratory at Charles River, Tranent, EH33 2NE, Scotland. Twenty-six rainbow trout (Oncorhynchus mykiss) *ca*. 2 years old were obtained from Belhaven Trout Company Ltd (Dunbar, Scotland, UK) and received on 02 May 2014. Fish were in good health and free from any apparent malformation. The fish were maintained at the test facility for 14 days prior to test item administration. The weight of each fish was *ca*. 250-300 g and the weight variation did not exceed +/- 25% of the average weight.

The study fish were housed in two separate tanks *ca*. 1400 L capacity, (1.5 m diameter x 0.8 m deep) covered with perspex lids with ventilation holes to prevent fish escaping, dust contamination and evaporation loss. The tanks were filled with reconstituted freshwater (RFW) to *ca*. 800 L and the volume maintained throughout the study. Holding and test tanks were maintained within a laboratory at a temperature of 13-17°C. A light cycle of 16 h light and 8 h dark was in operation throughout the test, illumination being provided by fluorescent tubes. Tanks were maintained under continuous flow conditions (*ca*. 2.45 L/ min per tank) and aerated to ensure that the dissolved oxygen concentration was at least 60% of air saturation, and filtration systems were employed to remove faeces during the testing period.

Before the dosing period fish were fed daily with standard fish feed (E Fico Alpha 717, 4.5mm, Biomar Limited, Grangemouth, UK). Fish were not fed in the 24 hrs prior to the initiation of this test. At commencement of the test a sample of feed was fortified with BAS 310 I and was used throughout the test period. Feed was offered at a rate of 2.0% estimated tank biomass per day, for fourteen consecutive days. Feeding was observed and any excess food was removed by siphoning within 10 mins of addition to the tank in order to prevent contamination of the water.

2. Sampling

Three fish were sampled from each radiolabelled experiment on day 10 and day 12 of the treatment regime. On day 14 of the administration phase all remaining fish were sacrificed and dissected into samples of muscle (without skin), skin (from muscle sample), liver, fillet, cleaned GI tract, hind gut contents (combined with faeces), pyloric caeca and residual carcass. Throughout the study all fish sampled were sacrificed with a blow to the back of the head followed by destruction of the brain. Daily samples of uneaten feed, faces and water were taken throughout the course of study.

3. Analytical procedures

Fish tissues were pulverised in dry ice which was allowed to sublime prior to determination of TRR by combustion analysis followed by liquid scintillation counting (LSC). Muscle, liver and skin samples from the 14 day sampling were extracted three times in acetonitrile followed by ultrapure water twice. Where necessary, further extractions were conducted with protease enzyme (37°C, for a total of 72 h). The unextracted radioactivity in the post-extraction solids was determined by combustion analysis followed by LSC. Extractability was determined for each sample by summing radioactivity in the extracts and unextracted residues.

Extracts were analysed using a reversed phase HPLC (Synergi Fusion – RP) column eluted with a gradient of water and acetonitrile containing 0.1% formic acid. The effluent was passed through a UV detector (230 nm) to detect reference standards. A second HPLC method and LC-MS was conducted to confirm the presence of metabolites.

4. Lipid Extraction and Determination

Lipid content of the tissues and feed were obtained by extracting in methanol and chloroform. The chloroform layer of each extract was dried by passing through anhydrous sodium sulphate (ca. 1-2 g) and the lipid content weighed after removing the solvent under a stream of nitrogen gas.

II. RESULTS AND DISCUSSSION

A. TOTAL RADIOACTIVE RESIDUES (TRRS)

Rainbow trout (*Oncorhynchus mykiss*) were administered fish feed fortified with [^{14}C]-BAS 310 I at a target dose of 10 mg/kg over a period of 14 days. Whole fish samples were taken at 10, 12 and 14 days. On day 14 fish were dissected into liver, residual carcass, faeces, pyloric caeca, hind gut content, cleaned GI tract and edible tissues (skin and muscle). Fish tissues were pulverised and extracted where required. The total radioactive residue (TRR) was determined as the sum of radioactivity in each extract and the unextracted residue.

1. Fillet

TRR in fillet treated were ≤0.105 mg/kg and ≤0.083 mg/kg for the [Benzyl-U-^{14}C]-BAS 310 I and [Cyclopropane-^{14}C]-BAS 310 I respectively. All values are presented below:

Table 6.2.5-1: Total Radioactive Residues of [^{14}C]-BAS 310 I in Trout Fillet

	Day 10 mg/kg	**Day 12 mg/kg**
Bz -^{14}C	0.070	0.090
Cy-^{14}C	0.083	0.053

Day 14	**Muscle (mg/kg)**	**Skin (mg/kg)**	**Total in natural proportions (mg/kg)**
Bz -^{14}C	0.094	0.184	0.105
Cy-^{14}C	0.076	0.127	0.083

2. Muscle, Liver and Skin

TRR data for muscle, liver and skin is provided below. Liver residues were the highest of all tissues extracted, 0.241 mg/kg and 0.299 mg/kg for the benzyl and cyclopropane labels respectively. The lowest residues were detected in the muscle, 0.094 mg/kg and 0.076mg/kg for the benzyl and cyclopropane labels respectively.

Table 6.2.5-2: Total Radioactive Residues of [^{14}C]-BAS 310 I in Muscle, Liver and Skin (after 14 Daily Doses of [^{14}C] BAS 310 I at 10 mg/kg Feed Nominal)

Fish Matrix	Interval	[Benzyl-U-^{14}C]-BAS 310 I (mg/kg)[1)]	[Cyclopropane-^{14}C]-BAS 310 I (mg/kg) [1)]
Muscle	14	0.094	0.076
Liver		0.241	0.299
Skin		0.184	0.127

1)TRR values were determined as the sum of the extracted + unextracted radioactivity.

3. Lipid Content

The measured lipid content of the muscle, skin, liver and feed are presented in Table 6.2.5-3. Muscle tissue contained 4.5 - 4.6 % lipid.

Table 6.2.5-3: Lipid Content of Tissues (after 14 Daily Doses of [^{14}C] BAS 310 I at 10 mg/kg Feed Nominal)

Tissue	[Benzyl-^{14}C] label % Lipid content	[Cyclopropane-^{14}C] label % Lipid content
Muscle	4.5	4.6
Skin	2.9	1.9
Liver	1.3	1.9
Feed	16.6	

B. EXTRACTION AND CHARACTERISATION OF RESIDUES

The extractability of the ^{14}C residues in fish samples was determined using acetonitrile followed by water. Where necessary, further extractions were conducted with protease enzyme (37°C, 72 h). The unextracted radioactivity in the post-extraction solids was determined by combustion analysis followed by LSC.

1. Extraction and characterisation of residues in muscle, liver and skin
   The total radioactive residues in muscle, liver and skin ranged from 0.076 to 0.299 mg/kg with BAS 310 I present in all samples (≤0.130 mg/kg).

   Muscle

   The parent BAS 310 I was the principal residue detected in both radiolabelled experiments at 75.9 and 78.0% TRR for the benzyl and cyclopropane labels, respectively. Hydroxylated BAS 310 I accountined for 9.6% and 7.9 %TRR (0.009 mg/kg and 0.006 mg/kg) for the benzyl and cyclopropane labels, respectively. All remaining residues were ≤3.0 %TRR (≤0.002 mg/kg).

   Skin

   The parent BAS 310 I was the principal residue detected in both radiolabelled experiments at 70.1 and 71.7 % TRR (0.130 and 0.091 mg/kg) for the benzyl and cyclopropane labels, respectively. Hydroxylated BAS 310 I accounted for 9.9 and 9.7 %TRR (0.018 mg/kg and 0.012 mg/kg) for the benzyl and cyclopropane labels, respectively. All remaining residues were ≤2.7 %TRR (≤0.003 mg/kg).

   Liver

   The parent BAS 310 I was detected in both radiolabelled experiments at 7.1% and 3.8 % TRR (0.017 and 0.011 mg/kg) for the benzyl and cyclopropane labels, respectively. The principal residue was multiple isomeric glucuronides of BAS 310 I detected at 25.1% and 19.7 % TRR (0.061 and 0.059 mg/kg) for the benzyl and cyclopropane labels, respectively. hydroxylated BAS 310 I accounted for 5.6 %and 4.3 %TRR (0.013 mg/kg and 0.013 mg/kg) for the benzyl and cyclopropane labels, respectively. All remaining residues were individually ≤4.0 %TRR (≤0.012 mg/kg) and were not possible to identify due to the low response in each case.

2. Storage stability of residues

   Samples were stored in a freezer set to maintain *ca*. -20°C and storage intervals recorded. All extraction, fractionation and HPLC analysis was completed within 6 months of sacrifice therefore storage stability was not performed.

3. Identification of BAS 310 I and metabolites

   The identities of the principal fish residue components were investigated by comparison of the extractable residues with different reference standards of BAS 310 I and its authenticated analytical reference standards. BAS 310 I was observed as the primary residue in skin and muscle of the fish (≤0.130 mg/kg) and this identification was confirmed by, MRM analysis. The residues corresponding to hydroxylated BAS 310 I and isomeric glucuronides of BAS 310 I were also confirmed by LC-MS.

4. Proposed metabolic pathway

BAS 310 I was observed as the primary residue in the skin and muscle of the fish with lower levels of hydroxylated BAS 310 I. BAS 310 I was metabolised more extensively in the liver. Glucuronic acid conjugatation of the hydroxylated BAS 310 I was the primary residue in liver together with hydroxylated BAS 310 I and unchanged BAS 310 I.

Table 6.2.5-4: Summary of Identified and Characterised 14C Residues Extracted in Composite Muscle (after 14 Daily Doses of [^{14}C]-BAS310 I at 10 mg/kg Feed Nominal)

Designation	[Benzyl-U-^{14}C]-BAS 310 I		[Cyclopropane-^{14}C]-BAS 310 I	
	mg/kg	**%TRR**	**mg/kg**	**%TRR**
Identified				
BAS 310 I (Reg. No. 4078193)	0.071	75.9	0.060	78.0
Hydroxylated BAS 310 I	0.009	9.6	0.006	7.9
Total Identified in Extractable Radioactivity	**0.080**	**85.8**	**0.066**	**85.9**
Total Characterized in Extractable Radioactivity	**0.003**	**3.0**	**0.005**	**6.7**
Total Identified and Characterized in Extractable Radioactivity	**0.083**	**88.5**	**0.071**	**92.5**
Post-Extraction Solid	**0.004**	**4.1**	**0.007**	**9.8**

Table 6.2.5-5: Summary of Identified and Characterised 14C Residues Extracted in Composite Skin (after 14 Daily Doses of [^{14}C]-BAS310 I at 10 mg/kg Feed Nominal)

Designation	[Benzyl-U-^{14}C]-BAS 310 I		[Cyclopropane-^{14}C]-BAS 310 I	
	mg/kg	**%TRR**	**mg/kg**	**%TRR**
Identified				
BAS 310 I (Reg. No. 4078193)	0.130	70.6	0.091	71.7
Hydroxylated BAS 310 I	0.018	9.9	0.012	9.7
Total Identified in Extractable Radioactivity	0.148	80.5	0.103	81.4
Characterised				
Organosoluble extractable residues following treatment with protease enzyme	0.005	~~2.1~~ 2.7	ND	ND
Aqueous soluble extractable residues following treatment with protease enzyme	0.002	1.0	ND	ND
Total Characterized in Extractable Radioactivity	**~~0.007~~ 0.019**	**~~5.2~~ 10.5**	**~~0.006~~ 0.009**	**~~5.4~~ 8.0**
Total Identified and Characterized in Extractable Radioactivity	**~~0.155~~ 0.167**	**~~85.7~~ 91.0**	**~~0.109~~ 0.112**	**~~86.8~~ 89.4**
Post-Extraction Solid	**0.004**	**2.2**	**0.008**	**6.2**

ND- Not detected

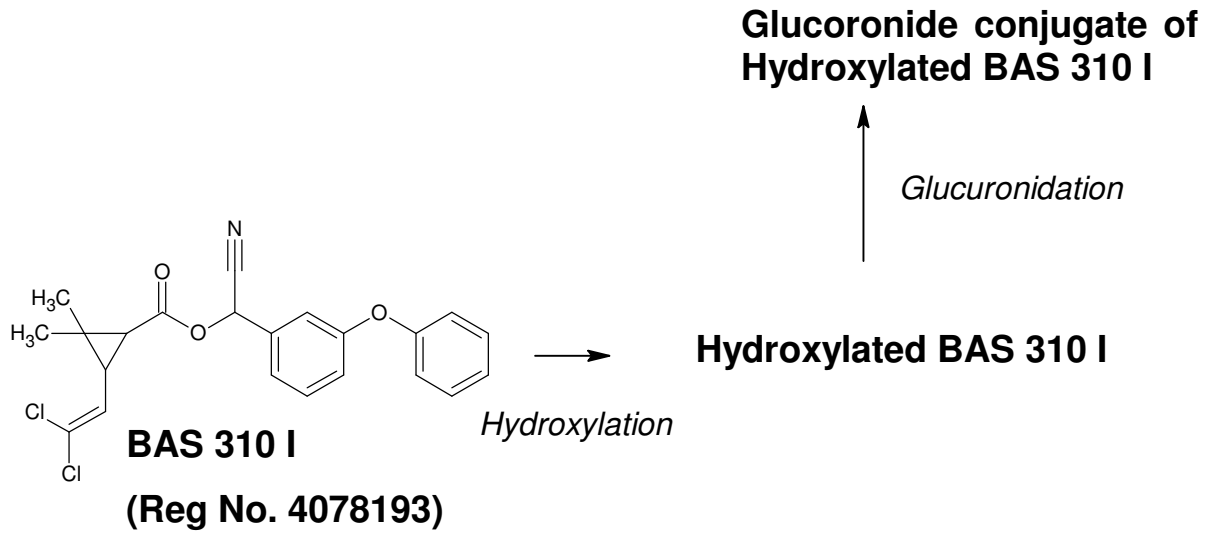
Table 6.2.5-6: Summary of Identified and Characterised 14C Residues Extracted in Trout Liver (after 14 Daily Doses of [^{14}C]-BAS310 I at 10 mg/kg Feed Nominal)

Designation	[Benzyl-U-^{14}C]-BAS 310 I		[Cyclopropane-^{14}C]-BAS 310 I	
	mg/kg	%TRR	mg/kg	%TRR
Identified				
BAS 310 I (Reg. No. 4078193)	0.017	7.1	0.011	3.8
Hydroxylated BAS 310 I	0.013	5.6	0.013	4.3
Glucuronide conjugate	0.061	25.1	0.059	19.7
Total Identified in Extractable Radioactivity	**0.091**	**37.8**	**0.083**	**27.8**
Total Characterized in Extractable Radioactivity	**0.043**	**18.1**	**0.113**	**37.2**
Total Identified and Characterized in Extractable Radioactivity	**0.134**	**55.9**	**0.196**	**65.0**
Post-Extraction Solid	**0.099**	**40.9**	**0.088**	**29.4**

III. CONCLUSION

Radioactivity in fillet rose steadily in the benzyl label to a maximum of 0.105 mg/kg at day 14, the cyclopropane labelled fillet was 0.083 mg/kg at both day 10 and day 14.
The solvent extractability of muscle and skin was high (≥90.2% TRR). Further extraction with protease enzyme was carried out on liver (both labels) and skin benzyl label to achieve extractability of ≥59.1%.
BAS 310 I was observed as the primary residue in the skin and muscle of the fish (70.1 to 78.0 %TRR, 0.060 to 0.130 mg/kg) with lower levels of hydroxylated BAS 310 I (7.9 to 9.9 %TRR, 0.006 to 0.018 mg/kg). BAS 310 I was metabolised more extensively in the. Glucuronic acid conjugatation of the hydroxylated BAS 310 I was the primary residue (19.7 to 25.1 %TRR, 0.059 to 0.061 mg/kg) together with hydroxylated BAS 310 I and unchanged BAS 310 I.

Figure 6.2.5-1: Proposed metabolic pathway for BAS 310 I in Rainbow Trout



Overall Summary Livestock Metabolism

The present Annex I renewal dossier provides new metabolism studies in goat, hen, fish and rat. The new studies mainly confirm the findings and degradation reactions known so far and complement additional details due to more sensitive technology.

Combining the findings of the older metabolism studies and the new ones, a system of degradation reactions results which can not only explain most of the facts gathered so far but also lead to a more complete picture.

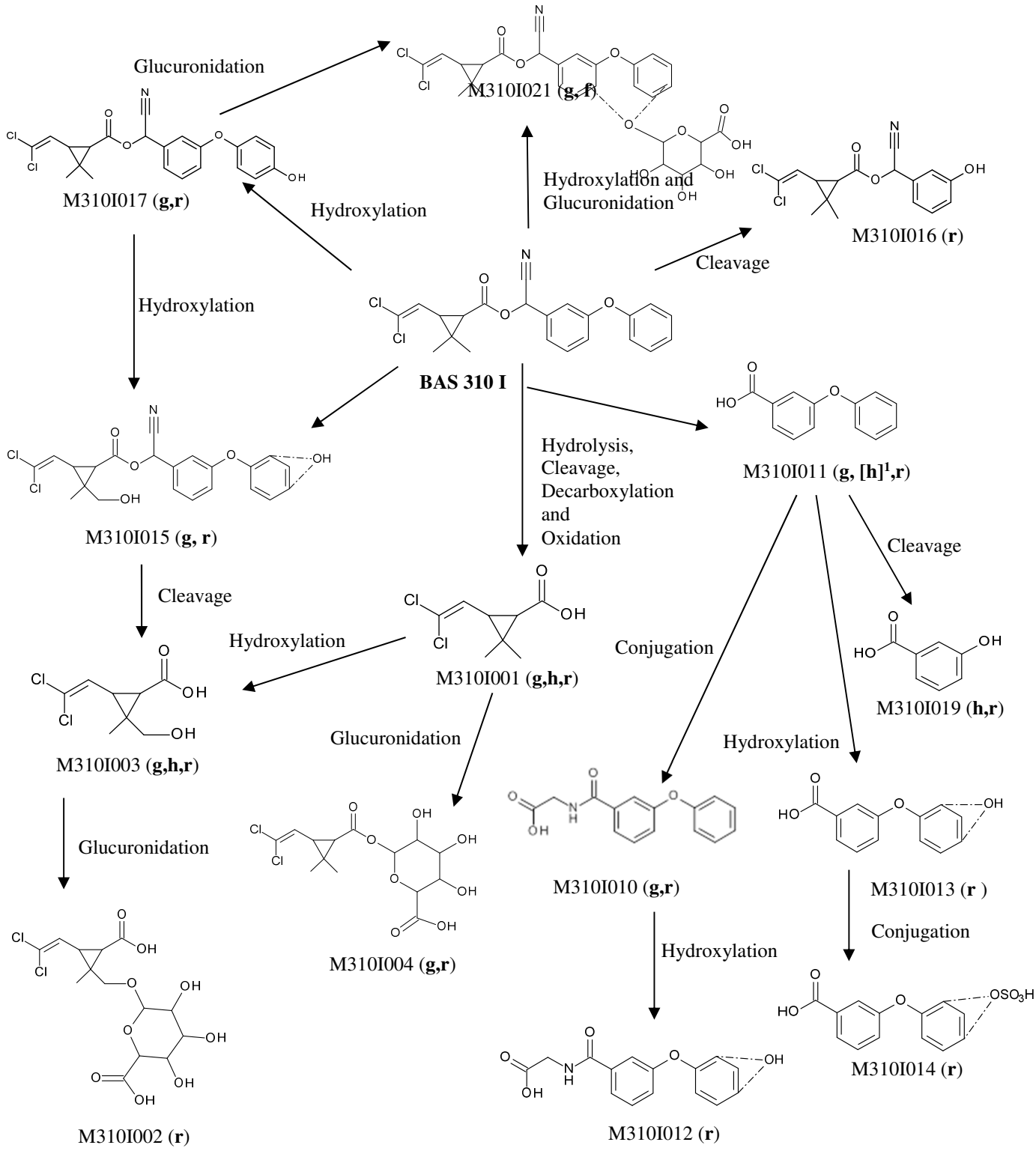
The general metabolic pathway in rats and ruminants was found to be comparable. Hydrolysis of the ester bond of BAS 310 I results in a cyclopropane and a phenoxybenzyl moiety of the molecule. The cyclopropane moiety corresponds to metabolite M310I001 (DCVA) which in **hen**, **goat** and **rat** is hydroxylated to give M310I003 and subsequently, via glucuronidation, is transformed into M310I002 (**rat**). Glucuronidation of M310I001 to yield M310I004 was shown in **goat** and **rat**.

Cleavage of the nitrile group of the phenoxybenzyl moiety and oxidation yields M310I011 (3-phenoxybenzoic acid), which was identified in the **rat** and **goat** metabolism studies. M310I011 can be cleaved to give M310I019 (3-Hydroxybenzoic acid; found in **hen**) or via hydroxylation and conjugation form M310I013 and M310I014 (**rat**). Conjugation of M310I011 with glycine leads to M310I010 (N-(3-phenoxybenzoyl)glycine, found in **goat** and **rat**), which can be hydroxylated to give M310I012 (rat).

A further metabolic route are hydroxylation and conjugation of the phenoxybenzyl and / or cyclopropyl moiety of the alpha-cypermethrin molecule without prior ester cleavage leading to metabolites M310I015 (**goat** and **rat**), M310I017 (4'hydroxy-alpha-cypermethrin; **goat** and **rat**), and M310I021 (Glucuronide conjugate of alpha-cypermethrin, **goat** and **fish**). Hydroxylation and glucuronidation of uncleaved alpha-cypermethrin to give the glucuronide conjugate M310I021 were the only metabolic reactions demonstrated in fish.

A complete picture including rat, goat, hen and fish results is shown in Figure 6.2.5-2.

Figure 6.2.5-2: Proposed metabolic pathway of BAS 310 I in animal (goat (g), hen (h), fish (f) and rat (r))



[1] postulated intermediate not detected in hen

CA 6.3 Magnitude of residues trials in plants

Alpha-cypermethrin is registered in multiple crops belonging to different EU crop groups. Within this dossier residue data are only provided for the representative uses supporting the renewal of approval. The ME formulation BAS 310 55 I has been selected as representative formulations.

Consequently, in this dossier section the relevant data for the following crops are summarized:

- Cucumber (for extrapolation to courgette)
  Leafy brassica
  Lettuce
  Oilseed rape
- Cereals (barley and wheat))

The studies provided below for cucumber, leafy brassica, lettuce, oilseed rape and cereals have not been evaluated within the MRL re-evaluation process according to Reg. 396/2005, Art. 12. Consequently they are not considered as peer-reviewed.

CA 6.3.1 Cucumber

Residue trials under glasshouse conditions were performed in the years 2004/2005. According to SANCO 7525/VI/95 - rev.9, March 2011, extrapolation is possible from cucumber to courgettes. Therefore, residue trials in cucumber that support the GAP for courgettes are presented in the following.

Table 6.3.1-1: cGAP for the use of BAS 310 I in/on cucumber

Crop	Maximum applied dose	Water volume	PHI	Application method	Application timing
Cucumber / Courgette, indoor (North, Central)	1 x 0.030 kg BAS 310 I/ha	200–1500	3	Overall spray	BBCH 10-89
Cucumber / Courgette, indoor (South)	2 x 0.015 kg BAS 310 I/ha	200–1500	3	Overall spray	BBCH 10-89
	1 x 0.030 kg BAS 310 I/ha	200–1500	3	Overall spray	BBCH 10-89

PHI = pre-harvest interval

Table 6.3.1-2: GAP information of residue trials conducted in cucumber in 2004-2005

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Indoor (EU N + S)	Denmark (1) Germany (1) Greece (1) France (2) Italy (1) Spain (1) The Netherlands (1) 2004	BAS 310 41 I SC	spray appl.	0.015	0.004	1 2	0±1 3±1 7±1 14±1	2004/ 5000720	No
	Belgium (1) Denmark (1) Germany (1) Greece (1) France (2) Italy (1) Spain (1) 2005	BAS 310 40 I EC	spray appl.	0.04	0.01	1	0 3 7 14-15	2006/ 1036934	No

0 Actual application rates varied by 10% at most
1 Days after last application

Report: CA 6.3.1/1
Leonard R.C., 2005a
Study on the residue behavior of BAS 310 I in cucumbers (glasshouse) after application of BAS 310 41 I in Germany, France (N), Denmark, Netherlands, France (S), Spain, Italy and Greece, 2004
2004/5000720
Guidelines: EEC 91/414 Annex II (Part A Section 6), EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 91/414 Annex III (Part A Section 8)
GLP: yes
(certified by United States Environmental Protection Agency)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31041I
Description: BAS 310 41 I (SC)
Lot/Batch #: 4000: 100 g/L
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01 and 1.0 mg/kg

2. Test Commodity:
Crop: Cucumber
Type: Cucurbits edible peel
Variety: Euphoria, Sheila, Naomi, Suxo, Solverde, Rawa, Palmera, Hiyield
Botanical name: *Cucumis sativus*
Crop parts(s) or processed commodity: fruit
Sample size: 0 DALA (days after last application): 12 fruit (min. 0.5 kg)
3±1 DALA: 12 fruit (min. 1.0 kg)
7±1 and 14±1 DALA: 12 fruit (min. 2.0 kg)

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2004 growing season, eight trials were conducted in cucumbers grown in glasshouses. A soluble concentrate formulation of alpha-cypermethrin (100 g a.s./L; BAS 310 41 I) was foliar applied to cucumbers on two different plots. In one variant, one application was made at a target rate of 0.015 kg alpha-cypermethrin/ha 7±1 days before harvest. This was compared to another variant in which two applications were made at the same target application rate, 14±1 and 7±1 days before harvest. The target water volume was 400 L/ha.

Cucumber fruit specimens were collected directly after the last application as well as 2-4, 7-8 and 13-14 days thereafter. There were some slight deviations from the targeted sampling parameters ("quantity"), none of which influenced the results of the study, which occurred at trial GRE/12/04. At 3 DALA, only eight cucumbers from plot 2 and six cucumbers from plot 3 were harvested, and at 14 DALA, only six cucumbers from plot 3 were collected.
Samples were stored frozen at or below -18°C including during transportation, until analysis, except for the samples denoted by an asterisk ("*") in Table 6.3.1-6 below (several samples from trials AGR/10/04 and ALB/07/04, and all samples from trial ITA/09/04). These samples were shipped frozen (packed in dry ice) by airfreight from BASF Aktiengesellschaft (Limburgerhof, Germany) on 29 November 2004 and were received unfrozen at BASF Agro Research (Research Triangle Park, North Carolina, U.S.A) on 08 December 2004. The samples defrosted for a brief period on account of delays incurred while in customs. The samples were cold and in good condition for residue analysis. The maximum frozen storage interval between sampling and extraction was 252 days.

Table 6.3.1-3: Target application rates and timings for cucumber

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2004	8	1	G	BAS 310 41 I (SC)	alpha-cypermethrin	0.015	400	7 ±1 days before harvest
		2	G	BAS 310 41 I (SC)	alpha-cypermethrin	0.015	400	1st appl. 14 ±1, 2nd appl. 7 ±1 days before harvest

2. Description of analytical procedures

The specimens were analysed by means of HPLC-MS/MS for total cypermethrin residues with BASF method No. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.
Principle of method: Residues of cypermethrin (alphacypermethrin or cypermethrin) are extracted with a mixture of methanol/water/2N hydrochloric acid (70:25:5). An aliquot of the extract is centrifuged, and cleaned by liquid/liquid partition against cyclohexane. The final determination of total cypermethrin residues is performed by HPLC-MS/MS.

Table 6.3.1-4: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in cucumber

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
fruit	0.01	4	80.7	14.3	17.7
	1.0	4	101.9	17.7	17.4
	Overall	8	91.3	18.7	20.5

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.1-5, detailed residue levels are shown in Table 6.3.1-6.
Immediately after one application of 0.015 kg alpha-cypermethrin/ha in the glasshouse, residues in cucumber fruit ranged from below the LOQ (<0.01 mg/kg) to 0.026 mg/kg. Residues were detected (0.023 and 0.026 mg/kg) at only two of the eight trial locations. Total cypermethrin residues were <0.01 mg/kg in all samples collected thereafter (2-14 days after application).
Immediately after the last of two applications at a rate of 0.015 kg alpha-cypermethrin/ha, total residues of cypermethrin in cucumber ranged from below the LOQ (<0.01 mg/kg) to 0.030 mg/kg. Residues decreased to <0.01-0.012 mg/kg at 2-4 days after the last application, and were <0.01 mg/kg in all samples collected thereafter (7-14 days after the last application).
No residues above the LOQ were found in any of the untreated samples.

Table 6.3.1-5: Summary of residues of BAS 310 I in cucumber after application of BAS 310 41 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
Indoor (EU N + S)	2004	BAS 310 41 I (SC) 1 x 0.015 kg a.s./ha	0	73-85	fruit	<0.01-0.026
			2-4	73-88	fruit	<0.01
			7-8	74-89	fruit	<0.01
			13-14	76-89	fruit	<0.01
		BAS 310 41 I (SC) 2 x 0.015 kg a.s./ha	0	73-85	fruit	<0.01-0.030
			2-4	73-88	fruit	<0.01-0.012
			7-8	74-89	fruit	<0.01
			13-14	76-89	fruit	<0.01

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Immediately after one application of 0.015 kg alpha-cypermethrin/ha in the glasshouse, residues in cucumber fruit ranged from below the LOQ (<0.01 mg/kg) to 0.026 mg/kg. Total cypermethrin residues were <0.01 mg/kg in all samples collected thereafter (2-14 days after application).
Immediately after the last of two applications at a rate of 0.015 kg alpha-cypermethrin/ha, total residues of cypermethrin in cucumber ranged from below the LOQ (<0.01 mg/kg) to 0.030 mg/kg. Residues decreased to <0.01-0.012 mg/kg at 2-4 days after the last application, and were <0.01 mg/kg in all samples collected thereafter (7-14 days after the last application).

Table 6.3.1-6: Residues of BAS 310 I in cucumber after one or two applications of BAS 310 41 I in greenhouse (EU N+S)

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**ACK/08/04 **GLP:**yes **Year**2004	Cucumber	Germany	BAS 310 41 I 1 x 0.0150	85	0	fruit	<0.01
				87	3	fruit	<0.01
				89	7	fruit	<0.01
				89	14	fruit	<0.01
			BAS 310 41 I 2 x 0.0150	85	0	fruit	<0.01
				87	3	fruit	<u>≤0.01</u>
				89	7	fruit	<0.01
				89	14	fruit	<0.01
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**AGR/10/04 **GLP:**yes **Year**2004	Cucumber	The Netherlands	BAS 310 41 I 1 x 0.0150	85	0	fruit	<0.01
				87	3	fruit	<0.01*
				89	7	fruit	<0.01
				89	13	fruit	<0.01*
			BAS 310 41 I 2 x 0.0150	85	0	fruit	<0.01*
				87	3	fruit	<u>≤0.01</u>
				89	7	fruit	<0.01
				89	13	fruit	<0.01*
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**ALB/07/04 **GLP:**yes **Year**2004	Cucumber	Denmark	BAS 310 41 I 1 x 0.0150	77	0	fruit	0.023
				84	4	fruit	<0.01*
				89	7	fruit	<0.01
				89	13	fruit	<0.01*
			BAS 310 41 I 2 x 0.0150	77	0	fruit	0.030
				84	4	fruit	<u>0.012*</u>
				89	7	fruit	<0.01
				89	13	fruit	<0.01*
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**ALO/17/04 **GLP:**yes **Year**2004	Cucumber	Spain	BAS 310 41 I 1 x 0.0150	73	0	fruit	<0.01
				73	4	fruit	<0.01
				74	7	fruit	<0.01
				76	13	fruit	<0.01
			BAS 310 41 I 2 x 0.0150	73	0	fruit	<0.01
				73	4	fruit	<u>≤0.01</u>
				74	7	fruit	<0.01
				76	13	fruit	<0.01
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**FAN/10/04 **GLP:**yes **Year**2004	Cucumber	France North	BAS 310 41 I 1 x 0.0150	73	0	fruit	<0.01
				74	2	fruit	<0.01
				76	8	fruit	<0.01
				78	14	fruit	<0.01
			BAS 310 41 I 2 x 0.0150	73	0	fruit	<0.01
				74	2	fruit	<u>≤0.01</u>
				76	8	fruit	<0.01
				78	14	fruit	<0.01

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**FBD/11/04 **GLP:**yes **Year**2004	Cucumber	France South	BAS 310 41 I 1 x 0.0150	81	0	fruit	<0.01
				85	3	fruit	<0.01
				89	7	fruit	<0.01
				89	14	fruit	<0.01
			BAS 310 41 I 2 x 0.0150	81	0	fruit	0.015
				85	3	fruit	<u><0.01</u>
				89	7	fruit	<0.01
				89	14	fruit	<0.01
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**GRE/12/04 **GLP:**yes **Year**2004	Cucumber	Greece	BAS 310 41 I 1 x 0.0150	81	0	fruit	0.026
				85	4	fruit	<0.01
				85	8	fruit	<0.01
				88	14	fruit	<0.01
			BAS 310 41 I 2 x 0.0150	81	0	fruit	0.014
				85	4	fruit	<u><0.01</u>
				85	8	fruit	<0.01
				88	14	fruit	<0.01
Study code:166297 **Doc ID:**2004/5000720 **Trial No.:**ITA/09/04 **GLP:**yes **Year**2004	Cucumber	Italy	BAS 310 41 I 1 x 0.0150	79	0	fruit	<0.01*
				88	3	fruit	<0.01*
				89	7	fruit	<0.01*
				89	14	fruit	<0.01*
			BAS 310 41 I 2 x 0.0150	79	0	fruit	<0.01*
				88	3	fruit	<u><0.01*</u>
				89	7	fruit	<0.01*
				89	14	fruit	<0.01*

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* sample defrosted for a brief period on account of delays incurred while in customs
_ underlined values were used for MRL calculation

Report: CA 6.3.1/2
Schulz H., 2007a
Study on the residue behaviour of Alpha-Cypermethrin in cucumber after treatment with BAS 310 40 I under greenhouse conditions in Southern France, Germany, Belgium, Denmark, Italy, Spain and Greece, 2005
2006/1036934

Guidelines: EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7

GLP: yes
(certified by Hessisches Ministerium fuer Umwelt, Laendlichen Raum und Verbraucherschutz)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I
Description: BAS 310 40 I (EC)
Lot/Batch #: 1171: 100.0 g/L
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01, 0.1 and 1.0 mg/kg

2. Test Commodity:
Crop: Cucumber
Type: Cucurbits edible peel
Variety: Defens, Avonis, Loretta, Naomi, Columbia, Jumbo, Suso, Luberon
Botanical name: *Cucumis sativus*
Crop parts(s) or processed commodity: fruit
Sample size: 12 items (2 kg)

B. STUDY DESIGN AND METHODS

1. Test procedure

During the growing season of 2005, eight greenhouse trials with cucumber were conducted in Southern France, Germany, Belgium, Denmark, Italy, Spain and Greece in order to determine the magnitude of the residues of alpha-cypermethrin.
Each greenhouse trial consisted of two plots, one untreated control plot and one plot treated once with 0.4 L/ha of BAS 310 40 I (100.0 g/L of alpha-cypermethrin, EC formulation) corresponding to 40 g of active substance per hectare. The application took place at 7 days before harvest with an application rate of 400 L/ha of spray.

Samples of cucumber fruit were collected 0, 3, 7 and 14-15 days after application.
The maximum storage interval was 492 days.

Table 6.3.1-7: Target application rates and timings for cucumber

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2005	8	1	G	BAS 310 40 I (EC)	alpha-cypermethrin	0.040	400	7 ±1 days before harvest

2. Description of analytical procedures

The specimens were analysed by means HPLC-MS/MS for total cypermethrin residues with BASF method No. 567/0.
Principle of method: Residues of cypermethrin (alpha-cypermethrin or cypermethrin) are extracted with a mixture of methanol/water/2N hydrochloric acid (70:25:5). After centrifugation, an aliquot of the extract is cleaned by liquid/liquid partition against cyclohexane. The residues are further purified (if needed) using a silica gel solid phase extraction (SPE) column. The final determination of total cypermethrin residues is performed by HPLC-MS/MS, monitoring the ion transition from m/z 433.2 / 191.1 (the ammonium adduct of cypermethrin). The limit of quantitation (LOQ) of the method for alpha-cypermethrin is 0.01 mg/kg.

Table 6.3.1-8: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in cucumber

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
fruit	0.01	4	79.3	9.0	11.4
	0.1	2	85.8	-	-
	1.0	1	102.0		
	Overall	7	84.4	11.9	14.1

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.1-9, detailed residue levels are shown in Table 6.3.1-10.
Initial residues between <0.01 mg/kg and 0.087 mg/kg were found in cucumber fruit immediately after application of 40 g alpha-cypermethrin/ha in the greenhouse. Residues declined to <0.01-0.037 mg/kg after 3 days and further to <0.01-0.014 mg/kg at 7 and <0.01-0.011 at 14-15 days after application, respectively.
No residues of alpha-cypermethrin above the LOQ were analysed in any of the untreated specimens.

Table 6.3.1-9: Summary of residues of BAS 310 I in cucumber after application of BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
Indoor (EU N+S)	2005	BAS 310 40 I (EC) 1 x 0.040 kg a.s./ha	0	69-89	fruit	<0.01-0.087
			3	71-89	fruit	<0.01-0.037
			7	72-89	fruit	<0.01-0.014
			14-15	73-89	fruit	<0.01-0.011

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Initial residues between <0.01 mg/kg and 0.087 mg/kg were found in cucumber fruit immediately after application of 40 g alpha-cypermethrin/ha in the greenhouse. Residues declined to <0.01-0.037 mg/kg after 3 days and further to <0.01-0.014 mg/kg at 7 and <0.01-0.011 at 14-15 days after application, respectively.

Table 6.3.1-10: Residues of BAS 310 I in in cucumber after one application of BAS 310 40 I in greenhouse (EU N+S)

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**05 I CL FR P32 **GLP:**yes **Year**2005	Cucumber	France South	BAS 310 40 I 1 x 0.040	71	0	fruit	0.027
				71	3	fruit	0.012
				72	7	fruit	0.012
				73	14	fruit	<0.01
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**AT-05/007-1 **GLP:**yes **Year**2005	Cucumber	Germany	BAS 310 40 I 1 x 0.040	69	0	fruit	<0.01
				75	3	fruit	<0.01
				75	7	fruit	<0.01
				75	15	fruit	<0.01
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**G022-05 I-B **GLP:**yes **Year**2005	Cucumber	Belgium	BAS 310 40 I 1 x 0.040	87-89	0	fruit	0.020
				87-89	3	fruit	0.014
				87-89	7	fruit	<0.01
				87-89	14	fruit	<0.01
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**ALB/190507-01 **GLP:**yes **Year**2005	Cucumber	Denmark	BAS 310 40 I 1 x 0.040	81	0	fruit	0.014
				85	3	fruit	0.020
				88	7	fruit	0.013
				89	14	fruit	<0.01
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**05 I CL FR P36 **GLP:**yes **Year**2005	Cucumber	France South	BAS 310 40 I 1 x 0.040	76	0	fruit	0.043
				76	3	fruit	0.031
				78	7	fruit	0.014
				79	14	fruit	0.011
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**IR05BASL11LG01 **GLP:**yes **Year**2005	Cucumber	Italy	BAS 310 40 I 1 x 0.040	88	0	fruit	0.087
				88	3	fruit	0.037
				89	7	fruit	0.012
				89	14	fruit	<0.01
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**05ES087R **GLP:**yes **Year**2005	Cucumber	Spain	BAS 310 40 I 1 x 0.040	75	0	fruit	<0.01
				77	3	fruit	<0.01
				79	7	fruit	<0.01
				79	14	fruit	<0.01
Study code:227122 **Doc ID:**2006/1036934 **Trial No.:**05RF044 **GLP:**yes **Year**2005	Cucumber	Greece	BAS 310 40 I 1 x 0.040	81	0	fruit	0.036
				84	3	fruit	0.014
				86	7	fruit	<0.01
				89	14	fruit	<0.01

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

CA 6.3.2 Leafy brassica

Table 6.3.2-1: cGAP for the use of BAS 310 I in/on leafy cabbage

Crop	Maximum applied dose	Water volume	PHI	Application method	Application timing
Leafy brassica, outdoor (North, Central)	2 x 0.010 kg BAS 310 I/ha	200-1000	3	Overall spray	BBCH 10-49
Leafy brassica, outdoor (South)	2 x 0.010 kg BAS 310 I/ha	200-1000	3	Overall spray	BBCH 10-49
	1 x 0.020 kg BAS 310 I/ha	200-1000	3	Overall spray	BBCH 10-49

PHI = pre-harvest interval

Table 6.3.2-2: GAP information of residue trials conducted in leafy cabbage in 2005-2007 in Northern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Northern EU	Germany (1) The United Kingdom (1) 2005	BAS 310 40 I EC	spray appl.	0.0125	0.003	1 2	0 3±1 7±1 14±1	2006/ 1026862	No
	France (1) The Netherlands (1)2006	BAS 310 40 I EC	spray appl.	0.0125 0.0125	0.003	1 2	0 3±1 7±1 14±1	2007/ 1013342	No
	Germany (4) 2006	Fastac SC	spray appl.	0.0125	0.0021	2	0 (2) 7 (4) 10 (2) 14 (4)	2007/ 1035745	No
	Germany (4) 2007	Fastac SC	spray appl.	0.0125	0.00208	2	0 7 10 14	2014/ 1083417	No

0 Actual application rates varied by 10% at most
1 Days after last application

Table 6.3.2-3: GAP information of residue trials conducted in leafy cabbage in 2005-2013 in Southern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Southern EU	Italy (1) Greece (1) 2005	BAS 310 40 I EC	spray appl.	0.025	0.006	1	0 3±1 7±1 14±1	2006/ 1026862	No
	France (1) Spain (1) 2006	BAS 310 40 I EC	spray appl.	0.025	0.006	1	0 3±1 7±1 14±1	2007/ 1013342	No
	France (1) Greece (1) Italy (1) Spain (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.00625	1	0 2-3 6-7 13-14	2013/ 1416285	No

0 Actual application rates varied by 10% at most
1 Days after last application

The studies not yet evaluated are summarized in the following chapter.

Report: CA 6.3.2/1
North L., 2007a
Study on the residue behaviour of Alpha-Cypermethrin in curly kale after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005
2006/1026862
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I
Description: BAS 310 40 I (EC)
Lot/Batch #: 1171, Alpha-Cypermethrin: 100 g/L
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01, 0.5 and 1.0 mg/kg

2. Test Commodity:
Crop: Curly kale
Type: Leafy brassica
Variety: Winterbox, Winneton, Black Toscano, Vates
Botanical name: *Brassica oleracea*
Crop parts(s) or processed commodity: Foliage
Sample size: 0 DALA (days after last application): 12 units (min. 1.0 kg)
3±1 DALA: 12 units (min. 1.0 kg)
7±1 DALA: 12 units (min. 2.0 kg)
14±1 DALA: 12 units (min. 2.0 kg)

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2005 and 2006 growing season, four field trials were conducted in different representative curly kale growing areas in the UK, Germany, Italy and Greece to determine the residue level of alpha-cypermethrin in or on Raw Agricultural Commodities (RAC).
At Trials AF/8815/BA/1 and 5 conducted in Northern Europe, BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g a.s./L), was foliar applied either once at 7 days before expected harvest (plot 2) or twice at 14 and 7 days before expected harvest (plot 3) at a rate of 0.125 L (equivalent to 0.0125 kg alpha-cypermethrin/ha) of formulated product /ha to curly kale. The nominal spray volume used was 400 L/ha.
At Trials AF/8815/BA/3 and 4 conducted in Southern Europe, BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g a.s./L), was foliar applied only once at 7 days before expected harvest (plot 2) at a rate of 0.25 L(equivalent to 0.025 kg alpha-cypermethrin/ha) of formulated product /ha to curly kale. The nominal spray volume used was 400 L/ha.
Specimens of kale leaves were collected at the day of the last application (0 DALA) from each plot as well as 3, 7 and 14 days thereafter.

Samples were stored frozen for a maximum of 213 days until analysis.

Table 6.3.2-4: Target application rates and timings for curly kale

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2005	2	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	7 ±1 days before harvest
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	1st appl. 7 ±1, 2nd appl. 14 ±1 days before harvest
	2	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.025	400	7 ±1 days before harvest

2. Description of analytical procedures

Specimens were analysed using BASF analytical method no. 567/0, which quantifies the residues of total cypermethrin by means of HPLC-MS/MS.
Principle of method:
Residues of cypermethrin (alpha-cypermethrin or cypermethrin) are extracted with a mixture of methanol/water/2N hydrochloric acid (70:25:5). After centrifugation, an aliquot of the extract is cleaned by liquid/liquid partition against cyclohexane. The residues are further purified (if needed) using a silica gel solid phase extraction (SPE) column. The final determination of total cypermethrin residues is performed by HPLC-MS/MS, monitoring the ion transition from m/z 433.2 / 191.1 (the ammonium adduct of cypermethrin).
The limit of quantitation (LOQ) of the method for alpha-cypermethrin is 0.01 mg/kg.

Table 6.3.2-5: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in curly kale

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Foliage	0.01	5	85.0	16.8	19.8
	0.5	2	91.5	-	-
	1.0	3	82.5	11.7	14.1
	Overall	10	85.6	12.9	15.1

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.2-6, detailed residue levels are shown in Table 6.3.2-7 and Table 6.3.2-8.
After a single broadcast foliar application of alpha-cypermethrin targeting 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in curly kale foliage ranged between 0.154 mg/kg and 0.159 mg/kg. Residues declined to levels between 0.203-0.227 mg/kg, 0.171-0.237 mg/kg and 0.083-0.116 mg/kg at 3, 7 and 14 days after application.

After two broadcast foliar applications of alpha-cypermethrin targeting at 0.0125 kg a.s./ha and application, initial residues were 0.241 mg/kg in kale foliage collected immediately after the last application. Residues declined to levels between 0.170-0.348 mg/kg, 0.084-0.383 mg/kg and 0.152-0.245 mg/kg at 3, 7 and 14 days after application.

After a single broadcast foliar application of alpha-cypermethrin targeting 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in leafy cabbage ranged between 0.349 mg/kg and 0.517 mg/kg. Residues declined to levels between 0.278-0.436 mg/kg, 0.257-0.263 mg/kg and 0.137-0.243 mg/kg at 3, 7 and 14 days after application

In the control specimens, no residues of total cypermethrin above the limit of quantitation were found, with the following exception: At the trial located in the United Kingdom (AF188151BA/5) apparent residues of cypermethrin were approximately at the limit of quantitation (0.01 mg/kg) in the control material collected at 0 and 3 DALA, but were <0.01 mg/kg in untreated controls collected at 7 and 14 DALA. These residues were relatively low compared with the residues in the associated treated specimens. The source of the contamination could not be determined.

Table 6.3.2-6: Summary of residues of BAS 310 I in leafy cabbage after application of BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2005-2006	BAS 310 40 I (EC) 1 x 0.125 kg a.s./ha	0	48-49	Foliage	0.154-0.159
			3	48-49	Foliage	0.203-0.227
			7	48-49	Foliage	0.171-0.237
			14	48-49	Foliage	0.083-0.116
		BAS 310 40 I (EC) 2 x 0.0125 kg a.s./ha	0	48-49	Foliage	0.241
			3	48-49	Foliage	0.170-0.348
			7	48-49	Foliage	0.084-0.383
			14	48-49	Foliage	0.152-0.245
S-EU	2005	BAS 310 40 I (EC) 1 x 0.025 kg a.s./ha	0	47-48	Foliage	0.349-0.517
			3	47-48	Foliage	0.278-0.436
			7	49	Foliage	0.257-0.263
			14	49	Foliage	0.137-0.243

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Immediately after the last of two treatments at a target rate of 0.0125 kg alpha-cypermethrin/ha, residues in leafy cabbage foliage were 0.241 mg/kg and ranged between 0.170-0.348 mg/kg and 0.084-0.383 mg/kg after 3 and 7 days, respectively.
Immediately after a single application of alpha-cypermethrin targeting 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in leafy cabbage foliage ranged between 0.349 mg/kg and 0.517 mg/kg. Residues declined to levels between 0.278-0.436 mg/kg and 0.257-0.263 mg/kg at 3 and 7 days after application, respectively.

Table 6.3.2-7: Residues of BAS 310 I in leafy cabbage after one or two applications of BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:181963 **Doc ID:**2006/1026862 **Trial No.:**AF/8815/BA/1 **GLP:**yes **Year**2005	Curly kale	Germany	BAS 310 40 I 1 x 0.0125	48-49	0	Foliage	0.154
				48-49	3	Foliage	0.227
				48-49	7	Foliage	0.171
				48-49	14	Foliage	0.083
			BAS 310 40 I 2 x 0.0125	48-49	0	Foliage	0.241
				48-49	3	Foliage	0.170
				48-49	7	Foliage	0.084
				48-49	14	Foliage	0.152
Study code:181963 **Doc ID:**2006/1026862 **Trial No.:**AF/8815/BA/5 **GLP:**yes **Year**2006	Curly kale	The United Kingdom	BAS 310 40 I 1 x 0.0125	48	0	Foliage	0.159
				49	3	Foliage	0.203
				49	7	Foliage	0.237
				49	14	Foliage	0.116
			BAS 310 40 I 2 x 0.0125	48	0	Foliage	0.241
				49	3	Foliage	0.348
				49	7	Foliage	0.383
				49	14	Foliage	0.245

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Table 6.3.2-8: Residues of BAS 310 I in leafy cabbage after one application of BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:181963 **Doc ID:**2006/1026862 **Trial No.:**AF/8815/BA/3 **GLP:**yes **Year**2005	Curly kale	Italy	BAS 310 40 I 1 x 0.025	47-48	0	Foliage	0.517
				47-48	3	Foliage	0.436
				49	7	Foliage	0.263
				49	14	Foliage	0.243
Study code:181963 **Doc ID:**2006/1026862 **Trial No.:**AF/8815/BA/4 **GLP:**yes **Year**2005	Curly kale	Greece	BAS 310 40 I 1 x 0.025	48	0	Foliage	0.349
				48	3	Foliage	0.278
				49	7	Foliage	0.257
				49	14	Foliage	0.137

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Report: CA 6.3.2/2
Diehl M., 2007a
Study on the residue behaviour of BAS 310 I in leafy cabbage after treatment with BAS 310 40 I under open field conditions in Southern and Northern Europe, 2006
2007/1013342

Guidelines: EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 96/68

GLP: yes
(certified by Swiss Federal Office of Public Health, Berne, Switzerland)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I

Description:	BAS 310 40 I (EC)
Lot/Batch #:	1171, Alpha-Cypermethrin: 100 g/L
Purity:	not reported
CAS#:	67375-30-8
Development code:	not reported
Spiking levels:	0.01, 0.10 and 1.0 mg/kg

2. Test Commodity:

Crop:	Leafy cabbage
Type:	Leafy brassica
Variety:	Reflex, Winnetou, Castelard, Manoko F-1
Botanical name:	*Brassica oleracea*
Crop parts(s) or processed commodity:	RAC leaves
Sample size:	min 2 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2006 growing season, four field trials were performed in leafy cabbage in France (Northern and Southern European region), Spain and The Netherlands.
In two trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to leafy cabbage plants either once or twice to separate plots at a target rate of 0.0125 kg alpha-cypermethrin/ha in a spray volume of 400 L/ha.

In two trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to leafy cabbage plants once at a target rate of 0.025 kg alpha-cypermethrin/ha in a spray volume of 400 L/ha.

Specimens of cabbage leaves were collected immediately after the last application from each plot, as well as 3-4, 6-8 and 14-15 days thereafter.

Samples were stored frozen a for a maximum of 136 days until analysis.

Table 6.3.2-9: Target application rates and timings for leafy cabbage

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2006	2	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	7 ±1 days before harvest
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	1st appl. 7 ±1, 2nd appl. 14 ±1 days before harvest
	2	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.025	400	7 ±1 days before harvest

2. Description of analytical procedures

Specimens were analysed using BASF analytical method no. 567/0, which has a limit of quantitation of 0.01 mg/kg in all sample materials.
Residues of alpha-cypermethrin were extracted from leafy cabbage (leaves) using methanol/water and hydrochloric acid. An aliquot of the extract was centrifuged and partitioned against cyclohexane/water. The cyclohexane phase was evaporated and the residue was dissolved in acidic methanol/water. Residues of alpha-cypermethrin were determined by high performance liquid chromatography (HPLC) with tandem mass spectrometry (LC-MS/MS).

Table 6.3.2-10: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in leafy cabbage

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Leaves	0.01	2	89.5	-	-
	0.10	1	83	-	-
	1.0	1	97	-	-
	0.01-1.0 mg/kg	4	89.8	9.0	10.0

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.2-11, detailed residue levels are shown in Table 6.3.2-12 and Table 6.3.2-13.
After one application at 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in leafy cabbage leaves were 0.26 mg/kg. Residues declined to levels between 0.18-0.26 mg/kg, 0.15-0.22 mg/kg and 0.074-0.17 mg/kg at 3-4, 7-8 and 14-15 days after application.

After two applications at 0.0125 kg alpha-cypermethrin/ha, initial residues ranged between 0.47 mg/kg and 0.48 mg/kg. Residues declined to levels between 0.35-0.59 mg/kg, 0.21-0.35 mg/kg and 0.19-0.24 mg/kg at 3-4, 7-8 and 14-15 days after application.

After a single application at 0.025 g a.s./ha, initial residues of alpha-cypermethrin in leafy cabbage ranged between 0.066 mg/kg and 0.13 mg/kg. Residues declined to levels between 0.046-0.056 mg/kg, 0.011-0.056 mg/kg and <0.01-0.011 mg/kg at 3, 6-7 and 14 days after application

No residues above the LOQ were found in any of the untreated samples.

Table 6.3.2-11: Summary of residues of BAS 310 I in leafy cabbage after application of BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2006	BAS 310 40 I (EC) 1 x 0.125 kg a.s./ha	0	48-49	Leaves	0.260
			3-4	48-49	Leaves	0.18-0.26
			7-8	48-49	Leaves	0.15-0.22
			14-15	48-49	Leaves	0.074-0.17
		BAS 310 40 I (EC) 2 x 0.0125 kg a.s./ha	0	48-49	Leaves	0.47-0.48
			3-4	48-49	Leaves	0.35-0.59
			7-8	48-49	Leaves	0.21-0.35
			14-15	48-49	Leaves	0.19-0.24
S-EU	2006	BAS 310 40 I (EC) 1 x 0.025 kg a.s./ha	0	47	Leaves	0.066-0.13
			3	47	Leaves	0.046-0.056
			6-7	47-48	Leaves	0.011-0.056
			14	48-49	Leaves	<0.01-0.011

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

After the last of two applications of BAS 310 40 I at a target rate of 0.0125 kg alpha-cypermethrin/ha, initial residues of alpha-cypermethrin ranged between 0.47 mg/kg and 0.48 mg/kg. Residues declined to levels between 0.35-0.59 mg/kg at 3-4 days after the last application.

After a single application of BAS 310 40 I at a target rate of 0.025 g a.s./ha, initial residues of alpha-cypermethrin in leafy cabbage ranged between 0.066 mg/kg and 0.13 mg/kg. Residues declined to levels between 0.046-0.056 mg/kg and 0.011-0.056 mg/kg at 3 and 6-7 days after application, respectively.

Table 6.3.2-12: Residues of BAS 310 I in leafy cabbage after one or two applications of BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:227098 **Doc ID:**2007/1013342 **Trial No.:**A/NF/I/06/115 **GLP:**yes **Year**2006	Leafy cabbage	France	BAS 310 40 I 1 x 0.0125	49	0	Leaves	0.26
				49	3	Leaves	0.26
				49	8	Leaves	0.22
				49	14	Leaves	0.17
			BAS 310 40 I 2 x 0.0125	49	0	Leaves	0.47
				49	3	Leaves	<u>0.59</u>
				49	8	Leaves	0.35
				49	14	Leaves	0.24
Study code:227098 **Doc ID:**2007/1013342 **Trial No.:**A/NL/I/06/116 **GLP:**yes **Year**2006	Leafy cabbage	The Netherlands	BAS 310 40 I 1 x 0.0125	48-49	0	Leaves	0.26
				48-49	4	Leaves	0.18
				48-49	7	Leaves	0.15
				48-49	15	Leaves	0.074
			BAS 310 40 I 2 x 0.0125	48-49	0	Leaves	0.48
				48-49	4	Leaves	<u>0.35</u>
				48-49	7	Leaves	0.21
				48-49	15	Leaves	0.19

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Table 6.3.2-13: Residues of BAS 310 I in leafy cabbage after one application of BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:227098 **Doc ID:**2007/1013342 **Trial No.:**A/SF/I/06/117 **GLP:**yes **Year**2006	Leafy cabbage	France	BAS 310 40 I 1 x 0.025	47	0	Leaves	0.066
				47	3	Leaves	<u>0.046</u>
				48	7	Leaves	0.011
				49	14	Leaves	<0.01
Study code:227098 **Doc ID:**2007/1013342 **Trial No.:**A/SP/I/06/118 **GLP:**yes **Year**2006	Leafy cabbage	Spain	BAS 310 40 I 1 x 0.025	47	0	Leaves	0.13
				47	3	Leaves	<u>0.056</u>
				47-48	6	Leaves	0.056
				48	14	Leaves	0.011

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Report: CA 6.3.2/3
Anonymous, 2007a
Residue behaviour of Alpha-Cypermethrin in/on fennel (bulbs formed by leafs stems), curly kale (leaves), blanched celery (stems) and spinach (leaves) outdoor after application of Fastac SC Super Contact (SC, 100 g/L) in Germany, 2006
2007/1035745
Guidelines: none
GLP: yes
(certified by Bayerisches Landesamt fuer Arbeitsschutz, Arbeitsmedizin und Sicherheitstechnik, Muenchen, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 41 I
Lot/Batch #: not reported (BAS 310 41 I, 100 g/L alpha-cypermethrin, SC)
Purity: not relevant
CAS#: alpha-cypermethrin: 67375-30-8
Development code: not applicable
Spiking levels: 0.01-0.1 mg/kg

2. Test Commodity:

Crop: Curly kale
Type: Brassica vegetables
Variety: Kobold, Winterbor, Winnetou
Botanical name: *Brassica oleracea* var. *sabellica* L.
Crop parts(s) or processed commodity: Leaves
Sample size: min. 0.5 kg

B. STUDY DESIGN

1. Test procedure

During the growing season 2006 residue decline, shortened decline and harvest trials, were performed in fennel (bulbs formed by leaf stems), curly kale (leaves), blanched celery (stems) and spinach (leaves) as field trials on several sites in Germany.
Fastac SC Super Contact (alpha-cypermethrin, SC 100) was applied foliar 2 times at rates of 0.125 L product per hectare (12.5 g/ha alpha-cypermethrin) during growing season. The trials were conducted in order to determine the magnitude of the residues of the active ingredients in or on fennel (bulbs formed by leaf stems), curly kale (leaves), blanched celery (stems) and spinach (leaves). The samples were collected 0 - 7 -10 -14 (decline curves), 0 - 7 -14 (shortened decline study) and 14 days (harvest trials) after last application, respectively.

Table 6.3.2-14: Target application rates and timings for curly kale

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2006	4	2	F	BAS 310 41 I (SC)	alpha-cypermethrin	0.0125	600-900	n.r.

n.r. = not reported

2. Description of analytical procedures

The samples were analysed for alpha-cypermethrin with modified method DFG S19. The limit of quantification (LOQ) was 0.010 mg/kg (alpha-cypermethrin).
Average recoveries from untreated samples, fortified with alpha-cypermethrin were 69 -105 % in fennel and 74 - 92 % in curly kale at fortification levels between 0.01 mg/kg and 0.10 mg/kg. In blanched celery the recoveries were 75 - 110 % at fortification levels between 0.01 mg/kg and 0.5 mg/kg. In spinach the recoveries were 90 - 100 % at fortification levels between 0.01 mg/kg and 1.0 mg/kg.
Alpha-cypermethrin: the samples were analysed for alpha-cypermethrin by the method DFG S 19 modified. The samples are extracted with acetone/water 2+1 (v+v) with subsequent extraction with cyclohexane/ethyl acetate 1+1 (v+v) and partition into acetone/cyclohexane/ethyl acetate. The extracts are cleaned up by gel permeation cbromatography on a Bio Beads SX-3 column followed by a silica gel fractionation. The residues are determined by gas chromatography using an electron capture detector. The quantification is done by external standardisation. The residues are not corrected for the recovery rates.

Table 6.3.2-15: Summary of recoveries of alpha-cypermethrin in curly kale leaves

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
DFG S 19 (modified)		**alpha-cypermethrin**			
curly kale leaves	**0.01 / 0.1 / 1.0**	**8**	**80**	**6**	**7**

II. RESULTS AND DISCUSSION

In context of this summary, only the results of alpha-cypermethrin for curly kale are reported. Residues of alpha-cypermethrin were at 0 DALA in leaves of curly kale in the range of 0.24 to 0.62 mg/kg. At 7 DALA, they ranged between 0.07 and 0.26 mg/kg, at 10 DALA they were again lower with values between 0.09 and 0.24 mg/kg. Finally, at 14 DALA they were in the range of 0.05 to 0.19 mg/kg.

Table 6.3.2-16: Summary of residues of BAS 310 I in curly kale from trials according to critical GAP after application of BAS 310 41 I

Region	Year	No. of Appl.	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin (BAS 310 I)
N-EU	2006	2	BAS 310 41 I (SC)	0	16-49	leaves	0.24 - 0.62
				7±1	18-49		0.07 - 0.26
				10±1	33-49		0.09 - 0.24
				14±1	33-49		0.05 - 0.19

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Immediately after the last of two applications of SC-formulation BAS 310 41 I at rates of 0.0125 kg alpha-cypermethrin/ha, residues in curly kale leaves were in the range of 0.24 to 0.62 mg/kg. Residues declined to 0.07-0.26 mg/kg and 0.09-0.24 mg/kg at 7±1 and 10±1 days after the last application, respectively.

Table 6.3.2-17: Residues of BAS 310 I after two applications of the formulation BAS 310 41 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:not reported **Doc ID:**2007/1035745 **Trial No.:**RU-I-05 06 NW BN 1/1 **GLP:**yes (only analytical phase) **Year**2006	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	16-17	0	leaves	0.24
				18-19	7	leaves	0.07
				33	10	leaves	0.09
				33	14	leaves	0.08
Study code:not reported **Doc ID:**2007/1035745 **Trial No.:**RU-I-05 06 NW BN 1/2 **GLP:**yes (only analytical phase) **Year**2006	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	16-17	0	leaves	0.62
				18-19	7	leaves	0.20
				33	10	leaves	0.24
				33	14	leaves	0.19
Study code:not reported **Doc ID:**2007/1035745 **Trial No.:**RU-I-05 06 NW BN 1/4 **GLP:**yes (only analytical phase) **Year**2006	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	39	7	leaves	0.24
				39	14	leaves	0.17
Study code:not reported **Doc ID:**2007/1035745 **Trial No.:**RU-I-05 06 HE WE 1/1 **GLP:**yes (only analytical phase) **Year**2006	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	48-49	0	leaves	0.54
				48-49	7	leaves	0.26
				49	10	leaves	0.16
				49	14	leaves	0.05

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Report: CA 6.3.2/4
Schneider D., 2008a
Determination of the residues of alpha-Cypermethrin in curly kale (leaves) and little radish (tuber)
2014/1083417
Guidelines: none
GLP: yes
(certified by Bayerisches Landesamt fuer Arbeitsschutz, Arbeitsmedizin und Sicherheitstechnik, Muenchen, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: BAS 310 41 I
Lot/Batch #: not reported (BAS 310 41 I, 100 g/L alpha-cypermethrin, SC)
Purity: not relevant
CAS#: alpha-cypermethrin: 67375-30-8
Development code: not applicable
Spiking levels: 0.01-0.5 mg/kg

2. Test Commodity:

Crop: Curly kale
Type: Brassica vegetables
Variety: Winnetou, Winterboer, Westländer
Botanical name: *Brassica oleracea* var. *sabellica* L.
Crop parts(s) or processed commodity: Leaves
Sample size: min. 1.0 kg

B. STUDY DESIGN

1. Test procedure

During the growing season 2007, four residue decline trials in curly kale and two residue decline trials in radish were conducted as field trials on several sites in Germany.
Fastac SC Super Contact (alpha-cypermethrin, SC 100) was applied foliar 2 times at rates of 0.125 L product per hectare (12.5 g/ha alpha-cypermethrin) during the growing season. The trials were conducted in order to determine the magnitude of the residues of the active ingredients in or on curly kale (leaves) and radish (tubers). In case of curly kale, specimens were sampled at 0, 7, 10 and 14 DALA, in case of radish samples were taken at 0, 3 and 7 DALA.

Table 6.3.2-18: Target application rates and timings for curly kale

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2007	4	2	F	BAS 310 41 I (SC)	alpha-cypermethrin	0.0125	600	n.r.

n.r. = not reported

2. Description of analytical procedures

The samples were analysed for alpha-cypermethrin with modified method DFG S19. The limit of quantification (LOQ) was 0.010 mg/kg (alpha-cypermethrin).
Recoveries from untreated samples, fortified with alpha-cypermethrin were between 70 and 93% in curly kale and between 81 and 105% in radish at fortification levels between 0.01 mg/kg and 0.5 mg/kg.
Alpha-cypermethrin: the samples were analysed for alpha-cypermethrin by the method DFG S 19 modified. The samples are extracted with acetone/water 2+1 (v+v) with subsequent extraction with cyclohexane/ethyl acetate 1+1 (v+v) and partition into acetone/cyclohexane/ethyl acetate. The extracts are cleaned up by gel permeation cbromatography on a Bio Beads SX-3 column followed by a silica gel fractionation. The residues are determined by gas chromatography using an electron capture detector. The quantification is done by external standardisation. The residues are not corrected for the recovery rates.

Table 6.3.2-19: Summary of recoveries of alpha-cypermethrin in curly kale leaves

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
DFG S 19 (modified)		alpha-cypermethrin			
curly kale leaves	0.01 / 0.02 / 0.1 / 0.5	11	81.5	6.8	8.4

II. RESULTS AND DISCUSSION

In context of this summary, only the results of alpha-cypermethrin for curly kale are reported. Residues of alpha-cypermethrin were at 0 DALA in leaves of curly kale in the range of 0.18 to 0.58 mg/kg. At 7 DALA, they ranged between 0.05 and 0.34 mg/kg, at 10 DALA they were on a comparable level with values between 0.07 and 0.34 mg/kg. Finally, at 14 DALA they were in the range of 0.03 to 0.26 mg/kg.

Table 6.3.2-20: Summary of residues of BAS 310 I in curly kale from trials according to critical GAP after application of BAS 310 41 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	Alpha-Cypermethrin (BAS 310 I)
N-EU	2007	BAS 310 41 I (SC) 2 x 0.0125 kg a.s./ha	0	44-49	leaves	0.18 - 0.58
			7±1	45-49	leaves	0.05 - 0.34
			10±1	45-49	leaves	0.07 - 0.34
			14±1	45-49	leaves	0.03 - 0.26

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Immediately after the last of two applications of SC-formulation BAS 310 41 I at rates of 0.0125 kg alpha-cypermethrin/ha, residues in curly kale leaves were in the range of 0.18 to 0.58 mg/kg. Residues declined to 0.05-0.34 mg/kg and 0.07-0.34 mg/kg at 7±1 and 10±1 days after the last application, respectively.

Table 6.3.2-21: Residues of BAS 310 I after two applications of the formulation BAS 310 41 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:not reported **Doc ID:**2014/1083417 **Trial No.:**RU-I-03 07 HE WE 2/1 **GLP:**yes **Year**2007	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	47-49	0	leaves	0.18
				47-49	7	leaves	0.05
				49	10	leaves	0.08
				49	14	leaves	0.06
Study code:not reported **Doc ID:**2014/1083417 **Trial No.:**RU-I-03 07 NW BN 2/1 **GLP:** yes **Year**2007	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	47	0	leaves	0.34
				47	7	leaves	0.06
				48	10	leaves	0.07
				48	14	leaves	0.03
Study code:not reported **Doc ID:**2014/1083417 **Trial No.:**RU-I-03 07 NW BN 2/2 **GLP:** yes **Year**2007	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	47	0	leaves	0.38
				48	7	leaves	0.23
				48	10	leaves	0.19
				48	14	leaves	0.26
Study code:not reported **Doc ID:**2014/1083417 **Trial No.:**RU-I-03 07 NW BN 2/3 **GLP:** yes **Year**2007	Curly kale	Germany (N)	BAS 310 41 I 2 x 0.0125	44	0	leaves	0.58
				45	7	leaves	0.34
				45	10	leaves	0.34
				45	14	leaves	0.25

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

~~**Report:** CA 6.3.2/5~~
~~Klimmek S.,Gizler A., 2014c~~
~~Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in cabbage (chinese) after two applications with BAS 310 55 I under field conditions in Southern France, Greece, Italy and Spain, 2013~~
~~2013/1416285~~
~~**Guidelines:** none~~
~~**GLP:** yes~~
~~(certified by Freie und Hansestadt Hamburg, Behoerde fuer Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg, Germany)~~

Report: CA 6.3.2/5
Klimmek S.,Gizler A., 2016 a
Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in cabbage (chinese) after two applications with BAS 310 55 I under field conditions in Southern France, Greece, Italy and Spain, 2013
2016/1154540
Guidelines: none
GLP: yes
(certified by Freie und Hansestadt Hamburg, Behoerde fuer Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1.

Test Material:	Alpha-Cypermethrin BAS 31055I
Description:	BAS 310 55 I (ME)
Lot/Batch #:	101198: 50 g/L nominal
Purity:	not reported
CAS#:	67375-30-8 (alpha-cypermethrin)
Development code:	not reported
Spiking levels:	0.01, 0.1 and 8.0 mg/kg

2. **Test Commodity:**

Crop:	Chinese cabbage
Type:	Leafy brassica
Variety:	Kaboko, Guest Star, Paranlo, Manoko (Bejo)
Botanical name:	*Brassica rapa* subsp. *pekinensis*
Crop parts(s) or processed commodity:	Head with wrapper leaves
Sample size:	Head with wrapper leaves: 0 DALA: ≥0.5 kg / ≥ 12 units 3±1, 7±1, 14±1 DALA: ≥2 kg / ≥ 12 units

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2013 growing season, four field trials were conducted under field conditions in representative cabbage (Chinese) growing areas in Southern France, Greece, Italy and Spain to determine the residue level of alpha-cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).

Two foliar applications with BAS 310 55 I, an ME formulation of alpha-cypermethrin (50.0 g a.s./L), were made to plot 2 to cabbage (Chinese), at a rate of 0.25 L formulated product/ha, equal to 12.5 g alpha-cypermethrin/ha, at 14 and 6-7 day(s) before harvest (DBH). The nominal spray volume used was 200 L/ha.

Treated cabbage (Chinese) (head with wrapper leaves) specimens of trials S13-00427-01, -02 and -03 were sampled at 0 DALA, at 3 DALA, at 6-7 DALA and at 13-14 DALA.
Treated cabbage (Chinese) (head with wrapper leaves) specimens of trial S13-00427-04 were sampled at 0 DALA, at 3 DALA and at 6 DALA. As the trial site had been ploughed prior to the 14 ± 1 DALA sampling point by mistake, the 14 ± 1 DALA could not be taken.

The maximum storage interval from harvest until analysis was 368 days.

Table 6.3.2-22: Target application rates and timings for Chinese cabbage

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2013	4	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	200	1st appl. 14, 2nd appl. 6-7 days before harvest

2. Description of analytical procedures

All Chinese cabbage specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
The residues of alpha-cypermethrin in Chinese cabbage specimens were extracted from plant matrices using a mixture of methanol / water / 2N hydrochloric acid (70:25:5, v/v/v). After centrifugation and partition of an aliquot of the extract into cyclohexane, evaporation to dryness and dissolving in methanol / water (80+20, v+v), the final determination of alpha-cypermethrin was performed by LC-MS/MS.
As a modification to optimize the recovery of alpha-cypermethrin during the liquid/liquid-partition with cyclohexane, the extract was partitioned three times in order to adapt the method to the laboratory situation.
The limit of quantitation (LOQ) was 0.01 mg/kg for alpha-cypermethrin.

Table 6.3.2-23: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in Chinese cabbage

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Chinese cabbage / Head with wrapper leaves	0.01	3	86.8	12.8	14.7
Chinese cabbage / Head with wrapper leaves	0.10	3	84.9	13.0	15.4
Chinese cabbage / Head with wrapper leaves	8.0	1	97.9	-	-
Overall		**7**	**87.6**	**11.5**	**13.2**

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.2-24. Detailed results are given in Table 6.3.2-25.

At 0 DALA the residues of alpha-cypermethrin (BAS 310 I) ranged between 0.090 mg/kg and 0.27 mg/kg. At 2-3 DALA the residues of alpha-cypermethrin (BAS 310 I) ranged between 0.019 mg/kg and 0.11 mg/kg. At 6-7 DALA residues between 0.013 mg/kg and 0.041 mg/kg of alpha-cypermethrin (BAS 310 I) were found. At 13-14 DALA the residues of alpha-cypermethrin (BAS 310 I) ranged between <0.01 mg/kg (below the LOQ) and 0.027 mg/kg.
No residues of alpha-cypermethrin (BAS 310 I) above the LOQ were found in the untreated specimens of this study. ~~, with the exception of sample L1300490001 of trial S13-00427-04 (L130049), taken at 0 DALA, where an alpha-cypermethrin (BAS 310 I) residue of 0.018 mg/kg was detected. The source of the contamination has not been determined. Taking into account the corresponding residues in the treated specimens the residue in the control sample had no impact on the study.~~

Table 6.3.2-24: Summary of residues of BAS 310 I in Chinese cabbage after application of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
S-EU	2013	BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	0	43-48	Head with wrapper leaves	0.090-0.27
			2-3	45-49	Head with wrapper leaves	0.019-0.11
			6-7	47-49	Head with wrapper leaves	0.013-0.041
			13-14	47-49	Head with wrapper leaves	<0.01-0.027

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Immediately after the last of two applications of alpha-cypermethrin at a target rate of 0.0125 g a.s./ha, the residues of alpha-cypermethrin in Chinese cabbage ranged between 0.090 mg/kg and 0.27 mg/kg. At 2-3 days after the last application the residues of alpha-cypermethrin (BAS 310 I) ranged between 0.019 mg/kg and 0.11 mg/kg.

Table 6.3.2-25: Residues of BAS 310 I in Chinese cabbage after two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408607 **Doc ID:**2013/1140312 **Trial No.:**S13-00427-01 / L130046 **GLP:**yes **Year**2013	Chinese cabbage	France	BAS 310 55 I 2 x 0.0125	47-48	0	Head with wrapper leaves	0.13
				47-49	3	Head with wrapper leaves	<u>0.086</u>
				47-49	7	Head with wrapper leaves	0.041
				47-49	14	Head with wrapper leaves	0.027
Study code:408607 **Doc ID:**2013/1140312 **Trial No.:**S13-00427-02 / L130047 **GLP:**yes **Year**2013	Chinese cabbage	Greece	BAS 310 55 I 2 x 0.0125	43 - 45	0	Head with wrapper leaves	0.27
				45 - 47	3	Head with wrapper leaves	<u>0.11</u>
				49	7	Head with wrapper leaves	0.036
				49	13	Head with wrapper leaves	<0.010
Study code:408607 **Doc ID:**2013/1140312 **Trial No.:**S13-00427-03 / L130048 **GLP:**yes **Year**2013	Chinese cabbage	Italy	BAS 310 55 I 2 x 0.0125	47	0	Head with wrapper leaves	0.10
				47	3	Head with wrapper leaves	<u>0.064</u>
				49	6	Head with wrapper leaves	0.029
				49	13	Head with wrapper leaves	0.013
Study code:408607 **Doc ID:**2013/1140312 **Trial No.:**S13-00427-04 / L130049 **GLP:**yes **Year**2013	Chinese cabbage	Spain	BAS 310 55 I 2 x 0.0125	47	0	Head with wrapper leaves	0.090
				47-48	2	Head with wrapper leaves	<u>0.019</u>
				48-49	6	Head with wrapper leaves	0.013

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

CA 6.3.3 Lettuce

Table 6.3.3-1: cGAP for the use of BAS 310 I in/on lettuce

Crop	Maximum applied dose	Water volume	PHI	Application method	Application timing
Lettuce, outdoor (North, Central)	2 x 0.010 kg BAS 310 I/ha	200-1000	3	Overall spray	BBCH 10 – 49
Lettuce, outdoor (South)	2 x 0.010 kg BAS 310 I/ha	200-1000	3	Overall spray	BBCH 10 – 49
	1 x 0.020 kg BAS 310 I/ha	200-1000	3	Overall spray	BBCH 10 – 49

PHI = pre-harvest interval

Table 6.3.3-2: GAP information of residue trials conducted in lettuce in 2005-2006 in Northern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Northern EU	France (1) Germany (1) The Netherlands (1) The United Kingdom (1) 2005	BAS 310 40 I EC	spray appl.	0.0125	0.003	1	0 3 7 14	2006/ 1026855	No
				0.0125		2			
	France North (1) The United Kingdom (1) 2006	BAS 310 40 I EC	spray appl.	0.0125	0.003	1	0 3 7 14	2007/ 1007938	No
				0.0125		2			
		BAS 310 08 I WG		0.0125		1			
				0.0125		2			
	Denmark (1) France North (1) Germany (1) The United Kingdom (1) 2006	BAS 310 40 I EC	spray appl.	0.0125	0.003	1	0 3 7 14	2007/ 1008496	No
				0.0125		2			

0 Actual application rates varied by 10% at most
1 Days after last application

Table 6.3.3-3: GAP information of residue trials conducted in lettuce in 2005-2013 in Southern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Southern EU	France (1) Greece (1) Italy (1) Spain (1) 2005	BAS 310 40 I EC	spray appl.	0.025	0.006	1	0 3 7 14	2006/ 1026855	No
	Italy (1) Spain (1) 2006	BAS 310 40 I EC BAS 310 08 I WG	spray appl.	0.025	0.006	1	0 3 7 14	2007/ 1007938	No
	France (1) Greece (1) Italy (1) Spain (1) 2006	BAS 310 40 I EC	spray appl.	0.025	0.006	1	0 3-4 7-8 14	2007/ 1008496	No
	France (2) Greece (2) Italy (2) Spain (2) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.006	2	0 1 2-3 6-8	2014/ 1140312	No

0 Actual application rates varied by 10% at most
1 Days after last application

The studies not yet evaluated are summarized in the following chapter.

In these studies, most lettuces used were open-leaf varieties. Supplemental information to support the leaf status of the varieties used for data generation can be found in BASF DocID 2016/1338439.

Report: CA 6.3.3/1
North L., 2007b
Study on the residue behaviour of Alpha-Cypermethrin in head lettuce after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2005
2006/1026855
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I

Description:	BAS 310 40 I (SC)
Lot/Batch #:	1171: 100 g/L
Purity:	not reported
CAS#:	67375-30-8
Development code:	not reported
Spiking levels:	0.01, 1.0 and 2.0 mg/kg

2. Test Commodity:

Crop:	Lettuce, head
Type:	Leafy vegetable
Variety:	Estelle, Little Gem, Tamburo, Nobilan, Sagesse, Gentilina, Filipus, Atraxion
Botanical name:	*Lactuca sativa*
Crop parts(s) or processed commodity:	Heads
Sample size:	12 units (min 1.0 kg)

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2005 growing season, eight field trials were conducted in head lettuce in France (Northern and Southern European region), Germany, Greece, Italy, Spain, the Netherlands and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to head lettuce plants on separate plots either once or twice at a target rate of 0.0125 kg alpha-cypermethrin/ha in a nominal spray volume of 400 L/ha.

In four trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to head lettuce plants once at a target rate of 0.025 kg alpha-cypermethrin/ha in an nominal spray volume of 400 L/ha.

Specimens of lettuce heads were collected immediately after the last application from each plot, as well as 2-3, 7-8 and 14-15 days thereafter.

Samples were stored frozen at or below -10°C for a maximum of 501 days until analysis.

Table 6.3.3-4: Target application rates and timings for lettuce

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2005	4	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	7 ±1 days before harvest
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	1st appl. 14 ±1, 2nd appl. 7 ±1 days before harvest
	4	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.025	400	7 ±1 days before harvest

2. Description of analytical procedures

Specimens were analysed using BASF analytical method No. 567/0, determines the underivatised cypermethrin by means of HPLC-MS/MS.
Principle of the method:
Residues of cypermethrin (alpha-cypermethrin or cypermethrin) are extracted with a mixture of methanol/water/2N hydrochloric acid (70:25:5). After centrifugation, an aliquot of the extract is cleaned by liquid/liquid partition against cyclohexane. The residues are further purified (if needed) using a silica gel solid phase extraction (SPE) column. The final determination of total cypermethrin residues is performed by HPLC-MS/MS, monitoring the ion transition from m/z 433.2 / 191.1 (the ammonium adduct of cypermethrin).
The limit of quantitation (LOQ) of the method for alpha-cypermethrin is 0.01 mg/kg.

Table 6.3.3-5: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in lettuce

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
head	0.01	3	88.4	13.0	15
	1.0	2	91.2	-	-
	2.0	1	86.8	-	-
	Overall	6	89	8	9

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.3-6, detailed residue levels are shown in Table 6.3.3-7 and Table 6.3.3-8.
After one application at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.282 mg/kg and 0.421 mg/kg. Residues declined to levels between 0.117-0.382 mg/kg, 0.042-0.243 mg/kg and <0.01-0.194 mg/kg at 2-3, 7 and 14 days after application.

After two applications at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.315 mg/kg and 0.654 mg/kg. Residues declined to levels between 0.125-0.722 mg/kg, 0.052-0.491 mg/kg and <0.01-0.412 mg/kg at 2-3, 7 and 14 days after application.

After a single application at a target rate of 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.299 mg/kg and 1.161 mg/kg. Residues declined to levels between 0.103-0.585 mg/kg, 0.062-0.520 mg/kg and 0.013-0.112 mg/kg at 3, 7-8 and 14-15 days after application.

In the control specimens, no residues of total cypermethrin above the limit of quantitation were found.

Table 6.3.3-6: Summary of residues of BAS 310 I in lettuce after application of BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2005	BAS 310 40 I (EC) 1 x 0.125 kg a.s./ha	0	45-49	Head	0.282-0.421
			2-3	45-49	Head	0.117-0.382
			7	47-49	Head	0.042-0.243
			14	48-49	Head	<0.01-0.194
		BAS 310 40 I (EC) 2 x 0.0125 kg a.s./ha	0	45-49	Head	0.315-0.654
			2-3	45-49	Head	0.125-0.722
			7	47-49	Head	0.052-0.491
			14	48-49	Head	<0.01-0.412
S-EU	2005	BAS 310 40 I (EC) 1 x 0.025 kg a.s./ha	0	41-47	Head	0.299-1.161
			3	41-48	Head	0.103-0.585
			7-8	43-49	Head	0.062-0.520
			14-15	45-51	Head	0.013-0.112

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

After two applications of alpha-cypermethrin at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.315 mg/kg and 0.654 mg/kg which declined to levels between 0.125-0.722 mg/kg and 0.052-0.491 mg/kg at 2-3 and 7 days, respectively, after the last application.

After a single application of alpha-cypermethrin at a target rate of 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.299 mg/kg and 1.161 mg/kg. Residues declined to levels between 0.103-0.585 mg/kg and 0.062-0.520 mg/kg at 3 and 7-8 days, respectively, after application.

Table 6.3.3-7: Residues of BAS 310 I in lettuce after one or two applications of BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/1 **GLP:**yes **Year**2005	Lettuce, head	France	BAS 310 40 I 1 x 0.0125	45-47	0	Head	0.421
				47	2	Head	0.382
				47-49	7	Head	0.243
				49	14	Head	0.194
			BAS 310 40 I 2 x 0.0125	45-47	0	Head	0.654
				47	2	Head	<u>0.722</u>
				47-49	7	Head	0.491
				49	14	Head	0.412
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/2 **GLP:**yes **Year**2005	Lettuce, head	The United Kingdom	BAS 310 40 I 1 x 0.0125	47	0	Head	0.295
				45-47	3	Head	<u>0.244</u>
				49	7	Head	0.064
				49	14	Head	0.025
			BAS 310 40 I 2 x 0.0125	47	0	Head	0.401
				45-47	3	Head	0.236
				49	7	Head	0.086
				49	14	Head	0.047
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/3 **GLP:**yes **Year**2005	Lettuce, head	The Netherlands	BAS 310 40 I 1 x 0.0125	45	0	Head	0.282
				45-46	3	Head	0.117
				48-49	7	Head	0.042
				49	14	Head	<0.01
			BAS 310 40 I 2 x 0.0125	45	0	Head	0.442
				45-46	3	Head	<u>0.125</u>
				48-49	7	Head	0.052
				49	14	Head	<0.01
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/4 **GLP:**yes **Year**2005	Lettuce, head	Germany	BAS 310 40 I 1 x 0.0125	48-49	0	Head	0.399
				48-49	3	Head	0.165
				48-49	7	Head	0.167
				48-49	14	Head	0.112
			BAS 310 40 I 2 x 0.0125	48-49	0	Head	0.315
				48-49	3	Head	<u>0.221</u>
				48-49	7	Head	0.176
				48-49	14	Head	0.085

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Table 6.3.3-8: Residues of BAS 310 I in lettuce after one application of BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/5 **GLP:**yes **Year**2005	Lettuce, head	France	BAS 310 40 I 1 x 0.025	41-43	0	Head	0.448
				41-43	3	Head	<u>0.103</u>
				43-45	7	Head	0.067
				45-49	14	Head	0.013
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/6 **GLP:**yes **Year**2005	Lettuce, head	Italy	BAS 310 40 I 1 x 0.025	45-46	0	Head	0.737
				47-48	3	Head	<u>0.305</u>
				49	8	Head	0.065
				49	14	Head	0.021
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/7 **GLP:**yes **Year**2005	Lettuce, head	Spain	BAS 310 40 I 1 x 0.025	47	0	Head	0.299
				47	3	Head	<u>0.210</u>
				47	7	Head	0.062
				51	14	Head	0.022
Study code:182014 **Doc ID:**2006/1026855 **Trial No.:**AF/8817/BA/8 **GLP:**yes **Year**2005	Lettuce, head	Greece	BAS 310 40 I 1 x 0.025	47	0	Head	1.161* (1.354/0.968)
				48	3	Head	<u>0.585</u>*(0.535/ 0.635)
				49	7	Head	0.520* (0.534/0.482/ 0.543)
				49	15	Head	0.112* (0.08/0.145)

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation
* mean of multiple determinations

Report: CA 6.3.3/2
Oxspring S., 2007a
Study on the residue behaviour of Alpha-Cypermethrin in head lettuce after treatment with BAS 310 40 I or BAS 310 08 I under field conditions in Northern and Southern Europe during 2006
2007/1007938
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I, BAS 31008I

Description:	BAS 310 40 I (EC), BAS 310 08 I (WG)
Lot/Batch #:	1171, BAS 310 40 I, Alpha-Cypermethrin: 100 g/L; 40000, BAS 310 08 I, Alpha-Cypermethrin: 150 g/kg
Purity:	not reported
CAS#:	67375-30-8
Development code:	not reported
Spiking levels:	0.01, 1.0 and 2.0 mg/kg

2. Test Commodity:

Crop:	Lettuce, head
Type:	Leafy vegetable
Variety:	Jambis, Igoma, Filipo, Gentilina
Botanical name:	*Lactuca sativa*
Crop parts(s) or processed commodity:	Heads
Sample size:	1.0 kg/12 units

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2006 growing season, four field trials were conducted in representative head lettuce growing areas in the United Kingdom, France, Spain and Italy to determine the residue level of alpha-cypermethrin in or on Raw Agricultural Commodities (RAC).

Trials AF/10503/BA/1 and 2 (France and the United Kingdom) consisted of four treated plots. Two plots were treated with BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g a.s./L). The test item was foliar applied at a rate of 0.125 L of formulated product/ha either once 7 days before expected harvest (plot 2) or twice 14 and 7 days before expected harvest (plot 3). Two further plots were treated with BAS 310 08 I, a WG formulation of alpha-cypermethrin (150 g a.s./kg). The test item was foliar applied at a rate of 0.08 kg of formulated product /ha either once 7 days before expected harvest (plot 4) or twice 14 and 7 days before expected harvest (plot 5).
Trials AF/10503/BA/3 and 4 (Italy and Spain) consisted of two treated plots. One plot was treated with BAS 310 40 1, an EC formulation of alpha-cypermethrin (100 g a.s./L), foliar applied at a rate of 0.25 L of formulated product /ha, 7 days before expected harvest (plot 2). A second plot was treated with BAS 310 08 I, a WG formulation of alpha-cypermethrin (150 g a.s./kg), foliar applied at a rate of 0.16 kg of formulated product /ha, 7 days before expected harvest (plot 3). The nominal spray volume used was 400 L/ha at all applications and all application timings were made within ± 1 day of that specified in the study plan.
Specimens of lettuce heads were collected immediately after the last application (0 DALA) from each plot as well as 2-3, 6-7 and 14 days thereafter.

Generally the specimens were frozen within 24 hours of being taken, and remained frozen at or below -18°C, including during transportation, until analysis.
The maximum storage interval from sample collection until extraction was 225 days.

Table 6.3.3-9: Target application rates and timings for lettuce

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2006	2	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	7 ±1 days before harvest
		1	F	BAS 310 08 I (WG)	alpha-cypermethrin	0.0125	400	7 ±1 days before harvest
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	1st appl. 14 ±1, 2nd appl. 7 ±1 days before harvest
		2	F	BAS 310 08 I (WG)	alpha-cypermethrin	0.0125	400	1st appl. 14 ±1, 2nd appl. 7 ±1 days before harvest
	2	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.025	400	7 ±1 days before harvest
		1	F	BAS 310 08 I (WG)	alpha-cypermethrin	0.025	400	7 ±1 days before harvest

2. Description of analytical procedures

For the analysis of alphacypermethrin, BASF method No. 567/0 was applied which determines the underivatised cypermethrin by means of HPLC-MS/MS.

Principle of method:
Residues of cypermethrin (alphacypermethrin or cypermethrin) are extracted with a mixture of methanol/water/2N hydrochloric acid (70:25:5). After centrifugation, an aliquot of the extract is cleaned by liquid/liquid partition against cyclohexane. The residues are further purified (if needed) using a silica gel solid phase extraction (SPE) column. The final determination of total cypermethrin residues is performed by HPLC-MS/MS, monitoring the ion transition from m/z 433.2 / 191.1 (the ammonium adduct of cypermethrin).

The limit of quantitation (LOQ) of the method for alpha-cypermethrin is 0.01 mg/kg.

Table 6.3.3-10: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in lettuce

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Head	0.01	3	90.2	5.3	5.9
	1.0	1	96.1	-	-
	2.0	1	98.0	-	-
	0.01-2.0	5	92.9	5.3	5.7

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.3-11, detailed residue levels are shown in Table 6.3.3-12 and Table 6.3.3-13.
After one application at a target rate of 0.0125 kg alpha-cypermethrin/ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.024 mg/kg and 0.131 mg/kg. Residues declined to levels between <0.01-0.036 mg/kg, <0.01-0.19 mg/kg and <0.01 mg/kg at 3, 7 and 14 days after application.

After two applications at a target rate of 0.0125 kg alpha-cypermethrin/ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.090 mg/kg and 0.193 mg/kg. Residues declined to levels between <0.01-0.072 mg/kg, <0.01-0.022 mg/kg and <0.01 mg/kg at 3, 7 and 14 days after application.

After one application at a target rate of 0.025 kg alpha-cypermethrin/ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.228 mg/kg and 0.512 mg/kg. Residues declined to levels between 0.139-0.387 mg/kg, 0.035-0.131 mg/kg and <0.01-0.013 mg/kg at 3, 7 and 14 days after application.

No difference in the residue levels was observed after application of the EC or the WG formulation. In the control specimens, no residues of total cypermethrin above the limit of quantitation were found.

Table 6.3.3-11: Summary of residues of BAS 310 I in lettuce after application of BAS 310 40 I or BAS 310 08 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2006	BAS 310 40 I (EC) 1 x 0.0125 kg a.s./ha	0	45-48	Head	0.088-0.131
			3	47-49	Head	<0.01-0.036
			7	49	Head	<0.01-0.018
			14	49	Head	<0.01
		BAS 310 08 I (WG) 1 x 0.0125 kg a.s./ha	0	45-48	Head	0.024-0.049
			3	47-49	Head	<0.01-0.028
			7	49	Head	<0.01-0.019
			14	49	Head	<0.01
		BAS 310 40 I (EC) 2 x 0.0125 kg a.s./ha	0	45-48	Head	0.134-0.193
			3	47-49	Head	<0.01-0.072
			7	49	Head	<0.01-0.018
			14	49	Head	<0.01
		BAS 310 08 I (WG) 2 x 0.0125 kg a.s./ha	0	45-48	Head	0.090-0.131
			3	47-49	Head	<0.01-0.057
			7	49	Head	<0.01-0.022
			14	49	Head	<0.01
S-EU	2006	BAS 310 40 I (EC) 1 x 0.025 kg a.s./ha	0	35-47	Head	0.424-0.512
			2-3	38-47	Head	0.139-0.387
			6-7	42-49	Head	0.065-0.106
			14	49	Head	<0.01
		BAS 310 08 I (WG) 1 x 0.025 kg a.s./ha	0	35-47	Head	0.228-0.475
			2-3	38-47	Head	0.195-0.289
			6-7	42-49	Head	0.035-0.131
			14	49	Head	<0.01-0.013

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

After two applications of alpha-cypermethrin at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.090 mg/kg and 0.193 mg/kg. Residues declined to levels between <0.01-0.072 mg/kg and <0.01-0.022 mg/kg at 3 and 7 days, respectively, after application.

After one application of alpha-cypermethrin at a target rate of 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.228 mg/kg and 0.512 mg/kg. Residues declined to levels between 0.139-0.387 mg/kg and 0.035-0.131 mg/kg at 2-3 and 6-7 days, respectively, after application.

No difference in the residue levels was observed after application of the EC or the WG formulation.

Table 6.3.3-12: Residues of BAS 310 I in lettuce after one or two applications of BAS 310 40 I or BAS 310 08 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:182062 **Doc ID:**2007/1007938 **Trial No.:**AF/10503/BA/1 **GLP:**yes **Year**2006	Lettuce, head	France	BAS 310 40 I 1 x 0.0125	47-48	0	Head	0.131
				48-49	3	Head	0.036
				49	7	Head	0.018
				49	14	Head	<0.01
			BAS 310 08 I 1 x 0.0125	47-48	0	Head	0.024
				48-49	3	Head	0.028
				49	7	Head	0.019
				49	14	Head	<0.01
			BAS 310 40 I 2 x 0.0125	47-48	0	Head	0.193
				48-49	3	Head	<u>0.072</u>
				49	7	Head	0.018
				49	14	Head	<0.01
			BAS 310 08 I 2 x 0.0125	47-48	0	Head	0.090
				48-49	3	Head	0.057
				49	7	Head	0.022
				49	14	Head	<0.01
Study code:182062 **Doc ID:**2006/1026855 **Trial No.:**AF/10503/BA/2 **GLP:**yes **Year**2006	Lettuce, head	The United Kingdom	BAS 310 40 I 1 x 0.0125	45	0	Head	0.088
				47	3	Head	<0.01
				49	7	Head	<0.01
				49	14	Head	<0.01
			BAS 310 08 I 1 x 0.0125	45	0	Head	0.049
				47	3	Head	<0.01
				49	7	Head	<0.01
				49	14	Head	<0.01
			BAS 310 40 I 2 x 0.0125	45	0	Head	0.134
				47	3	Head	<u><0.01</u>
				49	7	Head	<0.01
				49	14	Head	<0.01
			BAS 310 08 I 2 x 0.0125	45	0	Head	0.131
				47	3	Head	<0.01
				49	7	Head	<0.01
				49	14	Head	<0.01

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Table 6.3.3-13: Residues of BAS 310 I in lettuce after one application of BAS 310 40 I or BAS 310 08 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:182062 **Doc ID:**2007/1007938 **Trial No.:**AF/10503/BA/3 **GLP:**yes **Year**2006	Lettuce, head	Spain	BAS 310 40 I 1 x 0.025	35-37	0	Head	0.424
				38	3	Head	0.139
				42	6	Head	0.106
				49	14	Head	<0.01
			BAS 310 08 I 1 x 0.025	35-37	0	Head	0.228
				38	3	Head	0.195
				42	6	Head	0.131
				49	14	Head	0.013
Study code:182062 **Doc ID:**2007/1007938 **Trial No.:**AF/10503/BA/4 **GLP:**yes **Year**2006	Lettuce, head	Italy	BAS 310 40 I 1 x 0.025	46-47	0	Head	0.512
				47	2	Head	0.387
				49	7	Head	0.065
				49	14	Head	<0.01
			BAS 310 08 I 1 x 0.025	46-47	0	Head	0.475
				47	2	Head	0.289
				49	7	Head	0.035
				49	14	Head	<0.01

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Report:	CA 6.3.3/3 Diehl M., 2007b Study on the residue behaviour of BAS 310 I in head lettuce after treatment with BAS 310 40 I under open field conditions in Southern and Northern Europe, 2006 2007/1008496
Guidelines:	EEC 96/68, EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 1607/VI/97 rev. 2 10.06.1999
GLP:	yes (certified by Swiss Federal Office of Public Health, Berne, Switzerland)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I
Description: BAS 310 40 I (EC)
Lot/Batch #: 1171, Alpha-Cypermethrin: 100 g/L
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01, 0.1 and 1.0 mg/kg

2. Test Commodity:
Crop: Lettuce, head
Type: Leafy vegetable
Variety: Lucan, mixture of Dynnmite and Dolly, Matilda, Pinochio, Soleilan, Filippus, Carinos, Aberam
Botanical name: *Lactuca sativa*
Crop parts(s) or processed commodity: Head
Sample size: min. 2 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2006 growing season, eight field trials were conducted in head lettuce in Denmark, France (Northern and Southern European region), Germany, Greece, Italy, Spain and the United Kingdom.
In four trials located in the Northern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to head lettuce plants on separate plots either once or twice at a target rate of 125 mL/ha, equivalent to 0.0125 kg alpha-cypermethrin/ha. The spray volume targeted 400 L/ha.

In four trials located in the Southern European region, an emulsifiable concentrate formulation of alpha-cypermethrin (EC 100 g a.s./L; BAS 310 40 I), was foliar applied to head lettuce plants once at a target rate of 250 mL/ha, equivalent to 0.025 kg alpha-cypermethrin/ha in a target spray volume of 400 L/ha.
The actual application rates were within 10% of the nominal values presented below with the exception of single application on plot 102 and second application on plot 103 of trial A/GE/I/06/107 which deviated by 15-16% less from target amount.
Specimens of lettuce heads were collected immediately after the last application from each plot, as well as 2-4, 7-8 and 13-14 days thereafter.
Samples were stored frozen at or below -18°C for a maximum of 156 days until analysis below -18°C, including during transportation, until analysis. Only for short periods of time storage temperature peaks higher than -18°C occured - mainly caused by sample handling that did not affect the samples.

Table 6.3.3-14: Target application rates and timings for lettuce

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2006	4	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	7 ±1 days before harvest
		2	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.0125	400	1st appl. 14 ±1, 2nd appl. 7 ±1 days before harvest
	4	1	F	BAS 310 40 I (EC)	alpha-cypermethrin	0.025	400	7 ±1 days before harvest

2. Description of analytical procedures

Specimens were analysed using BASF analytical method No. 567/0.
Residues of alpha-cypermethrin were extracted from head lettuce (head) using methanol/water and hydrochloric acid. An aliquot of the extract was centrifuged and partitioned against cyclohexane/water. The cyclohexane phase was evaporated and the residue was dissolved in acidic methanol/water. Residues of alpha-cypermethrin were determined by high performance liquid chromatography (HPLC) with tandem mass spectrometry (LC-MS/MS). The validated sensitivity (LOQ, limit of quantitation) of the method was 0.01 mg/kg.

Table 6.3.3-15: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in lettuce

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Head	0.01	4	96	12	12
	0.1	3	92	2	2
	1.0	1	100	-	-
	Overall	8	95	8	9

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.3-16, detailed residue levels are shown in Table 6.3.3-17 and Table 6.3.3-18.
After one application at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.024 mg/kg and 0.38 mg/kg. Residues declined to levels between 0.014-0.13 mg/kg, <0.01-0.080 mg/kg and <0.01-0.025 mg/kg at 2-3, 7 and 13-14 days after application.

After two applications at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.030 mg/kg and 0.350 mg/kg. Residues declined to levels between 0.021-0.27 mg/kg, <0.01-0.12 mg/kg and <0.01-0.039 mg/kg at 2-3, 7 and 13-14 days after application.

After a single application at a target rate of 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.24 mg/kg and 1.1 mg/kg. Residues declined to levels between <0.01-0.41 mg/kg, <0.01-0.12 mg/kg and <0.01-0.031 mg/kg at 3-4, 7-8 and 13-14 days after application.
No residues of alpha-cypermethrin above the LOQ were found in the untreated specimens of head lettuce.

Table 6.3.3-16: Summary of residues of BAS 310 I in lettuce after application of BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2006	BAS 310 40 I (EC) 1 x 0.125 kg a.s./ha	0	45-47	Head	0.024-0.38
			2-3	45-47	Head	0.014-0.13
			7	45-49	Head	<0.01-0.080
			13-14	49-51	Head	<0.01-0.025
		BAS 310 40 I (EC) 2 x 0.0125 kg a.s./ha	0	45-47	Head	0.030-0.350
			2-3	45-47	Head	0.021-0.27
			7	45-49	Head	<0.01-0.12
			13-14	49-51	Head	<0.01-0.039
S-EU	2006	BAS 310 40 I (EC) 1 x 0.025 kg a.s./ha	0	30-47	Head	0.24-1.1
			3-4	35-47	Head	<0.01-0.41
			7-8	38-48	Head	<0.01-0.12
			13-14	40-49	Head	<0.01-0.031

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

After two applications of alpha-cypermethrin at a target rate of 0.0125 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.030 mg/kg and 0.350 mg/kg. Residues declined to levels between 0.021-0.27 mg/kg and <0.01-0.12 mg/kg at 2-3 and 7 days, respectively, after the last application.

After a single application of alpha-cypermethrin at a target rate of 0.025 kg a.s./ha, initial residues of alpha-cypermethrin in lettuce heads ranged between 0.24 mg/kg and 1.1 mg/kg. Residues declined to levels between <0.01-0.41 mg/kg and <0.01-0.12 mg/kg at 3-4 and 7-8 days, respectively, after application.

Table 6.3.3-17: Residues of BAS 310 I in lettuce after one or two applications of BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/NF/I/06/106 **GLP:**yes **Year**2006	Lettuce, head	France	BAS 310 40 I 1 x 0.0125	45	0	Head	0.38
				47	3	Head	0.13
				47	7	Head	0.080
				49	13	Head	0.025
			BAS 310 40 I 2 x 0.0125	45	0	Head	0.35
				47	3	Head	0.27
				47	7	Head	0.12
				49	13	Head	0.039
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/GE/I/06/107 **GLP:**yes **Year**2006	Lettuce, head	Germany	BAS 310 40 I 1 x 0.0125	45	0	Head	0.024
				46	2	Head	0.014
				48	7	Head	<0.01
				51	14	Head	<0.01
			BAS 310 40 I 2 x 0.0125	45	0	Head	0.030
				46	2	Head	0.021
				48	7	Head	<0.01
				51	14	Head	<0.01
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/DK/I/06/108 **GLP:**yes **Year**2006	Lettuce, head	Denmark	BAS 310 40 I 1 x 0.0125	47	0	Head	0.13
				47	3	Head	0.096
				49	7	Head	0.027
				49	14	Head	<0.01
			BAS 310 40 I 2 x 0.0125	47	0	Head	0.13
				47	3	Head	0.12
				49	7	Head	0.042
				49	14	Head	0.010
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/UK/I/06/109 **GLP:**yes **Year**2006	Lettuce, head	The United Kingdom	BAS 310 40 I 1 x 0.0125	45	0	Head	0.14
				45	3	Head	0.041
				45	7	Head	0.027
				49	14	Head	0.010
			BAS 310 40 I 2 x 0.0125	45	0	Head	0.13
				45	3	Head	0.055
				45	7	Head	0.055
				49	14	Head	0.013

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Table 6.3.3-18: Residues of BAS 310 I in lettuce after one application of BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/SF/I/06/110 **GLP:**yes **Year**2006	Lettuce, head	France	BAS 310 40 I 1 x 0.025	47	0	Head	0.36
				47	3	Head	<u>0.32</u>
				49	7	Head	0.10
				49	14	Head	0.021
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/SP/I/06/111 **GLP:**yes **Year**2006	Lettuce, head	Spain	BAS 310 40 I 1 x 0.025	45	0	Head	0.24
				45	3	Head	<u>0.11</u>
				47-48	8	Head	0.038
				48-49	13	Head	0.011
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/IT/I/06/112 **GLP:**yes **Year**2006	Lettuce, head	Italy	BAS 310 40 I 1 x 0.025	30-35	0	Head	1.1
				35-40	3	Head	<u>0.41</u>
				38-40	7	Head	0.12
				40-48	14	Head	0.031
Study code:227104 **Doc ID:**2007/1008496 **Trial No.:**A/GR/I/06/113 **GLP:**yes **Year**2006	Lettuce, head	Greece	BAS 310 40 I 1 x 0.025	44	0	Head	0.33
				46-47	4	Head	<u><0.01</u>
				49	8	Head	<0.01
				49	14	Head	<0.01

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

Report: CA 6.3.3/4
Klimmek S.,Gizler A., 2014a
Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in lettuce (open leaf varieties) after two applications with BAS 310 55 I under field conditions in Southern France, Greece, Italy and Spain, 2013
2014/1140312
Guidelines: none
GLP: yes
(certified by Freie und Hansestadt Hamburg, Behoerde fuer Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198, BAS 310 55 I, Alpha-Cypermethrin: 50 g/L;
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01, 0.1 and 4.0 mg/kg

2. Test Commodity:
Crop: Lettuce, open leaf
Type: Leafy vegetable
Variety: Kirina, Parris Island, Foglia di Quercia, Flavius, Ukulele, Magister F1, Analena, Filipo
Botanical name: *Lactuca sativa* var. *crispa*
Crop parts(s) or processed commodity: Heads
Sample size: 0 DALA: ≥ 0.5 kg / ≥ 12 units
1 DALA and later: ≥ 1 kg / ≥ 12 units

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2013 growing season, eight residue trials were conducted under field conditions in representative lettuce growing areas in Southern France, Greece, Italy and Spain to determine the residue level of alpha-Cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).

Two foliar applications with BAS 310 55 I, an ME formulation of alpha-Cypermethrin (50.0 g ai/L), were made to plot 2 to lettuce, at a rate of 0.25 L formulated product/ha, equal to 12.5 g alpha-Cypermethrin/ha, at 7 and 1 day(s) before harvest (DBH). The nominal spray volume used was 200 L/ha.
Untreated lettuce (head) specimens were sampled immediately pre application at 0 days before the last application (DBLA) and at 1, 2-3 and 6-8 days after last application (DALA).
Treated lettuce (head) specimens were sampled at 0 DALA and at 1, 2-3 and 6-8 DALA.
Generally the specimens were frozen within 12 hours of being taken, and remained frozen at or below -18°C, including during transportation, until analysis. The maximum storage interval from harvest until analysis of alpha-cypermethrin (BAS 310 I) was 320 days.

Table 6.3.3-19: Target application rates and timings for lettuce

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2013	8	2	F	BAS 310 55 I	alpha-cypermethrin	0.0125	200	7±1 days and 1 day before harvest

2. Description of analytical procedures

All lettuce specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
Principle of method: Samples are extracted with methanol/water/2N hydrochloric acid (70/25/5, v/v/v). After centrifugation and partition of an aliquot of the extract into cyclohexane, the samples are evaporated to dryness and dissolved in methanol / water (80+20, v+v).

The final determination of the analyte in the untreated and treated specimens was performed by single extraction and single injection with liquid chromatography and mass spectrometric detection (LC-MS/MS).
The limit of quantitation (LOQ) was 0.010 mg/kg for alpha-cypermethrin.

Table 6.3.3-20: Summary of recoveries for alpha-cypermethrin in lettuce

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (±)	RSD (%)
BASF Method No. 567/0		**alpha-cypermethrin (BAS 310 I)**			
Lettuce (head)	0.01	5	92.8	9.3	10.0
	0.10	5	86.9	8.6	9.9
	4.0	1	84.5	-	-
	Overall	11	89.4	8.7	9.7

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.3-21, detailed residue levels are shown in Table 6.3.3-22.

At 0 DALA the residues of alpha-cypermethrin (BAS 310 I) ranged between 0.063 mg/kg and 0.44 mg/kg. At 1 DALA (PHI) the residues of alpha-cypermethrin (BAS 310 I) ranged between 0.058 mg/kg and 0.42 mg/kg. At 3 DALA the residues of alpha-cypermethrin (BAS 310 I) ranged between 0.032 mg/kg and 0.24 mg/kg. At 7 DALA residues between 0.012 mg/kg and 0.094 mg/kg of alpha-cypermethrin (BAS 310 I) were found.
No residues above the LOQ of alpha-cypermethrin (BAS 310 I) were found in the untreated specimens of this study, except for the specimens of trial S13-00444-01 (L130050) taken at 0, 1 and 3 DALA.
In the untreated specimens of trial S13-00444-01 (L130050) taken at 0, 1 and 3 DALA, residues between 0.025 mg/kg and 0.027 mg/kg of alpha-cypermethrin (BAS 310 I) were found. The source of the contamination has not been determined. Taking into account the corresponding residues in the treated specimens the residues in the control samples had no impact on the study.

Table 6.3.3-21: Summary of residues in lettuce after application of BAS 310 55 I

Region	Year	No. of Appl.	Application	DALA[1]	Growth stage[2] BBCH	Range of Residues (mg/kg)	
						Matrix	alpha-cypermethrin
S-EU	2008	2	BAS 310 51 I (ME)	0	45-49	Head	0.063-0.44
				1	45-49	Head	0.058-0.42
				2-3	47-49	Head	0.032-0.24
				6-8	47-49	Head	0.012-0.094

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

Immediately after the last of two applications of the formulation BAS 310 55 I (ME) at a target rate of 0.0125 kg a.s./ha per application to open leaf lettuce varieties under field conditions in Southern Europe, residues of alpha-cypermethrin ranged between 0.063 mg/kg and 0.44 mg/kg. At 1 day after the last application, the residues of alpha-cypermethrin ranged between 0.058 mg/kg and 0.42 mg/kg and at 2-3 days after the last application the residues ranged between 0.032 mg/kg and 0.24 mg/kg.

Table 6.3.3-22: Residues of BAS 310 I in lettuce after two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130050 **GLP:**yes **Year**2013	Lettuce, open leaf	France	BAS 310 55 I 2 x 0.0125	45-47	0	Head	0.44
				45-47	1	Head	0.42
				47	3	Head	<u>0.24</u>
				47	7	Head	0.089
Study code:408662 **DocID:**2014/1140312 **Trial No.:**L130051 **GLP:**yes **Year**2013	Lettuce, open leaf	Greece	BAS 310 55 I 2 x 0.0125	47-49	0	Head	0.13
				47-49	1	Head	0.082
				47-49	2	Head	<u>0.075</u>
				49	8	Head	0.015
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130052 **GLP:**yes **Year**2013	Lettuce, open leaf	Italy	BAS 310 55 I 2 x 0.0125	47-49	0	Head	0.10
				47-49	1	Head	0.084
				47-49	3	Head	<u>0.032</u>
				49	7	Head	0.012
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130053 **GLP:**yes **Year**2013	Lettuce, open leaf	Spain	BAS 310 55 I 2 x 0.0125	49	0	Head	0.38
				49	1	Head	0.23
				49	3	Head	<u>0.21</u>
				49	7	Head	0.094
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130054 **GLP:**yes **Year**2013	Lettuce, open leaf	France	BAS 310 55 I 2 x 0.0125	47	0	Head	0.12
				47	1	Head	0.17
				47	3	Head	<u>0.049</u>
				47-49	7	Head	0.042
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130055 **GLP:**yes **Year**2013	Lettuce, open leaf	Greece	BAS 310 55 I 2 x 0.0125	47	0	Head	0.23
				49	1	Head	0.19
				49	3	Head	<u>0.11</u>
				49	7	Head	0.031
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130056 **GLP:**yes **Year**2013	Lettuce, open leaf	Italy	BAS 310 55 I 2 x 0.0125	48	0	Head	0.063
				48-49	1	Head	0.058
				48-49	2	Head	<u>0.037</u>
				49	6	Head	0.014
Study code:408662 **Doc ID:**2014/1140312 **Trial No.:**L130057 **GLP:**yes **Year**2013	Lettuce, open leaf	Spain	BAS 310 55 I 2 x 0.0125	49	0	Head	0.19
				49	1	Head	0.18
				49	3	Head	<u>0.091</u>
				49	7	Head	0.054

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL calculation

CA 6.3.4 Oilseed rape

Table 6.3.4-1: cGAP for the use of BAS 310 I in/on oilseed rape

Crop	Maximum applied dose	Water volume	PHI	Application method	Application timing
Oilseed rape, winter, outdoor (North, Central)	2 x 0.010 kg BAS 310 I/ha	100-400	28	Overall spray	BBCH 51-59
Oilseed rape, winter, outdoor (South)	2 x 0.010 kg BAS 310 I/ha	100-400	28	Overall spray	BBCH 51-59

PHI = pre-harvest interval

Table 6.3.4-2: GAP information of residue trials conducted in oilseed rape in 2007-2013 in Northern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Northern EU	France (2) 2007	BAS 310 40 I EC	spray appl.	0.0105	0.0035	2	0 14 21 28	2008/ 1019999	No
	Germany (1) France (1) 2008	BAS 310 40 I EC BAS 310 51 I ME	spray appl.	0.015	0.005	2	0 14±1 21±1 27	2009/ 1090702	No
	Belgium (1) Germany (1) 2012	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 14 21 29	2013/ 1037957	No
	Belgium (1) Germany (1) France (1) The Netherlands (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 13-14 20-22 27-29	2013/ 1416283	No

0 Actual application rates varied by 10% at most
1 Days after last application

Table 6.3.4-3: GAP information of residue trials conducted in oilseed rape in 2007-2013 in Southern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Southern EU	France (2) 2007	BAS 310 40 I EC	spray appl.	0.0105	0.0035	2	0 14±1 21 28±1	2008/ 1019999	No
	Italy (1) Spain (1) 2008	BAS 310 40 I EC BAS 310 51 I (ME)	spray appl.	0.030	0.01	1	0 14 21 28	2009/ 1090702	No
	France (1) Greece (1) Italy (1) Spain (1) 2012	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 14±1 21±1 28±1	2013/ 1037957	No
				0.025	0.0125	1			
	France (1) Greece (1) Italy (1) Spain (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 14-15 21-22 28-29	2013/ 1416283	No

0 Actual application rates varied by 10% at most
1 Days after last application

The studies not yet evaluated are summarized in the following chapter.

Report: CA 6.3.4/1
Oxspring S., 2008a
Study on the residue behaviour of Alpha-Cypermethrin in oilseed rape after treatment with BAS 310 40 I under field conditions in Northern and Southern Europe during 2007
2008/1019999
Guidelines: none
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31040I
Description: BAS 310 40 I (SC)
Lot/Batch #: 1209: 100 g/L nominal
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01-1.0 mg/kg

2. Test Commodity:
Crop: Oilseed rape
Type: Oilseeds
Variety: Adriana, Robuste, Corail, Standy
Botanical name: *Brassica napus*
Crop parts(s) or processed commodity: Whole plant w/o root, whole pods, seed
Sample size: 12 units/1.0 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2007 growing season, a total of six field trials were conducted on oilseed rape in order to determine the residue level of BAS 310 I in or on raw agricultural commodities (RAC). Trial AF/12151/BA/1 (L070763) in the UK was destroyed by the grower after the second sampling timing and was abandoned and Trial AF/121511BA/4 (L070766) in the UK was erroneously treated with the insecticide lambda-cyhalothrin. Due to these unexpected issues, only results from the remaining four winter oilseed rape trials were used for evaluation.
BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g a.s./L), was applied at a rate of 0.0105 kg alpha-cypermethrin/ha at each application in a spray volume of 300 L/ha. Applications were made at 10 day intervals approximately 31±1 and 21±1 days before expected harvest.

Whole plant specimens were collected immediately after the last application (0 DALA) from each plot at all trials. Oilseed rape seed was sampled at 14±1, 21±1 and 28±1 days after last application (DALA) at all the trials with the exception for Trial AF/12151/BA/3 where whole pods were sampled 14±1 DALA due to immaturity of the crop.
Generally the specimens were frozen within 24 hours of being taken, and remained frozen at or below -18°C, including during transportation, until analysis.
The maximum storage interval from harvest until analysis was 252 days.

Table 6.3.4-4: Target application rates and timings for oilseed rape

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (g a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2007	4	2	F	BAS 310 40 I	alpha-cypermethrin	10.5	300	1st appl. 21±1, 2nd appl. 31±1 days before harvest

2. Description of analytical procedures

Alpha-cypermethrin was extracted from specimens according to an adaption of BASF Method No. 567/0 (L0020/01). The extracts were analysed for alpha-cypermethrin using BASF Method No. 567/1 (L0071/01), which quantifies the analyte with a limit of quantitation (LOQ) of 0.01 mg/kg. BAS 310 I was extracted with a 95:5 methanol/hydrochloric acid mixture (solvent II of method 567/1). After centrifugation, an aliquot of the extract was partitioned twice into cyclohexane. The final determination was according to BASF method 567/0 with HPLC-MS/MS.

Table 6.3.4-5: Summary of recoveries for alpha-cypermethrin in oilseed rape

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
BASF Method No. 567/0		alpha-cypermethrin (BAS 310 I)		
whole plant w/o root, whole pods, seed	0.01/0.10/1.00	8	102	10

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.4-6, detailed residue levels are shown in Table 6.3.4-7 and Table 6.3.4-8.

No residues of alpha-cypermethrin at or above the LOQ were detected in the untreated specimens. Directly after the last application, alpha-cypermethrin ranged between 0.109 mg/kg and 0.390 mg/kg in oliseed rape whole plants. Approximately two weeks later, residues of alpha-cypermethrin were between <0.01 mg/kg and 0.012 mg/kg in seed, and 0.098 mg/kg in whole pods. At the intended harvest (21±1 DALA), no residues of alpha-cypermethrin above the limit of quantitation (0.01 mg/kg) were found in seed.

Table 6.3.4-6: Summary of residues in oilseed rape after application of BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2006	BAS 310 40 I 2 x 0.0105	0	79-80	Whole plant w/o root	0.109-0.159
			14	84-86	Whole pods	0.098
			14	88-89	Seed	<0.01
			21	89	Seed	<0.01
			28	89	Seed	<0.01
S-EU	2006	BAS 310 40 I 2 x 0.0105	0	87-89	Whole plant w/o root	0.327-0.390
			13-14	89	Seed	<0.01-0.012
			21	89	Seed	<0.01
			27-28	89	Seed	<0.01

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

No residues of alpha-cypermethrin above the limit of quantitation (0.01 mg/kg) were found in rape seed at 21 days or later after the last of two applications of BAS 310 40 I at a target rate of 0.0105 kg a.s./ha.

Table 6.3.4-7: Residues of alpha-cypermethrin after two applications of the formulation BAS 310 40 I in Northern Europe

Study Details		Crop	Country Trial No	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
							Matrix	Alpha-Cypermethrin
Study code:	311992	Oilseed Rape	France	2 x BAS 310 40 I 0.0105	79-80	0	Whole plant w/o root	0.159
Doc ID:	2008/1019999				88-89	14	Seed	<0.01
Trial No..	AF/12151/BA/2				89	21	Seed	<0.01
GLP:	yes				89	28	Seed	<u>0.01</u>
Year	2006							
Study code:	311992	Oilseed Rape	France	2 x BAS 310 40 I 0.0105	79-80	0	Whole plant w/o root	0.109
Doc ID:	2008/1019999				84-86	14	Whole pods	0.098
Trial No..	AF/12151/BA/3				89	21	Seed	<0.01
GLP:	yes				89	28	Seed	<u>0.01</u>
Year	2006							

DALA = Days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL determination

Table 6.3.4-8: Residues of alpha-cypermethrin after two applications of the formulation BAS 310 40 I in Southern Europe

Study Details		Crop	Country Trial No	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
							Matrix	Alpha-Cypermethrin
Study code:	311992	Oilseed Rape	France	2 x BAS 310 40 I 0.0105	89	0	Whole plant w/o root	0.327
Doc ID:	2008/1019999				89	13	Seed	<0.01
Trial No..	AF/12151/BA/6				89	21	Seed	<0.01
GLP:	yes				89	27	Seed	<u>≤0.01</u>
Year	2006							
Study code:	311992	Oilseed Rape	France)	2 x BAS 310 40 I 0.0105	87	0	Whole plant w/o root	0.390
Doc ID:	2008/1019999				89	14	Seed	0.012
Trial No..	AF/12151/BA/7				89	21	Seed	<0.01
GLP:	yes				89	28	Seed	<u>≤0.01</u>
Year	2006							

DALA = Days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL determination

Report: CA 6.3.4/2
Schulz H., 2009a
Study on the residue behaviour of Alpha-Cypermethrin in rape after treatment with BAS 310 51 I and BAS 310 40 I under field conditions in Germany, Northern France, Italy and Spain, 2008
2009/1090702
Guidelines: EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7
GLP: yes
(certified by Hessisches Ministerium fuer Umwelt, Laendlichen Raum und Verbraucherschutz)

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:** Alpha-Cypermethrin BAS 31051I, BAS 31040I
Description: BAS 310 51 I (ME)
BAS 310 40 I (SC)
Lot/Batch #: 101156, BAS 310 51 I, Alpha Cypermethrin: 50.0 g/L; 1209, BAS 310 40 I, Alpha Cypermethrin: 100.0 g/L
Purity: not reported
CAS#: 67375 30 8
Development code: not reported
Spiking levels: 0.01 1.00 mg/kg

2. **Test Commodity:**
Crop: Oilseed rape
Type: Oilseed
Variety: Elektra, Campo, Fantasio, Kabel
Botanical name: *Brassica napus*
Crop parts(s) or processed commodity: Whole plant, pods with seed, seed
Sample size: 0.5 1.0 kg

B. STUDY DESIGN AND METHODS

1. **Test procedure**

During the 2008 growing season, a total of four open field trials were conducted in rape in Germany, Northern France, Italy and Spain.
The objective of the study was to determine the magnitude of alpha-cypermethrin residues in rape after application of the formulations BAS 310 51 I (ME formulation) and BAS 310 40 I (EC formulation). The additional objective was the comparison of the residue levels after the application of the EC and the ME formulation. Each field trial consisted of three plots:
Trials L080185 (Germany) and L080186 (Northern France):

Plot 1: Control
Plot 2: Treated twice with 0.3 L/ha of BAS 310 51 I (15.0 g/ha of BAS 310 I, ME formulation)
Plot 3: Treated twice with 0.15 L/ha of BAS 310 40 I (15.0 g/ha of BAS 310 I, EC formulation)
Trials L080187 (Italy) and L080188 (Spain):
Plot 1: Control
Plot 2: Treated once with 0.6 L/ha of BAS 310 51 I (30.0 g/ha of BAS 310 I, ME formulation)
Plot 3: Treated once with 0.3 L/ha of BAS 310 40 I(30.0 g/ha of BAS 310 I, EC formulation)
The applications took place 30-31 days before harvest (DBH) and 20-22 DBH on trials L080185 and L080186 and 21 DBH on trials L080187 and L080188 with a spray volume of 300 L/ha of spray.
Both formulations were applied with the same GAP.

0 DALA (days after last application) sampling in the untreated plot was performed immediately prior to the last application of the treated plots. The treated rape specimens (whole plant and seed) were taken immediately after the last application (0 DALA). Untreated and treated specimens were additionally collected 14±1, 21±1 and 28±1 DALA.
The maximum storage interval from harvest until start of analysis was 266 days.

Table 6.3.4-9: Target application rates and timings for oilseed rape

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (g a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2008	4	2 1	F	BAS 310 51 I	alpha-cypermethrin	15 30	300	21±1, 31±1 days before harvest
	4	2 1	F	BAS 310 40 I	alpha-cypermethrin	15 30	300	21±1, 31±1 days before harvest

2. Description of analytical procedures

Alpha-cypermethrin was extracted from specimens according to an adaption of BASF Method No. 567/0 (L0020/01) with a limit of quantitation (LOQ) of 0.01 mg/kg.
The residues of alpha-cypermethrin in whole plant no root specimens were extracted from plant matrices using a mixture of methanol, water and HCl (2 mol/L). For clean-up a liquid/liquid partition against cyclohexane was used. The final determination of alpha-cypermethrin was performed by LC-MS/MS.
The residues of alpha-cypermethrin in seed specimens were extracted from plant matrices using a mixture of acetonitrile and n-hexane. An aliquot of acetonitrile phase was taken. For clean-up a liquid/liquid partition against n-hexane was used. The final determination of alpha-cypermethrin was performed by LC-MS/MS.

Table 6.3.4-10: Summary of recoveries for alpha-cypermethrin in oilseed rape

Matrix	Fortification Level (mg/kg)	Summary Recoveries		
		n	Mean (%)	RSD (%)
BASF Method No. 567/0		alpha-cypermethrin (BAS 310 I)		
rape (whole plant no root)	0.01-1.00	2	94	n. a.
rape (seed)	0.01-1.00	3	78	29
rape (rows/pods with seed)	0.01-1.00	2	104	n. a.

n.a. = not applicable

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.4-11, detailed residue levels are shown in Table 6.3.4-12 and Table 6.3.4-13.

Directly after the last application (0 DALA) of BAS 310 51 I (plot 2), alpha-cypermethrin residues were 0.20-0.41 mg/kg in whole plant specimens. At 13-14 days after the last application, residues were <0.01-0.02 mg/kg in rape seed and 0.05 mg/kg in pods with seed. At 20-22 and at 27-28 days after the last application, residues in seed were <0.01-0.01 mg/kg.
Directly after the last application (0 DALA) of BAS 310 40 I (plot 3), alpha-cypermethrin residues were 0.12-0.54 mg/kg in whole plant specimens. At 13-14 days after the last application residues were <0.01-0.01 mg/kg in rape seed and 0.03 mg/kg in pods with seed. At 20-22 and at 27-28 days after the last application, residues in seed were <0.01-0.01 mg/kg.

Table 6.3.4-11: Summary of residues in oilseed rape after application of BAS 310 51 I or BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2008	BAS 310 51 I 2 x 0.0150 kg a.s./ha	0	78-87	whole plant	0.20-0.26
			13	89	seed	<0.01
			15	82	pods with seed	0.05
			20-22	89	seed	<0.01
			27	89-99	seed	<0.01
		BAS 310 40 I 2 x 0.0150 kg a.s./ha	0	78-87	whole plant	0.16-0.17
			13	89	seed	<0.01
			15	82	pods with seed	0.03
			20-22	89	seed	<0.01-0.01
			27	89-99	seed	<0.01
S-EU	2008	BAS 310 51 I 1 x 0.0300 kg a.s./ha	0	65-75	whole plant	0.33-0.41
			14	84-87	seed	0.01-0.02
			21	88-89	seed	<0.01-0.01
			28	88-89	seed	<0.01-0.01
		BAS 310 40 I 1 x 0.0300 kg a.s./ha	0	65-75	whole plant	0.12-0.54
			14	84-87	seed	<0.01-0.01
			21	88-89	seed	<0.01
			28	88-89	seed	<0.01-0.01

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

At 27-28 days after the last of two applications of alpha-cypermethrin formulations at a target rate of 0.015 kg a.s./ha or after a single application at 0.03 mg a.s./ha, residues in oilseed rape seed ranged between <0.01-0.01 mg/kg.
The analytical results demonstrate that the treatment with BAS 310 51 I and BAS 310 40 I did not lead to significantly different residue levels in the raw agricultural commodity at harvest.

Table 6.3.4-12: Residues of alpha-cypermethrin after application of the formulations BAS 310 51 I and BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin
Study code: 319697 Doc ID: 2009/1090702 Trial No.: L080185 GLP: yes Year: 2007	Oilseed Rape	Germany	2 x BAS 310 51 I 0.0150	87	0	whole plant	0.20
				89	13	seed	<0.01
				89	20	seed	<0.01
				99	27	seed	<0.01
			2 x BAS 310 40 I 0.0150	87	0	whole plant	0.17
				89	13	seed	<0.01
				89	20	seed	<0.01
				99	27	seed	<0.01
Study code: 319697 Doc ID: 2009/1090702 Trial No.: L080186 GLP: yes Year: 2007	Oilseed Rape	France	2 x BAS 310 51 I 0.0150	78	0	whole plant	0.26
				82	15	pods with seed	0.05
				89	22	seed	<0.01
				89	27	seed	<0.01
			2 x BAS 310 40 I 0.0150	78	0	whole plant	0.16
				82	15	pods with seed	0.03
				89	22	seed	0.01
				89	27	seed	<0.01

DALA = Days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL determination

Table 6.3.4-13: Residues of alpha-cypermethrin after application of the formulations BAS 310 51 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	Alpha-Cypermethrin
Study code: 319697 Doc ID: 2009/1090702 Trial No.: L080187 GLP: yes Year: 2007	Oilseed Rape	Italy	1 x BAS 310 51 I 0.0300	75 87 89 89	0 14 21 28	whole plant seed seed seed	0.41 0.02 0.01 <0.01
			1 x BAS 310 40 I 0.0300	75 87 89 89	0 14 21 28	whole plant seed seed seed	0.54 0.01 <0.01 0.01
Study code: 319697 Doc ID: 2009/1090702 Trial No.: L080188 GLP: yes Year: 2007	Oilseed Rape	Spain	1 x BAS 310 51 I 0.0300	65 84 88 88	0 14 21 28	whole plant seed seed seed	0.33 0.01 <0.01 0.01
			1 x BAS 310 40 I 0.0300	65 84 88 88	0 14 21 28	whole plant seed seed seed	0.12 <0.01 <0.01 <0.01

DALA = Days after last application
BBCH = Growth stage (GS) at respective sampling
_ underlined values were used for MRL determination

Report:	CA 6.3.4/2 Plier S., 2014a Determination of residues of BAS 310 I (Alpha-Cypermethrin) in oilseed rape after application of BAS 310 55 I in Germany, Belgium, France (South), Greece, Italy and Spain, 2012 2013/1037957
Guidelines:	EEC 91/414, SANCO/3029/99 rev. 4 (11 July 2000), EEC 7525/VI/95 rev. 9 (March 2011), OECD 509 Crop Field Trial (2009), EEC 79/117, EC 1107/2009 of the European Parliament and of the Council of 21 Oct 2009, EEC 7029/VI/95 rev. 5 Appendix B (July 22 1997)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Report:	CA 6.3.4/3 Plier S., 2017 a Amendment No. 1 - Determination of residues of BAS 310 I (Alpha-Cypermethrin) in oilseed rape after application of BAS 310 55 I in Germany, Belgium, France (South), Greece, Italy and Spain, 2012 2017/1134337
Guidelines:	EEC 91/414, SANCO/3029/99 rev. 4 (11 July 2000), EEC 7525/VI/95 rev. 9 (March 2011), OECD 509 Crop Field Trial (2009), EEC 79/117, EC 1107/2009 of the European Parliament and of the Council of 21 Oct 2009, EEC 7029/VI/95 rev. 5 Appendix B (July 22 1997)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:** Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50.0 g/L nominal
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01-1.0 mg/kg

2. **Test Commodity:**
Crop: Oilseed rape
Type: Oilseeds
Variety: Heros, DK Exquisite, CSZ8992 + ES Alias, PR46W31, PR44W29, Kabel
Botanical name: *Brassica napus*
Crop parts(s) or processed commodity: Whole plant w/o root, whole pods, seed
Sample size: Whole plant w/o root: 0.50-2.3 (>0.50) kg
Pods with seeds: 0.51-0.96 (>0.50) kg
Seed: 0.50-1.2 (>0.50) kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2012 growing season, a total of six field trials were conducted in oilseed rape in order to determine the residue level of BAS 310 I in or on raw agricultural commodities (RAC).
The trials were performed in Northern Europe (Germany and Belgium) and in Southern Europe (France, Greece, Italy and Spain).

The trials performed in Northern Europe consisted of two plots: one untreated plot (control, plot 1) and one plot treated twice with BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha (plot 2).
In Southern Europe (France, Greece, Italy), the experimental set-up consisted of three plots: one plot untreated (plot 1), a second plot treated twice with BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha (plot 2) and a third plot treated once with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha (plot 3). The applications were made at 28 and 20-22 days before harvest (plot 2) and at 20-22 days before harvest (plot 3) using a spray volume of 200 L/ha.
In all trials the applications were made at crop stages BBCH between 75 and 87.
For the analysis samples of rape plant (without roots) were taken directly after the last application (0 DALA). Seed and/or pod with seed specimens and rest of plant (without roots) were taken at 13-14, 20-22, and 28-29 DALA, depending on the crop maturity.
The maximum storage interval from harvest until analysis for plant samples was 416 days.
A deviation has to be noted for L120462: Two forbidden pesticides were applied on the crops before, lambda-cyhalothrin and tau-fluvalinate. With regard to the use of tau-fluvalinate and lambda-cyhalthrin, these two compounds could theoretically interfere with the analysis of alpha-cypermethrin if one of the MS transitions were identical to the Alpha-cypermethrin and were not detected. In the case of trial L120462, these two products were applied to the crop on 1st March and 26th March, whereas the trial started on 24 May, 8 to 11 weeks after the crop treatment with these two products. Reviewing the results of the control specimens collected, it is apparent, that no residue of this crop treatment was potentially interfering with the specimen analysis of the trial plots.

Table 6.3.4-14: Target application rates and timings for oilseed rape

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2012	6	2	F	BAS 310 55 I (EC)	alpha-cypermethrin	0.0125	200	1st appl.: 28±1 DBH 2nd appl.: 21±1 DBH
2012	4	1	F	BAS 310 55 I (EC)	alpha-cypermethrin	0.025	200	1st appl.: 21±1 DBH

DBH: days before harvest

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).
The residues of alpha-cypermethrin in rape specimens were extracted with acetonitrile/n-hexane. After centrifugation and two further partitioning steps with n-hexane, an aliquot of the extract was diluted with acetonitrile/water (80:20, v/v) for LC/MS/MS determination. Final determination was performed by LC/MS/MS using the ammonium adduct of cypermethrin.
The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery in rape specimens averaged 86.3±10.6% (mean ± SD) for alpha-cypermethrin at fortification levels of 0.01-1.0 mg/kg.

Table 6.3.4-15: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in oilseed rape

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
whole plant w/o root	0.01-1.0	9	80.1	7.3	9.2
seed	0.01-1.0	6	75.8	2.5	3.3
pods with seed	0.01-1.0	6	94.3	11.3	12.0
rest of plant without roots	0.01-1.0	9	94.2	5.4	5.7

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.4-16, detailed residue levels are shown in Table 6.3.4-17 and Table 6.3.4-18.
Directly after the last of two applications of BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha, residues of BAS 310 I (alpha-cypermethrin) in whole plant (without root) specimens ranged between 0.17-0.49 mg/kg. At 13-14 days after the last application residues were 0.014-0.037 mg/kg in pods with seeds and 0.033-0.42 in rest of plant (without roots). At 20-22 DALA residues were 0.039 mg/kg in pods with seeds and 0.011-0.22 mg/kg in rest of plant (without roots). After a longer PHI (28-29 DALA), residues in rest of plant (without roots) were 0.018-0.19 mg/kg.
Residues in seeds after two treatments with BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha were below the limit of quantitation (LOQ; 0.01 mg/kg) at all sampling times (13-14, 20-22 and 28-29 DALA).
Directly after a single treatment with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha, residues of BAS 310 I (alpha-cypermethrin) in whole plant (without root) specimens ranged between 0.25-0.44 mg/kg. At 13-14 days after application residues were 0.021-0.064 mg/kg in pods with seeds and 0.013-0.14 in rest of plant (without roots). At 20-22 DALA, and at 28-29 DALA, residues in rest of plant (without roots) were 0.010-0.17 mg/kg and <0.01-0.22 mg/kg, respectively.
Residues in seeds after a single treatment with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha were below the limit of quantitation (LOQ; 0.01 mg/kg) at all sampling times (13-14, 20-22 and 28-29 DALA).

No residues above the LOQ were found in any of the analysed untreated specimens generated from trials L120460 through L120465.

Table 6.3.4-16: Summary of residues of BAS 310 I in oilseed rape after one or two applications of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2012	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	79-87	Whole plant*	0.19-0.49
			14	81-87	Rest of plant*	0.063-0.42
			14	81	Pods with seeds	0.025
			14	87	Seed	<0.01
			21	87-89	Rest of plant	0.059-0.22
			21	87-89	Pods with seeds	0.039
			21	87-89	Seed	<0.01
			29	89	Rest of plant*	0.11-0.16
			29	89	Seed	<0.01
S-EU	2012	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	77-80	Whole plant*	0.17-0.28
			13-14	81-89	Rest of plant*	0.033-0.10
			13-14	81-87	Pods with seeds	0.014-0.037
			14	87/89	Seed	<0.01
			20-22	85-89	Rest of plant*	0.011-0.12
			20-22	85-89	Seed	<0.01
			28-29	89	Rest of plant*	0.018-0.19
			28-29	89	Seed	<0.01
		BAS 310 55 I 1 x 0.025 kg a.s./ha	0	77/79-80	Whole plant*	0.25-0.44
			13-14	81-87/89	Seed	<0.01
			13-14	81-87/89	Pods with seeds	0.021-0.064
			13-14	81-87/89	Rest of plant*	0.013-0.14
			20-22	85-89	Seed	<0.01
			20-22	85-89	Rest of plant*	0.010-0.17
			28-29	89	Seed	<0.01
			28-29	89	Rest of plant*	<0.01-0.22

1 Days after last application
2 Growth stage at respective sampling
* = without roots

III. CONCLUSION

After two treatments with BAS 310 55 I at a target rate of 2 x 0.0125 kg alpha-cypermethrin/ha or a single treatment at 0.025 kg alpha-cypermethrin/ha, no residues of alpha-cypermethrin above the limit of quantitation (LOQ, 0.01 mg/kg) were found in seed at 21±1 days or later after the (last) application.

Table 6.3.4-17: Residues of BAS 310 I after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408612	Oilseed rape	Germany	BAS 310 55 I 2 x 0.0125	87	0	Whole plant*	0.49
Doc ID:2013/1037957				87	14	Seed	<0.01
Trial No.:L120460				87	14	Rest of plant*	0.42
GLP:yes				87-89	21	Seed	<0.01
Year2012				87-89	21	Rest of plant*	0.22
				89	29	Seed	<u>≤0.01</u>
				89	29	Rest of plant*	<u>0.16</u>
Study code:408612	Oilseed rape	Belgium	BAS 310 55 I 2 x 0.0125	79	0	Whole plant*	0.19
Doc ID:2013/1037957				81	14	Rest of plant*	0.063
Trial No.:L120461				81	14	Pods with seeds	0.025
GLP:yes				87-89	21	Rest of plant*	0.059
Year2012				87-89	21	Pods with Seeds	0.039
				89	29	Seed	<u>≤0.01</u>
				89	29	Rest of plant*	<u>0.11</u>

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 6.3.4-18: Residues of BAS 310 I after one or two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408612 **Doc ID:**2013/1037957 **Trial No.:**L120462 **GLP:**yes **Year**2012	Oilseed rape	France	BAS 310 55 I 2 x 0.0125	80	0	Whole plant*	0.17
				87	13	Rest of plant*	0.087
				87	13	Pods with Seeds	0.037
				89	20	Seed	<0.01
				89	20	Rest of plant*	0.093
				89	28	Seed	<0.01
				89	28	Rest of plant*	0.067
			BAS 310 55 I 1 x 0.025	80	0	Whole plant*	0.25
				87	13	Rest of plant*	0.14
				87	13	Pods with Seeds	0.064
				89	20	Seed	<0.01
				89	20	Rest of plant*	0.096
				89	28	Seed	<0.01
				89	28	Rest of plant*	0.061
Study code:408612 **Doc ID:**2013/1037957 **Trial No.:**L120463 **GLP:**yes **Year**2012	Oilseed rape	Greece	BAS 310 55 I 2 x 0.0125	77/79	0	Whole plant*	0.28
				87/89	14	Seed	<0.01
				87/89	14	Rest of plant*	0.033
				89	21	Seed	<0.01
				89	21	Rest of plant*	0.011
				89	28	Seed	<0.01
				89	28	Rest of plant*	0.018
			BAS 310 55 I 1 x 0.025	77/79	0	Whole plant*	0.44
				87/89	14	Seed	<0.01
				87/89	14	Rest of plant*	0.013
				89	21	Seed	<0.01
				89	21	Rest of plant*	0.010
				89	28	Seed	<0.01
				89	28	Rest of plant*	<0.01
Study code:408612 **Doc ID:**2013/1037957 **Trial No.:**L120464 **GLP:**yes **Year**2012	Oilseed rape	Italy	BAS 310 55 I 2 x 0.0125	80	0	Whole plant*	0.20
				81	13	Rest of plant*	0.10
				81	13	Pods with Seeds	0.033
				87-89	21	Seed	<0.01
				87-89	21	Rest of plant*	0.090
				89	28	Seed	<0.01
				89	28	rest of plant*	0.19
			BAS 310 55 I 1 x 0.025	80	0	Whole plant*	0.30
				81	13	Rest of plant*	0.12
				81	13	Pods with Seeds	0.028
				87-89	21	Seed	<0.01
				87-89	21	Rest of plant*	0.14
				89	28	Seed	<0.01
				89	28	Rest of plant*	0.22

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408612 **Doc ID:**2013/1037957 **Trial No.:**L120465 **GLP:**yes **Year**2012 **Study code:**408612	Oilseed rape	Spain	BAS 310 55 I 2 x 0.0125	79	0	Whole plant*	0.17
				81	14	Rest of plant*	0.082
				81	14	Pods with Seeds	0.014
				85	22	Seed	<0.01
				85	22	Rest of plant*	0.12
				89	29	Seed	<u><0.01</u>
				89	29	Rest of plant*	<u>0.13</u>
			BAS 310 55 I 1 x 0.025	79	0	Whole plant*	0.28
				81	14	Rest of plant*	0.13
				81	14	Pods with Seeds	0.021
				85	22	Seed	<0.01
				85	22	Rest of plant*	0.17
				89	29	Seed	<0.01
				89	29	Rest of plant*	0.17

DALA = days after last application;
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Report: CA 6.3.4/4
Klimmek S.,Gizler A., 2014 c
Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in oilseed rape after two applications with BAS 310 55 I under field conditions in Germany, Belgium, N-France, The Netherlands, S-France, Greece, Italy and Spain, 2013
2013/1416283
Guidelines: none
GLP: yes
(certified by Freie und Hansestadt Hamburg, Behoerde fuer Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg, Germany)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50 g/L nominal
Purity: not reported
CAS#: 67375-30-8 (alpha-cypermethrin)
Development code: not reported
Spiking levels: 0.01, 0.10 and 8.0 mg/kg

2. Test Commodity:
Crop: Oilseed rape
Type: Oilseeds
Variety: Marquis, DK Esquisite, Safran, msl559c x ccc621205, Anderson, Nelson, Bagira, Frilola
Botanical name: *Brassica napus*
Crop parts(s) or processed commodity: Whole plant w/o roots, rest of plant without roots, pods with seeds, seed
Sample size: Whole plant without roots: ≥ 0.5 kg / 12 units
Rest of plant without roots: ≥ 0.5 kg / 12 units
Pods with seeds: ≥ 0.5 kg
Seed: ≥ 0.5 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2013 growing season, eight field trials were conducted under field conditions in representative oilseed rape growing areas in Germany, Belgium, Northern France, the Netherlands, Southern France, Greece, Italy and Spain to determine the residue level of alpha-cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).

Two foliar applications with BAS 310 55 I, an ME formulation of alpha-cypermethrin (50.0 g a.s./L), were made to plot 2 at a rate of 0.25 L of formulated product/ha (equivalent to 0.0125 kg a.s./ha) to oilseed rape at 28 days before harvest (DBH) and at 21 DBH. The nominal spray volume used was 200 L/ha.

Untreated and treated oilseed rape (whole plant (no roots)) specimens were sampled immediately pre application at 0 days before the last application (0 DBLA, GS 75-81) or immediately after the last application (0 DALA, GS 75-81), respectively.
Untreated and treated oilseed rape (seed) specimens were sampled at 14 DALA (GS 88-89), at 21-22 DALA (GS 87-89) and at 27-29 DALA (GS 89-90).
Untreated and treated oilseed rape (rest of plant without roots) specimens were sampled at 13-15 DALA (GS 79-89), at 20-22 DALA (GS 83-89) and at 27-29 DALA (GS 89-90).
Untreated and treated oilseed rape (pods with seeds) specimens were sampled at 13-15 DALA (GS 79-89) and at 20-22 DALA (GS 83-88).

The maximum storage interval from harvest until analysis of alpha-cypermethrin (BAS 310 I) was 439 days.

Table 6.3.4-19: Target application rates and timings for oilseed rape

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2013	8	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	200	1st appl. 28, 2nd appl. 21 days before harvest

2. Description of analytical procedures

All oilseed rape specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
The residues of alpha-cypermethrin in wheat specimens were extracted from plant matrices using a mixture of methanol / water / 2N hydrochloric acid (70:25:5, v/v/v). The extract was centrifuged and an aliquot was partitioned of into cyclohexane. The final determination of alpha-cypermethrin was performed by LC-MS/MS.
As a modification to optimize the recovery of alpha-cypermethrin during the liquid/liquid-partition with cyclohexane, the extract was partitioned three times in order to adapt the method to the laboratory situation.

The limit of quantitation (LOQ) was 0.01 mg/kg for alpha-cypermethrin.

Table 6.3.4-20: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in oilseed rape

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Oilseed rape / whole plant no roots	0.01	1	96.2	6.3	6.6
	0.1	1			
	8.0	1			
Oilseed rape / rest of plant without roots	0.01	3	83.9	16.4	19.6
	0.1	3			
	8.0	1			
Oilseed rape / pods with seeds	0.01	1	69.8	-	-
	0.10	1			
Oilseed rape / seed	0.01	2	86.3	16.9	19.6
	0.1	2			
Overall		**16**	**85.0**	**15.3**	**18.0**

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are given in Table 6.3.4-21, detailed residue levels are shown in Table 6.3.4-22 and Table 6.3.4-23.

At 0 DALA (GS 75-83) the residues of alpha-cypermethrin (BAS 310 I) in oilseed rape (whole plant no roots) specimens ranged between 0.20 mg/kg and 0.31 mg/kg.
At 14 DALA (GS 88-89) the residue of alpha-cypermethrin (BAS 310 I) in oilseed rape (seed) specimen was below the LOQ (<0.01 mg/kg).
At 13-15 DALA (GS 79-89) the residues of alpha-cypermethrin (BAS 310 I) in oilseed rape (rest of plant without roots) specimens ranged between 0.015 mg/kg and 0.15 mg/kg.
At 13-15 DALA (GS 79-89) the residues of alpha-cypermethrin (BAS 310 I) in oilseed rape (pods with seeds) specimens ranged between <0.01 mg/kg and 0.16 mg/kg.
At 21-22 DALA (GS 87-89) the residues of alpha-cypermethrin (BAS 310 I) in the oilseed rape (seed) specimens were all below the LOQ (<0.01 mg/kg).
At 20-22 DALA (GS 83-89) the residues of alpha-cypermethrin (BAS 310 I) in the oilseed rape (rest of plant without roots) specimens ranged between <0.01 mg/kg and 0.13 mg/kg.
At 20-22 DALA (GS 83-89) the residues of alpha-cypermethrin (BAS 310 I) in the oilseed rape (pods with seeds) specimens ranged between 0.036 mg/kg and 0.12 mg/kg.
At 27-29 DALA (GS 89-90) the residues of alpha-cypermethrin (BAS 310 I) in the oilseed rape (seed) specimens ranged between <0.01 mg/kg and 0.015 mg/kg.
At 27-29 DALA (GS 89-90) the residues of alpha-cypermethrin (BAS 310 I) in the oilseed rape (rest of plant without roots) specimens ranged between 0.040 mg/kg and 0.27 mg/kg.
No residues above the LOQ of alpha-cypermethrin (BAS 310 I) were found in any of the untreated specimens.

Table 6.3.4-21: Summary of residues of BAS 310 I in oilseed rape after one or two applications of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2013	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	78-83	Whole plant*	0.20-0.31
			13-14	81-87	Rest of plant*	0.055-0.15
			13-14	81-87	Pods with seeds	0.057-0.16
			20-22	87-89	Rest of plant*	<0.01-0.13
			20	88	Pods with seeds	0.12
			21-22	87-89	Seed	<0.01
			27-29	89-90	Rest of plant*	0.066-0.27
			27-29	89-90	Seed	<0.01
SEU	2013	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	75-81	Whole plant*	0.25-0.29
			14	88-89	Seed	<0.01
			14-15	79-87	Pods with seeds	<0.01-0.14
			14-15	79-89	Rest of plant*	0.015-0.078
			21-22	83-89	Rest of plant*	0.019-0.13
			21-22	83-87	Pods with seeds	0.036-0.056
			21-22	87-89	Seed	<0.01
			28-29	89	Seed	<0.01-0.015
			28-29	89	Rest of plant*	0.04-0.092

1 Days after last application
2 Growth stage at respective sampling
* = without roots

III. CONCLUSION

After treatment with BAS 310 55 at a target rate of two applications at 0.0125 kg a.s./ha, alpha-cypermethrin residues in seeds were < 0.01-0.015 mg/kg at 27-29 days after the last application.

Table 6.3.4-22: Residues of BAS 310 I after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408616 **Doc ID:**2013/1416283 **Trial No.:**S13-00443-01 / L130038 **GLP:**yes **Year**2013	Oilseed rape	Germany	BAS 310 55 I 2 x 0.0125	79-81	0	Whole plant*	0.20
				86-87	14	Rest of plant*	0.10
				86-87	14	Pods with Seeds	0.057
				89	22	Seed	<0.01
				89	22	Rest of plant*	0.10
				89-90	29	Seed	<u><0.01</u>
				89-90	29	Rest of plant*	<u>0.14</u>
Study code:408616 **Doc ID:**2013/1416283 **Trial No.:**S13-00443-02/ L130039 **GLP:**yes **Year**2013	Oilseed rape	Belgium	BAS 310 55 I 2 x 0.0125	80	0	Whole plant*	0.31
				81	14	Rest of plant*	0.15
				81	14	Pods with Seeds	0.13
				87-89	21	Seed	<0.01
				87-89	21	Rest of plant*	0.056
				89	28	Seed	<u><0.01</u>
				89	28	Rest of plant*	<u>0.066</u>
Study code:408616 **Doc ID:**2013/1416283 **Trial No.:**S13-00443-03/ L130040 **GLP:**yes **Year**2013	Oilseed rape	France	BAS 310 55 I 2 x 0.0125	78	0	Whole plant*	0.24
				82	14	Rest of plant*	0.14
				82	14	Pods with Seeds	0.16
				89	21	Seed	<0.01
				89	21	Rest of plant*	0.13
				89	28	Seed	<u><0.01</u>
				89	28	Rest of plant*	<u>0.27</u>
Study code:408616 **Doc ID:**2013/1416283 **Trial No.:**S13-00443-04/ L130041 **GLP:**yes **Year**2013 **Study code:**408616	Oilseed rape	The Netherlands	BAS 310 55 I 2 x 0.0125	83	0	Whole plant*	0.21
				87	13	Rest of plant*	0.055
				87	13	Pods with Seeds	0.14
				88	20	Rest of plant*	<0.01
				88	20	Pods with Seeds	0.12
				89	27	Seed	<u><0.01</u>
				89	27	Rest of plant*	<u>0.13</u>

DALA = days after last application;
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 6.3.4-23: Residues of BAS 310 I after two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408616	Oilseed rape	France	BAS 310 55 I 2 x 0.0125	80-81	0	Whole plant*	0.29
Doc ID:2013/1416283				85-87	14	Rest of plant*	0.078
Trial No.:S13-00443-05/ L130042				85-87	14	Pods with Seeds	0.14
GLP:yes				87	22	Seed	<0.01
Year2013				87	22	Rest of plant*	0.038
				89	29	Seed	<u><0.01</u>
				89	29	Rest of plant*	<u>0.040</u>
Study code:408616	Oilseed rape	Greece	BAS 310 55 I 2 x 0.0125	75	0	Whole plant*	0.27
Doc ID:2013/1416283				79	14	Rest of plant*	0.015
Trial No.:S13-00443-06/ L130043				79	14	Pods with Seeds	<0.01
GLP:yes				83	22	Rest of plant*	0.019
Year2013				83	22	Pods with Seeds	0.036
				89	29	Seed	<u>0.015</u>
				89	29	Rest of plant*	<u>0.092</u>
Study code:408616	Oilseed rape	Italy	BAS 310 55 I 2 x 0.0125	80	0	Whole plant*	0.28
Doc ID:2013/1416283				82	15	Rest of plant*	0.067
Trial No.:S13-00443-07/ L130044				82	15	Pods with Seeds	0.038
GLP:yes				87	21	Rest of plant*	0.047
Year2013				87	21	Pods with Seeds	0.056
				89	29	Seed	<u>0.012</u>
				89	29	Rest of plant*	<u>0.084</u>
Study code:408616	Oilseed rape	Spain	BAS 310 55 I 2 x 0.0125	78-79	0	Whole plant*	0.25
Doc ID:2013/1416283				88-89	14	Seed	<0.01
Trial No.:S13-00443-08/ L130045				88-89	14	Rest of plant*	0.057
GLP:yes				89	21	Seed	<0.01
Year2013				89	21	Rest of plant*	0.13
Study code:408616				89	28	Seed	<u><0.01</u>
				89	28	Rest of plant*	<u>0.060</u>

DALA = days after last application;
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

CA 6.3.5 Barley

Table 6.3.5-1: cGAP for the use of BAS 310 I in/on barley

Crop	Maximum applied dose	Water volume	PHI	Application method	Application timing
Cereals, outdoor (North; Central)	2 x 0.010 kg BAS 310 I/ha	100-400	28	Overall spray	BBCH 51 – 83
Cereals, outdoor (South)	2 x 0.010 kg BAS 310 I/ha	100-400	28	Overall spray	BBCH 51 – 83

PHI = pre-harvest interval

Table 6.3.5-2: GAP information of residue trials conducted in barley in 2012-2014 in Northern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Northern EU	France (1) Germany (1) The Netherlands (1) The United Kingdom (1) 2012	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 14±1 21±1 28±1	2013/ 1388974	No
	Belgium (1) Germany (1) The Netherlands (1) The United Kingdom (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.006	2	0 (4) 14-15 (4) 20-22 (4) 27-28 (2)	2013/ 1416284	No
	The United Kingdom (1) 2014	BAS 310 55 I ME	spray appl.	0.0125	0.006	2	0 14±1 21±1 28±1	2014/ 1173599	No

0 Actual application rates varied by 10% at most
1 Days after last application

Table 6.3.5-3: GAP information of residue trials conducted in barley in 2012-2013 in Southern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Southern EU	France (1) Greece (1) Italy (1) 2012	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 (3) 14 (3) 21±1 (3) 27±1 (2)	2013/ 1388974	No
				0.025	0.0125	1			
	Spain (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 14 21 28	2013/ 1416281	No
				0.025	0.0125	1			
	France (1) Greece (1) Italy (1) Spain (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.006	2	0 (4) 13-14 (4) 20-22 (4) 26-28 (4) 36 (1)	2013/ 1416284	No

0 Actual application rates varied by 10% at most
1 Days after last application

The studies not yet evaluated are summarized in the following chapter.

Report: CA 6.3.5/1
Tandy R., 2014a
Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in barley after treatment with BAS 310 55 I in Northern and Southern Europe during 2012
2013/1388974
Guidelines: SANCO/3029/99 rev. 4 (11 July 2000), EEC 91/414, EEC 1607/VI/97 rev. 2 10.06.1999
GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50.0 g/L nominal; 101209: 50.0 g/L nominal
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01-1.0 mg/kg

2. Test Commodity:
Crop: Barley
Type: Cereals
Variety: Pelican, Westminster, Naomi, Sebastian, Baracca, Loverde, Arta
Botanical name: *Hordeum vulgare*
Crop parts(s) or processed commodity: Whole plant w/o roots, ears, rest of plant, grain, straw
Sample size: Whole plant without roots: 0 DALA: ≥1.0kg / 12 units
Rest of plant without roots: ≥1.0kg
Grain: ≥1.0kg
Straw: ≥0.5kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the growing season of 2012, a total of seven trials were conducted in representative barley growing areas in Northern Europe (France, Germany, The Netherlands and The United Kingdom) and Southern Europe (France, Greece, Italy) to determine the magnitude and decline of residues of alpha-cypermethrin (BAS 310 I) in or on raw agricultural commodities (RAC).

The trials performed in Northern Europe (France, Germany, The Netherlands and The United Kingdom) consisted of two plots: one untreated plot (control) and one plot treated twice with BAS 310 55 I, a ME formulation of alpha-cypermethrin (50.0 g/L), at a target rate of 0.0125 kg alpha-cypermethrin/ha (plot 2).
In Southern Europe (France, Greece, Italy), the experimental set-up consisted of three plots: one plot untreated, a second plot treated twice with BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha (plot 2) and a third plot treated once with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha (plot 3). Plot 2 was treated 28-36 days before harvest and 21-29 days before harvest, respectively, while plot 3 was treated 21-29 days before harvest. The nominal spray volume used was 200 L/ha. In all trials the applications were made at crop stages BBCH 71-85.
In trial L120445, a higher rate of 0.0153 kg/a.s./ha (+22.3% above target) was applied to plot 2 at the first treatment. In trial L120450, the second application in plot 2 was at a rate of 0.014 kg a.s./ha (+12.22% above target) and the treatment rate for plot 3 was 0.0297 kg a.s./ha (+18.89% above target). These rates are still within 25% that is considered comparable in SANCO 7525/VI/95 - rev.9 and can be regarded as a worst case scenario.

For the analysis samples of barley plant (without roots) were taken directly after the last application (0 DALA). Ears and rest of plant (without roots) or grain and straw were taken at 13-14, 20-21, and 26-29 DALA, depending on the crop maturity.
The maximum storage interval from sampling until analysis was 359 days.

Table 6.3.5-4: Target application rates and timings for barley

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2012	7	2	F	BAS 310 55 I (EC)	alpha-cypermethrin	0.0125	200	1st appl.: 28±1 DBH 2nd appl.: 21±1 DBH
2012	3	1	F	BAS 310 55 I (EC)	alpha-cypermethrin	0.025	200	1st appl.: 21±1 DBH

DBH:days before harvest

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).
The residues of alpha-cypermethrin in barley specimens were extracted from plant matrices using a mixture of methanol/water/hydrochloric acid (70:25:5, v/v/v). After centrifugation, an aliquot of the extract was partitioned into cyclohexane. An aliquot of the cyclohexane phase was evaporated and the residue was taken up into methanol/water (80:20, v/v). The final determination of alpha-cypermethrin was performed by LC-MS/MS using the ammonium adduct of cypermethrin. The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery averaged 81±9% (mean±SD) for alpha-cypermethrin at fortification levels of 0.01 mg/kg -1.0 mg/kg.

Table 6.3.5-5: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in barley

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
whole plant w/o root	0.01-1.0	9	87.4	10.6	12.1
ears	0.01-0.4	6	78.9	7.7	9.8
rest of plant	0.01-0.5	6	82.4	7.2	8.8
grain	0.01-0.2	9	77.9	7.3	9.4
straw	0.01-0.5	6	74.6	5.5	7.3

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.5-6, detailed residue levels are shown in Table 6.3.5-7 and Table 6.3.5-8.
Directly after the last of two applications of BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha, residues of BAS 310 I (alpha-cypermethrin) in whole plant (without root) specimens ranged between 0.27-0.57 mg/kg. At 13-14 days after the last application residues were 0.099-0.33 mg/kg in ears, 0.14-0.45 in rest of plant (without roots) or 0.052 mg/kg and 0.43 mg/kg in grain and straw, respectively. At 20-21 DALA residues were 0.10-0.29 mg/kg in ears, 0.19-0.39 mg/kg in rest of plant without roots or 0.023-0.066 mg/kg and 0.24-0.41 mg/kg in grain and straw, respectively. After a longer PHI (26-29 DALA), residues in ears and rest of plant (without roots) were 0.12 and 0.40 mg/kg, respectively or 0.026-0.053 and 0.21-0.49 mg/kg in grain and straw, respectively.

Directly after a single treatment with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha, residues of BAS 310 I (alpha-cypermethrin) in whole plant (without root) specimens ranged between 0.32-0.61 mg/kg. At 14 days after the last application residues were 0.14-0.23 mg/kg in ears, 0.28-0.35 in rest of plant without roots or 0.099 mg/kg and 0.32 mg/kg in grain and straw, respectively. At 20-21 DALA residues were 0.13-0.21 mg/kg in ears, 0.031-0.39 mg/kg in rest of plant without roots or 0.061 mg/kg and 0.20 mg/kg in grain and straw, respectively. After a longer PHI (26-28 DALA) residues were 0.040-0.043 and 0.29-0.33 mg/kg in grain and straw, respectively.

No residues above LOQ were found in any of the analysed untreated specimens generated from trials L120444 through L120448 and L120450-L120451, respectively.

Table 6.3.5-6: Summary of residues of BAS 310 I in barley after one or two applications of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2012	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	76-83	Whole plant*	0.32-0.57
			13-14	78-87	Ears	0.099-0.33
			13-14	78-87	Rest of plant*	0.14-0.45
			20-21	85-87	Ears	0.10-0.18
			20-21	85-87	Rest of plant*	0.19-0.39
			20	85-87	Grain	0.029
			20	85-87	Straw	0.44
			28	89	Ears	0.12
			28	89	Rest of plant*	0.40
			28-29	89	Grain	0.031-0.053
			28-29	89	Straw	0.25-0.49
S-EU	2012	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	73-85	Whole plant*	0.27-0.51
			14	77-85	Ears	0.19-0.31
			14	77-85	Rest of plant*	0.22-0.25
			14	87	Grain	0.052
			14	87	Straw	0.43
			20-21	85-87	Ears	0.12-0.29
			20-21	85-87	Rest of plant*	0.23-0.37
			21	89	Grain	0.066
			21	89	Straw	0.24
			26-28	89	Grain	0.026-0.050
			26-28	89	Straw	0.21-0.40
		BAS 310 55 I 1 x 0.025 kg a.s./ha	0	73-85	Whole plant*	0.32-0.61
			14	77-85	Ears	0.14-0.23
			14	77-85	Rest of plant*	0.28-0.35
			14	87	Grain	0.099
			14	87	Straw	0.32
			20-21	85-87	Ears	0.13-0.21
			20-21	85-87	Rest of plant*	0.031-0.39
			20-21	89	Grain	0.061
			20-21	89	Straw	0.20
			26-28	89	Grain	0.040-0.043
			26-28	89	Straw	0.29-0.33

1 Days after last application
2 Growth stage at respective sampling
* = without roots

III. CONCLUSION

The residue levels of BAS 310 I in barley specimens taken directly after the last of two applications at a target rate of 0.0125 kg alpha-cypermethrin/ha (0 DALA) ranged between 0.27-0.57 mg/kg. In cereal grains from the same plots collected at 26-29 DALA, the residues of BAS 310 I were between 0.026-0.053 mg/kg.
Immediately after a single application of 0.025 kg alpha-cypermethrin/ha, the residue levels of BAS 310 I in barley whole plant ranged between 0.32-0.61 mg/kg. In cereal grains from the same plots collected at 26-28 DALA, the residues of BAS 310 I were between 0.040-0.043 mg/kg.

Table 6.3.5-7: Residues of BAS 310 I after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120444 **GLP:**yes **Year**2012	Barley	Germany	BAS 310 55 I 2 x 0.0125	81	0	Whole plant*	0.32
				85-87	13	Ears	0.18
				85-87	13	Rest of plant*	0.21
				87	20	Ears	0.18
				87	20	Rest of plant*	0.19
				89	29	Grain	0.035
				89	29	Straw	0.25
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120445 **GLP:**yes **Year**2012	Barley	UK	BAS 310 55 I 2 x 0.0125	79	0	Whole plant*	0.57
				83-85	13	Ears	0.33
				83-85	13	Rest of plant*	0.45
				85-87	20	Grain	0.029
				85-87	20	Straw	0.44
				89	29	Grain	0.053
				89	29	Straw	0.49
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120446 **GLP:**yes **Year**2012	Barley	The Netherlands	BAS 310 55 I 2 x 0.0125	76	0	Whole plant*	0.34
				78	14	Ears	0.16
				78	14	Rest of plant*	0.14
				85	21	Ears	0.15
				85	21	Rest of plant*	0.20
				89	28	Grain	0.032
				89	28	Straw	0.26
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120447 **GLP:**yes **Year**2012	Barley	France	BAS 310 55 I 2 x 0.0125	83	0	Whole plant*	0.37
				85	14	Ears	0.099
				85	14	Rest of plant*	0.34
				87	21	Ears	0.10
				87	21	Rest of plant*	0.39
				87	21	Grain	0.023
				87	21	Straw	0.41
				89	28	Ears	0.12
				89	28	Rest of plant*	0.40
				89	28	Grain	0.031
				89	28	Straw	0.47

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 6.3.5-8: Residues of BAS 310 I after one or two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120448 **GLP:**yes **Year**2012	Barley	Italy	BAS 310 55 I 2 x 0.0125	75	0	Whole plant*	0.27
				83-85	14	Ears	0.31
				83-85	14	Rest of plant*	0.25
				87	20	Ears	0.29
				87	20	Rest of plant*	0.37
				89	26	Grain	<u>0.050</u>
				89	26	Straw	<u>0.40</u>
			BAS 310 55 I 1 x 0.025	75	0	Whole plant*	0.36
				83-85	14	Ears	0.14
				83-85	14	Rest of plant*	0.35
				87	20	Ears	0.13
				87	20	Rest of plant*	0.031
				89	26	Grain	0.040
				89	26	Straw	0.29
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120450 **GLP:**yes **Year**2012	Barley	France	BAS 310 55 I 2 x 0.0125	85	0	Whole plant*	0.51
				87	14	Grain	0.052
				87	14	Straw	0.43
				89	21	Grain	<u>0.066</u>
				89	21	Straw	<u>0.24</u>
			BAS 310 55 I 1 x 0.025	85	0	Whole plant*	0.32
				87	14	Grain	0.099
				87	14	Straw	0.32
				89	21	Grain	0.061
				89	21	Straw	0.20
Study code:408610 **Doc ID:**2013/1388974 **Trial No.:**L120451 **GLP:**yes **Year**2012	Barley	Greece	BAS 310 55 I 2 x 0.0125	73	0	Whole plant*	0.51
				77	14	Ears	0.19
				77	14	Rest of plant*	0.22
				85	21	Ears	0.12
				85	21	Rest of plant*	0.23
				89	28	Grain	<u>0.026</u>
				89	28	Straw	<u>0.21</u>
			BAS 310 55 I 1 x 0.025	73	0	Whole plant*	0.61
				77	14	Ears	0.23
				77	14	Rest of plant*	0.28
				85	21	Ears	0.21
				85	21	Rest of plant*	0.39
				89	28	Grain	0.043
				89	28	Straw	0.33

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Report:	CA 6.3.5/2 Tandy, R., 2014c Study on the residue behaviour of alpha-cypermethrin (BAS 310 I) in barley after treatment with BAS 310 55 I in Southern Europe during 2013 2013/1416281
Guidelines:	SANCO/3029/99,EEC 1607/VI/97 rev. 2 10.06.1999,EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8)
GLP:	yes (certified by ENAC, Entidad Nacional de Acreditación, Madrid Spain)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I

Description:	BAS 310 55 I (ME)
Lot/Batch #:	101198: 50 g/L nominal
Purity:	not reported
CAS#:	67375-30-8 (alpha-cypermethrin)
Development code:	not reported
Spiking levels:	0.01, 0.10 and 8.0 mg/kg

2. Test Commodity:

Crop:	Barley
Type:	Cereals
Variety:	Unia
Botanical name:	*Hordeum vulgare*
Crop parts(s) or processed commodity:	Plant w/o roots, rest of plant without roots, ears, grain, straw
Sample size:	Whole plant without roots: ≥ 1 kg / 12 units Rest of plant without roots: ≥ 1 kg Ears: ≥ 1 kg Grain: ≥ 1 kg Straw: ≥ 0.5 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2013 growing season, one field trial was conducted in a representative barley growing area in Spain to determine the residue level of alpha-cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).

Plot 2 was treated with BAS 310 55 I, a ME formulation of alpha-cypermethrin (50.0 g/L) and was foliar applied twice at a rate of 0.25 L of formulated product/ha (equal to 0.0125 kg a.s./ha alpha-cypermethrin) to barley 28 days before harvest, and 21 days before harvest. The nominal spray volume used was 200 L/ha.

Plot 3 was treated with BAS 310 55 I, a ME formulation of alpha-cypermethrin (50.0 g/L) and was foliar applied at a rate of 0.5 L of formulated product/ha (equal to 0.025 kg a.s/ha alpha-cypermethrin) to barley 21 days before harvest. The nominal spray volume used was 200 L/ha.
Whole plant specimens were collected immediately before (plot U1) and after (plots 2 and 3) the last application. Ears and rest of plant specimens were collected 14 days after last application (DALA) from the untreated plot (U1) and treated plots 2 and 3. Grain and straw specimens were collected 21 and 28 days after last application (DALA) from the untreated plot (U1) and treated plots 2 and 3.

The maximum storage interval from harvest to first measurement was 296 days for barley specimens.

Table 6.3.5-9: Target application rates and timings for barley

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2013	1	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	200	1st appl. 28, 2nd appl. 21 days before harvest

2. Description of analytical procedures

All barley specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
The residues of alpha-cypermethrin in wheat specimens were extracted from plant matrices using a mixture of methanol / water / 2N hydrochloric acid (70:25:5, v/v/v). After centrifugation and partition of an aliquot of the extract into cyclohexane, evaporation to dryness and dissolving in methanol / water (80+20, v+v), the final determination of alpha-cypermethrin was performed by LC-MS/MS.
The limit of quantitation (LOQ) was 0.01 mg/kg for alpha-cypermethrin.

Table 6.3.5-10: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in barley

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Barley / whole plant no roots	0.01	1	94.7	22	23
	0.1	1			
	8.0	1			
Barley / ears	0.01	1	102.1	14	13
	0.1	1			
	8.0	1			
Barley / rest of plant without roots	0.01	1	82.0	17	20
	0.1	1			
	8.0	1			
Barley / grain	0.01	1	94.7	-	-
	0.10	1			
Barley / straw	0.01	2	94.9	16	17
	0.1	1			
	8.0	1			
Overall		**15**	**93.6**	**16**	**18**

II. RESULTS AND DISCUSSION

The residue results are shown in Table 6.3.5-11 and Table 6.3.5-12.
Residues of alpha-cypermethrin for plot 2 (two treatments at a target rate of 0.0125 kg a.s./ha), 0 days after last application were 0.39 mg/kg for whole plant (no roots) specimens. At 14 days after last application, plot 2, residues of alpha-cypermethrin were 0.38 mg/kg for ears and 0.38 mg/kg for rest of plant (without roots). At 21 days after last application, plot 2, residues of alpha-cypermethrin were 0.067 mg/kg for grain and 0.52 mg/kg for straw specimens. At 28 days after last application, plot 2, residues of alpha-cypermethrin were 0.083 mg/kg for grain and 0.43 mg/kg for straw specimens.

Residues of alpha-cypermethrin for plot 3 (a single treatment at 0.025 kg a.s./ha), 0 days after last application were 0.34 mg/kg for whole plant (no roots) specimens. At 14 days after last application, plot 3, residues of alpha-cypermethrin were 0.42 mg/kg for ears and 0.41 mg/kg for rest of plant (without roots). At 21 days after last application, plot 3, residues of alpha-cypermethrin were 0.12 mg/kg for grain and 0.57 mg/kg for straw specimens. At 28 days after last application, plot 3, residues of alpha-cypermethrin were 0.092 mg/kg for grain and 0.46 mg/kg for straw specimens.
No residues of alpha-cypermethrin at or above the limit of quantitation (0.010 mg/kg) were detected in the untreated barley specimens of this study.

Table 6.3.5-11: Summary of residues of BAS 310 I in barley after application of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
S-EU	2013	BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	0	79-83	Whole plant no roots	0.39
			14	87-89	Ears	0.38
					Rest of plant without roots	0.38
			21	89	Grain	0.067
					Straw	0.52
			28	89	Grain	0.083
					Straw	0.43
		BAS 310 55 I (ME) 1 x 0.025 kg a.s./ha	0	79-83	Whole plant no roots	0.34
			14	87-89	Ears	0.42
					Rest of plant without roots	0.41
			21	89	Grain	0.12
					Straw	0.57
			28	89	Grain	0.092
					Straw	0.46

1 Days after last application
2 Growth stage at respective sampling

III. CONCLUSION

At 28 days after treatment with BAS 310 55 I at a target rate of either twice 0.0125 kg a.s./ha or once 0.025 kg a.s./ha, the residues of alpha-cypermethrin (BAS 310 I) in barley straw ranged between 0.43 mg/kg and 0.46 mg/kg while in grain residues between 0.083-0.092 mg/kg were detected.

Table 6.3.5-12: Residues of BAS 310 I in barley after one or two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408610_1 **Doc ID:**2013/1416281 **Trial No.:**S13-00446-01 / L130003 **GLP:**yes **Year**2013	Barley	Spain	BAS 310 55 I 2 x 0.0125	79-83	0	Whole plant*	0.39
				87-89	14	Ears	0.38
				87-89	14	Rest of plant*	0.38
				89	21	Grain	0.067
				89	21	Straw	0.52
				89	28	Grain	<u>0.083</u>
				89	28	Straw	<u>0.43</u>
			BAS 310 55 I 1 x 0.025	79-83	0	Whole plant*	0.34
				87-89	14	Ears	0.42
				87-89	14	Rest of plant*	0.41
				89	21	Grain	0.12
				89	21	Straw	0.57
				89	28	Grain	0.092
				89	28	Straw	0.46

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Report: CA 6.3.5/3
Klimmek, S., Gizler, A., 2014c
Study on the residue behaviour of alpha-cypermethrin (BAS 310 I) in barley after two applications with BAS 310 55 I under field conditions in Germany, United Kingdom, The Netherlands, Belgium, Italy, Spain, Southern France and Greece, 2013
2013/1416284
Guidelines: none
GLP: yes
(certified by Freie und Hansestadt Hamburg, Behoerde für Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50 g/L nominal
Purity: not reported
CAS#: 67375-30-8 (alpha-cypermethrin)
Development code: not reported
Spiking levels: 0.01-8.0 mg/kg

2. Test Commodity:
Crop: Barley
Type: Cereals
Variety: Semper, Saffron, Malabar, Meridian, Atomo, Meseta, Ketos, Mutso
Botanical name: *Hordeum vulgare*
Crop parts(s) or processed commodity: Whole plant w/o roots, ears, rest of plant, grain, straw
Sample size: Whole plant without roots: ≥1.0 kg / ≥ 12 units
Rest of plant without roots: ≥1.0 kg / ≥ 12 units
Ears: ≥1.0 kg / ≥ 12 units
Grain: ≥1.0 kg / ≥ 12 units
Straw: ≥0.5 kg / ≥ 12 units

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2013 growing season, eight field trials were conducted under field conditions in representative barley growing areas in Germany, Belgium, the United Kingdom, the Netherlands, Italy, Spain, Greece and Southern France to determine the residue level of alpha-cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).

Two foliar applications with BAS 310 55 I, an ME formulation of alpha-cypermethrin (50.0 g a.s./L), were made to plot 2 at a rate of 0.25 L of formulated product/ha (equivalent to 0.0125 kg a.s./ha) to barley at 28 days before harvest (28 DBH) and at 21 DBH. The nominal spray volume used was 200 L/ha.

For trial S13-00441-02 (L130027), the actual application rates for plot 2, applications A1 and A2, were more than 25% below the nominal rate due to the incorrect amount of chemical weighed out. The results of this trial are reported separately.

At the day of application, the untreated barley (whole plant (no roots)) specimens were sampled immediately pre application (0 DBLA) and the treated barley (whole plant (no roots)) were taken immediately after the last application (0 DALA).

Untreated and treated barley (ears and rest of plant) specimens were sampled at 13-15 DALA, at 20-21 DALA and at 28 DALA.
Untreated and treated barley (grain and straw) specimens were sampled at 14 DALA, at 20-22 DALA, 27-28 DALA and at 26-36 DALA (BBCH 89, normal commercial harvest).

The maximum storage interval from harvest until analysis was 343 days.

Table 6.3.5-13: Target application rates and timings for barley

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2013	8	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	200	1st appl. 28±1, 2nd appl. 21±1 days before harvest

2. Description of analytical procedures

All barley specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
The residues of alpha-cypermethrin in barley specimens were extracted from plant matrices using a mixture of methanol/water/hydrochloric acid (70:25:5, v/v/v). After centrifugation and partition of an aliquot of the extract into cyclohexane, the final determination of alpha-cypermethrin was performed by LC-MS/MS.
As a modification to optimize the recovery of alpha-cypermethrin during the liquid/liquid-partition with cyclohexane, the extract was partitioned three times in order to adapt the method to the laboratory situation.
The limit of quantitation (LOQ) was 0.01 mg/kg for alpha-cypermethrin.

Table 6.3.5-14: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in barley

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Whole plant w/o root	0.01, 0.1 and 4.0	3	70.7	13.7	19.4
Ears	0.01 and 0.10	4	76.1	15.4	20.3
Rest of plant without roots	0.01, 0.1 and 8.0	6	79.0	10.1	12.8
Grain	0.01 and 0.10	7	80.2	15.2	19.0
Straw	0.01, 0.1 and 8.0	7	87.2	14.0	16.1
Overall		**27**	**80.1**	**13.7**	**17.1**

II. RESULTS AND DISCUSSION

The residue ranges for the different trials (excluding trial S13-00441-02 / L130027) are shown inTable 6.3.5-15, detailed residue levels are shown in Table 6.3.5-17 and Table 6.3.5-18. The results for trial S13-00441-02 / L130027 are shown in Table 6.3.5-16 and Table 6.3.5-19.

At 0 DALA the residues of alpha-cypermethrin (BAS 310 I) in barley (whole plant (no roots)) specimens ranged between 0.25 mg/kg and 0.78 mg/kg.
Alpha-cypermethrin (BAS 310 I) residues in barley (ears) specimens ranged between 0.048 mg/kg and 0.29 mg/kg at 13-14 DALA and between 0.15 mg/kg and 0.20 mg/kg at 20-21 DALA. At 28 DALA, the alpha-cypermethrin (BAS 310 I) in the barley (ears) specimen was 0.14 mg/kg.
Alpha-cypermethrin (BAS 310 I) residues in barley (rest of plant (without roots)) ranged between 0.16 mg/kg and 0.55 mg/kg at 13-14 DALA and between 0.20 mg/kg and 0.45 mg/kg at 20-21 DALA. At 28 DALA, the alpha-cypermethrin (BAS 310 I) in the barley (rest of plant (without roots)) specimen was 0.38 mg/kg.

Alpha-cypermethrin (BAS 310 I) residues in barley (grain) specimens ranged between 0.024 mg/kg and 0.075 mg/kg at 14 DALA and between 0.027 mg/kg and 0.11 mg/kg at 20-22 DALA.
Alpha-cypermethrin (BAS 310 I) residues in barley (grain) specimens ranged between 0.020 mg/kg and 0.079 mg/kg at 27-28 DALA and between 0.024 mg/kg and 0.079 mg/kg at 26-36 DALA.
Alpha-cypermethrin (BAS 310 I) residues in barley (straw) specimens ranged between 0.37 mg/kg and 0.61 mg/kg at 14 DALA and between 0.37 mg/kg and 0.92 mg/kg at 20-22 DALA. Alpha-cypermethrin (BAS 310 I) residues in barley (straw) specimens ranged between 0.35 mg/kg and 0.68 mg/kg at 27-28 DALA and between 0.40 mg/kg and 0.47 mg/kg at 26-36 DALA.
No residues above the LOQ of alpha-cypermethrin (BAS 310 I) were found in any of the untreated specimens.

Due to application rates being more than 25% below the target rate, the trial S13-00441-02 (L130027) is reported separately.
At 0 DALA the residue of alpha-cypermethrin (BAS 310 I) in the barley (whole plant (no roots)) specimen of trial S13-00441-02 was 0.13 mg/kg.
The alpha-cypermethrin (BAS 310 I) residue in the barley (ears) specimen of trial S13-00441-02 was 0.19 mg/kg at 15 DALA.
The alpha-cypermethrin (BAS 310 I) residue in the barley (rest of plant (without roots)) of trial S13-00441-02 was 0.34 mg/kg at 15 DALA.
The alpha-cypermethrin (BAS 310 I) residue in the barley (grain) specimen of trial S13-00441-02 was 0.026 mg/kg at 18 DALA.
The alpha-cypermethrin (BAS 310 I) residue in the barley (straw) specimen of trial S13-00441-02 was 0.35 mg/kg at 18 DALA.
No residues above the LOQ of alpha-cypermethrin (BAS 310 I) were found in any of the untreated specimens.

Table 6.3.5-15: Summary of residues of BAS 310 I in barley after application of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2013	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	77-85	Whole plant*	0.26-0.51
			14	85-87	Ears	0.048-0.29
			14	85-87	Rest of plant*	0.38-0.44
			14	87	Grain	0.075
			14	87	Straw	0.37
			20-22	89	Grain	0.027-0.11
			20-22	89	Straw	0.37-0.48
			27-28	89-90	Grain	0.020-0.079
			27-28	89-90	Straw	0.35-0.41
S-EU	2013	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	73-85	Whole plant*	0.25-0.78
			13-14	77-87	Ears	0.15-0.23
			13-14	77-87	Rest of plant*	0.16-0.55
			14	89	Grain	0.024
			14	89	Straw	0.61
			20-21	83-87	Ears	0.15-0.20
			20-21	83-87	Rest of plant*	0.20-0.45
			20-22	89	Grain	0.034-0.051
			20-22	89	Straw	0.46-0.92
			28	87	Ears	0.14
			28	87	Rest of plant*	0.38
			26-28	89	Grain	0.035-1.4**
			26-28	89	Straw	0.47-18.7**
			36	89	Grain	0.024
			36	89	Straw	0.40

1 Days after last application
2 Growth stage at respective sampling
* = without roots
** these results were deemed not plausible

Table 6.3.5-16: Summary of residues of BAS 310 I in barley after application of BAS 310 55 I - Trial S13-00441-02 / L130027

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2013	BAS 310 55 I 2 x 0.0125 kg a.s./ha	0	75-77	Whole plant*	0.13
			15	87	Ears	0.019
			15	87	Rest of plant*	0.34
			18	89	Grain	0.026
			18	89	Straw	0.35

III. CONCLUSION

At 26-28 days or later after the last of two applications of BAS 310 55 I at a target rate of 0.0125 kg a.s./ha, residues of alpha-cypermethrin in barley grain ranged between 0.020-0.079 mg/kg, while in straw 0.35-0.68 mg/kg were detected.

Table 6.3.5-17: Residues of BAS 310 I in barley after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 408615 **Doc ID:** 2013/1416284 **Trial No.:** S13-00441-01 / L130026 **GLP:** yes **Year** 2013	Barley	Germany	BAS 310 55 I 2 x 0.0125	83	0	Whole plant*	0.31
				87	14	Ears	0.048
				87	14	Rest of plant*	0.38
				89	20	Grain	0.027
				89	20	Straw	0.37
				90	27	Grain	<u>0.020</u>
				90	27	Straw	<u>0.35</u>
Study code: 408615 **Doc ID:** 2013/1416284 **Trial No.:** S13-00441-03 / L130028 **GLP:** yes **Year** 2013	Barley	The Netherlands	BAS 310 55 I 2 x 0.0125	77	0	Whole Plant*	0.51
				85	14	Ears	0.29
				85	14	Rest Of Plant*	0.44
				89	22	Grain	<u>0.077</u>
				89	22	Straw	<u>0.48</u>
Study code: 408615 **Doc ID:** 2013/1416284 **Trial No.:** S13-00441-04 / L130029 **GLP:** yes **Year** 2013	Barley	Belgium	BAS 310 55 I 2 x 0.0125	85	0	Whole Plant*	0.26
				87	14	Grain	0.075
				87	14	Straw	0.37
				89	21	Grain	0.11
				89	21	Straw	0.43
				89	28	Grain	<u>0.079</u>
				89	28	Straw	<u>0.41</u>

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 6.3.5-18: Residues of BAS 310 I in barley after two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408615 **Doc ID:**2013/1416284 **Trial No.:**S13-00441-05 / L130030 **GLP:**yes **Year**2013	Barley	Italy	BAS 310 55 I 2 x 0.0125	85	0	Whole plant*	0.78
				87	13	Ears	0.15
				87	13	Rest of plant*	0.55
				89	20	Grain	0.034
				89	20	Straw	0.92
				89	27	Grain	1.4**
				89	27	Straw	18.7**
Study code:408615 **Doc ID:**2013/1416284 **Trial No.:**S13-00441-06 / L130031 **GLP:**yes **Year**2013	Barley	Spain	BAS 310 55 I 2 x 0.0125	73-75	0	Whole Plant*	0.25
				83	13	Ears	0.18
				83	13	Rest of plant*	0.16
				85-87	20	Ears	0.20
				85-87	20	Rest of plant*	0.20
				89	26	Grain	0.079
				89	26	Straw	0.47
Study code:408615 **Doc ID:**2013/1416284 **Trial No.:**S13-00441-07 / L130032 **GLP:**yes **Year**2013	Barley	France	BAS 310 55 I 2 x 0.0125	83-85	0	Whole plant*	0.41
				89	14	Grain	0.024
				89	14	Straw	0.61
				89	22	Grain	0.051
				89	22	Straw	0.46
				89	28	Grain	0.035
				89	28	Straw	0.68
Study code:408615 **Doc ID:**2013/1416284 **Trial No.:**S13-00441-08 / L130033 **GLP:**yes **Year**2013	Barley	Greece	BAS 310 55 I 2 x 0.0125	73	0	Whole plant*	0.49
				77	14	Ears	0.23
				77	14	Rest of plant*	0.39
				83	21	Ears	0.15
				83	21	Rest of plant*	0.45
				87	28	Ears	0.14
				87	28	Rest of plant*	0.38
				89	36	Grain	0.024
				89	36	Straw	0.40

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation
** these results were deemed not plausible

Table 6.3.5-19: Residues of BAS 310 I in barley after two applications of BAS 310 55 I in Northern Europe – trial S13-00441-02 / L130027

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408615	Barley	UK	BAS 310 55 I 2 x 0.0125	76-77	0	Whole plant*	0.13
Doc ID:2013/1416284				87	15	Ears	0.019
Trial No.:S13-00441-02 / L130027				87	15	Rest of plant*	0.34
GLP:yes				89	18	Grain	0.026
Year2013				89	18	Straw	0.35

Report: CA 6.3.5/4
Klimmek, S., 2014
Study on the residue behaviour of alpha-cypermethrin (BAS 310 I) in barley after two applications with BAS 310 55 I under field conditions in United Kingdom, 2014
2014/1173599
Guidelines: none
GLP: yes
(certified by Freie und Hansestadt Hamburg, Behoerde für Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50 g/L nominal
Purity: not reported
CAS#: 67375-30-8 (alpha-cypermethrin)
Development code: not reported
Spiking levels: 0.01-1.0 mg/kg

2. Test Commodity:
Crop: Barley
Type: Cereals
Variety:
Botanical name: *Hordeum vulgare*
Crop parts(s) or processed commodity: Whole plant w/o roots, ears, rest of plant without roots, grain, straw
Sample size: Whole plant without roots: ≥1.0 kg
Rest of plant without roots: ≥1.0 kg / ≥ 12 units
Ears: ≥1.0 kg / ≥ 12 units
Grain: ≥1.0 kg / ≥ 12 units
Straw: ≥0.5 kg / ≥ 12 units

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2014 growing season, one field trial was conducted under field conditions in a representative barley growing area in the United Kingdom to determine the residue level of alpha-cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).

Two foliar applications with BAS 310 55 I, an ME formulation of alpha-cypermethrin (50.0 g a.s./L), were made to plot 2 at a rate of 0.25 L of formulated product/ha to barley at 28±1 days before harvest (28±1 DBH) and at 21±1 DBH. The nominal spray volume used was 200 L/ha.

Untreated and treated barley (whole plant (no roots)) specimens were sampled immediately pre application at 0 days before the last application (0 DBLA) or immediately after the last application (0 DALA), respectively.
Untreated and treated barley specimens were sampled at 14±1 DALA, at 21±1 DALA and at 28±1 DALA.

Table 6.3.5-20: Target application rates and timings for barley

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2014	1	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	200	1st appl. 28±1, 2nd appl. 21±1 days before harvest

2. Description of analytical procedures

All barley specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
The residues of alpha-cypermethrin in barley specimens were extracted from plant matrices using a mixture of methanol/water/hydrochloric acid (70:25:5, v/v/v). After centrifugation and partition of an aliquot of the extract into cyclohexane, the final determination of alpha-cypermethrin was performed by LC-MS/MS.
As a modification to optimize the recovery of alpha-cypermethrin during the liquid/liquid-partition with cyclohexane, the extract was partitioned three times in order to adapt the method to the laboratory situation.
The limit of quantitation (LOQ) was 0.01 mg/kg for alpha-cypermethrin.

Table 6.3.5-21: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in barley

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Whole plant w/o root	0.01	1	72.3	-	-
	0.1	1			
	1.0	1			
Ears	0.01	1	81.6	4.3	5.2
	0.1	1			
	1.0	1			
Rest of plant without roots	0.01	1	73.0	9.6	13.1
	0.1	1			
	1.0	1			
Grain	0.01	1	81.4	-	-
	0.1	1			
Straw	0.01	1	72.4	11.6	16.0
	0.1	1			
	1.0	1			
Overall			**73.7**	**10.0**	**13.5**

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.5-22, detailed residue levels are shown in Table 6.3.5-23.

At 0 DALA the residues of alpha-cypermethrin (BAS 310 I) in barley (whole plant (no roots)) specimens were 0.18 mg/kg.
Alpha-cypermethrin (BAS 310 I) residues in barley (ears) specimens were 0.16 mg/kg and in rest of plant without roots 0.21 mg/kg at 14±1 DALA. After 21±1 days, residues in grain were 0.05 mg/kg, while in straw 0.23 mg/kg were detected. After 28 days, residues in grain were 0.03 mg/kg and in straw 0.20 mg/kg, respectively.

Table 6.3.5-22: Summary of residues of BAS 310 I in barley after application of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] (BBCH)	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
EU-N	2014	BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	0		Whole plant no roots	0.18
			14±1		Ears	0.16
			14±1		Rest of plant without roots	0.21
			21±1		Grain	0.05
			21±1		Straw	0.23
			28±1		Grain	0.03
			28±1		Straw	0.20

1 Days after last application
2 At harvest

III. CONCLUSION

At 28 days after the last of two applications of BAS 310 55 I, residues of alpha-cypermethrin in barley grain lay at 0.03 mg/kg, while in straw 0.2 mg/kg were found.

Table 6.3.5-23: Residues of BAS 310 I in barley after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code: 408615_1 **Doc ID:** 2014/1173599 **Trial No.:** S14-00678 / L140092 **GLP:** yes **Year** 2014	Barley	UK	BAS 310 55 I 2 x 0.0125		0	Whole plant*	0.18
					14±1	Ears	0.16
					14±1	Rest of plant*	0.21
					21±1	Grain	0.05
					21±1	Straw	0.23
					28±1	Grain	<u>0.03</u>
					28±1	Straw	<u>0.20</u>

DALA = days after last application
BBCH = growth stage at respective sampling
* = without roots
_ underlined values were used for MRL calculation

CA 6.3.6 Wheat

Table 6.3.6-1: cGAP for the use of BAS 310 I in/on wheat

Crop	Maximum applied dose	Water volume	PHI	Application method	Application timing
Cereals, outdoor (North; Central)	2 x 0.010 kg BAS 310 I/ha	100-400	28	Overall spray	BBCH 51 – 83
Cereals, outdoor (South)	2 x 0.010 kg BAS 310 I/ha	100-400	28	Overall spray	BBCH 51 – 83

PHI = pre-harvest interval

Table 6.3.6-2: GAP information of residue trials conducted in wheat in 2007-2013 in Northern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Northern EU	Belgium (1) Denmark (1) Germany (1) The Netherlands (1) 2007	BAS 310 70 00 I SL	spray appl.	0.0125	0.00417	2	0 28±1 35±1 42±1	2008/ 1002701	no
		BAS 310 40 I EC		0.015	0.005				
	France (1) Germany (1) The United Kingdom (2) 2012	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 (4) 14±1 (4) 21±1 (4) 28±1 (4) 35 (1) 65 (1)	2014/ 1028112	no
	France (1) Germany (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.006	2	0 14 20-21 27-28	2013/ 1416282	no

0 Actual application rates varied by 10% at most
1 Days after last application

Table 6.3.6-3: GAP information of residue trials conducted in wheat in 2007-2013 in Southern Europe

Region	Country (No of trials) Year	Formu-lation	Application[0]				DALA[1]	DocID	EU accepted
			Method	Rate[0] (kg a.s./ha)	Spray conc. (kg a.s./hL)	No			
Southern EU	France (1) Greece (1) Italy (1) Spain (1) 2007	BAS 310 70 00 I SL	spray appl.	0.0125	0.00417	2	0 28±1 35±1 42±1	2008/ 1002701	no
		BAS 310 40 I EC		0.015	0.005				
	France (1) Greece (1) Italy (1) Spain (1) 2012	BAS 310 55 I ME	spray appl.	0.0125	0.00625	2	0 (4) 14±1 (4) 21±1 (4) 28 (4) 36 (1)	2014/ 1028112	no
				0.025	0.0125	1			
	Italy (1) Spain (1) 2013	BAS 310 55 I ME	spray appl.	0.0125	0.006	2	0 14 21 27-28	2013/ 1416282	no

0 Actual application rates varied by 10% at most
1 Days after last application

The studies not yet evaluated are summarized in the following chapter.

Report:	CA 6.3.6/1 Kreke N.,Gehl J., 2008b Residues of Alpha-Cypermethrin and Acetamiprid in wheat (RAC whole plant, straw and grain) following two treatments with BAS 370 00 I, BAS 310 40 I or BAS 911 10 I from eight open field trials in Northern and Southern Europe in 2007 2008/1002701
Guidelines:	EEC 96/68,EEC 1607/VI/97 rev. 2 10.06.1999,EEC 7029/VI/95 rev. 5,EEC 7525/VI/95 rev. 7,EEC 91/414 Annex II (Part A Section 6),EEC 91/414 Annex III (Part A Section 8)
GLP:	yes (certified by Swiss Federal Office of Public Health, Berne, Switzerland)

Data from supplemental decline trials reported in chapter 6 (supplemental information) indicate that after a more critical application of 1x 15g/ha, residues after 7 days were already <0.01 mg/kg in wheat grains. The last application is therefore more important as the interval between the two applications as residues do not accumulate. Therefore, the applicant considers the results of this study still acceptable even though the interval between two applications is 14 days instead of 7 days.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description:	BAS 370 00 I BAS 310 40 I BAS 9111 0 I
Lot/Batch #:	1463 (BAS 370 00 I, 25 g/L alpha-cypermethrin, 100 g/L acetamiprid, SL) 1209 (BAS 310 40 I, 100 g/L alpha-cypermethrin, EC) FRE-000429 (BAS 9111 0 I, 200 g/kg acetamiprid)
Purity:	not relevant
CAS#:	alpha-cypermethrin: 67375-30-8 acetamiprid: 160430-64-8
Development code:	not applicable
Spiking levels:	0.01-1.0 mg/kg (alpha-cypermethrin) 0.01-5.0 mg/kg (acetamiprid)

2. Test Commodity:

Crop:	Wheat
Type:	Cereals
Variety:	Opus, Bussard, Limes, Einstein, Dakter, Eva, Arcancelo, Galera
Botanical name:	*Triticum aestivum*
Crop parts(s) or processed Commodity:	Whole plant w/o root, rest of plant w/o root, straw, grain, ears
Sample size:	0.5-1.0 kg

B. STUDY DESIGN

1. Test procedure

During the 2007 growing season, a total of eight trials were conducted on wheat in order to determine the magnitude of residues of active ingredient(s) in or on Raw Agricultural Commodities (RAC).
Plot 101: Untreated (control)
Plot 102: BAS 370 00 I, a SL formulation of alpha-cypermethrin (25 g/L) and acetamiprid (100 g/L), was applied twice at the rate of 500 mL/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).
Plot 103: BAS 310 40 I, an EC formulation of alpha-cypermethrin (100 g/L), was applied twice at the rate of 150 mL/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).
Plot 104: BAS 9111 0 I, a WP formulation of acetamiprid (200 g/kg), was applied twice at the rate of 250 g/ha, in order to determine the magnitude of the residues of active ingredient in or on Raw Agricultural Commodities (RAC).
In context of this summary, only the results of alpha-cypermethrin (plot 102 & plot 103) are reported.
Wheat specimens were collected immediately after the last application and 28 ± 1, 35 ± 1 and 42 ± 1 days after the last application.
The maximum storage interval from first sampling until analysis was 321 days.

Table 6.3.6-4: Target application rates and timings for wheat

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2007	8	2	F	BAS 370 00 I (SL)	alpha-cypermethrin acetamiprid	0.0125 0.05	300	49±1 DBH 35±1 DBH
			F	BAS 310 40 I (EC)	alpha-cypermethrin	0.015		
			F	BAS 9111 0 I (WP)	acetamiprid	0.05		

2. Description of analytical procedures

The specimens were analyzed according to BASF analytical method number 567/0 for alpha-cypermethrin and to Nippon Soda CO. LTD method RD-9991 N2 for acetamiprid.
Both methods have a limit of quantification of 0.01 mg/kg in all sample materials. Procedural recoveries averaged 88% for alpha-cypermethrin and 83% for acetamiprid, respectively, at fortification levels between 0.01 and 1 mg/kg (alpha-cypermethrin) or between 0.01 and 5 mg/kg (acetamiprid).
Alpha-cypermethrin: the residues of alpha-cypermethrin are extracted from plant matrices using a mixture of methanol, water and HCl 2 mol/L. For clean-up a liquid/liquid partition against cyclohexane is used. The final determination of alpha-cypermethrin is performed by LC/MS/MS. The limit of quantification (LOQ) ofthe method is 0.01 mg/kg.

Acetamiprid: The residues of acetamiprid are extracted from plant matrices using a mixture of methanol and water. For clean-up a liquid/liquid partition against dichloromethane is used. The final determination of acetamiprid is performed by LC/MS/MS. The limit of quantification (LOQ) ofthe method is 0.01 mg/kg.

Table 6.3.6-5: Summary of recoveries of alpha-cypermethrin and acetamiprid in wheat matrices

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
wheat grain	0.01 / 1.0	20	88	11	13
RD-9991 N2		acetamiprid			
wheat grain, ears, rest of plant w/o roots	0.01 / 0.1 / 5.0	23	83	8	10

II. RESULTS AND DISCUSSION

For alpha-cypermethrin, at all samplings from plot 102 and 103, in wheat grain only residues below or at the LOQ (0.01 mg/kg) were determined. At 0 DALA, in whole plants residues up to 0.82 mg/kg were observed. In straw or rest of plant specimens without roots, sampled at about 28 DALA, residues up to 1.04 mg/kg were determined. In straw specimens sampled at about 35 or 42 DALA, residues up to 0.96 were observed.
An overall summary of the residues is given in Table 6.3.6-6, detailed results are listed in Table 6.3.6-7 and Table 6.3.6-8.

Table 6.3.6-6: Summary of residues of BAS 310 I in wheat from trials according to critical GAP after application of BAS 370 00 I and BAS 310 40 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2007	BAS 370 00 I 2 x 0.0125 kg a.s./ha	0	71-75	Whole plant*	0.18-0.59
			28	85	Ears	0.06-0.27
			28	85	Rest of plant*	0.34-0.73
			27	87	Grain	<0.01
			27	87	Straw	0.16-0.51
			34-36	88-89	Grain	<0.01
			34-36	88-89	Straw	0.28-0.73
			41-42	89-91	Grain	<0.01
			41-42	89-91	Straw	0.23-0.82
		BAS 310 40 I 2 x 0.015 kg a.s./ha	0	71-75	Whole plant*	0.22-0.42
			28	85	Ears	0.06-0.26
			28	85	Rest of plant*	0.30-0.46
			27	87	Grain	<0.01
			27	87	Straw	0.26-0.46
			34-36	88-89	Grain	<0.01
			34-36	88-89	Straw	0.28-0.56
			41-42	89-91	Grain	<0.01-0.01
			41-42	89-91	Straw	0.24-0.96
S-EU	2007	BAS 370 00 I 2 x 0.0125 kg a.s./ha	0	69-83	Whole plant*	0.25-0.82
			27-28	83-87	Ears	0.04-0.31
			27-28	83-87	Rest of plant*	0.15-0.99
			27-28	87-92	Grain	<0.01
			27-28	87-92	Straw	0.49-1.04
			87-93	34-36	Grain	<0.01
			87-93	34-36	Straw	0.20-0.80
			41-42	89-99	Grain	<0.01
			41-42	89-99	Straw	0.15-0.82
		BAS 310 40 I 2 x 0.015 kg a.s./ha	0	69-83	Whole plant*	0.28-0.69
			27-28	83-87	Ears	0.04-0.17
			27-28	83-87	Rest of plant*	0.09-0.52
			27-28	87-92	Grain	<0.01
			27-28	87-92	Straw	0.57-1.04
			87-93	34-36	Grain	<0.01
			87-93	34-36	Straw	0.16-0.81
			41-42	89-99	Grain	<0.01
			41-42	89-99	Straw	0.15-0.83

1 Days after last application
2 Growth stage at respective sampling
* = without roots

III. CONCLUSION

Directly after the last application of BAS 370 00 I or BAS 310 40 I, the residue of alpha-cypermethrin ranged between 0.18 and 0.82 mg/kg in wheat specimens (whole plants). At 28±1 days (and later) the residues found in grain ranged between <0.01 and 0.01 mg/kg.
The analytical results obtained demonstrate that the treatment with two applications of either BAS 370 00 I or BAS 310 40 I did not lead to significantly different residue results in the raw agricultural commodity at harvest.

Table 6.3.6-7: Residues of BAS 310 I after two applications of the formulation BAS 370 00 I and BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070422 **GLP:**yes **Year**2007	Wheat	Denmark	BAS 370 00 I 2 x 0.0125	73	0	Whole plant*	0.38
				85	28	Ears	0.06
				85	28	Rest of plant	0.34
				89	35	Grain	<u><0.01</u>
				89	35	Straw	<u>0.36</u>
				89	42	Grain	<0.01
				89	42	Straw	0.23
			BAS 310 40 I 2 x 0.015	73	0	Whole plant*	0.29
				85	28	Ears	0.06
				85	28	Rest of plant	0.30
				89	35	Grain	<0.01
				89	35	Straw	0.28
				89	42	Grain	<0.01
				89	42	Straw	0.33
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070423 **GLP:**yes **Year**2007	Wheat	Germany	BAS 370 00 I 2 x 0.0125	75	0	Whole plant*	0.37
				85	28	Ears	0.27
				85	28	Rest of plant	0.73
				88	36	Grain	<u><0.01</u>
				88	36	Straw	<u>0.73</u>
				91	41	Grain	<0.01
				91	41	Straw	0.70
			BAS 310 40 I 2 x 0.015	75	0	Whole plant*	0.25
				85	28	Ears	0.26
				85	28	Rest of plant	0.46
				88	36	Grain	<0.01
				88	36	Straw	0.48
				91	41	Grain	0.01
				91	41	Straw	0.56

Table 6.3.6-7: Residues of BAS 310 I after two applications of the formulation BAS 370 00 I and BAS 310 40 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070424 **GLP:**yes **Year**2007	Wheat	The Netherlands	BAS 370 00 I 2 x 0.0125	71	0	Whole plant*	0.18
				87	27	Grain	<u><0.01</u>
				87	27	Straw	<u>0.51</u>
				89	34	Grain	<0.01
				89	34	Straw	0.28
				89	41	Grain	<0.01
				89	41	Straw	0.25
			BAS 310 40 I 2 x 0.015	71	0	Whole plant*	0.22
				87	27	Grain	<0.01
				87	27	Straw	0.46
				89	34	Grain	<0.01
				89	34	Straw	0.28
				89	41	Grain	<0.01
				89	41	Straw	0.24
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070425 **GLP:**yes **Year**2007	Wheat	Belgium	BAS 370 00 I 2 x 0.0125	71	0	Whole plant*	0.59
				87	27	Grain	<0.01
				87	27	Straw	0.16
				89	34	Grain	<u>0.01</u>
				89	34	Straw	0.48
				89	42	Grain	<0.01
				89	42	Straw	<u>0.82</u>
			BAS 310 40 I 2 x 0.015	71	0	Whole plant*	0.42
				87	27	Grain	<0.01
				87	27	Straw	0.26
				89	34	Grain	<0.01
				89	34	Straw	0.56
				89	42	Grain	<0.01
				89	42	Straw	0.96

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 6.3.6-8: Residues of BAS 310 I after two applications of the formulation BAS 370 00 I and BAS 310 40 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070426 **GLP:**yes **Year**2007	Wheat	France	BAS 370 00 I 2 x 0.0125	83	0	Whole plant*	0.82
				87	28	Ears	0.31
				87	28	Rest of plant	0.99
				89	35	Grain	<0.01
				89	35	Straw	0.80
				97	42	Grain	<0.01
				97	42	Straw	0.50
			BAS 310 40 I 2 x 0.015	83	0	Whole plant*	0.53
				87	28	Ears	0.17
				87	28	Rest of plant	0.52
				89	35	Grain	<0.01
				89	35	Straw	0.81
				97	42	Grain	<0.01
				97	42	Straw	0.83
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070427 **GLP:**yes **Year**2007	Wheat	Greece	BAS 370 00 I 2 x 0.0125	71	0	Whole plant*	0.54
				87	27	Grain	<0.01
				87	27	Straw	0.49
				89	34	Grain	<0.01
				89	34	Straw	0.58
				89	42	Grain	<0.01
				89	42	Straw	0.82
			BAS 310 40 I 2 x 0.015	71	0	Whole plant*	0.69
				87	27	Grain	<0.01
				87	27	Straw	0.57
				89	34	Grain	<0.01
				89	34	Straw	0.56
				89	42	Grain	<0.01
				89	42	Straw	0.62
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070428 **GLP:**yes **Year**2007	Wheat	Italy	BAS 370 00 I 2 x 0.0125	71-75	0	Whole plant*	0.58
				89-92	28	Grain	<0.01
				89-92	28	Straw	1.04
				92-93	35	Grain	<0.01
				92-93	35	Straw	0.66
				97-99	41	Grain	<0.01
				97-99	41	Straw	0.79
			BAS 310 40 I 2 x 0.015	71-75	0	Whole plant*	0.38
				89-92	28	Grain	<0.01
				89-92	28	Straw	1.04
				92-93	35	Grain	<0.01
				92-93	35	Straw	0.70
				97-99	41	Grain	<0.01
				97-99	41	Straw	0.77

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:253702 **Doc ID:**2008/1002701 **Trial No.:**L070429 **GLP:**yes **Year**2007	Wheat	Spain	BAS 370 00 I 2 x 0.0125	69	0	Whole plant*	0.25
				83-85	27	Ears	0.04
				83-85	27	Rest of plant	0.15
				87-89	36	Grain	<u><0.01</u>
				87-89	36	Straw	<u>0.20</u>
				89	42	Grain	<0.01
				89	42	Straw	0.15
			BAS 310 40 I 2 x 0.015	69	0	Whole plant*	0.28
				83-85	27	Ears	0.04
				83-85	27	Rest of plant	0.09
				87-89	36	Grain	<0.01
				87-89	36	Straw	0.16
				89	42	Grain	<0.01
				89	42	Straw	0.15

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Report: CA 6.3.6/2
Tandy R., 2014d
Study on the residue behaviour of Alpha-Cypermethrin (BAS 310 I) in wheat after treatment with BAS 310 55 I in Northern and Southern Europe during 2012
2014/1028112

Guidelines: SANCO/3029/99 rev. 4 (11 July 2000), EEC 7029/VI/95 rev. 5, EEC 91/414, EU Regulation 1107/2009 with Regulation 283/2013, EU Regulation 1107/2009 with Regulation 284/2013

GLP: yes
(certified by Department of Health of the Government of the United Kingdom, United Kingdom)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50.0 g/L nominal
Purity: not reported
CAS#: 67375-30-8
Development code: not reported
Spiking levels: 0.01-1.5 mg/kg

2. Test Commodity:
Crop: Wheat
Type: Cereals
Variety: Tabasco, AC Barrie, Granary, Gabriel, Imgeuio, Simeto, Dylan, Bastide
Botanical name: *Triticum aestivum*
Crop parts(s) or processed commodity: Whole plant w/o root, rest of plant, straw, grain, ears
Sample size: Whole plant w/o root ≥1.0 kg / 12 units; ears, rest of plant (w/o roots), grain ≥1.0 kg; straw ≥0.5 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

During the growing season of 2012, a total of eight trials were conducted in representative wheat growing areas in Northern and Southern Europe to determine the magnitude and decline of residues of alpha-cypermethrin (BAS 310 I) in or on raw agricultural commodities (RAC).

The trials performed in Northern Europe (France, Germany, and The United Kingdom-2 trials) consisted of two plots: one untreated plot (control) and one plot treated twice with BAS 310 55 I (ME, 50 g a.s./L) at a target rate of 0.0125 kg alpha-cypermethrin/ha.

In Southern Europe (France, Greece, Italy and Spain), the experimental set-up consisted of three plots: one plot untreated, a second plot treated twice with BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha and a third plot treated once with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha.
The applications to plot 2 (two applications at 0.0125 kg alpha-cypermethrin/ha) were made 35 - 72 days before harvest, and 28 - 65 days before harvest. Plot 3 (single application at 0.025 kg alpha-cypermethrin/ha) was treated 28 - 36 days before harvest. The nominal spray volume used was 200 L/ha for both variants.
For the analysis samples of wheat plant (without roots) were taken directly after the last application (0 DALA). Ears and rest of plant (without roots) or grain and straw were taken at 13-15, 20-22, 28-29 and 35-65 DALA, depending on the crop maturity.
The maximum storage interval from sampling until analysis was 393 days.

Table 6.3.6-9: Target application rates and timings for wheat

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2012	8	2	F	BAS 310 55 I (EC)	alpha-cypermethrin	0.0125	200	1st appl.: 28±1 DBH 2nd appl.: 21±1 DBH
2012	4	1	F	BAS 310 55 I (EC)	alpha-cypermethrin	0.025	200	1st appl.: 21±1 DBH

DBH: days before harvest

2. Description of analytical procedures

The method BASF No. 567/0 was used in this study for the determination of BAS 310 I (alpha-cypermethrin).
The residues of alpha-cypermethrin in wheat specimens were extracted from plant matrices using a mixture of methanol/water/hydrochloric acid (70:25:5, v/v/v). After centrifugation, an aliquot of the extract was partitioned into cyclohexane. An aliquot of the cyclohexane phase was evaporated and the residue was taken up into methanol/water (80:20, v/v). The final determination of alpha-cypermethrin was performed by LC-MS/MS using the ammonium adduct of cypermethrin. The limit of quantitation (LOQ) of the method is 0.01 mg/kg. The mean procedural recovery averaged 80.9±10.8% (mean±SD) for alpha-cypermethrin at fortification levels of 0.01 mg/kg -1.5 mg/kg.

Table 6.3.6-10: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in wheat

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
whole plant w/o root	0.01-1.0	6	91.7	4.0	4.4
ears	0.01-0.6	9	79.1	6.3	8.0
rest of plant (without roots)	0.01-0.5	6	72.6	4.4	6.1
grain	0.01-0.5	11	78.4	9.3	11.9
straw	0.01-1.5	9	84.3	15.6	18.6

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.6-11, detailed residue levels are shown in Table 6.3.6-12 and Table 6.3.6-13.
Directly after the last of two applications of BAS 310 55 I at a target rate of 0.0125 kg alpha-cypermethrin/ha, residues of BAS 310 I (alpha-cypermethrin) in whole plant (without root) specimens ranged between 0.17-0.86 mg/kg. At 13-15 days after the last application residues were 0.088-0.40 mg/kg in ears, 0.11-0.44 in rest of plant (without roots) or <0.01 mg/kg and 0.346 mg/kg in grain and straw, respectively. At 20-22 DALA, residues were 0.069-0.41 mg/kg in ears, 0.13-0.38 mg/kg in rest of plant (without roots) or <0.01-0.013 mg/kg and 0.26-0.71 mg/kg in grain and straw, respectively. At 28-29 DALA, residues in ears and rest of plant (without roots) were 0.047-0.24 and 0.11-0.33 mg/kg, respectively or <0.01 and 0.29-0.53 mg/kg in grain and straw, respectively. After a longer PHI (35-65 DALA), residues were <0.01 mg/kg in grain and 0.18-0.35 in straw.
Directly after a single treatment with BAS 310 55 I at a target rate of 0.025 kg alpha-cypermethrin/ha, residues of BAS 310 I (alpha-cypermethrin) in whole plant (without root) specimens ranged between 0.44-0.89 mg/kg. At 14 days after application, residues were 0.098-0.32 mg/kg in ears, 0.12-0.27 in rest of plant (without roots) or <0.01 mg/kg and 0.45 mg/kg in grain and straw respectively. At 21-22 DALA residues were 0.11-0.27 mg/kg in ears, 0.11-0.32 mg/kg in rest of plant (without roots) or <0.01 mg/kg and 0.46-0.84 mg/kg in grain and straw, respectively. At 28 DALA, residues in ears and rest of plant (without roots) were 0.064-0.21 and 0.12-0.19 mg/kg, respectively or <0.01 and 0.36-0.74 mg/kg in grain and straw, respectively.After a longer PHI (36 DALA), residues were <0.01 and 0.26 mg/kg in grain and straw, respectively.

No residues above the LOQ were found in any of the analysed untreated specimens generated from trials L120452 through L120459.

Table 6.3.6-11: Summary of residues of BAS 310 I in wheat after one or two applications of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2012	BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	0	69-83	Whole plant*	0.30-0.86
			13-15	75-89	Ears	0.088-0.34
			13-15	75-89	Rest of plant*	0.21-0.44
			21-22	77-89	Ears	0.077-0.41
			21-22	77-89	Rest of plant*	0.21-0.38
			20-21	85-89	Grain	<0.01-0.013
			20-21	85-89	Straw	0.26-0.71
			28-29	77-87	Ears	0.047-0.12
			28-29	77-87	Rest of plant*	0.13-0.33
			28-29	87-89	Grain	<0.01
			28-29	87-89	Straw	0.29-0.53
			35-65	89	Grain	<0.01
			35-65	89	Straw	0.18-0.35
S-EU	2012		0	69-85	Whole plant*	0.17-0.61
			14-15	77-87	Ears	0.088-0.40

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
		BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	14-15	77-87	Rest of plant*	0.11-0.25
			14	87	Grain	<0.01
			14	87	Straw	0.35
			21-22	83-89	Ears	0.069-0.27
			21-22	83-89	Rest of plant*	0.13-0.32
			21-22	87-89	Grain	<0.01
			21-22	87-89	Straw	0.38-0.56
			28	83-89	Ears	0.05-0.24
			28	83-89	Rest of plant*	0.11-0.20
			28	89	Grain	<0.01
			28	89	Straw	0.33-0.54
			36	89	Grain	<0.01
			36	89	Straw	0.18
		BAS 310 55 I (ME) 1 x 0.025 kg a.s./ha	0	69-85	Whole plant*	0.44-0.89
			14-15	77-87	Ears	0.098-0.32
			14-15	77-87	Rest of plant*	0.12-0.27
			14	87	Grain	<0.01
			14	87	Straw	0.45
			21-22	83-89	Ears	0.11-0.27
			21-22	83-89	Rest of plant*	0.11-0.32
			21-22	87-89	Grain	<0.01
			21-22	87-89	Straw	0.46-0.84
			28	83-89	Ears	0.064-0.21
			28	83-89	Rest of plant*	0.12-0.19
			28	89	Grain	<0.01
			28	89	Straw	0.36-0.74
			36	89	Grain	<0.01
			36	89	Straw	0.26

1 Days after last application
2 Growth stage at respective sampling
* = without roots

III. CONCLUSION

The residue levels of BAS 310 I in wheat specimens taken directly after the last of two applications at a target rate of 0.0125 kg alpha-cypermethrin/ha (0 DALA) ranged between 0.17-0.86 mg/kg. In wheat grains from the same plots collected at 28±1 DALA and later, no residues above the LOQ were detected (<0.01 mg/kg).
Immediately after a single application of 0.025 kg alpha-cypermethrin/ha, the residue levels of BAS 310 I in wheat whole plant (without roots) ranged between 0.44-0.89 mg/kg. In wheat grains from the same plots collected at 28±1 DALA and later, no residues above the LOQ were detected (<0.01 mg/kg).

Table 6.3.6-12: Residues of BAS 310 I in wheat after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120452 **GLP:**yes **Year**2012	Wheat	Germany	BAS 310 55 I 2 x 0.0125	83	0	Whole plant*	0.46
				85	13	Ears	0.24
				85	13	Rest of plant*	0.35
				89	20	Grain	0.013
				89	20	Straw	0.71
				89	28	Grain	<u><0.01</u>
				89	28	Straw	<u>0.53</u>
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120453 **GLP:**yes **Year**2012	Wheat	UK	BAS 310 55 I 2 x 0.0125	77-79	0	Whole plant*	0.86
				87-89	13	Ears	0.34
				87-89	13	Rest of plant*	0.44
				89	22	Ears	0.41
				89	22	Rest of plant*	0.38
				89	29	Grain	<u><0.01</u>
				89	29	Straw	<u>0.29</u>
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120454 **GLP:**yes **Year**2012	Wheat	UK	BAS 310 55 I 2 x 0.0125	69-71	0	Whole plant*	0.60
				75	15	Ears	0.12
				75	15	Rest of plant*	0.21
				77-83	22	Ears	0.077
				77-83	22	Rest of plant*	0.21
				77-83	29	Ears	0.047
				77-83	29	Rest of plant*	0.13
				89	65	Grain	<u><0.01</u>
				89	65	Straw	<u>0.181</u>
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120455 **GLP:**yes **Year**2012	Wheat	France	BAS 310 55 I 2 x 0.0125	75	0	Whole plant*	0.30
				83	14	Ears	0.088
				83	14	Rest of plant*	0.25
				85	21	Ears	0.078
				85	21	Rest of plant*	0.25
				85	21	Grain	<0.01
				85	21	Straw	0.26
				87	28	Ears	0.12
				87	28	Rest of plant*	0.33
				87	28	Grain	<u><0.01</u>
				87	28	Straw	<u>0.40</u>
				89	35	Grain	<0.01
				89	35	Straw	0.35

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Table 6.3.6-13: Residues of BAS 310 I after one or two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120456 **GLP:**yes **Year**2012	Wheat	France	BAS 310 55 I 2 x 0.0125	83-85	0	Whole plant*	0.30
				87	14	Grain	<0.01
				87	14	Straw	0.35
				89	21	Grain	<0.01
				89	21	Straw	0.56
				89	28	Grain	<u><0.01</u>
				89	28	Straw	<u>0.54</u>
			BAS 310 55 I 1 x 0.025	83-85	0	Whole plant*	0.44
				87	14	Grain	<0.01
				87	14	Straw	0.45
				89	21	Grain	<0.01
				89	21	Straw	0.84
				89	28	Grain	<0.01
				89	28	Straw	0.74
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120457 **GLP:**yes **Year**2012	Wheat	Greece	BAS 310 55 I 2 x 0.0125	69-71/73	0	Whole plant*	0.61
				77	14	Ears	0.12
				77	14	Rest of plant*	0.25
				85	21	Ears	0.088
				85	21	Rest of plant*	0.19
				89	28	Grain	<u><0.01</u>
				89	28	Straw	<u>0.33</u>
			BAS 310 55 I 1 x 0.025	69-71/73	0	Whole plant*	0.89
				77	14	Ears	0.18
				77	14	Rest of plant*	0.26
				85	21	Ears	0.15
				85	21	Rest of plant*	0.32
				89	28	Grain	<0.01
				89	28	Straw	0.38
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120458 **GLP:**yes **Year**2012	Wheat	Italy	BAS 310 55 I 2 x 0.0125	69-71	0	Whole plant*	0.17
				79	14	Ears	0.088
				79	14	Rest of plant*	0.11
				83	21	Ears	0.069
				83	21	Rest of plant*	0.13
				83-85	28	Ears	0.050
				83-85	28	Rest of plant*	0.11
				89	36	Grain	<u><0.01</u>
				89	36	Straw	<u>0.18</u>
			BAS 310 55 I 1 x 0.025	69-71	0	Whole plant*	0.50
				79	14	Ears	0.098
				79	14	Rest of plant*	0.12
				83	21	Ears	0.11
				83	21	Rest of plant*	0.11
				83-85	28	Ears	0.064
				83-85	28	Rest of plant*	0.12
				89	36	Grain	<0.01
				89	36	Straw	0.26

Study Details	Crop	Country	Formulation, Application Rate (kg a.s./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408613 **Doc ID:**2014/1028112 **Trial No.:**L120459 **GLP:**yes **Year**2012	Wheat	Spain	BAS 310 55 I 2 x 0.0125	75-77	0	Whole plant*	0.45
				85-87	15	Ears	0.40
				85-87	15	Rest of plant*	0.17
				87-89	22	Ears	0.27
				87-89	22	Rest of plant*	0.32
				87-89	22	Grain	<0.01
				87-89	22	Straw	0.38
				89	28	Ears	0.24
				89	28	Rest of plant*	0.20
				89	28	Grain	<u><0.01</u>
				89	28	Straw	<u>0.35</u>
			BAS 310 55 I 1 x 0.025	75-77	0	Whole plant*	0.49
				85-87	15	Ears	0.32
				85-87	15	Rest of plant*	0.27
				87-89	22	Ears	0.27
				87-89	22	Rest of plant*	0.27
				87-89	22	Grain	<0.01
				87-89	22	Straw	0.46
				89	28	Ears	0.21
				89	28	Rest of plant*	0.19
				89	28	Grain	<0.01
				89	28	Straw	0.36

DALA = days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

Report: CA 6.3.6/3
Klimmek, S., Gizler, A., 2014d
Study on the residue behaviour of alpha-cypermethrin (BAS 310 I) in wheat after two applications with BAS 310 55 I under field conditions in Germany, Northern France, Italy and Spain, 2013
2013/1416282
Guidelines: none
GLP: yes
(certified by Freie und Hansestadt Hamburg, Behoerde für Soziales, Familie, Gesundheit und Verbraucherschutz, Hamburg)

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material: Alpha-Cypermethrin BAS 31055I
Description: BAS 310 55 I (ME)
Lot/Batch #: 101198: 50 g/L nominal
Purity: not reported
CAS#: 67375-30-8 (alpha-cypermethrin)
Development code: not reported
Spiking levels: 0.01, 0.1 and 8.0 mg/kg

2. Test Commodity:
Crop: Wheat
Type: Cereals
Variety: Tabasco, Courtot, Blasco, Burgos
Botanical name: *Triticum aestivum*
Crop parts(s) or processed commodity: Plant w/o roots, rest of plant without roots, ears, grain, straw
Sample size: Plant without roots: 0 DALA ≥ 1 kg / ≥ 12 units
14±1, 21±1, 28±1 DALA:
Rest of plant without roots: ≥ 1 kg / ≥ 12 units
Ears: ≥ 1 kg / ≥ 12 units
Grain: ≥ 1 kg / ≥ 12 units
Straw: ≥ 0.5 kg / ≥ 12 units

B. STUDY DESIGN AND METHODS

1. Test procedure

During the 2013 growing season, four field trials were conducted under field conditions in representative wheat growing areas in Germany, Northern France, Italy and Spain to determine the residue level of alpha-cypermethrin (BAS 310 I) in or on Raw Agricultural Commodities (RAC).
Two foliar applications with BAS 310 55 I, an ME formulation of alpha-cypermethrin (50.0 g a.s./L), were made to plot 2 to wheat, at a rate of 0.25 L formulated product/ha, equal to 12.5 g alpha-cypermethrin/ha. The nominal spray volume used was 200 L/ha.
Untreated and treated wheat specimens of all trials were sampled immediately pre application at 0 days before the last application (0 DBLA, untreated whole plant no roots specimens only), at 0 days after the last application (0 DALA, treated whole plant no roots specimens only) and at 14 (ears and rest of plant or grain and straw, according to the stage of the crop), 20-21 (ears and rest of plant or grain and straw, according to the stage of the crop) and 27-28 (grain and straw) days after the last application.

The maximum storage interval from harvest until analysis was 372 days.

Table 6.3.6-14: Target application rates and timings for wheat

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2013	4	2	F	BAS 310 55 I (ME)	alpha-cypermethrin	0.0125	200	1st appl. 28±1, 2nd appl. 21±1 days before harvest

2. Description of analytical procedures

All wheat specimens were analyzed for alpha-cypermethrin according to BASF analytical method no. 567/0, which determines the analytes by means of LC-MS/MS.
The residues of alpha-cypermethrin in wheat specimens were extracted from plant matrices using a mixture of methanol / water / 2N hydrochloric acid (70:25:5, v/v/v). After centrifugation and partition of an aliquot of the extract into cyclohexane, evaporation to dryness and dissolving in methanol / water (80+20, v+v), the final determination of alpha-cypermethrin was performed by LC-MS/MS.
As a modification to optimize the recovery of alpha-cypermethrin during the liquid/liquid-partition with cyclohexane, the extract was partitioned three times in order to adapt the method to the laboratory situation.
The limit of quantitation (LOQ) was 0.01 mg/kg for alpha-cypermethrin.

Table 6.3.6-15: Summary of recoveries of BAS 310 I (alpha-cypermethrin) in wheat

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
Wheat / whole plant no roots	0.01	1	75.7	14.3	18.8
	0.1	1			
	8.0	1			
Wheat / ears	0.01	1	74.9	-	-
	0.1	1			
Wheat / rest of plant without roots	0.01	2	73.9	12.2	16.6
	0.1	2			
	8.0	1			
Wheat / grain	0.01	1	79.9	-	-
	0.10	1			
Wheat / straw	0.01	1	71.0	10.5	14.7
	0.1	1			
	8.0	1			
Overall		**15**	**74.6**	**9.7**	**13.0**

II. RESULTS AND DISCUSSION

The residue ranges for the different trials are shown in Table 6.3.6-16. Detailed results are shown in Table 6.3.6-17 and Table 6.3.6-18.

At 0 DALA the residues of alpha-cypermethrin (BAS 310 I) in wheat (whole plant no roots) specimens ranged between 0.35 mg/kg and 0.56 mg/kg. At 14 days after the last application, residues in wheat (ears) specimens ranged between 0.026 mg/kg and 0.095 mg/kg and between 0.25 mg/kg and 0.40 mg/kg wheat (rest of plant without roots) specimens. At the same sampling point, the residue of alpha-cypermethrin (BAS 310 I) in the wheat (grain) specimen was below the LOQ (<0.01 mg/kg), while a residue of 0.32 mg/kg was determined in straw. After 20 days, the residue of alpha-cypermethrin (BAS 310 I) in the wheat (ears) specimen was 0.082 mg/kg and 0.23 mg/kg in the wheat (rest of plant no roots) specimen. Residues in wheat (straw) specimens ranged between 0.32 mg/kg and 0.38 mg/kg at 21 days after the last application. In wheat (grain) specimens the residues were all below the LOQ (<0.01 mg/kg) at 21 days after the last application. At harvest (27-28 DALA, GS 89) the residues of alpha-cypermethrin (BAS 310 I) in wheat (grain) specimens were all below the LOQ (<0.01 mg/kg), while the residues of alpha-cypermethrin (BAS 310 I) in wheat (straw) specimens ranged between 0.36 mg/kg and 0.47 mg/kg.
No residues of alpha-cypermethrin (BAS 310 I) above the LOQ were found in the untreated specimens of this study, with the exception of rest of plant without roots specimen L1300360003 of trial S13-00445-03 (L130036), taken at 14 DALA (GS 87), where an alpha-cypermethrin (BAS 310 I) residue of 0.019 mg/kg was detected. The source of the contamination has not been determined. Taking into account the corresponding residues in the treated specimens the residue in the control sample had no impact on the study.

Table 6.3.6-16: Summary of residues of BAS 310 I in wheat after application of BAS 310 55 I

Region	Year	Application	DALA[1]	Growth stage[2] BBCH	Residues Found (mg/kg)	
					Matrix	alpha-cypermethrin (BAS 310 I)
N-EU	2013	BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	0	75-77	Whole plant*	0.35-0.37
			14	83-87	Ears	0.026-0.095
			14	83-87	Rest of plant*	0.25-0.34
			20	87	Ears	0.082
			20	87	Rest of plant*	0.23
			21	89	Grain	<0.01
			21	89	Straw	0.38
			27-28	89	Grain	<0.01
			27-28	89	Straw	0.36-0.40
S-EU	2013	BAS 310 55 I (ME) 2 x 0.0125 kg a.s./ha	0	81-87	Whole plant*	0.47-0.56
			14	87	Ears	0.048
			14	87	Rest of plant*	0.40
			14	87-89	Grain	<0.01
			14	87-89	Straw	0.32
			21	87-89	Grain	<0.01
			21	87-89	Straw	0.32-0.35
			27-28	89	Grain	<0.01
			27-28	89	Straw	0.37-0.47

1 Days after last application
2 Growth stage at respective sampling
* = without roots

III. CONCLUSION

At 27-28 days after the last of two applications of BAS 310 55I at a target rater of 0.0125 kg a.s./ha, the residues of alpha-cypermethrin (BAS 310 I) in straw ranged between 0.36 mg/kg and 0.47 mg/kg. No residues above the LOQ were detected in any grain samples at the PHI or at earlier time points (all <0.01 mg/kg).

Table 6.3.6-17: Residues of BAS 310 I in wheat after two applications of BAS 310 55 I in Northern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408665 **Doc ID:**2013/1416282 **Trial No.:**S13-00445-01 / L130034 **GLP:**yes **Year**2013	Wheat	Germany	BAS 310 55 I 2 x 0.0125	75	0	Whole plant*	0.37
				83	14	Ears	0.026
				83	14	Rest of plant*	0.25
				87	20	Ears	0.082
				87	20	Rest of plant*	0.23
				89	27	Grain	<u><0.01</u>
				89	27	Straw	<u>0.36</u>
Study code:408665 **Doc ID:**2013/1416282 **Trial No.:**S13-00445-02 / L130035 **GLP:**yes **Year**2013	Wheat	France	BAS 310 55 I 2 x 0.0125	77	0	Whole plant*	0.35
				87	14	Ears	0.095
				87	14	Rest of plant*	0.34
				89	21	Grain	<0.01
				89	21	Straw	0.38
				89	28	Grain	<u><0.01</u>
				89	28	Straw	<u>0.40</u>

Table 6.3.6-18: Residues of BAS 310 I in wheat after two applications of BAS 310 55 I in Southern Europe

Study Details	Crop	Country	Formulation, Application Rate (kg as./ha)	GS BBCH	DALA	Residues Found (mg/kg)	
						Matrix	BAS 310 I
Study code:408665 **Doc ID:**2013/1416282 **Trial No.:**S13-00445-03 / L130036 **GLP:**yes **Year**2013	Wheat	Italy	BAS 310 55 I 2 x 0.0125	83	0	Whole plant*	0.47
				87	14	Ears	0.048
				87	14	Rest of plant*	0.40
				87-89	21	Grain	<0.01
				87-89	21	Straw	0.35
				89	27	Grain	<u><0.01</u>
				89	27	Straw	<u>0.37</u>
Study code:408665 **Doc ID:**2013/1416282 **Trial No.:**S13-00445-04 / L130037 **GLP:**yes **Year**2013	Wheat	Spain	BAS 310 55 I 2 x 0.0125	81-87	0	Whole plant*	0.56
				87-89	14	Grain	<0.01
				87-89	14	Straw	0.32
				89	21	Grain	<0.01
				89	21	Straw	0.32
				89	28	Grain	<u><0.01</u>
				89	28	Straw	<u>0.47</u>

DALA = Days after last application
BBCH = Growth stage (GS) at respective sampling
* = without roots
_ underlined values were used for MRL calculation

In addition to the studies presented in CA 6.3 for the representative crops cereals, oil seed rape, lettuce, courgette and leafy cabbage, supervised field trials on other crops for which registrations of alpha-cypermethrin based products exist are presented in a supplement document (DocID 2014/1314800). These studies were conducted at the currently registered GAP for the respective crops and are presented to provide additional information on the derivation of crop related MRLs and STMRs used in the chronic dietary risk assessment presented in section CA 6.7 of this dossier and for the determination of metabolite relevance. These calculations are presented in detail in the supplement document and the results are summarized in section CA 6.9. The additional studies are partly peer reviewed, but also new studies are presented. These data are not meant to be fully evaluated but to provide further information as outline above.

CA 6.4 Feeding studies

Intended uses with the representative formulation BAS 310 55 I covered in this submission include kale, oilseed rape and cereals which are relevant feed items.
As a chronic consumer risk assessment based on only these crops does not allow a realistic overall assessment a second scenario was calculated comprising all uses intended to be defended in future. Feeding studies necessary as basis for these calculations were all submitted during the peer review under Directive 91/414/EEC. No new studies were conducted.

CA 6.4.1 Poultry

During the peer review under Directive 91/414/EEC, a feeding study with laying hens (AL-440-018) was submitted. This study was assessed as not being required as the feed burden estimate performed resulted in a maximum intake of 0.085 mg/kg diet which was below the trigger (3rd Addendum to the Monograph: Addendum Chapter B-7 (Residue data), January 2003). Therefore, a review was not performed at this time. This study was not included in the application as it had been submitted already with the alpha-cypermethrin addendum dossier.

Report:	CA 6.4.1/1 [redacted] 2001a Fastac insecticide (Alphacypermethrin - BAS 310 I): Magnitude of BAS 310 I residues in laying hen eggs, muscle, liver and abdominal fat after oral administration of BAS 310 I for 28 consecutive days AL-440-018
Guidelines:	EPA 860.1480, EEC 91/414, EEC 7031/VI/95 Appendix G rev. 4, SANCO/3029/99 rev. 4 (11 July 2000), 8046/VI/97-rev 4 (15/12/1998)
GLP:	yes (certified by <none>)

Executive Summary

The objective of the present study was to investigate the magnitude of alpha-cypermethrin residues in poultry tissues and eggs. In this study, 56 acclimated laying hens were divided randomly into 6 treatment groups: placebo (A, 12 hens), 1.2 mg/kg feed (B, 12 hens), 6.1 mg/kg feed (C, 12 hens) and 12 mg/kg feed (D, 12 hens / E, 4 hens / F, 4 hens). The nominal doses were equivalent to approximately 0.076, 0.38 and 0.76 mg a.s./kg bw/animal/day. Each hen/group was orally dosed once daily for 28 consecutive days using gelatin capsules. Alpha-cypermethrin accumulation was monitored in composite egg and edible tissue samples (muscle, liver and abdominal fat) from each treatment group. Egg samples were collected approximately 3 times each week during the dosing period, and edible tissues were collected immediately upon sacrifice. Group A-D animals were sacrificed within 24 hours of receiving their final (28th) dose. Group E and F animals were sacrificed at 7 days after the final dose and at 14 days after the final dose, respectively.

No residues of alpha-cypermethrin above the LOQ (0.01 mg/kg) were detected in eggs from treated hens in the 1.2 mg/kg group. In the 6.1 mg/kg dose group, residues slightly above the LOQ from day 9 until the end of the dosing period (0.0101-0.0125 mg/kg between day 9-28).
Alpha-cypermethrin residues in eggs from treated hens in the high dose group reached a plateau averaging approximately 27 mg/kg within approximately 21-24 days of dosing. There were no reportable residues after 7 days' depuration.
Alpha-cypermethrin residues did not accumulate in laying hen liver or muscle after 28 daily doses. Residues were <0.05 mg/kg in liver and muscle tissue sampled from the high dose animals. Alpha-cypermethrin residues in laying hen abdominal fat averaged approximately 0.235 mg/kg after 28 days of dosing at the high rate and 0.085 mg/kg at the medium rate. No residues above the LOQ (0.05 mg/kg) were detected in fat tissue from the low dose group. A residue decline in abdominal fat of approximately 61% was noted in the high dose group after 7-14 days' depuration.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: Alpha-cypermethrin (BAS 310 I, former code AC 900049)
Lot/Batch #: AC12395-18
Purity: 96.1 %
CAS#: 67375-30-8
Development code: not reported
Dose levels: equivalent to 0 (placebo) / 1.21 / 6.05 / 12.1 mg/kg in feed (nominal)
Spiking levels: 0.01-0.5

2. Test Commodity:

Organism: Poultry
Type: Chicken
Variety: Babcock B-300 White Leghorn (Hens)
Systematical name: *Gallus gallus domesticus*
commodity: Eggs, muscle, abdominal fat, liver
Sample size: Eggs: Eggs were collected twice per sampling interval. Each egg sampling interval consisted of all eggs from the PM collection of the day previous plus all eggs from the AM collection of that sampling day. All eggs from the four hens per treatment subgroup were collected at each sampling timepoint.
Sacrifice within 24 h after final dosing:

Muscle:	332-539 g (composite samples, 4 animals)
Abdominal fat:	83-156 g (composite samples, 4 animals)
Liver:	112-153 g (composite samples, 4 animals)

B. STUDY DESIGN

1. Test procedure

Fifty-six acclimated laying hens were divided randomly into 6 treatment groups: placebo (A, 12 hens), 1.2 mg/kg feed (B, 12 hens), 6.1 mg/kg feed (C, 12 hens) and 12 mg/kg feed (D, 12 hens / E, 4 hens / F, 4 hens). The nominal doses were equivalent to approximately 0.076, 0.38 and 0.76 mg a.s./kg bw/animal/day.
Each hen/group was orally dosed once daily for 28 consecutive days using gelatin capsules. The test material was a Certified Toxicological sample at 96.1 % purity.

Lock ring gelatin capsules (Torpac, size 0, batch 1610892, expiration October 2003) filled with were used. The lactose was delivered into capsules using a capsule-filling machine.
Test substance dosing solutions were prepared in 1: 5 acetone: n-hexane, such that every capsule would receive the same volume of working solution independent of the dose rate. Each capsule received 100 µl of the appropriate dosing solution. Solvent was permitted to evaporate from the capsules at room temperature for at least 3 hours prior to packaging. Each dosing capsule was placed into a labeled vial, packaged together according to dose group in appropriately labeled boxes, and stored refrigerated until use in the study.

BAS 310 I accumulation was monitored in composite egg and edible tissue samples (muscle, liver and abdominal fat) from each treatment group. Egg samples were collected approximately 3 times each week during the dosing period. Eggs were collected twice per sampling interval. Each egg sampling interval consisted of all eggs from the PM collection of the day previous plus all eggs from the AM collection of that sampling day. All eggs from the four hens per treatment subgroup were collected at each sampling timepoint.
Edible tissues were collected immediately upon sacrifice. Group A-D animals were sacrificed within 24 hours of receiving their final (28th) dose. Group E and F animals were sacrificed at 7 days after the final dose and at 14 days after the final dose, respectively.

All samples from each sampling interval were hand-delivered to the Sample Preparation and Handling Area (SPHA) at BASF in Princeton, NJ. Egg samples were homogenized within 2 hours of receipt. The resulting homogenate was divided into three 50-ml polypropylene conical tubes and were placed into a freezer at approximately ≤-10°C, where they were maintained until transfer into analytical laboratory custody.

Edible tissue samples were logg placed into a freezer maintained at approximately ≤-10°C until preparation for analysis. During preparation, individual tissue samples were thoroughly ground and homogenized in a Hobart food chopper surrounded by dry ice. The resulting homogenates were divided into two sub-samples, packed into cardboard containers, and were placed back into the freezer until transfer to the analytical laboratory.

Egg sub-samples were removed from SPHA storage and transferred to the analytical laboratory for processing. Edible tissue sub-samples were shipped by Federal Express to the contract analytical laboratory (MAXIM Technologies, Incorporated in Middleport, NY).

Statistics:
Other than calculation of means, standard deviations and CVs, no statistical analysis was performed on the data from this study. This study was properly controlled throughout its in-life and analytical phases to minimize bias.

2. Description of analytical procedures

Remark: Although analytical method SAMS 461-1 has already been assessed, it is briefly described for completeness of the study summary.

BASF method SAMS 461-1 for hen tissues:
Edible tissues were collected and analyzed for ~~BAS 310 I~~total cypermethrin residues according to BASF method SAMS 461-1, which was validated for hen muscle, liver and abdominal fat as part of this study (LOQ, 0.05 mg/kg). Method SAMS 461-1 was peer-reviewed under Directive 91/414/EEC earlier (Alpha-cypermethrin Monograph, 1999).

Principle of method SAMS 461-1:
Residues of BAS 310 I were extracted from poultry tissues by boiling (three times) with acetone:hexane (1 :2; v/v). The extract was concentrated to dryness and redissolved in hexane. For fat and meat an aliquot of the extract is partitioned with acetonitrile using an Extrelut cartridge. For liver, an aliquot was removed and a liquid/liquid, hexane/acetonitrile partition was used. The extract was further cleaned up by SPE using a Florisil cartridge. Residues were quantitated using a GC equipped with an ECD. Results were calculated by the direct comparison of peak heights to those of external standards. The validated sensitivity (LOQ, limit of quantitation) of the method is 0.05 mg/kg.

Table 6.4.1-1: Summary of Validation data of alpha-cypermethrin in hen´s tissues

Matrix Poultry	Fortification Level (mg/kg)	Summary Recoveries				
		Individual recoveries (%)	n	Mean (%)	SD	RSD (%)
BASF Method SAMS 461-1						
Meat	0.05	104 / 110 / 95 / 98 / 115	5	104	8.3	7.9
	0.50	74 / 74 / 95 / 102 / 74	5	84	13.6	16.2
Liver	0.05	84 / 79 / 84 / 87 / 83	5	83	2.9	3.5
	0.50	89 / 81 / 80 / 84 / 74	5	82	5.5	6.7
Fat	0.05	109 / 101 / 104 / 89 / 106	5	102	7.7	7.6
	0.50	100 / 98 / 108 / 93 / 93	5	98	6.2	6.3

Table 6.4.1-2: Method SAMS 461-1 - Concurrent recoveries

Compound	Matrix Poultry	Range		
		Fortification Level (mg/kg)	Recovery (%)	Average recovery (%)
BAS 310 I	Meat	0.05	115	NA
	Liver	0.05-0.50	78-88	83 (n=2)
	Fat	0.05-0.50	86-107	93 (n=3)

BASF method M 3466 for hen eggs:
Egg samples were analyzed for BAS 310 I residues according to BASF method M 3466 (limit of quantitation, LOQ, 0.01 mg/kg) which was validated in this study. Method M 3466 as a part of this poultry-feeding study was not peer-reviewed under Directive 91/414/EEC earlier because the whole study was regarded as not required due to an anticipated feed burden below the trigger value. Therefore, the validation results and method performance parameters are summarized in detail below.

Principle of the method M 3466:
In the validated method, residues of BAS 310 I are extracted from homogenized eggs with a mixture of hexane and tetrahydrofuran (THF) (1:2 v/v). Aliquots of the organic extracts are evaporated and the residue is dissolved in a mixture of hexane and dichloromethane (1:1 v/v). Extracts are purified using adsorption chromatography on a one-gram silica SPE cartridge, followed by liquid-liquid partitioning using hexane/acetonitrile. Quantitative determination is carried out by gas chromatography/electron capture negative ion chemical ionization tandem mass spectrometry (GC/MS-ECNICI). Results are calculated as BAS 310 I by the direct comparison of the sample peak response to the peak responses of bracketing standards. The validated sensitivity (LOQ, limit of quantitation) of the method is 0.01 mg/kg.

Detailed results of the method validation are given in Table 6.4.1-1. Concurrent recoveries from egg sample analysis, extract and freezer stability analyses are presented in Table 6.4.1-2.

Table 6.4.1-3: Method M 3466 - Summary of method accuracy and precision (method validation)

Commodity	Fortification level (mg/kg)	Recovery (%)	Recovery range (%)	Mean recovery (%)	SD (%)	RSD (%)
Hen egg	0.01	119 / 102 / 117 / 106 / 106	102-119	110	8	7
	0.1	96 / 97 / 95 / 74 / 98	74-98	92	10	11
Overall (n=10)			**74-119**	**101**	**13**	**13**

Table 6.4.1-4: Method M 3466 - Concurrent recoveries from egg sample analysis, extract and freezer stability analyses

Fortification level (mg/kg)	Recovery range (%)	Mean recovery (%)
0.01	71-119	101 (n=15)
0.1	83-116	97 (n=15)
Overall	**71-119**	**99 (n=30)**

Recovery findings:
The method was validated at two fortification levels (0.01 and 0.1 mg/kg) in hen eggs. For each fortification level, five replicates were analyzed. Additionally, two replicates of unfortified samples were examined. It was proven that the method M 3466 is suitable to determine residues of BAS 310 I in hen eggs´samples. As shown in Table 6.4.1-1, the mean recovery values were found to be within the acceptable range of 70-120% for all methods tested. The overall relative standard deviations (RSD, %) for all fortification levels were below 20%.

Linearity:
Good linearity was demontrated with r=0.9997.

Specificity:
Specificity was demonstrated by the absence of interferences in the control samples.

Limit of Quantitation:
The limit of quantitation (LOQ) was defined by the lowest fortification level successfully tested. LOQ is 0.01 mg/kg for BAS 310 I.

Repeatability:
The overall relative standard deviations (RSD, %) for all fortification levels were below 20%. The detailed values are shown in Table 6.4.1-1.

Reproducibility:
Reproducibility of the method was not determined within this validation study.

II. RESULTS AND DISCUSSION

Observation:
Actual BAS 310 I doses to the hens in this study averaged approximately 100, 132, 119, 124, 140, and 149 percent of the intended daily rates/hen in treatment Groups A-F, respectively. There were no significant differences observed in average animal weights, egg production (total egg weights or numbers), feed intake or general health across treatment groups during the in-life phase. There were no treatment-related side effects noted. A summary is shown in Table 6.4.1-1.

Table 6.4.1-5: Actual Average Daily BAS 310 I Doses/Hen

Group	No. of animals	Average hen weight per group (kg)[b, d]	µg a.s. /hen/day[a]	Average daily feed intake /hen (g dry matter)	Average mg a.s./kg bw/day[b, d]	Average mg/kg BAS 310 I in daily diet (µg a.s. per gram dry feed/day)[c, d, e]
A	12	1.54	0	101	0	0
B	12	1.53	151	94	0.0987	1.6
C	12	1.61	757	105	0.470	7.2
D	12	1.56	1506	99	0.965	15
E	4	1.58	1506	90	0.953	17
F	4	1.41	1506	86	1.07	18

Small discrepancies in dose estimates due to significant figures.

a Based on nominal values as calculated by the analyst who prepared working capsule dosing solutions. Values are adjusted for 96.1% potency of the test substance. Not adjusted for dose verification assay results.

b Based upon the average of pre-treatment and at sacrifice weights across the entire treatment group.

c Based on average daily feed consumption (adjusted for 90% feed dry matter) across the 28 day treatment period of 112, 104, 117, 110, 100 and 95 grams dry matter/hen/day for groups A-F respectively.

d There were no significant differences noted across treatment groups for average total egg production/hen, average total egg weight/hen, average dry matter consumption/hen or average general heath/hen during the course of this study. Average and standard deviation of pre-treatment and at sacrifice hen weights across all treatment groups was approximately 1.54 ± 0.07 kg.

e Actual average daily doses administered to study AP00PT06 were 100, 132, 119, 124, 140, and 149 percent of intended on a ppm (mg/kg feed) basis.

No residues of alpha-cypermethrin above the LOQ (0.01 mg/kg) were detected in eggs from treated hens in the 1.2 mg/kg group. In the 6.1 mg/kg dose group, residues slightly above the LOQ were found from day 9 until the end of the dosing period (0.0101-0.0125 mg/kg between day 9-28). BAS 310 I residues in eggs from treated hens in the high dose group reached a plateau averaging approximately 0.027 mg/kg within approximately 21-24 days of dosing. There were no reportable residues after 7 days' depuration. Apparent BAS 310 I residues in eggs from untreated animals were <0.002 mg/kg.
A summary of egg results is shown in Table 6.4.1-2.

Table 6.4.1-6: Summary of BAS 310 I Residues Found in Eggs in Study AP00PT06

Dose Day	Group A: Control BAS 310 I mg/kg		Group B: 1.21 mg/kg feed dose BAS 310 I mg/kg		Group C: 6.05 mg/kg feed dose BAS 310 I mg/kg		Group D: 12.1 mg/kg feed dose BAS 310 I mg/kg		Group E: 12.1 mg/kg feed dose (Depuration) BAS 310 I mg/kg		Group F: 12.1 mg/kg feed dose (Depuration) BAS 310 I mg/kg	
	Sample #		Sample #		Sample #		Sample #		Sample #		Sample #	
	0101	<0.002	1101	<0.010	2101	<0.010	3101	<0.010	4101	<0.010	5101	<0.010
-1	0102	<0.002	1102	<0.010	2102	<0.010	3102	<0.010				
	0103	<0.002	1103	<0.010	2103	<0.010	3103	<0.010				
	Average	<0.002		<0.010		<0.010		<0.010				
	0104	<0.002	1104	<0.010	2104	<0.010	3104	<0.010	4102	<0.010	5102	<0.010
1	0105	<0.002	1105	<0.010	2105	<0.010	3105	<0.010				
	0106	<0.002	1106	<0.010	2106	<0.010	3106	<0.010				
	Average	<0.002		<0.010		<0.010		<0.010				
	0107	<0.002	1107	<0.010	2107	<0.010	3107	<0.010	4103	<0.010	5103	<0.010
3	0108	<0.002	1108	<0.010	2108	<0.010	3108	<0.010				
	0109	<0.002	1109	<0.010	2109	<0.010	3109	<0.010				
	Average	<0.002		<0.010		<0.010		<0.010				
	0110	<0.002	1110	<0.010	2110	<0.010	3110	0.0163	4104	0.0122	5104	0.0133
6	0111	<0.002	1111	<0.010	2111	<0.010	3111	0.0232				
	0112	<0.002	1112	<0.010	2112	<0.010	3112	0.0140				
	Average	<0.002		<0.010		<0.010		0.0178				
	0113	<0.002	1113	<0.010	2113	<0.010	3113	0.0173	4105	<0.010	5105	0.0135
9	0114	<0.002	1114	<0.010	2114	0.0113	3114	0.0214				
	0115	<0.002	1115	<0.010	2115	<0.010	3115	0.0224				
	Average	<0.002		<0.010		<0.010		0.0204				
	0116	<0.002	1116	<0.010	2116	<0.010	3116	0.0176	4106	<0.010	5106	0.0277
12	0117	<0.002	1117	<0.010	2117	0.0105	3117	0.0184				
	0118	<0.002	1118	<0.010	2118	<0.010	3118	0.0197				
	Average	<0.002		<0.010		<0.010		0.0186				
	0119	<0.002	1119	<0.010	2119	<0.010	3119	0.0221	4107	0.0122	5107	0.0271
15	0120	<0.002	1120	<0.010	2120	0.0107	3120	0.0207				
	0121	<0.002	1121	<0.010	2121	<0.010	3121	0.0226				
	Average	<0.002		<0.010		<0.010		0.0218				
	0122	<0.002	1122	<0.010	2122	<0.010	3122	0.0145	4108	0.0107	5108	0.0241
18	0123	<0.002	1123	<0.010	2123	0.0109	3123	0.0206				
	0124	<0.002	1124	<0.010	2124	0.0101	3124	0.0234				
	Average	<0.002		<0.010		<0.010		0.0195				
	0125	<0.002	1125		2125	<0.010	3125	0.0213	4109	0.0199	5109	0.0242
21	0126	<0.002	1126		2126	<0.010	3126	0.0272				
	0127	<0.002	1127		2127	<0.010	3127	0.0208				
	Average	<0.002				<0.010		0.0231				
	0128	<0.002	1128	<0.010	2128	<0.010	3128	0.0215	4110	0.0229	5110	0.0210
24	0129	<0.002	1129	<0.010	2129	<0.010	3129	0.0353				
	0130	<0.002	1130	<0.010	2130	<0.010	3130	0.0227				
	Average	<0.002		<0.010		<0.010		0.0265				
	0131	<0.002	1131		2131	0.0106	3131	0.0251	4111	0.0186	5111	0.0224
28	0132	<0.002	1132		2132	0.0108	3132	0.0281				
	0133	<0.002	1133		2133	0.0125	3133	0.0214				
	Average	<0.002				0.0113		0.0249	4112	<0.010	5112	<0.010

Notes:
Dosing was stopped on day 28. Sample 4112 and 5112 were collected on day 35 and 42, respectively.

Protocol section 11.4 directed the analyst to analyze egg samples from the highest treatment group first. If there were no quantifiable residues, samples from lower treatment groups did not need to be analyzed. Blanks indicate egg samples not analyzed. Refer to protocol deviation #4 regarding samples 1131, 1132 and 1133

BAS 310 I residues did not accumulate in laying hen liver or muscle after 28 daily doses. Residues were <0.05 mg/kg in liver and muscle tissue sampled from the high dose animals. BAS 310 I residues in laying hen abdominal fat averaged approximately 0.235 mg/kg after 28 days of dosing at the high rate and 0.085 mg/kg at the medium rate. No residues above the LOQ (0.05 mg/kg) were detected in fat tissue from the low dose group. A residue decline in abdominal fat of approximately 61% was noted in the high dose group after 7-14 days' depuration. BAS 3101 was stable in refrigerated muscle, liver and abdominal fat extracts for at least 7, 8 and 5 days, respectively. There was no need to collect storage stability data in laying hen tissues, since samples were analyzed within 30 days of collection. Apparent BAS 310 I residues in edible tissues from untreated animals were <0.01 mg/kg.
A summary of edible tissue results is shown in Table 6.4.1-3.

Table 6.4.1-7: Summary of BAS 310 I Residues Found in Edible Tissues in Study AP00PT06

Tissue	Group A: Control BAS 310 I mg/kg		Group B: 1.21 mg/kg feed dose BAS 310 I mg/kg		Group C: 6.05 mg/kg feed dose BAS 310 I mg/kg		Group D: 12.1 mg/kg feed dose BAS 310 I mg/kg		Group E: 12.1 mg/kg feed dose (Depuration)[1] BAS 310 I mg/kg		Group F: 12.1 mg/kg feed dose (Depuration)[2] BAS 310 I mg/kg	
	Sample#		Sample #		Sample #		Sample #		Sample #		Sample #	
	0134	<0.01	1134	<0.05	2134	0.0856	3134	0.205	4113	0.0875	5113	0.092
Fat	0135	<0.01	1135	<0.05	2135	0.0877	3135	0.260				
	0136	<0.01	1136	<0.05	2136	0.0819	3136	0.240				
	Average	<0.01		<0.05		0.0851		0.235				
	0137	<0.01	1137	NA	2137	NA	3137	<0.05	4114	NA	5114	NA
Muscle	0138	<0.01	1138	NA	2138	NA	3138	<0.05				
	0139	<0.01	1139	NA	2139	NA	3139	<0.05				
	Average	<0.01						<0.05				
	0140	<0.01	1140	NA	2140	NA	3140	<0.05	4115	NA	5115	NA
Liver	0141	<0.01	1141	NA	2141	NA	3141	<0.05				
	0142	<0.01	1142	NA	2142	NA	3142	<0.05				
	Average	<0.01						<0.05				

Method LOQ 0.05 mg/kg all matrices.

[1] Tissues collected on 7DAT28.

[2] Tissues collected on 14DAT28.

Protocol section 11.4 directed the analyst to analyze tissue samples from the highest treatment group first. If there were no quantifiable residues, samples from lower treatment groups did not need to be analyzed. NA indicates tissue samples not analyzed.

Extract stability:
Final extracts from method validation were used for extract stability study. BAS 310 I was stable in egg extracts for at least 7 days.
The results are summarized in Table 6.4.1-1.

Table 6.4.1-8: Summary of Stability of BAS 310 I Residues in Extract of Hen Egg:

	Fortification Level (mg/kg)	Recovery (%)					Mean Recovery (%)	SD (%)	RSD (%)
fresh extract	0.01	119	102	117	106	106	110	8	7
extract stored for 7 days	0.01	123	100	112	105	110	110	9	8
fresh extract	0.1	96	97	95	74	98	92	10	11
extract stored for 7 days	0.1	109	98	97	75	99	96	12	13
fresh extract	overall						101	13	13
extract stored for 7 days	overall						103	13	13

Freezer Storage Stability:
BAS 310 I was stable in frozen egg for at least 45 days.
Freezer storage stability data in hen eggs are summarized in Table 6.4.1-2.

Table 6.4.1-9: Summary of the Freezer Storage Stability Data of BAS 310 I Residues in Hen Egg

Sample Number	Interval (day)	mg/kg found	recovery (%)	corrected Rec. (%)
AP00PT06-001	30	<0.0001	--	--
AP00PT06-002	30	0.0866	87	--
AP00PT06-003	30	0.0877	88	101
AP00PT06-004	30	0.0816	82	94
AP00PT06-009	45	<0.00005	--	--
AP00PT06-010	45	0.0894	89	--
AP00PT06-011	45	0.0790	79	89
AP00PT06-012	45	0.0841	84	94

III. CONCLUSION

No residues of alpha-cypermethrin above the LOQ (0.01 mg/kg) were detected in eggs from treated hens in the 1.2 mg/kg group. In the 6.1 mg/kg dose group, residues slightly above the LOQ were found from day 9 until the end of the dosing period (0.0101-0.0125 mg/kg between day 9-28).
Alpha-cypermethrin residues in eggs from treated hens in the high dose group reached a plateau averaging approximately 27 mg/kg within approximately 21-24 days of dosing. There were no reportable residues after 7 days' depuration.
Alpha-cypermethrin residues did not accumulate in laying hen liver or muscle after 28 daily doses. Residues were <0.05 mg/kg in liver and muscle tissue sampled from the high dose animals. Alpha-cypermethrin residues in laying hen abdominal fat averaged approximately 0.235 mg/kg after 28 days of dosing at the high rate and 0.085 mg/kg at the medium rate. No residues above the LOQ (0.05 mg/kg) were detected in fat tissue from the low dose group. A residue decline in abdominal fat of approximately 61% was noted in the high dose group after 7-14 days' depuration.

CA 6.4.2 Ruminants

During the peer review under Directive 91/414/EEC, four studies on the magnitude of alpha-cypermethrin residues in ruminants were submitted. Three of these studies (Al-870-020, Al-870-021 and Al-870-022) were related to a dermal exposure and were assessed as being out of interest.

One feeding study with lactating cows (AL-705-006) was peer-reviewed. In this study, 3 groups of 3 dairy cows were fed with oral doses of alpha-cypermethrin incorporated in capsules for 28 days at dosages of 77, 231 and 769 mg alphacypermethrin/cow/day (corresponding to about 3.9, 11.6 and 38.5 mg/kg in diet/day). Milk samples were collected for determination of alpha-cypermethrin residues on study days –1, 1, 2, 3, 6, 8, 10, 13, 15, 17, 20, 22, 24, and 27. Animals were sacrificed within 24 hours after the final dose. Cattle muscle, liver, kidney, and fat samples were taken for alpha-cypermethrin analysis.

At the lowest feeding level, residues of alphacypermethrin were present only in fat (0.058-0.064 mg/kg). All samples of whole milk from the 1 x dose rate group were below the limit of determination of the analytical method (0.010 mg/kg) for alpha-cypermethrin.

Alpha-cypermethrin residues did not exceed 0.02 mg/kg in the milk of dairy cattle exposed to 231 mg alpha-cypermethrin/animal/day. Alpha-cypermethrin residues in muscle, liver, and kidney were less than the limit of determination of the analytical method (0.05 mg/kg); the average alpha-cypermethrin residue found in the fat of dairy cattle fed at this level was approximately 0.16 mg/kg.

Alpha-cypermethrin residues did not exceed 0.08 mg/kg in the milk of dairy cattle exposed to 769 mg alpha-cypermethrin/animal/day. Alpha-cypermethrin residues in muscle, liver, and kidney were less than the limit of determination of the analytical method (0.05 mg/kg); the average alpha-cypermethrin residue found in the fat of dairy cattle fed at this level was approximately 0.87 mg/kg.

CA 6.4.3 Pigs

No feeding studies in pigs were performed.

CA 6.4.4 Fish

No feeding studies in fish were performed.

CA 6.5 Effects of Processing

In the framework of the original inclusion into Annex I according to Directive 91/414/EEC, the effect of processing on alpha-cypermethrin residues in barley and oilseed rape as well as cabbage and plum had been submitted.
Additional processing studies in barley and gherkins have been performed. These new studies will be summarized in the following. In addition, a published study from open peer-reviewed literature on the effect of processing in tomato is included.

An overview on the processing studies is presented in Table 6.5-1.

Table 6.5-1: Overview on processing studies

Crop / Test system	DocID / Reference	EU reviewed
Fruit (plum), vegetable(cabbage)	CY-790-060	Yes
Barley	AL-730-061 1993/7002041	Yes
Oilseed rape	AL-750-038	Yes
Oilseed rape	AL-750-042 2002/1004088	Yes
Processing hydrolysis	AL-790-046	Yes
Tomato	2005/1043620 Lin H.-M.et al., 2004a[1]	No
Barley	2007/1013068	No
Gherkins	2007-1011647	No

1 peer-reviewed open literature

CA 6.5.1 Nature of the residue

The effect of processing on the nature of alpha-cypermethrin residues was investigated during the EU Review. Studies were conducted using three test conditions: 20 minutes at 90°C and pH 4, simulating pasteurization; 60 minutes at 100°C and pH 5, simulating baking, brewing and boiling; and 20 minutes at 120°C, simulating sterilization.
Alpha-cypermethrin was found to be hydrolytically stable under the simulated processing conditions at pH 4 and 5 at 90°C and 100°C. However, alphacypermethrin was found to be susceptible to hydrolysis at the ester linkage at simulated processing conditions at pH 6 at 120°C resulting in the breakdown of the ester linkage to 3-phenoxybenzaldehyde (M310I018, CL 206969) and dichlorovinydimethylcarboxylic acid (DCVA, M310I001, CL 912554) as major degradates exceeding 10% of the applied dose. The major component of the residue was unchanged parent (83% - 90%).

Data on the fate of cypermethrin residues and formation of 3-phenoxybenzaldehyde during processing of tomatoes are available in peer reviewed open literature and submitted in the context of this dossier. A summary is given below.

Report: CA 6.5.1/1
Lin H.-M. et al., 2004a
Stability of the insecticide Cypermethrin during tomato processing and implications for endocrine activity
2005/1043620
Guidelines: none
GLP: no

Executive Summary

The thermal and pH stabilities of cypermethrin during food processing were investigated using tomato as a model food system and high-performance liquid chromatography as the analytical method. Cypermethrin was thermally unstable in aqueous conditions, where the hydrolysis of the pesticide was accelerated by heat. The mean proportion remaining after heating cypermethrin in water for 10 min was 66%, falling to 27% after 1 h. Similarly, thermal processing of canned tomatoes caused cypermethrin to degrade, with remaining levels in the final product ranging from 30 to 60% of the original. Cypermethrin was unstable at extreme pHs, with acid hydrolysis occurring faster than alkaline hydrolysis in phosphate buffers. The acidity of tomato paste (pH 4.3) caused cypermethrin levels to decrease by 30% within 12 days at 5°C. The studies indicate that cypermethrin residues are likely to degrade by hydrolysis during food processing, thus reducing the exposure of consumers to cypermethrin. 3-Phenoxybenzaldehyde, a hydrolysis breakdown product of cypermethrin, was detected in the tomato paste and from the heating of cypermethrin in water at 100°C.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material:

Description: Cypermethrin
Lot/Batch #: com. source, not stated
Purity: 96.8%
CAS#: 52315-07-8
Development code: not applicable
Spiking levels: 0.5, 1.5, 2 and 3.5 µg ml^{-1}

2. Test Commodity:

Crop: Tomato
Type: Fruiting vegetables, solanacea
Variety: fruit: Roma, paste and canned tomatoes: not stated
Botanical name: *Lycopersicon esculentum*
Crop parts(s) or processed commodity: fruit, paste, canned tomatoes
Sample size: not stated

B. STUDY DESIGN

1. Test procedure

Thermal stability of cypermethrin in aqueous conditions:
A 1.5 µg ml^{-1} cypermethrin aqueous solution was prepared by diluting required amounts of a cypermethrin standard solution with de-ionized water, where the percentage of methanol in the aqueous solution was not more than 2% (water solubility of cypermethrin is 9 $x10^{-3}$ mg l^{-1}). A total of 25 ml aqueous solution was poured into a 50ml capped Schott bottle, which was then placed in a steam bath (100°C). The bottle was allowed to equilibrate for 8 min before duplicate samples of 1 ml each were taken every 10 min for 1 h. The samples were analysed by HPLC, together with standard solutions of the 3-phenoxybenzyl compounds and an aqueous standard of 3-phenoxybenzaldehyde (prepared by diluting the standard solution with de-ionized water).

pH stability of cypermethrin in aqueous conditions:
A total of 2 µg ml^{-1} cypermethrin pH solutions were prepared by diluting required amounts of a cypermethrin standard solution with phosphate buffers of various pH. Samples of the solutions were measured by HPLC within 8 h of preparation, together with a 2 µg ml^{-1} cypermethrin aqueous solution. The solutions were analysed again after 12 days of storage in the dark at 5°C. The pH solutions, except for pH 7, were always adjusted to the pH range 6.5–7.5 by dropwise additions of concentrated hydrochloric acid or NaOH, before being subjected to HPLC analysis.

Effects of tomato paste acidity on stability of cypermethrin during tomato processing:
A total of 60 g commercial tomato paste (22–24% tomato solids) was fortified with 2.4 ml cypermethrin standard solution to obtain a spike concentration of 3.5 µg g^{-1}. The concentration of cypermethrin in the paste was determined before and after 12 days storage in the dark at 5°C.

Effects of thermal processing on stability of cypermethrin during tomato processing:
Thermal processing of canned peeled tomatoes was simulated by autoclaving unsealed cans of peeled tomatoes. The cans were emptied cans of whole peeled tomatoes (400 g), and were cleaned by boiling in water for 1 h, followed by drying in an oven for approximately 10 min. Peeled tomatoes were prepared by hand-peeling tomatoes, of the Roma variety, that were placed in boiling water for 1 min followed by immersion in cold water. The cans were filled with the peeled tomatoes and tomato juice that was liberated from tomatoes using a food mill. The filled cans were spiked with 0.2 ml cypermethrin standard solution to obtain a spike concentration of 0.5 µg g^{-1}, and with lids pushed shut they were autoclaved in a 40 litre Autoclave for 20 min at 110°C. A 50 ml Schott bottle containing 30ml 5 µg ml^{-1} cypermethrin aqueous solution was included in the autoclave. The autoclave was allowed to depressurize for approximately 5 h before the cans were removed.

Table 6.5.1-1: Conditions for thermal and pH stability study of cypermethrin

Processing represented	Matrix	pH	Temperature (°C)	Time
Model system aqueous stability	Water/≤2% methanol	Not stated	100	68 min
Model system pH stability	Phosphate buffer	2	5	12 days
	Phosphate buffer	4	5	12 days
	Phosphate buffer	7	5	12 days
	Phosphate buffer	10	5	12 days
Effect of tomato paste acidity	Tomato paste	4.3	5	12 days
Effect of thermal processing	Tomatoes, peeled and canned	Not stated	110	20 min
	Aqueous	Not stated	110	20 min

2. Description of analytical procedures

Analysis of cypermethrin aqueous and pH solutions was carried out by HPLC with diode array detection at 225 nm (UV). Separation was achieved on a SPHERI-5 octadecylsilane column, 220x4.6mm i.d., particle size 5 mm, with the mobile phase consisting of 70/30 (v/v) acetonitrile/water with a flow rate of 2ml min^{-1}. Sample injection volume was 50 µl. The limit of detection for cypermethrin in the aqueous solutions was 0.5 µg ml^{-1}. Recoveries were not reported for this method.

Tomato paste and autoclaved tomatoes were homogenized with acetonitrile, filtered and salted out with sodium chloride. The aqueous phase was discarded and an aliquot of the remaining acetonitrile was mixed with de-ionized water, followed by clean-up on a C18 SPE cartridge and a subsequent clean-up on a Florisil cartridge. The solvent of the eluate was evaporated , the residue redissolved in methanol and filtered. The determination of cypermethrin levels in processed tomatoes was carried out by HPLC with diode array detection at 225 nm (UV), as described for the aqueous and pH solutions. Recoveries and limit of quantitation were not reported for this method.

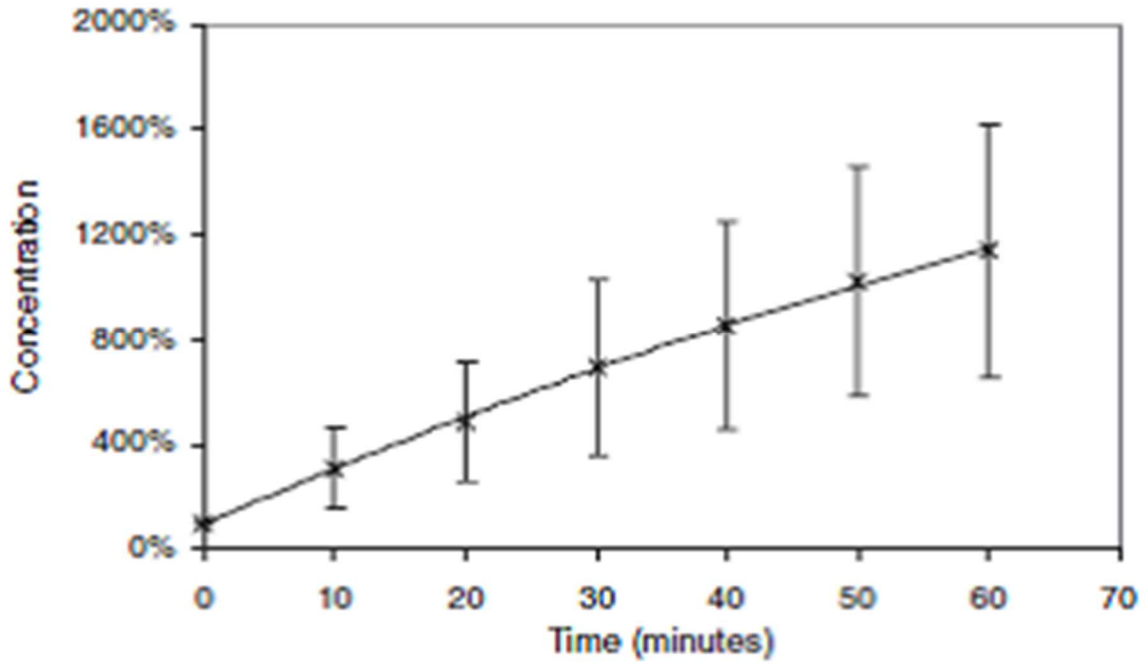
II. RESULTS AND DISCUSSION

Thermal stability of cypermethrin in aqueous conditions

Cypermethrin was thermally unstable in aqueous conditions, where the hydrolysis of the pesticide was accelerated by heat. The mean proportion remaining after heating cypermethrin in water at 100°C for 10 min was 66%, falling to 27% after 1 h.

Two breakdown products, compounds A and B, were observed in the HPLC chromatograms of cypermethrin. Compound B appeared only in some chromatograms of cypermethrin after 1 h of heating, while compound A was already present at a low level in cypermethrin aqueous solutions before heating, and its concentration increased over time during heating. The increase in the concentration of compound A during 60 minutes is shown in Figure 6.5.1-1.

Figure 6.5.1-1: Increase in the concentration of compound A from heating 1.5 µg ml^{-1} cypermethrin aqueous solution at 100°C.



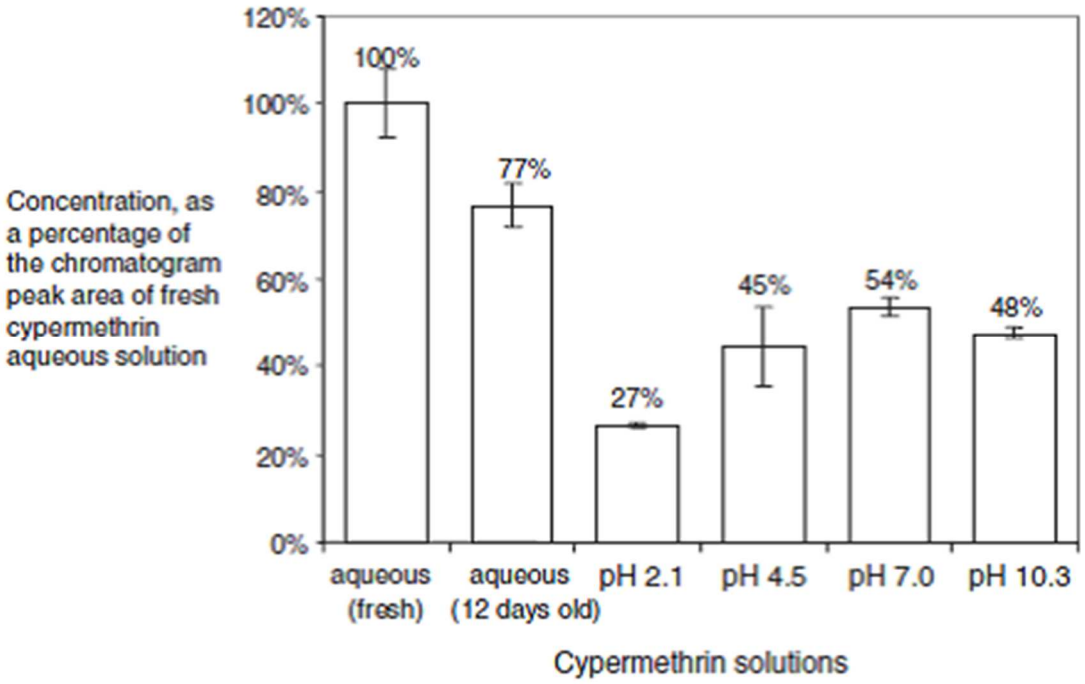
Error bars represent the standard deviation of the data from three experiments. (from Lin H.-M- *et al*., Food Additives & Contaminants: Part A, 22: 1, 15 - 22)

Compound A was identified as 3-phenoxybenzaldehyde, based on retention time and UV spectra comparisons with a 3-phenoxybenzaldehyde standard. The retention time of compound B did not match 3-phenoxybenzoic acid or 3-phenoxbenzyl alcohol.

pH stability of cypermethrin in aqueous conditions

The concentration of cypermethrin decreased by at least 46% at all pH after 12 days at 5°C (Fig. Figure 6.5.1-2 with the greatest decline at pH 2, followed by pH 4, pH 10 then pH 7. Therefore, acid hydrolysis of cypermethrin occurred to a greater extent than alkaline hydrolysis.
Phosphate appeared to influence the pH stability of cypermethrin because the decline in the concentration of cypermethrin after 12 days was higher in the phosphate buffer solutions compared with the aqueous solution. No 3-phenoxybenzaldehyde was found in the pH 4.5 phosphate buffer; no data on 3-phenoxybenzaldehyde were given for the other buffer solutions. The concentration of cypermethrin found after 12 days in buffered solutions at different pHs is shown in Figure 6.5.1-2.

Figure 6.5.1-2: Concentration of cypermethrin pH solutions after 12 days at 5°C.



Error bars represent the standard deviation of the data from three experiments three replicate solutions, except for the error bar for the fresh aqueous solution, which represents the standard deviation of duplicate HPLC measurements. (from Lin H.-M- *et al*., Food Additives & Contaminants: Part A, 22: 1, 15 - 22)

Stability of cypermethrin during tomato processing

The concentration of cypermethrin remaining in tomato paste of pH 4.3, after 12 days at 5°C, was 70% of the initial concentration, while in a pH 4.5 phosphate buffer after 12 days at 5°C only 45±9% of the initial concentration were found. Although the decrease in the concentration of cypermethrin in tomato paste was less than that of the aqueous model system, the breakdown product 3-phenoxybenzaldehyde was detected, according to its UV spectrum and retention time. The peak was significantly larger than that found in control tomato paste freshly spiked with cypermethrin. Table 6.5.1-2 shows the concentration of cypermethrin in tomato paste after 12 days at 5°C.

Table 6.5.1-2: Concentration of cypermethrin in tomato paste after 12 days at 5°C

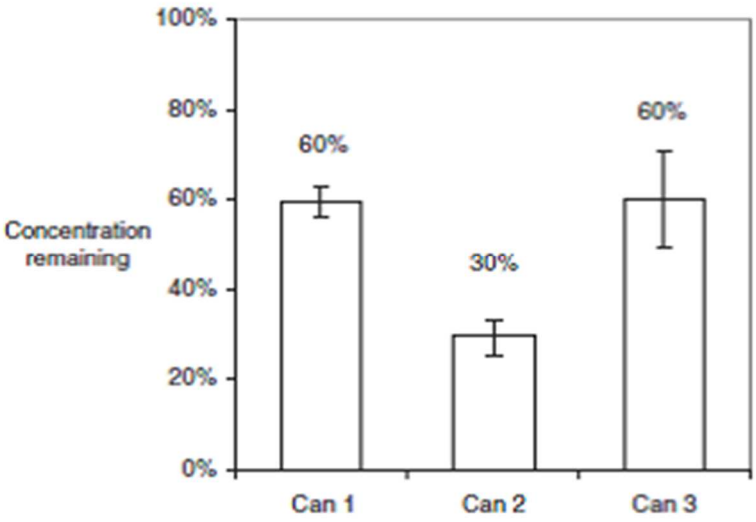
	Concentration (µ g^{-1})	Number of sample determinations
Before 12 days	3.5±0.2	3
After 12 days	2.4±0.2	2
Difference (%)	30±6	

Initial spike level 3.5 µg g^{-1}. The concentration after 12 days was significantly lower than the initial concentration; P=0.0037, one tail t-test, confidence level of 95%.

Effects of thermal processing

Peeled tomatoes were spiked with cypermethrin at a spike level of 0.5 µg g^{-1}. A significant loss of cypermethrin was observed in three cans autoclaved simultaneously, where remaining concentrations ranged from 30 to 60%. A 5 µg ml^{-1} cypermethrin aqueous solution that was autoclaved together with the cans was completely degraded (limit of detection for cypermethrin in the solution was 0.5 µg ml^{-1}). Chromatogram peaks matching the retention time of 3-phenoxybenzaldehyde were detected at very low levels in the autoclaved tomatoes. 3-Phenoxybenzaldehyde was detected in the aqueous solution, indicating that the aldehyde did not decompose under the high temperatures. The cypermethrin levels after autoclaving cans of peeled tomatoes at 110°C for 20 minutes are shown in Figure 6.5.1-3.

Figure 6.5.1-3: Cypermethrin levels after autoclaving cans of peeled tomatoes



Initial spike level 0.5 µg g^{-1}. Error bars for cans 1 and 3 represent the standard deviation of the data from three replicate sample determinations. The error bar for can 2 is the standard deviation of the data from duplicate determinations because an outlier concentration of <0.01 µg g^{-1} was excluded. (from Lin H.-M- *et al.*, Food Additives & Contaminants: Part A, 22: 1, 15 - 22)

III. CONCLUSION

The degradation rates of cypermethrin in the aqueous model systems differ from those of the tomato model system, where the hydrolysis rates of cypermethrin in tomato paste and during thermal processing were slower than that of the aqueous model systems. The results indicate that one major degradation product is 3-phenoxybenzaldehyde, which has a much lower acute oral toxicity in rats than the parent compound cypermethrin.

CA 6.5.2 Distribution of the residue in inedible peel and pulp

Not relevant for the intended uses in cucumber, leafy cabbage, lettuce, oilseed rape and cereals.

CA 6.5.3 Magnitude of residues in processed commodities

Report: CA 6.5.3/1
Schulz H., 2007c
Study on the residue behaviour of Alpha-Cypermethrin in gherkins and processed products after treatment with BAS 310 41 I under greenhouse conditions in Germany and The Netherlands, 2004
2007/1011647

Guidelines: EEC 91/414 Annex II (Part A Section 6), EEC 91/414 Annex III (Part A Section 8), EEC 7029/VI/95 rev. 5, EEC 7525/VI/95 rev. 7, EEC 1607/VI/97 rev. 2 10.06.1999, EEC 7035/VI/95 rev. 5

GLP: yes
(certified by Hessisches Ministerium fuer Umwelt, Laendlichen Raum und Verbraucherschutz)

Executive Summary
During the 2004 growing season, four greenhouse trials in gherkins were conducted in Germany and the Netherlands in order to determine the magnitude and distribution of alpha-cypermethrin residues in a number of intermediate and end products after processing into canned gherkins. Gherkin plants were foliar sprayed twice with a soluble concentrate formulation of alpha-cypermethrin (code: BAS 310 41 I) at an exaggerated target rate of 0.075 kg alpha-cypermethrin/ha. The applications were made 14(±1) and 7(±1) days before harvest. The spray volume used was 400 l/ha. Gherkin fruit samples were collected immediately after the second application at growth stages between 69-87 and 6-8 days thereafter at growth stages between 71-89. The specimen were processed into canned gherkins and the intermediate fractions washed gherkins, wash water and vegetable stock using using simulated household and industrial processing procedures.

At 0 days after the last application, the alpha-cypermethrin residues in gherkin fruit were between 0.012 and 0.066 mg/kg. In the gherkins taken for processing 6-8 days after the last application, these residues declined to 0.015 - 0.038 mg/kg. In wash water, washed gherkins and vegetable stock, the transfer factors were below 1. In two out of four canned gherkin specimens, a slight increase (transfer factors 1.33 and 1.07, respectively) of the alpha-cypermethrin residues was observed.

I. MATERIAL AND METHODS

A. MATERIALS

1.

Test Material:	Alpha-cypermethrin BAS 31041I
Description:	BAS 310 41 I (SC)
Lot/Batch #:	4000, Alpha-cypermethrin: 100 g/L
Purity:	not reported
CAS#:	67375-30-8
Development code:	not reported
Spiking levels:	0.01,0.1 and 1.0 mg/kg

2. Test Commodity:

Crop:	Gherkin
Type:	Cucurbits edible peel
Variety:	Harmonie, Serena F1
Botanical name:	*Cucumis sativus*
Crop parts(s) or processed commodity:	Gherkins unwashed and washed, wash water, canned gherkins, vegetable stock
Sample size:	0 DALA: 12 fruits (min. 0.5 kg) 7±1 DALA: 12 fruits (min. 1.0 kg) 7±1 DALA: 2-10 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

Field phase

During the 2004 growing season, four greenhouse trials in gherkins were conducted in Germany (Brandenburg; ACK/01/04 and ACK/25/04) and the Netherlands (Limburg; AGR/01/04 and AGR/02/04) in order to determine the magnitude and distribution of alpha-cypermethrin residues in a number of intermediate and end products after processing (washed gherkins, wash water, canned gherkins, vegetable stock).

Two of the trials consisted of two plots, one untreated and one treated (ACK/01/04 and AGR/01/04); the other two (ACK/25/04 and AGR/02/04) consisted of one treated plot each.

Gherkin plants (varieties: Serena F1 in the German trials and Harmonie in the Dutch trials) were foliar sprayed twice with a soluble concentrate formulation of alpha-cypermethrin (code: BAS 310 41 I) at an exaggerated target rate of 0.075 kg alpha-cypermethrin/ha. Actual rates were within +/10 % of the nominal rate. This exaggerated treatment rate was used in an effort to generate finite alpha-cypermethrin residues in gherkins so that the distribution of the residue in the processed fractions could be better quantified.

The applications were made 14(±1) and 7(±1) days before harvest. The spray volume used was 400 I/ha. Gherkin fruit samples were collected immediately after the second application at growth stages between 69-87 and 6-8 days thereafter at growth stages between 71-89. All specimens were frozen within 24 hours of being taken, and remained frozen at or below -18°C, including during transportation, until analysis. The maximum storage interval from sampling until extraction was 278 days.

Target application rates and timings for gherkins

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2004	4	2	G	BAS 310 41 I (SC)	alpha-cypermethrin	0.075	400	14±1 and 7 ±1 days before harvest

Processing phase

Sorting
Damaged and rotten gherkins were sorted out by hand and subsequently discarded. Only healthy gherkins were used for processing.

Washing
A quantity of gherkins between 2 - 10 kg from each field trial was cleaned with tap water (20° C) using a brush. For that, the gherkins of every plot were divided into 3 batches. For the washing of each batch, about 3 L of tap water were used, out of which 1 L was put aside after the washing of each batch. 1 L of fresh water was used for the rinsing of the cleaned gherkins in a sieve. This rinsing water was added to the remaining 2 L of water for the washing of the next batch of gherkins. This procedure was repeated for the 3 portions of each gherkin specimen. The wash water and the rinsing water of each specimen were combined and indicated as "wash water". This procedure resulted in approx. 6 L of wash water used for the washing of the gherkin specimens.
Specimens of wash water for residue analysis were taken, filled into 1 L PE bottles with screw caps and stored deep-frozen. Also specimens of washed gherkins for residue analysis were taken, filled into PE bags and stored deep-frozen. The remaining washed gherkins were used for the processing to canned gherkins.

Canning
The washed gherkins were filled into 1 L jars (approx. 450 g per jar). Throughout the study, the term "canned gherkins" is used, although gherkins were filled into jars.
A hot solution of tap water (3.75 kg), table salt (150 g), sugar (200 g) and vinegar (1000 g, 10 %) was prepared, which was used to fill the jars to the brim (approx. 450 g) at a temperature of about 85°C. The jars were steamed at the top for 15 seconds (head space vaporization) and then immediately closed by a lid. The subsequent pasteurization was carried out by heating the jars to 90°C and keeping them at that temperature for 20 minutes. They were then allowed to cool down in an autoclave to 75 °C, which took about 13 to 27 minutes, after which the jars were placed in a water bath and allowed to cool down to ambient temperature (35 to 90 minutes).
The jars were then opened and the gherkins were separated from the vegetable stock using a sieve. A specimen each of the canned gherkins and the vegetable stock was taken for residue analysis and stored deep-frozen.

2. Description of analytical procedures

The specimens were analysed for residues of alpha-cypermethrin using BASF method No. 567/0. The limit of quantification (LOQ) of the method was 0.01 mg/kg.
Principle of the method:
BAS 310 I (alpha-cypermethrin) is extracted with a 70:25:5 methanol/water/hydrochloric acid mixture. After centrifugation, an aliquot of the extract is partitioned into cyclohexane. Silica gel SPE column is used for further purification, if required. Final determination is performed by LC/MS/MS.

Table 6.5.3-1: Procedural recoveries

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
gherkin fruit, wash water, vegetable stock, washed gherkins, canned gherkins	0.01-1.0	15	91.1	10.3	11.3

II. RESULTS AND DISCUSSION

Directly after application, the alpha-cypermethrin residues in the gherkin specimens ranged from 0.012 to 0.066 mg/kg and declined to 0.015 to 0.038 mg/kg in the gherkins taken for processing, at 6 - 8 days after the last application. In washed gherkins, a reduction of alpha-cypermethrin residues was observed, with levels ranging between 0.009-0.013 mg/kg (mean transfer factor 0.60), while in wash water, residues of alpha-cypermethrin between <0.01-0.016 mg/kg were detected (mean transfer factor 0.59).

In two out of four canned gherkin specimens, a slight increase (transfer factors 1.33 and 1.07, respectively) of the alpha-cypermethrin residues was observed; the residue levels ranged between 0.013-0.020 mg/kg, and the mean transfer factor was 0.86.
In vegetable stock, no residues above the limit of quantitation were detected (all <0.01 mg/kg); the mean transfer factor was <0.53 mg/kg.

No residues at or above the LOQ were detected in the untreated RAC specimens or in the processed fractions.

The alpha-cypermethrin residues in gherkins and processed products as well as the corresponding transfer factors are summarized in Table 6.5.3-2, whereby the transfer factors in the gherkins (RAC) were set as 1:

Table 6.5.3-2: Summary of alpha-cypermethrin residues and transfer factors

Matrix	Residue alpha-cypermethrin mg/kg				Transfer factor alpha-cypermethrin				
	ACK/ 01/04	ACK/ 25/04	AGR/ 01/04	AGR/ 02/04	ACK/ 01/04	ACK/ 25/04	AGR/ 01/04	AGR/ 02/04	mean
fruit (0 DALA[1)])	0.057	0.012	0.066	0.065	N/A	N/A	N/A	N/A	N/A
fruit (RAC, 6-8 DALA[1)])	0.015	0.015	0.038	0.020	1	1	1	1	1
wash water	< 0.01	< 0.01	0.016	0.012	<0.67	<0.67	0.42	0.60	0.59
washed gherkins	0.013	0.011	0.009[2)]	0.011	0.87	0.73	0.24	0.55	0.60
canned gherkins	0.020	0.016	0.015	0.013	1.33	1.07	0.39	0.65	0.86
vegetable stock	<0.01	<0.01	<0.01	<0.01	<0.67	<0.67	<0.26	<0.50	<0.53

1) days after last application
2) 0.009 mg/kg is reported as the actual calculated residue, not as < 0.01.

III. CONCLUSION

At 0 days after the last application, the alpha-cypermethrin residues in gherkin fruit were between 0.012 and 0.066 mg/kg. In the gherkins taken for processing 6-8 days after the last application, these residues declined to 0.015 - 0.038 mg/kg. In wash water, washed gherkins and vegetable stock, the transfer factors were below 1. In two out of four canned gherkin specimens, a slight increase (transfer factors 1.33 and 1.07, respectively) of the alpha-cypermethrin residues was observed.

Report:	CA 6.5.3/2 Mamouni A., 1993a Residue analysis of Alphacypermethrin (SAG 30505) in summer barley and its processed fractions after brewing (field study SKG-9249) AL-730-061
Guidelines:	BBA IV 2, BBA Merkblatt Nr. 58 (1983)
GLP:	yes (certified by Eidgenoessisches Departement des Innern, Bern, Schweiz)

Executive Summary

Two trials in spring barley were performed in Germany in the year 1992. Barley plants were treated with alpha-cypermethrin 100 g a.s./L OD (OSK, "ölhaltiges Suspensionskonzentrat") three times at a rate of 15 g a.s./ha. The growth stage of the plants at the last application was between 71 (Watery ripe: first grains have reached half their final size) – 75 (Medium milk: grain content milky, grains reached final size, still green). Samples were taken 0 and 35 days after the last application.

A part of the harvested barley was used to brew beer and to obtain by-products of the brewing process such as malt, malt germ, beer yeast, spent barley and trub.

The results of this study clearly demonstrate that residues in or on the raw agricultural commodity barley grain are not transferred to spent yeast and the final consumer product beer. No residues above limit of quantitation (0.01 mg/kg) of alpha-cypermethrin could be found in beer. The mean transfer factor for beer was <0.35 for alpha-cypermethrin residues.

I. MATERIAL AND METHODS

A. MATERIALS

1.

Test Material:	Alpha-Cypermethrin SAG 305 05 I
Description:	SAG 305 05 I (OSK)
Lot/Batch #:	not reported, Alpha-Cypermethrin: 100.0 g/L nominal
Purity:	not reported
CAS#:	67375-30-8
Development code:	not reported
Spiking levels:	0.02-0.7 mg/kg

2.

Test Commodity:	
Crop:	Spring barley
Type:	Cereals
Variety:	Alexis, Apex
Botanical name:	*Hordeum vulgare*

Crop parts(s) or processed commodity: ears, grain, spent barley, malt, malt germs, trub, yeast, beer

Sample size: ear, rest of plant: 1-2.3 kg;
grain: 10-22 kg
malt: approx.. 800 mL, equivalent to 350 g
malt germ: all germ material from individual malt sample

spent barley: 1.3-1.6 kg
trub material: all trub separated from individual sample
yeast: all yeast remaining after fermentation of individual sample
beer: 9-10 bottles à 0.33 L

B. STUDY DESIGN AND METHODS

1. Test procedure

Field phase

During the growing season of 1992, two trials in spring barley were performed in Germany. Barley plants were treated with alpha-cypermethrin 100 g a.s./L OSK three times at a rate of 15 g a.s./ha. The applications were made 86-87, 58-67 and 35 days before harvest in a spray volume of 400 litres.
The growth stage of the plants at the last application was between 71 (Watery ripe: first grains have reached half their final size) – 75 (Medium milk: grain content milky, grains reached final size, still green). Samples were taken 0 and 35 days after the last application.

Table 6.5.3-3: Target application rates and timings for barley

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
1992	2	3	F	Alpa-cypermethrin 100 g/L OSK	alpha-cypermethrin	0.0150	400	86-87, 58-67 and 35days before harvest

Processing phase:

Malting and beer production was conducted in the pilot plants of Weissheimer Malz in D-56610 Andernach and in the pilot plant of Binding Brauerei AG in D-60598 Frankfurt am Main. The pilot plants were designed to perform all steps of malting and beer brewing on a small scale and to simulate the same process principles as used in industrial malthouses and breweries.

Beer was obtained from barley by malting and brewing of the malt. Each batch of samples was processed separately to minimize the risk of cross contamination.

Malting:
The complete barley sample was passed through the grader for size grading(>2.2->2.5 mm. The untreated barley was graded before the barley treated with alpha-cypermethrin was fractionated. The graded barley was weighed into sample cylinders, the cylinders were closed with the sieve and placed into the steeping tank. Steeping was performed for 44 hours at a target temperature of 14°C. At the end the product was allowed to rest for an additional 4 hours.
The samples were then germinated under aeration with air adjusted to a temperature of 14°C for 96 -144 hours. After germination, the samples underwent kilning at max. 80°C for 15 hours. Finally, the malt was degerminated sieve to remove the malt germ.

Brewing:
The malt was ground with a two-roll mill and mashed into decarbonised water which was heated to 52°C. The total time of the mashing procedure was between 150 and 163 minutes.
At the end, the mash was drained off into the heated lauter tub (76°C). The wort was separated from the spent barley in the lauter tub by filtration, the spent grain was washed and the washings were combined with the filtrate. The wort was heated by passing steam through the heat exchanger and hop extract was added to the wort within 5 minutes before and 2 minutes after beginning of boiling. 75 minutes after the beginning of boiling, trub material had usually coagulated sufficiently and the trub was separated from the wort. The trub was either separated in the whirlpool or in case of insufficient coagulation by filtration.
The wort was cooled down to 18.2 to 24.5°C within 110 to 203 minutes and then pumped off into a sterilized fermentation vessel. Air was passed in to saturate the wort with oxygen. When the wort had reached a temperature of 14-18°C, yeast slurry was added and the fermentation process allowed to proceed under dark conditions for seven days at approximately 10°C. After removal of the yeast, carbon dioxide was forced under pressure into the storage container before the container was placed into the cooling box at approximately 5-6.3°C for five days. After this period the container was stored at 0°C for ten days. Finally, the beer was clarified by filtration and filled into bottles.

All procedures were conducted using the same technical principles as commonly used in industrial malthouses and in the brewing industry.

2. Description of analytical procedures

All specimens were analysed using an adaptation of analytical method SAMS 351-1.
The analytical material was homogenized with acetone/n-hexane (1 + 1 v/v). This mixture was partitioned into n-hexane, and the n-hexane layer was rotary-evaporated to dryness. The residue was dissolved in n-hexane and cleaned-up by column chromatography on activated Florisil®. In addition, a second clean-up step on silica gel 60/activated carbon was performied for trub, yeast, malt and beer samples. The eluates were evaporated to dryness and dissolved in dodecane/acetone (9 + 1 v/v). Alpha-cypermethrin was determined by electron capture detection (ECD) with a LOQ of 0.01 mg/kg.

Table 6.5.3-4: Procedural recoveries

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. SAMS 351-1 (adapted)		alpha-cypermethrin			
Ear	0.02-0.7	3	89.7	8.9	9.9
Grain	0.02-0.1	2	85.2	-	-
Rest of plant (above ground)	0.02-0.5	3	93.1	8.1	8.7
Malt	0.1	1	92.8	-	-
Malt germs	0.02	1	92.1	-	-
Beer	0.02	1	92.1	-	-
Yeast	0.02	1	89.4*	-	-
Trub	0.1	1	84.6*	-	-
Spent barley	0.1x	1	87.0	-	-
Overall	**0.02-0.7**	**14**	**89.8**	**5.7**	**6.4**

* Corrected = Signal of fortified sample minus signal of control sample used for fortification. Corrected recovery was used for calculation of overall mean recovery.

II. RESULTS AND DISCUSSION

The control (untreated) samples of the raw agricultural commodity (RAC) and of the processing products contained residue concentrations below the routine limit of determination of 0.01 mg/kg for both trials.

Immediately after the last of three applications at 0.015 kg alpha-cypermethrin/ha, residues in treated ears were 0.416 and 0.700 mg/kg, while 0.319 and 0.517 mg/kg, respectively, were found in barley plant. Residues in grain were 0.019 and 0.060 mg/kg, respectively, at35 days after last application.

In the malt / beer production the following residues were observed: In malt, residues of alpha-cypermethrin were reduced compared to RAC (0.013 and 0.014 mg/kg). In malt germs, residues of 0.011 and 0.026 mg/kg were observed. In spent barley, residues of 0.014 and 0.032 mg/kg were determined. The highest mean transfer factors were determined malt germs and spent barley (0.78 and 0.96, respectively), followed by malt (0.48).

No residues above the limit of quantitation were found in treated trub, yeast and beer samples from both trials (all <0.01 mg/kg). The transfer factors ranged between 0.17 and 0.53 (mean 0.35)

The residue levels detected in the treated specimens and their processed fractions as well as the calculated transfer factors are summarized in Table 6.5.3-5.

Table 6.5.3-5: Summary of alpha-cypermethrin residues in barley and transfer factors

Matrix	Residue alpha-cypermethrin mg/kg		Transfer factor** alpha-cypermethrin		
Trial	**9249-01**	**9249-02**	**9249-01**	**9249-02**	**Mean**
Ear (0 days after last application)	0.700	0.416	-	-	
Rest of plant (0 days after last application)	0.517	0.319	-	-	
Grain (35 days after last appl.)	0.060	0.019	-	-	
Malt	0.013	0.014	0.22	0.74	0.48
Malt germs	0.011	0.026	0.18	1.37	0.78
Spent barley	0.014	0.032	0.23	1.68	0.96
Trub	<0.01	<0.01	0.17	0.53	0.35
Yeast	<0.01	<0.01	0.17	0.53	0.35
Beer	<0.01	<0.01	0.17	0.53	0.35

* For calculation purposes, "< 0.01" is set as 0.01

III. CONCLUSION

The results of this study clearly demonstrate that residues in or on the raw agricultural commodity barley grain are not transferred to spent yeast and the final consumer product beer. No residues above limit of quantification of alpha-cypermethrin could be found in these fractions. The transfer factors ranged between 0.17 to 0.53 for alpha-cypermethrin residues.

Report:	CA 6.5.3/3 Harant H., 2007a Determination of residues of Alpha-Cypermethrine in spring barley and its processing products after two applications of BAS 310 41 I in Germany 2007/1013068
Guidelines:	EEC 7029/VI/95 rev. 5, BBA IV 3-3, IVA Guideline I-III (1992)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary
During the growing season 2004, four field trials were conducted in representative spring barley growing areas in Germany in order to generate specimens for analysis to determine the residues of alpha-cypermethrin in plants without roots and grain as well as after processing into beer and pot barley and the respective intermediate fractions.
The test item BAS 310 41 I, a SC formulation of alpha-cypermethrin (100 g alpha-cypermethrin/L) was foliar applied twice at an exaggerated target rate of 1.5 L product/ha (nominal a.s./ha: 0.150 kg alpha-cypermethrin) for each application, resulting in a seasonal target rate of 300 g a.s./ha (nominal). The applications were made ~~42~~ 49 (±1) days and ~~35~~ 42 (±1) days before the planned harvest date, using a spray volume of 300 L/ha.
Specimen were collected at the day of the last application and at harvest (41-43 days after the last application) for analysis of the raw.agricultural commodity as well as for processing.
The specimen were processed into beer and pot barley and the intermediate fractions brewing malt, malt culms, spent grain, spent hops (flocs), spent yeast, pearling dust/bran and flour using simulated industrial processing procedures.
The results of this study clearly demonstrate that residues in or on the raw agricultural commodity barley grain are not transferred to spent yeast and the final consumer product beer. No residues above limit of quantitation of alpha-cypermethrin could be found in this fraction. The mean transfer factor was 0.05 for alpha-cypermethrin residues.
In pot barley residue values of alpha-cypermethrin were found ranging from 0.01 to 0.04 mg/kg with a mean transfer factor of 0.10.

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:** Alpha-Cypermethrin BAS 31041I
   Description: BAS 310 41 I (SC)
   Lot/Batch #: 4000, Alpha-Cypermethrin: 100.0 g/L nominal
   Purity: not reported
   CAS#: 67375-30-8
   Development code: not reported
   Spiking levels: 0.01, 0.1 and 1.0

2. Test Commodity:

Crop:	Spring barley	
Type:	Cereals	
Variety:	Barke, Ursa, Pasadena, Scarlett	
Botanical name:	*Hordeum vulgare*	
Crop parts(s) or processed commodity:	plants without roots, grain, brewing malt, malt culms, spent grain, spent hops (flocs), spent yeast, beer, pot barley, pearling dust/bran, flour	
Sample size:	plant without roots:	1.16-1.66 kg
	grain:	44.12-77.80 kg
	malt:	0.53-0.61 kg
	malt culms:	0.38-0.51 kg
	spent grain:	1.0-1.1 kg
	spent hops (flocs):	0.10-0.12 kg
	spent yeast:	0.20-0.24 kg
	beer:	1.14-1.36 kg
	pot barley:	1.20-1.56 kg
	pearling dust/bran:	0.20-0.22 kg
	flour:	0.30-0.37 kg

B. STUDY DESIGN AND METHODS

1. Test procedure

Field phase

During the growing season 2004, four field trials were conducted in representative spring barley growing areas in Germany in order to generate specimens for analysis to determine the residues of alpha-cypermethrin in plants without roots and grain as well as after processing in brewing malt, malt culms, spent grain, spent hops (flocs), spent yeast, beer, pot barley, pearling dust/bran and flour.

The test item BAS 310 41 I, a SC formulation of alpha-cypermethrin (100 g alpha-cypermethrin/L) was foliar applied twice at an exaggerated target rate of 1.5 L product/ha (nominal a.s./ha: 0.150 kg alpha-cypermethrin) for each application, resulting in a seasonal target rate of 300 g a.s./ha (nominal). This exaggerated rate was used in an attempt to generate residue levels sufficiently above the method limit of quantification (LOQ) in the raw agricultural commodity. The applications were made ~~42~~ 49 (±1) days and ~~35~~ 42 (±1) before the planned harvest date, using a spray volume of 300 L/ha.

Two of the trials consisted of two plots, one untreated and one treated (FR/01/04/40 and FR/01/04/70); the other two (FR/01/04/20 and FR/01/04/50) consisted of one treated plot each.

Specimen of barley grain were collected at the day of the last application and at harvest (at growth stage 89) for analysis of the raw.agricultural commodity as well as for processing.

The specimen for RAC analysis were frozen within a maximum of 6 hours. The specimens for processing were stored at room temperature until the processing start.

Table 6.5.3-6: Target application rates and timings for barley

Year	No. of Trials	No. of Appl.	F P or G	Test Item	Active Substance	Appl. Rate (kg a.s./ha)	Water Volume (L/ha)	Target Date/ Timing
2004	4	2	F	BAS 310 41 I (SC)	alpha-cypermethrin	0.150	300	42 ±1 and 35 ±1 days before harvest

Processing phase: Beer processing

Malting:

Before malting was started, all specimens were sieved to remove contaminants and to grade the barley grain into kernels with the same minimum size (sieve mesh: 2.5 mm). The steeping process was conducted as a combined wet and dry steeping. Sieved barley grain was transferred in a special steeping vessel. The steeping temperature was 13-15 °C and the steeping degree was 44.8-44.9% (water content). The subsequent germination was performed for approximately 118-119 hours at 13-15 °C and a relative air humidity of 80-100% under continuous turning. After kiln-drying to a residual water content of 4.0% and removal of the sprouts by a trimmer, the treated malt was stored for 35-43 days at ambient conditions (room temperature) until brewing was started.

Mashing, boiling and fermentation:

Mashing was conducted with tap water in a heatable tun using the infusion method (whole amount of water and malt was processed within one step). The mash was heated in a stepwise temperature increase process to a final temperature of 76-78°C. The whole mashing process took 102-110 minutes. After mashing, the wort was separated from the insoluble malt components (spent grains). The extract remaining in those spent grains was extracted by washing them with hot water. The wort separation was done using a refining fat. After the addition of commercial hop, the wort was boiled (approx. 90 min at normal pressure), then the trub was separated in a whirlpool, causing the sludge to deposit on the bottom in the shape of a cone. For cooling and ventilating the wort, plate heat exchangers and an intra-plant circulation were used. By adding oxygen until saturation, the conditions were prepared for fermentation.

In the pilot plant the classical primary fermentation (low fermentation) was carried out using bottom fermenters (approx. 50 L capacity). Fermentation heat was dissipated by means of room ventilation.

As soon as the extract content of the fermenting young beer was 2 % higher than the final attenuation, it was transferred into casks through hoses. After fermentation, the yeast deposited on the tank bottom.

For maturation, the young beer was stored for 2 days at room temperature (warm maturation) in casks. Then, under pressure the young beer was stored at approx. 0-2 °C for about 4 weeks (cold maturation). Finally, the rack beer was filtered using a filter combination.

Processing phase: Pot barley processing

The optimal moisture to mill barley grains is approx. 14 %. In order to obtain acceptable milling results a moisture content of up to 16 % is acceptable. The moisture content of the specimens of the special processing part ranged in a lower moisture level (12.8-13.0%, treated samples). Therefore, the grain was processed after damping which resulted in a moisture content of 14.9-15.1% (treated samples).
The specimens were hulled using a "Schule-Vertikal-Schälmaschine". Each specimen was hulled until the stipulated abrasion for pot barley of 20-25 % was reached. The degree of abrasion was determined by the proportion of pot barley with respect to the total portion of purified grain used for the hulling process. As a side product from hulling pearling dust/bran and flour were derived.

2. Description of analytical procedures

Samples of barley grain, malt, pearling dust and pot barley were analysed for alpha-cypermethrin with BASF analytical method no. 567/0 using HPLC-MS/MS for quantitative determination. All other matrices were analysed with BASF analytical method no. 567/1 using GC/MS for quantitative determination.

Principle of BASF method no. 567/0:
BAS 310 I (alpha-cypermethrin) is extracted with a 70:25:5 methanol/water/hydrochloric acid mixture. After centrifugation, an aliquot of the extract is partitioned into cyclohexane. A silica gel SPE column is used for further purification, if required. Final determination is performed by HPLC-MS/MS using the ammonium adduct of cypermethrin.
The limit of quantitation (LOQ) of the method for alpha-cypermethrin is 0.01 mg/kg.

Principle of BASF Method No. 567/1:
BAS 310 I (alpha-cypermethrin) is extracted with a 70:25:5 methanol/water/hydrochloric acid mixture in case of beer and spent hops and with 95:5 methanol/hydrochloric acid in case of plant w/o root, spent grain, malt culms, spent yeast and flour. After centrifugation, an aliquot of the extract is partitioned into cyclohexane. A silica gel SPE column is used for further purification. Final determination is performed by GC-MS. The limit of quantitation (LOQ) of the method for alpha-cypermethrin is 0.01 mg/kg.

Table 6.5.3-7: Procedural recoveries

Matrix	Fortification Level (mg/kg)	Summary Recoveries			
		n	Mean (%)	SD (+/-)	RSD (%)
Method No. 567/0		alpha-cypermethrin			
grain, malt, pot barley, pearling dust/bran	0.01, 0.1	8	89.9	13.9	15.4
Method No. 567/1		alpha-cypermethrin			
Plant w/o roots, malt culms, spent grain, spent hops, spent yeast, beer, flour	0.01, 0.1, 1.0	64	94.3	11.3	12.0

II. RESULTS AND DISCUSSION

In the control samples, two cases occurred where residues were found: in trial FR 01/04/40 alpha-cypermethrin was found in plant w/o roots (0.02 mg/kg) and spent hops (0.07 mg/kg). In all other control samples no residues of alpha-cypermethrin at or above the limit of quantitation were found, which proves that in general no interferences of the specimen material with the analytical procedure occurred.

The amounts of alpha-cypermethrin in spring barley grain treated at an exaggerated rate and collected 41 to 43 days after last application ranged from 0.11 to 0.33 mg/kg.
In the malt / beer production the following residues were observed: alpha-cypermethrin in amounts comparable to the RAC was found in malt (0.12 to 0.19 mg/kg). With residues between 0.32 and 1.12 mg/kg, a slight concentration could be seen in malt culms. Low amounts of alpha-cypermethrin were found in spent grain and spent hops (0.13 to 0.26 mg/kg and 0.02 to 0.07 mg/kg respectively). Comparable to the residues found the highest mean transfer factor was determined for malt culms (3.18) followed by spent grain (0.95), brewing malt (0.72) and spent hops (0.22).
In spent yeast and the final consumer product beer, no residues above the limit of quantitation of alpha-cypermethrin could be found. The transfer factors ranged between 0.03 to 0.09 for alpha-cypermethrin residues.
In pot barley residue values of alpha-cypermethrin were found ranging from 0.01 to 0.04 mg/kg with a mean transfer factor of 0.10. Pearling dust/bran and flour had higher residue values with 0.72 to 1.62 mg/kg as well as 1.35 to 2.17 mg/kg, respectively. The mean transfer factor determined for pearling dust/bran is 5.79 and for flour 8.67.

The residue levels detected in the treated specimens and their processed fractions as well as the calculated transfer factors are summarized in Table 6.5.3-8.

Table 6.5.3-8: Summary of alpha-cypermethrin residues in barley and transfer factors

Matrix	DALA	Residue alpha-cypermethrin mg/kg				Transfer factor alpha-cypermethrin				
Trial*		FR 01/04/40	FR 01/04/70	FR 01/04/20	FR 01/04/50	FR 01/04/40	FR 01/04/70	FR 01/04/20	FR 01/04/50	mean
Plant without roots	0	3.13	1.79	4.41	2.82	-	-	-	-	-
Grain, RAC	41-43	0.33	0.26	0.22	0.11	1	1	1	1	1
Brewing malt		0.15	0.19	0.13	0.12	0.45	0.73	0.59	1.09	0.72
Malt culms		0.73	0.65	1.12	0.32	2.21	2.50	5.09	2.91	3.18
Spent grain		0.25	0.26	0.19	0.13	0.76	1.00	0.86	1.18	0.95
Spent hops		0.06	0.05	0.07	0.02	0.18	0.19	0.32	0.18	0.22
Spent yeast		< 0.01*	< 0.01*	< 0.01*	< 0.01*	0.03	0.04	0.05	0.09	0.05
Beer		< 0.01*	< 0.01*	< 0.01*	< 0.01*	0.03	0.04	0.05	0.09	0.05
Flour		2.14	2.17	1.67	1.35	6.48	8.35	7.59	12.27	8.67
Pearling dust/bran		1.62	1.61	1.21	0.72	4.91	6.19	5.50	6.55	5.79
Pot barley		0.04	0.01	0.03	0.01	0.12	0.04	0.14	0.09	0.10

* For calculation purposes, "< 0.01" is set as 0.01

III. CONCLUSION

The results of this study clearly demonstrate that residues in or on the raw agricultural commodity barley grain are not transferred to spent yeast and the final consumer product beer. No residues above limit of quantification of alpha-cypermethrin could be found in these fractions. The transfer factors ranged between 0.03 to 0.09 for alpha-cypermethrin residues. With residues between 0.32 and 1.12 mg/kg, a slight concentration could be seen in malt culms. Low amounts of alpha-cypermethrin were found in spent grain and spent hops (0.13 to 0.26 mg/kg and 0.02 to 0.07 mg/kg, respectively). Comparable to the residues found the highest mean transfer factor was determined for malt culms (3.18) followed by spent grain (0.95), brewing malt (0.72) and spent hops (0.22).
In pot barley residue values of alpha-cypermethrin were found ranging from 0.01 to 0.04 mg/kg with a mean transfer factor of 0.10. Pearling dust/bran and flour had higher residue values with 0.72 to 1.62 mg/kg as well as 1.35 to 2.17 mg/kg, respectively. The mean transfer factor determined for pearling dust/bran is 5.79 and for flour 8.67.

This entry is taken from public literature and was not included in the study list of the application.

Report: CA 6.5.3/4
Kang S.-M.,Lee M.-G., 2005a
Fate of some pesticides during brining and cooking of chinese cabbage and spinach
2005/1043600
Guidelines: none
GLP: no

Executive Summary

In 2002, Chinese cabbages and spinach were cultivated in a greenhouse in two separate field lots according to good agricultural practices and subjected to pesticide application. Pesticide solutions were sprayed onto the crops once a week. Cypermethrin was applied at a rate of 0.24 kg as/ha or 0.30 kg as/ha on Chinese cabbage and at a rate of 0.13 kg as/ha or 0.17 kg as/ha on spinach. Chinese cabbages were harvested 3 days after ninth and eleventh pesticide applications. Spinach was harvested on the 7th and 2nd day after three applications.
The crops were subjected to brining, heat-cooking and blanching to determine residue or transfer ratios of the pesticides.
The reduction of cypermethrin residues in the brined Chinese cabbage was 13% and in the heat-cooked Chinese cabbage 0%. The reduction of cypermethrin residues in the blanched spinach was 13% and in the heat-cooked spinach 8%.

I. MATERIAL AND METHODS

A. MATERIALS

1. **Test Material:** Cypermethrin
   Description: Cypermethrin 5% (EC)
   Lot/Batch #: not reported
   Purity: not reported
   CAS#: 52315-07-8
   Development code: not applicable
   Spiking levels: between 0.5 and 2.0 mg/kg

2. **Test Commodity:** Brassica vegetables, leafy vegetables
   Crop: Chinese cabbage, spinach
   Type: not reported
   Variety: not reported
   Botanical name: Brassica rapa, spinacia oleracea L.
   Crop parts(s) or processed
   Commodity: Chinese cabbage (RAC) and processed commodities: brining and head-cooking; spinach (RAC) and processed commodities: blanching and heat-cooking
   Sample size: Brining of Chinese cabbage: 500 g, heat-cooking of Chinese cabbage: 300 g, blanching of spinach: 200 g, heat-cooking of spinach 200 g

B. STUDY DESIGN AND METHODS

1. Test procedure

In 2002, Chinese cabbages were cultivated in a greenhouse in two separate field lots according to good agricultural practices and subjected to pesticide application. Mixed pesticide solutions, including two or three pesticides, were sprayed onto the cabbages once a week. Cypermethrin was applied at a rate of 0.24 kg as/ha or 0.30 kg as/ha. The vegetables were harvested 3 days after ninth and eleventh pesticide applications to obtain low and high pesticide-level samples, respectively.
In 2002, spinach was cultivated in a greenhouse in two separate field lots according to the good agricultural practices and subjected to pesticide application. Pesticide solutions were sprayed onto the spinach once a week. Cypermethrin was applied at a rate of 0.13 kg as/ha or 0.17 kg as/ha. To obtain low and high pesticide-level samples, the vegetables were harvested on the 7^{th} and 2^{nd} day after three applications, respectively.
Other pesticide formulations (EC) applied during the growth of Chinese cabbage and spinach were chlorpyrifos 20%, diazinon 34%, dichlorvos 50%, EPN 45%, endosulfan 35%, deltamethrin 1% and fenvalerate 5%.

Brining and heat-cooking of Chinese cabbage: A head of Chinese cabbage weighing about 2.5 kg was longitudinally cut into eight portions. Two portions (equivalent to 500 g) were then soaked in 2 L of 8% saline at 20 °C, after 4 hours drained on a sieve for 30 min, washed by hands in 1 L tap water for 10 seconds, and drained again for 30 minutes. The brine-soaking resulted in 428 g brined cabbage, 1.932 mL waste brine, and 1.010 mL water-washings.
One portion of the longitudinally-cut cabbage weighing about 300 g was further cut into 5-cm pieces and cooked for 20 minutes with two stirred in 1.2 L covered boiling water pot. The pot was left standing for 30 minutes to cool down to 60 °C. After draining for 30 minutes, 240 g cabbage solids and 1.013 mL cooked fluid were obtained.

Blanching and heat-cooking of spinach: About 200 g spinach leaves were dipped into 1 L boiling water for 2 minutes and drained on a sieve for 30 minutes. The blanching resulted in 228 g spinach solids and 875 mL blanched fluids.
For heat-cooking, about 200 g of spinach bunches were cut into four portions cross-wise and cooked in 1 L tap water for 15 minutes as with the Chinese cabbage. The heat-cooking resulted in 218 g spinach solids and 810 mL cooked fluid.

2. Description of analytical procedures

The raw vegetable samples were cleaned by removing roots and spoiled leaved, and subjected to residue analysis. Solid liquid samples obtained in duplicate runs were extracted using appropriate solvent systems and subjected to residue analysis in triplicates.
Gas chromatograph equipped with an HP-5 column was used to detect organophosphorus pesticides with a flame photometric detector, and organochlorine and synthetic pyrethroids with an electron capture detector.
Samples for residue analysis were spiked with standard pesticides at 0.5-2.0 mg/kg of vegetable or liquid samples and subjected to routine analysis in triplicates. The recoveries for cypermethrin were between 94 and 110%.

II. RESULTS AND DISCUSSION

The residue levels of cypermethrin in the treated Chinese cabbage and spinach specimens are shown in Table 6.5.3-9. Out of two different samples, those with residue concentrations nearer the Korea MRLs were chosen and subjected to reduction studies.

Brining process of Chinese cabbage:
Residue concentrations of raw and brined cabbages, and brine obtained after soaking and washings of brined cabbages were determined. Weight or volumes of the samples were measured, and the total amount of residues in analyzed samples was calculated. The residue ratios of samples were calculated as the total amount of residues on the basis of raw cabbage as 100, and difference from 100% was regarded as the loss during brining process.
The residue ratios of cypermethrin in the brined cabbage was 112%. The transfer ratio of cypermethrin into the soaking brine or washings after brining was 10% and 7%, respectively. No loss of residues was observed during brining.

Heat-cooking of Chinese cabbage
Residue concentrations of the solid cabbage and cooking fluid were analyzed. The residue ratios were calculated in the same way as for the brining process.
The residue ratio of cypermethrin in the heat-cooked cabbage was 119%, the transfer ratio into the fluid or loss during cooking was zero, suggesting that cypermethrin do not decompose or vaporize during cooking.

Blanching of spinach
Residue concentrations of the solid spinach and blanching fluid were analyzed. The residue ratios were calculated in the same way as for the brining process.
The residue ratio of cypermethrin in blanched spinach was 87%, the transfer ratio into the blanching water was zero and the loss in blanching was 13%.

Heat-cooking of spinach:
Residue concentrations of the solid spinach and cooking fluid were analyzed. The residue ratios were calculated in the same way as for the brining process.
The residue of cypermethrin in heat-cooked spinach was 92%, the transfer ratio into the fluid was zero and the loss in cooking was 8%.

The residue and transfer ratios of cypermethrin in processed fractions of Chinese cabbage and spinach are presented in Table 6.5.3-10.

Table 6.5.3-11 shows the % disappearance of cypermethrin residues in the raw vegetables during processing. The reduction of cypermethrin residues in the brined Chinese cabbage was 13% and in the heat-cooked Chinese cabbage 0%. The reduction of cypermethrin residues in the blanched spinach was 13% and in the heat-cooked spinach 8%.

Table 6.5.3-9: Residue levels of Cypermethrin in the treated Chinese cabbage and spinach specimens

Matrix	Concentration level	Application rate (kg as/ha)	Residue concentration (mg/kg)
Chinese Cabbage	Low	0.24	0.176
Chinese Cabbage	High	0.3	0.195*
Spinach	Low	0.13	1.32
Spinach	High	0.17	2.61*

*) Samples having residues nearer to legal Korea MRLs (5.0 mg/kg for Chinese cabbage and 2.0 mg/kg for spinach) were subjected to cooking test

Table 6.5.3-10: Residue and transfer ratios of Cypermethrin in processed fractions of Chinese cabbage and Spinach

Processed Fraction	Residue concentration (mg/kg)	Residue and transfer ratio (%)
Chinese cabbage - brining and washing		
Raw cabbage	0.20	100
Brined cabbage		112
Soaking brine		10
Washing after brining		7
Loss in brining		0
Chinese cabbage - cooking		
Raw cabbage	0.20	100
Cooked cabbage		119
Cooking water		1
Loss in cooking		0
Spinach - blanching		
Raw spinach	2.6	100
Blanched spinach		87
Blanching water		0
Loss in blanching		13
Spinach - heat-cooking		
Raw spinach	2.6	100
Cooked spinach		92
Cooking water		0
Loss in heat-cooking		8

Table 6.5.3-11: Disappearance of Cypermethrin in cooking of Chinese cabbage or Spinach

Chinese cabbage			Spinach		
Disappearance of cypermethrin (%)			Disappearance of cypermethrin (%)		
Brining-washing	Cooking	Average removal	Blanching	Cooking	Average removal
13*	0	6.5	13	8	10.5

*according to the report
Data are % of residues which are discarded in cooking process, out of total in the raw materials. Edible and inedible portions were regarded as follows. An edible portion in brining of Chinese cabbage is brined cabbage; its inedible portions are soaking brine, washings after brining and loss in brining. Edible portions in cooking of Chinese cabbage are cooked cabbage and cooking water; its inedible portion is loss in cooking. An edible portion in blanching of spinach is blanched spinach; its inedible portions are blanching water and loss in blanching. Edible portions in cooking of spinach are cooked spinach and cooking water; its inedible portion is loss in cooking. Residues in edible and inedible portinos were regarded as 100%.

III. CONCLUSION

The reduction of cypermethrin residues in the brined Chinese cabbage was 13% and in the heat-cooked Chinese cabbage 0%. The reduction of cypermethrin residues in the blanched spinach was 13% and in the heat-cooked spinach 8%.

The RMS and co-RMS consider the loss – values in Table 6.5.3-10 as erroneous/misleading and as insufficiently reliable. Therefore, Table 6.5.3-11 was to be removed in the RAR.

CA 6.6 Residues in Rotational Crops

CA 6.6.1 Metabolism in rotational crops

Residues in succeeding or rotational crops have been investigated for cypermethrin. Sufficient information regarding the metabolites found in soil has been generated from a confined ([^{14}C] radiolabeled) and non-radiolabeled crop rotational studies conducted with cypermethrin, aerobic soil metabolism studies conducted with alpha-cypermethrin and cypermethrin, and from alpha-cypermethrin photodegradation and cypermethrin anaerobic soil metabolism studies.

The fate of alpha-cypermethrin and cypermethrin in soil (aerobic soil metabolism) has been well defined and described in the dossiers submitted in 1995 by American Cyanamid Company for EU reregistration of these active substances. The data related to fate and behavior of alpha-cypermethrin and cypermethrin were presented together in Annex B of the alpha-cypemethrin monograph (September 1999) since several studies were presented in parallel for both active substances. This allowed extrapolation of the effects of one active substance to another.
Based on these studies and the fact that cypermethrin and alpha-cypermethrin behave similarly, succeeding crops in an alpha-cypermethrin treated field soil would be exposed mainly to the unchanged alpha-cypermethrin and to a much lesser extent to their soil and photo degradates, 3-phenoxybenzoic acid (3PBA) and 3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylic acid (DCVA).

A summary of a 1993 study of alpha-cypermethrin photodegradation on soil (Van Dyk *et al.*) is presented in Section B. 7.1.1.3, Soil photolysis (Annex IIA 7.1.1.1.2) of the alpha-cypermethrin monograph. [^{14}C-benzyl] alpha-cypermethrin was used in this GLP study. The results indicate that alpha-cypermethrin is subjected to photolysis with subsequent formation of the metabolites, phenoxybenzoic acid (17% after 30 days) and phenoxybenzyl alcohol (2.7% after 30 days), bound residue (13.3% after 30 days) and mineralization (6.2% after 30 days). DT_{50} of 31 days (irradiated samples) and 193 days (dark samples) were calculated based on first-order reaction kinetics.

A summary of the anaerobic degradation of cypermethrin in soil [Section B. 7.1.2 Anaerobic degradaton in soil (Annex IIA 7.1.1.1.2)] is provided in the alpha-cypermethrin monograph. The findings showed that the degradation of cypermethrin under water-logged conditions was almost as rapid as in aerobic soil conditions. However, under anaerobic conditions, much greater amounts (52-72%) of the 3-PBA metabolite were found to accumulate in the soil. No mineralization occurred in this study (Standen, 1976).

In a crop rotational study (Jackson C., 1978) conducted to demonstrate the metabolism of [^{14}C] labeled cypermethrin (BAS 311 I) residues in potatoes grown as a rotation crop to lettuce, test substance [^{14}C] cypermethrin was labeled on the benzyl and cyclopropyl rings. At the time of lettuce harvest, the soil contained a total residue of 0.18 mg/kg and 0.10 mg/kg respectively, for the benzyl and cyclopropyl labeled cypermethrin. Similar residues were present in the soil when the rotation crop (potato) was sown. The cypermethrin metabolites, 3PBA and DCVA, were found and their presence confirmed. The mature potatoes contained very low levels of residues in tubers (≤ 0.01 mg/kg total residue) and haulms (≤ 0.007 mg/kg total residue) and no residues were detected in most of the samples. According to the conclusions in the cypermethrin monograph (October 1999), "The residues present in the soil 15 months after treatment were strongly absorbed to the soil."

Furthermore, the soil and photo degradates of cypermethrin have occurred at low levels (or have not been detected) as cypermethrin parent (<0.01 mg/kg), 3PBA and its conjugate (<0.05 ppm) and DCVA and its conjugate (<0.05 ppm) in non-radiolabeled succeeding crop studies on sugar beets, wheat, and field beans (peer-reviewed in the framework of the original inclusion into Annex I according to Directive 91/414/EEC; see below CA 6.6.2).

Based on the results of the cypermethrin confined crop rotational studies (using [^{14}C]) and unlabeled cypermethrin), alpha-cypermethrin and cypermethrin aerobic soil metabolism studies, alpha-cypermethrin photodegradation study, and cypermethrin anaerobic soil metabolism study, we would not expect to find metabolites other than those already identified in these studies (namely 3-PBA and DCVA), when conducting a crop rotational study with alpha-cypermethrin. As the 3-PBA and DCVA metabolites have occurred at low levels, we propose that the residue definition for rotational crops should only be alpha-cypermethrin parent (BAS 310 I). Thus, a non-radiolabeled crop rotational field study will adequately define the alpha-cypermethrin exposure in rotational crops.

CA 6.6.2 Magnitude of residues in rotational crops

In the framework of the original inclusion into Annex I according to Directive 91/414/EEC studies on the magnitude of the residue in rotational crops were peer-reviewed (Monograph Alpha-cypermethrin, September 1999, and 3rd Addendum: Addendum Chapter B-7 (Residue data), January 2003). No new studies have been performed.

The characteristics of the crop rotation studies are summarised in Table 6.6.2-1.

Table 6.6.2-1: Summary of studies in rotational crops previously available

Crop group	Crop	Test substance	Application and sampling details				Year	DocID
			Method, F or G	Rate (kg a.s./ha)	Interval between application and planting	Harvest time		
Legume vegetables	Field beans	Cypermethrin 40% EC RIPCORD	F	Pre-planting; 0.5 kg a.s./ha; half of treated area incorporated to a depth of 5 cm.	Planting: 58 weeks	71 weeks after treatment	1976-78 (field part)	CY-790-057 BLGR. 79.112
Root and tuber vegetables	Sugar beet				Planting: 58 weeks	99 weeks after treatment		
Cereals	Wheat				Planting: 47 weeks	108 weeks after treatment		
Root and tuber vegetables	Sugar beet	Cypermethrin 40% EC RIPCORD	F	Pre-planting; 0.5 kg a.s./ha; half of treated area incorporated to a depth of 5 cm.	Planting: 58 weeks	99 weeks after treatment	1976-78 (field part)	AL-790-032 CY-790-061 BEGR. 80.197
Cereals	Wheat				Planting: 47 weeks	108 weeks after treatment		
Brassica vegetables	Cabbage	Alpha-cypermethrin 150 g a.s./kg WG RLM 11203	F	1 x 0.02 to bare ground plot or 3 x 0.02 to bare ground plot	Planting: 14 days	33, 55, 76 DAP; 47, 69, 90 DALT	2000	AL-790-049 2002/5004754
Root and tuber vegetables	Carrot			1 x 0.02 to bare ground plot or 3 x 0.02 to bare ground plot	Planting: 12 days	46, 67, 82 DAP; 58, 79, 94 DALT		
Leafy vegetables	Lettuce			1 x 0.02 to bare ground plot or 3 x 0.02 to bare ground plot	Planting: 13 days	14, 31, 76 DAP; 27, 44, 89 DALT		
Cereals	Wheat			1 x 0.02 to bare ground plot or 3 x 0.02 to bare ground plot	Planting: 19 days	105, 179, 238 DAP; 124, 198, 257 DALT		

Crop group	Crop	Test substance	Application and sampling details				Year	DocID
			Method, F or G	Rate (kg a.s./ha)	Interval between application and planting	Harvest time		
Brassica vegetables	Cabbage	Alpha-cypermethrin 150 g/kg WG BAS 310 08 I	F	1 x 0.02 to bare ground plot or 2 x 0.02 to bare ground plot	Planting: 11 days	22, 42, 49 DAP; 33, 53, 60 DALT	2001	2002/ 1011569
					Planting: 14 days	36, 77, 155 DAP; 49, 91, 169 DALT		
					Planting: 13 days	41, 62, 64 DAP; 54, 65, 77 DALT		
Root and tuber vegetables	Carrot			1 x 0.02 to bare ground plot or 2 x 0.02 to bare ground plot	Planting: 11 days	49, 76, 110 DAP; 60, 87, 121 DALT		
					Planting: 11 days	49, 72, 87 DAP; 60, 83, 98 DALT		
					Planting: 14 days	91, 113, 125 DAP; 105, 127, 139 DALT		

Crop group	Crop	Test substance	Application and sampling details				Year	DocID
			Method, F or G	Rate (kg a.s./ha)	Interval between application and planting	Harvest time		
Leafy vegetables	Lettuce	Alpha-cypermethrin 150 g/kg WG BAS 310 08 I	F	1 x 0.02 to bare ground plot or 2 x 0.02 to bare ground plot	Planting: 11 days	17,35,42 DAP; 28, 46, 53 DALT	2001	2002/ 1011569
					Planting: 14 days	35, 56, 70 DAP; 49, 70, 84 DALT		
					Planting: 14 days	34, 42, 52 DAP; 48, 59, 66 DALT		
Cereals	Wheat			1 x 0.02 to bare ground plot or 2 x 0.02 to bare ground plot	Planting: 11 days	27, 189 DAP; 38, 200 DALT; wheat did not mature sufficiently to provide harvest wheat grain		
					Planting: 12 days	44, 106, 114 DAP; 56, 118, 126 DALT		
					Planting: 13 days	52, 120, 129 DAP; 65, 133, 142 DALT		

F Field/outdoor
G Glasshouse/protected
DAP days after planting
DALT days after (last) treatment

In the studies performed with cypermethrin (Ripcord ® 40 EC; Doc IDs CY-790-057 and AL-790-032), cypermethrin was applied at a rate of 0.5 kg a.s./ha pre-planting. No residues of cypermethrin above the LOQ (0.01 mg/kg; bean foliage: 0.02 mg/kg) were detected in sugar beet (root and foliage) or wheat (straw) after approximately 2 years or in field beans (beans and foliage) after approximately 16 months.
No residues of the metabolites 3-phenoxybenzoic acid and 3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylic acid above the LOQ (0.05 mg/kg) were detected in sugar beet roots and wheat straw approximately 2 years after treatment of soil with 0.5 kg cypermethrin/ha.

The magnitude of alpha-cypermethrin residues in rotational crops was investigated in two studies performed in Southern Europe (Italy and Southern France) and Northern Europe (Northern France and the United Kingdom).
In the first study performed in Italy (AL-790-049), prior to seeding or planting of representative crops (lettuce, head cabbage, carrot and wheat), alpha-cypermethrin was applied to 2 plots of bare soil at a dose rate of 20 g a.s./ha, once and 3 times respectively with 7 days interval between each application.
Rotational crops were sampled at various intervals between final application and harvest. The different samples were extracted and partitioned with solvents and were analysed by GC-MS which was validated for each isomer pair (cis 1, cis 2, trans 3 and trans 4). The limit of quantitation of the analytical method was 0.01 mg/kg. No detectable cypermethrin isomer residues (cis-1, cis-2, trans-3, trans-4) were found above the validated method LOQ (0.01 mg/kg) in any untreated specimen or in any rotational crop planted in BAS 310 I-treated soil at any interval sampled.
These data showed that the rotation crops took up no alpha-cypermethrin from soil. The level of residues at harvest didn't exceed the LOQ of the analytical method for monitoring (0.01 mg/kg for each isomer).

The second study was performed in France (North and South) and the United Kingdom (DocID 2002/1011569, AL-790-048). Prior to seeding or planting of representative crops (lettuce, cabbage, carrot and wheat), bare soil was treated either once or twice with alpha-cypermethrin at a dose rate of 20 g a.s./ha. The first and the second applications were made 19 days and 12 days before seeding or planting respectively. No ageing period of the soil was performed.
Cabbage, lettuce, carrots and wheat were sampled at different growth stages. The different samples were extracted with successive solvents and partitioned with water and the levels of residues were determined using GLC with mass spectrometric detection (Limit of quantitation : 0.01 mg/kg). The residues were determined as cis 1, cis 2, trans 3 and trans 4 isomers of cypermethrin.

No detectable cypermethrin isomer residues (cis-1, cis-2, trans-3, trans-4) were found above the validated method LOQ (0.01 mg/kg) in any untreated specimen.
No detectable cypermethrin isomer residues (cis-1, cis-2, trans-3, trans-4) were found above the validated method LOQ (0.01 mg/kg) in any cabbage or wheat planted in alpha-cypermethrin-treated soil at any interval sampled.
With one exception, no detectable cypermethrin isomer residues (cis-1, cis-2, trans-3, trans-4) were found above the validated method LOQ (0.01 mg/kg) in any lettuce planted in BAS 310 I-treated soil at any interval sampled. A trace trans-4 (0.01 mg/kg) residue was detected in the immature lettuce (BBCH 16) in one trial 48 days following the second of two treatments at 20 g a.s./ha.

0.02 mg/kg trans-4 and 0.03 mg/kg trans-4 were found in two immature whole carrot plants (BBCH 16) in one trial (Southern France) 60 days following the 1 x 20 g a.s./ha and 2 x 20 g a.s./ha, respectively. There were no other detectable cypermethrin isomer residues (cis-1, cis-2, trans-3, trans-4) found above the validated method LOQ (0.01 mg/kg) in any remaining carrot specimens from the BAS 310 I-treated soil.
It was concluded that the rotation crops took up very little alpha-cypermethrin from the soil. The level of residues at harvest didn't exceed the LOQ of the analytical method for monitoring (0.01 mg/kg) No MRL was proposed for rotational crops.

Overall conclusion on the magnitude of residues in rotational crops

The data available from rotational crop studies show that residues of alpha-cypermethrin can be expected to be very low. In cabbage, carrot, lettuce and wheat planted on alpha-cypermethrin treated soil approximately two weeks after the final of 1-3 treatments at 20 g a.s./ha/treatment no residues of cypermethrin isomers (cis-1, cis-2, trans-3, trans-4) above the limit of quantitation (0.01 mg/kg) were found in the edible parts at harvest.

It was shown that the application of alpha-cypermethrin at a rate exceeding the critical GAP did not lead to a significant uptake in edible parts of vegetables with short development times even when planted after a short re-plant interval of approximately two weeks. This scenario can be regarded as worst case situation and it can therefore be concluded that after longer waiting periods or in plants with longer development times no residues of alpha-cypermethrin will be found.

CA 6.7 Proposed residue definitions and maximum residue levels

CA 6.7.1 Proposed residue definitions

Plant Matrices

For proposing a suitable residue definition in plant and animal matrices, multiple investigations were performed. As presented in sections 6.2, 6.3, 6.5 and 6.6, plant and animal studies were performed in which alpha-cypermethrin was applied according to the intended use patterns.

For deriving a suitable **residue definition for food of plant origin**, five crop metabolism studies in three different crop categories using ^{14}C-benzyl-, ^{14}C-vinyl- and/or ^{14}C-cyclopropane-labeled alpha-cypermethrin were performed covering the categories of leafy vegetables, brassica vegetables and cereals. The effect of processing on the nature of the residue was investigated in the framework of the original inclusion into Annex I according to Directive 91/414/EEC using test conditions simulating pasteurization, baking, brewing, boiling and sterilization. Information on the residue situation in succeeding or rotational crops is available from a confined ([^{14}C] radiolabeled) and non-radiolabeled crop rotational studies conducted with cypermethrin, aerobic soil metabolism studies conducted with alpha-cypermethrin and cypermethrin, and from alpha-cypermethrin photodegradation and cypermethrin anaerobic soil metabolism studies.

In general, metabolism of alpha-cypermethrin in plants comprises

-hydrolysis at the ester linkage with the formation of two moieties :
a) the phenoxybenzoyl portion which leads to a multiplicity of metabolites as acids, aldehydes, alcohols and converted into amino acids or glucose conjugates;
b) the dichlorovinyl cyclopropane acid portion of the molecule (cis- and trans-DCVA isomers and conjugates)

-oxidative ring hydroxylation of the phenyl ring which was observed only in wheat and cabbage.

The metabolism studies indicate that parent alpha-cypermethrin is usually the predominant component. Slight isomeric conversion from cis to trans configuration was observed in wheat forage, hay and straw.
Alpha-cypermethrin was found to be susceptible to hydrolysis at the ester linkage at simulated processing conditions at pH 6 at 120°C resulting in the breakdown of the ester linkage to 3-phenoxybenzaldehyde (M310I018, CL 206969) and dichlorovinydimethylcarboxylic acid (DCVA, M310I001, CL 912554) which are also found in the metabolism studies (see CA 6.5.1). Formation of 3-Phenoxybenzaldehyde (M310I018) was also reported in a published study (Lin *et al.*, 2005/1043620, see CA 6.5.1/1)

It was shown that the application of alpha-cypermethrin at a rate exceeding the critical GAP did not lead to a significant uptake in edible parts of vegetables with short development times even when planted after a short re-plant interval of approximately two weeks. This scenario can be regarded as worst case situation and it can therefore be concluded that after longer waiting periods or in plants with longer development times no residues of alpha-cypermethrin will be found.

Based on these findings from all studies summarised above the following residue definitions for alpha-cypermethrin in plant matrices are proposed:

For dietary risk assessment: alpha-cypermethrin

For MRL setting alpha-cypermethrin

Animal matrices

The main biotransformation reactions were observed in rats, goats and laying hens, so there is a consistent picture of the metabolism of ^{14}C-BAS 310 I in all animal species investigated.

Ensuing from the degradation routes as explained for plant metabolism, cleavage of the ester linkage, loss of the nitrile group and oxidation is observed with the formation of two moieties, the phenoxybenzoyl portion and the dichlorovinyl cyclopropane acid portion of the molecule. Hydroxylation and / or conjugation occur with both parts of the molecule.

The metabolism studies indicate that parent alpha-cypermethrin is usually the predominant component.

In order to analyze whether one isomer of alpha-cypermethrin was preferably metabolized, enantiomer-specific analysis of the parent compound BAS 310 I, isolated from selected matrices, was performed in goat and hen. In goats for both labels, matrix-specific differences were observed for the isomer ratio. While in feces the ratio of both isomers of BAS 310 I was found to be approximately 1:1 for both labels, the relative amount of the (S)-cyano-(1R,3R) (isomer 1) was lower compared to the (R)-cyano-(1S,3S) (isomer 2) in the other investigated matrices (milk and renal fat in the case of the benzyl label, milk, composite fat and composite muscle in the case of the cyclopropane label). The relative amounts of isomer 1 : isomer 2 ranged from values of 16.0% : 84.0% to 22.2% : 77.8%. In hens the parent compound BAS 310 I was also isolated from egg yolk and fat extracts in order to determine the enantiomer ratio. For both labels, the enantiomer ratio of BAS 310 I in egg yolk and fat was approximately 1:1, similar to the ratio of the application solutions.
Based on these findings from all studies summarised above the following residue definitions for alpha-cypermethrin in animal matrices are proposed:

For risk assessment

in milk and kidney: alpha-cypermethrin

in other animal matrices: alpha-cypermethrin

For enforcement alpha-cypermethrin

Metabolites

In order to address the relevance of the main plant and animal metabolites, the approach as laid down in the Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment, (EFSA Journal 2012;10(07): 2799) was mainly followed.
For the all metabolites identified in the metabolism studies cited in this dossier the exposure assessment showed that the Cramer class III trigger of 1.5 µg/kg bw/day was not exceeded (see MCA, chapter 6.9). Therefore, these metabolites were not included into the residue definition for risk assessment.

CA 6.7.2 Proposed maximum residue levels (MRLs) and justification of the acceptability of the levels proposed

Table 6.7.2-1 shows the existing EU MRLs as well as the MRLs proposed in this dossier:

Table 6.7.2-1: Existing and proposed MRLs for alpha-cypermethrin

Code Number	Commodity	Existing EU MRL (mg/kg)	New proposals by BASF (mg/kg)
Enforcement residue definition:		**Cypermethrin (sum of all constituent isomers)**	**Alpha-cypermethrin**
232010	Cucumber	0.2	~~0.06~~ 0.05
232030	Courgette	0.2	~~0.06~~ 0.05
243010	Chinese cabbage	1	~~1.5~~ 1.0
243020	Kale	1	~~1.5~~ 1.0
251020	Lettuce	2	1.0
~~251030~~	~~Scarole~~	~~2~~	~~1.0~~
~~251060~~	~~Rocket, Rucola~~	~~2~~	~~1.0~~
401060	Oilseed rape	0.2	0.08
500010	Barley	2	0.15
500050	Oats		0.15
500070	Rye		0.015
500090	Wheat	2	0.015

a indicates the lower limit of analytical determination

MRLs in this dossier are based on residue trials as described in chapter CA 6.3 with application rates according to the current cGAPs. MRLs are proposed based on the rounded MRLs of parent alpha-cypermethrin derived with the OECD calculator (OECD calculator spreadsheet: http://www.oecd.org/document/34/0,3746,en_2649_37465_48447010_1_1_1_37465,00.html).

Cucumber / Courgette

For cucumber 16 indoor trials are available which were performed according to the cGAP for courgette (2 indoor applications at 0.015 kg/ha, PHI 3 days for EU South or a single indoor application at 0.03 kg/ha, PHI 3 days for EU North, Central and South). The trials are described in chapter CA 6.3.1. As the residue trials in cucumbers support the critical GAP for courgette, according to European Community Guideline 7525/VI/95 rev. 9 dated March 2011 extrapolation from cucumber to courgette is adequate. The MRL calculations are based on the results of the study reported in DocID 2006/1036934 which represents the worst-case scenario (a single application at a target rate of 0.04 kg a.s./ha, target PHI 3±1 days; 8 trials). Despite the application rate deviating more than +25% from the cGAP rate, the 8 independent trials at 0.04 kg a.s./ha, are considered acceptable to support the critical GAP. Additionally, applying the proportionality concept to the dataset allowed to derive STMR and HR values and to calculate an additional MRL closer to realistic conditions.

DocID	Residues at target PHI (mg/kg)	Residues at target PHI (mg/kg) applying proportionality concept (x 0.75)	Region
2006/1036934	<0.01 (2x), 0.012, 0.014 (2x), 0.02, 0.031, 0.037	<0.01 (2x), 0.009, 0.011 (2x), 0.015, 0.023, 0.028	Europe (indoor)

Table 6.7.2-2: MRL calculation for cucumber

OECD Calculator	Alpha-cypermethrin [mg/kg]	Alpha-cypermethrin [mg/kg] applying proportionality concept
	EU, indoor	EU, indoor
Highest residue	0.037	0.028
Mean + 4 SD	0.059	0.043
CF x 3 Mean	0.046	0.037
Rounded MRL	**0.06**	**0.05**
STMR	0.014	0.011

Based on these calculations the following EU MRL is proposed according to the residue definition for enforcement (alpha-cypermethrin) and the present residue data reflecting the worst-case scenario:

0.06 mg/kg for courgettes (group 232030).

Leafy brassica

For leafy brassica 12 outdoor trials are available which were performed according to the cGAP (2 outdoor application at 0.01 kg/ha with a PHI of 3 days in Northern, Central and Southern Europe or a single outdoor application at 0.02 kg/ha with a PHI of 3 days in Southern Europe). 4 of these trials were done in EU North; in EU South, 8 trials were conducted. The trials are described in chapter CA 6.3.2. The MRL calculations are based on the results of the studies reported in DocIDs 2006/1026862, 2007/1013342 and 2013/1416285, which represent the worst-case scenario (two applications at a target rate of 0.0125 kg/ha (N-EU and S-EU) or a single application at 0.025 kg a.s./ha (S-EU), target PHI 3±1 days). At a few occasions, a higher residue level was measured at a later PHI and in other cases, at a certain PHI a higher residue level was observed after the single treatment than after the double treatment. These more critical residue values were considered for MRL calculation purposes. Despite the application rate deviating more than +25% from the cGAP rate, the independent trials at (1)-2x 0.0125 kg a.s./ha, are considered acceptable to support the critical GAP. Additionally, applying the proportionality concept to the dataset allowed to derive STMR and HR values and to calculate an additional MRL closer to realistic conditions.

DocID	Residues at target PHI (mg/kg)	Residues at target PHI (mg/kg) applying proportionality concept (x 0.8)	Region
2006/1026862, 2007/1013342	~~0.17~~ 0.23, 0.35, 0.38, 0.59	0.18, 0.28, 0.30, 0.47	Northern Europe
2006/1026862, 2007/1013342, 2013/1416285	0.019, 0.046, 0.056, 0.064, 0.086, 0.11, 0.278, 0.436	0.015, 0.037, 0.045, 0.051, 0.069, 0.088, 0.22, 0.35	Southern Europe

Table 6.7.2-3: MRL calculation for leafy brassica

OECD Calculator	Alpha-cypermethrin [mg/kg]		
	N-EU, outdoor	S-EU, outdoor	N + S
Highest residue	0.59	0.436	0.59
Mean + 4 SD	1.062 0.991	0.716	0.964
CF x 3 Mean	1.120 1.163	0.411	0.647
Rounded MRL	**1.5***	**0.8**	**1.0**
STMR	0.367	0.075	0.140
OECD Calculator	**Alpha-cypermethrin applying proportionality concept [mg/kg]**		
	N-EU, outdoor	**S-EU, outdoor**	**N + S**
Highest residue	0.470	0.350	0.470
Mean + 4 SD	0.789	0.573	0.769
CF x 3 Mean	0.923	0.328	0.526
Rounded MRL	**1.0***	**0.6**	**0.8**
STMR	0.290	0.060	0.134

*High uncertainty of MRL estimate due to small dataset

Based on the worst-case scenario calculations an EU MRL of

1.5 mg/kg for leafy brassica (group 243000)

is proposed according to the residue definition for enforcement (alpha-cypermethrin) and the present residue data.

Lettuce

For lettuce 29 outdoor trials are available which were performed according to the cGAP (2 appl. at 0.01 kg/ha with a PHI of 3 days in Northern, Central and Southern Europe or a single application at 0.02 kg/ha with a PHI of 3 days in Southern Europe, outdoor). 10 of these trials were done in EU North; in EU South, 19 trials were conducted. The trials that have not been peer-reviewed are described in chapter CA 6.3.3. One trial is considered that had already been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 (AL-726-004).

The MRL calculations are based on the results of the studies reported in DocIDs AL-726-004, 2006/1026855, 2007/1008496, 2007/1007938 and 2014/1140312, which represent the worst-case scenario (two applications at a target rate of 0.0125 kg/ha (N-EU and S-EU) or a single application at 0.025 kg a.s./ha (S-EU), target PHI 3±1 days; one application at 0.015 kg a.s./ha with a PHI of 3 days in study AL-726-004).

DocID	Residues at target PHI (mg/kg)	Region
2006/1026855, 2007/1008496, 2007/1007938	<0.01, 0.021, 0.055, 0.072, 0.12, 0.125, 0.221, 0.244[1], 0.27, 0.722	Northern Europe
2006/1026855, 2007/1008496, 2007/1007938, 2014/1140312, AL-726-004	<0.01, 0.032, 0.037, 0.049, 0.075, 0.091, 0.103, 0.11 (2x), 0.195, 0.21 (2x), 0.24, 0.28, 0.305, 0.32, 0.387, 0.41, 0.585	Southern Europe

[1] result from plot treated once with 0.0125 kg a.s./ha was higher than result from plot treated twice with 0.0125 kg a.s./ha (0.236 mg/kg)

Table 6.7.2-4: MRL calculation for lettuce

OECD Calculator	Alpha-cypermethrin [mg/kg]		
	N-EU, outdoor	**S-EU, outdoor**	**N + S**
Highest residue	0.722	0.585	0.722
Mean + 4 SD	1.024	0.814	0.880
CF x 3 Mean	0.521	0.573	0.555
Rounded MRL	**1.0**	**0.9**	**0.9**
STMR	0.123	0.195	0.125

Based on these calculations an EU MRL of

1.0 mg/kg for lettuce and salad plants (group 0251000)

is proposed according to the residue definition for enforcement (alpha-cypermethrin) and the present residue data.

Oilseed rape

For oilseed rape 29 trials were considered. 13 of these trials were done outdoor in EU North; in EU South, 16 outdoor trials were conducted. The trials that have not been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 are described in chapter CA 6.3.4.

The MRL calculations are based on the results of the studies reported in DocIDs 2008/1019999, 2013/1037957 and 2013/1416283, which represent the worst-case scenario (two applications at a target rate of 0.0105 kg/ha (N-EU and S-EU), target PHI 28±1 days). 11 trials that had already been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 were also considered (AL-750-004 – one treatment at 0.02 kg a.s./ha, AL-750-021 - one treatment at 0.02 kg a.s./ha, and 2002/1004087 – 2 treatments at 0.01 kg a.s./ha).

DocID	Residues at target PHI (mg/kg)	Region
2008/1019999, 2013/1037957, 2013/1416283, AL-750-004	<0.01 (13 x)	Northern Europe
2008/1019999, 2013/1037957, 2013/1416283, AL-750-021, 2002/1004087	<0.01 (12 x), 0.012, 0.015, <0.05, 0.06	Southern Europe

Table 6.7.2-5: MRL calculation for oilseed rape

OECD Calculator	Alpha-cypermethrin [mg/kg]		
	N-EU, outdoor	**S-EU, outdoor**	**N + S**
Highest residue	0.01	0.06	0.06
Mean + 4 SD	0.01	0.078	0.06
CF x 3 Mean	0.01	0.022	0.016
Rounded MRL	**0.01***	**0.08***	**0.06**
STMR	0.01	0.01	0.01

* High uncertainty of MRL estimate due to high level of censoring.

Based on these calculations an EU MRL of

0.08 mg/kg for oilseed rape seed (group 401060)

is proposed according to the residue definition for enforcement (alpha-cypermethrin) and the present residue data.

Barley

For barley 17 trials trials were considered. 8 of these trials were done in EU North; in EU South, 9 trials were conducted. The trials that have not been peer-reviewed within the framework of inclusion of alpha-cypermethrin into Annex I of Directive 91/414 are described in chapter CA 6.3.5. One trial is considered that had already been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 (AL-730-029).
The MRL calculations are based on the results of the studies reported in DocIDs 2013/1388974, 2013/1416281, 2013/1416284 and 2014/1173599 which represent the worst-case scenario (two applications at a target rate of 0.0125 kg/ha (N-EU and S-EU), target PHI of 28±1 days (or BBCH 89). One trial that had already been peer-reviewed was also considered - one application at 0.015 kg a.s./ha in study AL-730-029).

DocID	Residues at target PHI (mg/kg)	Region
2013/1388974, 2013/1416284, 2014/1173599	0.02, 0.03, 0.031, 0.032, 0.035, 0.053,0.077, 0.079	Northern Europe
2013/1388974, 2013/1416284, AL-730-029	<0.01, 0.024, 0.026, 0.034, 0.035, 0.05, 0.066, 0.079, 0.083,	Southern Europe

Table 6.7.2-6: MRL calculation for barley

OECD Calculator	Alpha-cypermethrin [mg/kg]		
	N-EU, outdoor	**S-EU, outdoor**	**N + S**
Highest residue	0.079	0.083	0.083
Mean + 4 SD	0.135	0.148	0.139
CF x 3 Mean	0.134	0.126	0.129
Rounded MRL	**0.15**	**0.15**	**0.15**
STMR	0.033	0.035	0.035

Based on these calculations an EU MRL of

0.15 mg/kg for barley grain (group 500010)
0.15 mg/kg for oats grain (group 500050, by extrapolation from barley according to SANCO 7525/VI/95 - rev.9, March 2011)

is proposed according to the residue definition for enforcement (alpha-cypermethrin) and the present residue data.

Wheat

For wheat 35 trials are available which were performed according to the cGAP (2 appl. at 0.01 kg/ha with a PHI of 28 days in Northern, Central and Southern Europe, outdoor). 16 of these trials were done in EU North; in EU South, 19 trials were conducted. The trials that have not been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 are described in chapter CA 6.3.6. 15 trials are considered that had already been peer-reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414 (AL-730-001, AL-730-003, AL-730-029, AL-730-030, AL-730-031).
The MRL calculations are based on the results of the studies reported in DocIDs 2008/1002701, 2013/1416282, and 2014/1028112, which represent the worst-case scenario (two applications at a target rate of 0.0125 kg/ha (N-EU and S-EU), target PHI of 28±1 days (or BBCH 89). 15 trials are considered that had already been peer-reviewed – one treatment at 0.015-0.0175 kg a.s./ha (AL-730-001, AL-730-003, AL-730-029, AL-730-030, AL-730-031).

DocID	Residues at target PHI (mg/kg)	Region
2008/1002701, 2013/1416282, 2014/1028112, AL-730-001, AL-730-003[1)], AL-730-031	<0.01 (15x), 0.01	Northern Europe
2008/1002701, 2013/1416282, 2014/1028112, AL-730-001, AL-730-003[1)], AL-730-029, AL-730-030	<0.01 (19x)	Southern Europe

[1)] AL-730-003 is an updated report of study AL-730-004 taking into account crop water content and as such presenting worse case residue data compared to AL-730-004.

Table 6.7.2-7: MRL calculation for wheat

OECD Calculator	Alpha-cypermethrin [mg/kg]		
	N-EU, outdoor	S-EU, outdoor	N + S
Highest residue	0.010	0.01	0.010
Mean + 4 SD	0.010	0.01	0.010
CF x 3 Mean	0.011	0.01	0.011
Rounded MRL	**0.015***	**0.01***	**0.015***
STMR	0.01	0.01	0.01

*High uncertainty of MRL estimate due to high level of censoring

Based on these calculations an EU MRL of

0.015 mg/kg for wheat (spelt, triticale, group 500090)
0.015 mg/kg for rye (group 500070, by extrapolation from wheat according to SANCO 7525/VI/95 - rev.9, March 2011)

is proposed according to the residue definition for enforcement (alpha-cypermethrin) and the present residue data.

Animal matrices

Feed burden calculations

Intended uses with the representative formulation BAS 310 55 I covered in this submission include the crops leafy brassica, oilseed rape and cereals (barley and wheat). These crops are relevant as feed items.

In this submission, also a risk assessment describing the uses intended by BASF in future is enclosed to provide a realistic overview beyond the representative uses. Therefore, also a new feed burden calculation based on these data is necessary.

The input values used for this calculation are summarized in Table 6.7.2-8. Detailed information on the studies and the individual residue values used for the estimation of the livestock feed burden is given in the supplement dossier.

The targeted crop treatment timing is actually excluding the timing for forage production of cereal crops (except barley). However, the current GAPs do not explicitly exclude cereal cultivation for fodder /hay/silage production, which would manly concern barley, if at all. However, the extrapolation to other cereals does reflect a worst-case situation and is therefore applied here.

Table 6.7.2-8: Input values used for feed burden calculation

Feedstuff Crop	Commodity	IFN Code	Classification	Residue input (mg/kg)				DM (%)
				max		mean		
Forages								
Barley	forage	2-00-511	R	0.38[1]	HR	0.38[1]	HR	30
Barley	hay	1-00-495	R	0.92	HR	0.40	STMR	88
Barley	straw	1-00-498	R	0.92	HR	0.40	STMR	89
Barley	silage	3-00-512	R	0.38[1]	HR	0.38[1]	HR	40
Bean	vines	2-14-388	R	1.013	HR	0.614	STMR	35
Beet, mangel	fodder	2-00-632	R	0.292	HR	0.128	STMR	15
Beet, sugar	tops	2-00-649	R	0.292	HR	0.128	STMR	23
Cabbage, heads	leaves	2-01-046	R	0.105	HR	0.034	STMR	15
Corn, field	forage/silage	3-28-345	R	0.46	HR	0.18	STMR	40
Corn, field	stover	3-28-251	R	0.46	HR	0.18	STMR	83
Corn, pop	stover	2-02-963	R	0.46	HR	0.18	STMR	85
Corn, sweet	forage	1-08-407	R	0.46	HR	0.18	STMR	48

Feedstuff Crop	Commodity	IFN Code	Classification	Residue input (mg/kg)				DM (%)
				max		mean		
Corn, sweet	stover	1-01-405	R	0.46	HR	0.18	STMR	83
Cowpea	forage	2-01-655	R	0.431	HR	0.275	STMR	30
Cowpea	hay	1-01-645	R	1.008	HR	0.589	STMR	86
Kale	leaves	2-02-446	R	0.590	HR	0.367	STMR	15
Oat	forage	2-03-292	R	0.38	HR	0.38	HR	30
Oat	hay	1-03-280	R	0.92	HR	0.40	STMR	90
Oat	straw	1-03-283	R	0.92	HR	0.40	STMR	90
Oat	silage	3-03-298	R	0.38	HR	0.38	HR	35
Pea	vines	3-03-596	R	0.431	HR	0.275	STMR	25
Pea	hay	1-03-572	R	1.008	HR	0.589	STMR	88
Pea	silage	3-03-590	R	0.431	HR	0.275	STMR	40
Rape	forage	2-03-867	R	0.270	HR	0.135	STMR	30
Rice	straw	1-03-925	R	0.354	HR	0.136	STMR	90
Rice	whole crop silage	NA	R	0.354[2]	HR	0.136[2]	STMR	40
Rye	forage	2-04-018	R	0.99	HR	0.335	STMR	30
Rye	straw	1-04-007	R	1.040	HR	0.33	STMR	88
Rye	silage	3-04-020	R	0.99	HR	0.335	STMR	28
Triticale	forage	2-02-647	R	0.99	HR	0.335	STMR	30
Triticale	hay	NA	R	1.040	HR	0.33	STMR	88
Triticale	straw	NA	R	1.040	HR	0.33	STMR	90
Triticale	silage	3-26-208	R	0.99	HR	0.335	STMR	35
Turnip	tops (leaves)	2-05-063	R	0.292	HR	0.128	STMR	30
Wheat	forage	2-08-078	R	0.99	HR	0.335	STMR	25
Wheat	hay	1-05-172	R	1.040	HR	0.33	STMR	88
Wheat	straw	1-05-175	R	1.040	HR	0.33	STMR	88
Wheat	silage	3-05-186	R	0.99	HR	0.335	STMR	30
Roots & Tubers								
Carrot	culls	2-01-146	CC	0.01	HR	0.01	STMR	12
Potato	culls	4-03-787	CC	0.01	HR	0.01	STMR	20
Swede	roots	4-04-001	CC	0.01	HR	0.01	STMR	10
Turnip	roots	4-05-067	CC	0.01	HR	0.01	STMR	15
Cereal Grains/Crops Seeds								
Barley	grain	4-00-549	CC	0.035	STMR	0.035	STMR	88
Bean	seed	4-00-515	PC	0.01	STMR	0.01	STMR	88
Corn, field	grain	4-20-698	CC	0.01	STMR	0.01	STMR	88
Corn, pop	grain	4-02-964	CC	0.01	STMR	0.01	STMR	88
Cotton	undelinted seed	5-01-614	PC	0.01	STMR	0.01	STMR	88
Cowpea	seed	5-01-661	PC	0.01	STMR	0.01	STMR	88
Lupin	seed	5-02-707	PC	0.01	STMR	0.01	STMR	88
Oat	grain	4-03-309	CC	0.035	STMR	0.035	STMR	89
Pea	seed	5-03-600	PC	0.01	STMR	0.01	STMR	90
Rice	grain	4-03-939	CC	0.092	STMR	0.092	STMR	88
Rye	grain	4-04-047	CC	0.01	STMR	0.01	STMR	88
Triticale	grain	4-20-362	CC	0.01	STMR	0.01	STMR	89
Wheat	grain	4-05-211	CC	0.01	STMR	0.01	STMR	89

Feedstuff Crop	Commodity	IFN Code	Classification	Residue input (mg/kg)				DM (%)
				max		mean		
By-Products								
Barley	bran fractions	4-00-515	CC	0.205[3]	STMR-P	0.205[3]	STMR-P	90
Beet, sugar	dried pulp	4-29-307	R	0.099[4]	STMR-P	0.099[4]	STMR-P	88
Beet, sugar	ensiled pulp	4-00-662	R	0.03	STMR	0.03	STMR	15
Beet, sugar	molasses	4-30-289	CC		STMR		STMR	75
Brewer's grain	dried	5-00-516	CC	0.033[5]	STMR-P	0.033[5]	STMR-P	92
Canola	meal	5-08-136	PC	0.01	STMR	0.01	STMR	88
Cotton	meal	5-01-617	PC	0.01	STMR	0.01	STMR	89
Distiller's grain	dried	5-00-518	CC	0.033[5]	STMR	0.033[5]	STMR	92
Flaxseed/ linseed	meal	5-02-043	PC	0.01	STMR	0.01	STMR	88
Grape	pomace, wet	2-02-206	CC	0.157[6]	STMR-P	0.157[6]	STMR-P	15
Lupin seed	meal	NA	PC	0.01	STMR	0.01	STMR	85
Potato	dried pulp	4-03-775	CC	0.033[7]	STMR-P	0.033[7]	STMR-P	88
Rape	meal	5-26-093	PC	0.01	STMR	0.01	STMR	88
Safflower	meal	5-26-095	PC	0.01	STMR	0.01	STMR	91
Tomato	pomace, wet	NA	CC	0.233[8]	STMR-P	0.233[8]	STMR-P[6]	20

1 DocID 2013/1416284; single data point, no other data for PHI=28
2 HR/STMR for straw, no data for plant at PHI
3 calculated using STMR of barley grain (0.035 mg/kg) x median TF (5.845) of pearling dust/bran from barley processing study 2007/1013068 (see MCA 6.5)
4 calculated using STMR of sugar beet root (0.03 mg/kg) x median TF (3.285) from raisin production as estimate for residue concentration during drying (DocID 2005/1014175, see supplement dossier)
5 calculated using STMR of barley grain (0.03 mg/kg) x median TF (0.93) of spent grain from barley processing studies AL-730-061 and 2007/1013068 (see MCA 6.5)
6 calculated using STMR of grapes (0.049 mg/kg) x median TF (3.2) for pomace derived from grape processing stuy (DocID 2005/1014175, see supplement dossier)
7 calculated using STMR of potatoes (0.01 mg/kg) x median TF (3.285) from raisin production as estimate for residue concentration during drying (DocID 2005/1014175, see supplement dossier)
8 calculated using STMR of tomatoes (0.037 mg/kg, glasshouse)) x median TF (6.3) from remainder of straining from tomato juice processing as estimate for residue concentration in tomato pomace (DocID 2006/1021295, see supplement dossier)

Feed commodities related to representative uses are underlined

The feed burden calculations for Europe were performed using the OECD calculator.
Two scenarios were calculated using the data listed in Table 6.7.2-8: Scenario 1 considering only the representative uses and scenario 2 considering all uses to be defended in future.

Scenario 1:
A summary is given in Table 6.7.2-9 and Table 6.7.2-10. In Table 6.7.2-11 and Table 6.7.2-12 the results of the total maximum and mean dietary burden calculations for cattle (beef and dairy), sheep (ram/ewe and lamb), swine (breeding and finishing) and poultry (broiler, layer, turkey) for the European Union (EU) are presented in detail.

Table 6.7.2-9: Summary: Maximum Feed burden calculation results for Europe (Reasonable Worst Case Feeding Levels (RWCFL)) – Scenario 1, only representative uses considered

	Beef	Dairy	Ram/ewe	Lamb	Swine breeding	Swine finishing	Poultry broiler	Poultry layer	Poultry turkey
Regions	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**
Body weight (kg)	500	650	75	40	260	100	1.7	1.9	7
Daily intake (kg DM)	12	25	2.5	1.7	6	3	0.12	0.13	0.5
Dietary Burden (mg/kg bw)	0.020	0.031	0.054	0.057	0.019	0.001	0.002	0.030	0.002
Feed Burden (mg/kg DM)	0.823	0.813	1.607	1.344	0.824	0.039	0.031	0.432	0.023

Table 6.7.2-10: Summary: Mean Feed burden calculation results for Europe (Reasonable Worst Case Feeding Levels (RWCFL)) – Scenario 1, only representative uses considered

	Beef	Dairy	Ram/ewe	Lamb	Swine breeding	Swine finishing	Poultry broiler	Poultry layer	Poultry turkey
Regions	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**
Body weight (kg)	500	650	75	40	260	100	1.7	1.9	7
Daily intake (kg DM)	12	25	2.5	1.7	6	3	0.12	0.13	0.5
Dietary Burden (mg/kg bw)	0.012	0.020	0.022	0.028	0.007	0.001	0.002	0.012	0.002
Feed Burden (mg/kg DM)	0.521	0.511	0.653	0.653	0.300	0.039	0.031	0.170	0.023

Table 6.7.2-11: Total maximum dietary burden with highest contributors for commodity groups– Scenario 1

EU								
Category	**Crop**	**Feedstuff**	**% of Diet**	**Dietary Contribution (mg/kg bw)**	**Cumulative % Diet**	**Adjusted % Diet**	**Adjusted Dietary Contribution (mg/kg bw)**	**Feed Burden (mg/kg DM)**
Beef								
Forages	Wheat	forage	20	0.019	20	20	0.019	0.792
Cereal grains	Barley	grain	70	0.001	90	70	0.001	0.028
By-products	Distiller's grain	dried	10	0.000	100	10	0.000	0.004
Roots/tubers	Turnip	roots	20	0.000	100	0	0.000	0.000
Total			120	0.020	-	100	0.020	0.823
Dairy								
Forages	Wheat	forage	20	0.030	20	20	0.030	0.792
Cereal grains	Barley	grain	40	0.001	60	40	0.001	0.016
By-products	Brewer's grain	dried	15	0.000	75	15	0.000	0.005
Roots/tubers	Turnip	roots	20	0.000	95	20	0.000	0.000
Total			**95**	**0.031**	**-**	**95**	**0.031**	**0.813**
Ram/Ewe								
Forages	Wheat	forage	40	0.053	40	40	0.053	1.584
Cereal grains	Barley	grain	40	0.001	80	40	0.001	0.016
By-products	Brewer's grain	dried	30	0.000	100	20	0.000	0.007
Roots/tubers	Turnip	roots	30	0.000	100	0	0.000	0.000
Total			**140**	**0.054**	**-**	**100**	**0.054**	**1.607**
Lamb								
Forages	Rye	forage	40	0.056	40	40	0.056	1.320
Cereal grains	Barley	grain	60	0.001	100	60	0.001	0.024
By-products	Distiller's grain	dried	10	0.000	100	0	0.000	0.000
Roots/tubers	Turnip	roots	30	0.000	100	0	0.000	0.000
Total			**140**	**0.057**	**-**	**100**	**0.057**	**1.344**
Swine breeding								
Forages	Wheat	forage	20	0.018	20	20	0.018	0.792
Cereal grains	Barley	grain	80	0.001	100	80	0.001	0.032
By-products	Distiller's grain	dried	20	0.000	100	0	0.000	0.000
Roots/tubers	Turnip	roots	40	0.000	100	0	0.000	0.000
Total			**160**	**0.019**	**-**	**100**	**0.019**	**0.824**
Swine finishing								
Cereal grains	Barley	grain	80	0.001	80	80	0.001	0.032
By-products	Distiller's grain	dried	20	0.000	100	20	0.000	0.007
Roots/tubers	Turnip	roots	40	0.000	100	0	0.000	0.000
Forages	Wheat	silage	0	0.000	100	0	0.000	0.000
Total			**140**	**0.001**	**-**	**100**	**0.001**	**0.039**
Broiler								
Cereal grains	Barley	grain	70	0.002	70	70	0.002	0.028
By-products	Distiller's grain	dried	10	0.000	80	10	0.000	0.004
Roots/tubers	Turnip	roots	10	0.000	90	10	0.000	0.000
Forages	Wheat	silage	0	0.000	90	0	0.000	0.000
Total			**90**	**0.002**	**-**	**90**	**0.002**	**0.031**
Layer								
Forages	Wheat	forage	10	0.027	10	10	0.027	0.396
Cereal grains	Barley	grain	100	0.003	100	90	0.002	0.036
By-products	Distiller's grain	dried	10	0.000	100	0	0.000	0.000
Roots/tubers	Turnip	roots	10	0.000	100	0	0.000	0.000
Total			**130**	**0.030**	**-**	**100**	**0.030**	**0.432**
Turkey								
Cereal grains	Barley	grain	50	0.001	50	50	0.001	0.020
By-products	Distiller's grain	dried	10	0.000	60	10	0.000	0.004
Roots/tubers	Turnip	roots	10	0.000	70	10	0.000	0.000
Forages	Wheat	silage	0	0.000	70	0	0.000	0.000
Total			**70**	**0.002**	**-**	**70**	**0.002**	**0.023**

Table 6.7.2-12: Total mean dietary burden with highest contributors for commodity groups– Scenario 1

EU								
Category	**Crop**	**Feedstuff**	**% of Diet**	**Dietary Contribution (mg/kg bw)**	**Cumulative % Diet**	**Adjusted % Diet**	**Adjusted Dietary Contribution (mg/kg bw)**	**Feed Burden (mg/kg DM)**
Beef								
Forages	Kale	leaves	20	0.012	20	20	0.012	0.489
Cereal grains	Barley	grain	70	0.001	90	70	0.001	0.028
By-products	Distiller's grain	dried	10	0.000	100	10	0.000	0.004
Roots/tubers	Turnip	roots	20	0.000	100	0	0.000	0.000
Total			**120**	**0.012**	**-**	**100**	**0.012**	**0.521**
Dairy								
Forages	Kale	leaves	20	0.019	20	20	0.019	0.489
Cereal grains	Barley	grain	40	0.001	60	40	0.001	0.016
By-products	Brewer's grain	dried	15	0.000	75	15	0.000	0.005
Roots/tubers	Turnip	roots	20	0.000	95	20	0.000	0.000
Total			**95**	**0.020**	**-**	**95**	**0.020**	**0.511**
Ram/Ewe								
Forages	Barley	forage	50	0.021	50	50	0.021	0.633
Cereal grains	Barley	grain	40	0.001	90	40	0.001	0.016
By-products	Brewer's grain	dried	30	0.000	100	10	0.000	0.004
Roots/tubers	Turnip	roots	30	0.000	100	0	0.000	0.000
Total			**150**	**0.022**	**-**	**100**	**0.022**	**0.653**
Lamb								
Forages	Barley	forage	50	0.027	50	50	0.027	0.633
Cereal grains	Barley	grain	60	0.001	100	50	0.001	0.020
By-products	Distiller's grain	dried	10	0.000	100	0	0.000	0.000
Roots/tubers	Turnip	roots	30	0.000	100	0	0.000	0.000
Total			**150**	**0.028**	**-**	**100**	**0.028**	**0.653**
Swine breeding								
Forages	Wheat	forage	20	0.006	20	20	0.006	0.268
Cereal grains	Barley	grain	80	0.001	100	80	0.001	0.032
By-products	Distiller's grain	dried	20	0.000	100	0	0.000	0.000
Roots/tubers	Turnip	roots	40	0.000	100	0	0.000	0.000
Total			**160**	**0.007**	**-**	**100**	**0.007**	**0.300**
Swine finishing								
Cereal grains	Barley	grain	80	0.001	80	80	0.001	0.032
By-products	Distiller's grain	dried	20	0.000	100	20	0.000	0.007
Roots/tubers	Turnip	roots	40	0.000	100	0	0.000	0.000
Forages	Wheat	silage	0	0.000	100	0	0.000	0.000
Total			**140**	**0.001**	**-**	**100**	**0.001**	**0.039**
Broiler								
Cereal grains	Barley	grain	70	0.002	70	70	0.002	0.028
By-products	Distiller's grain	dried	10	0.000	80	10	0.000	0.004
Roots/tubers	Turnip	roots	10	0.000	90	10	0.000	0.000
Forages	Wheat	silage	0	0.000	90	0	0.000	0.000
Total			**90**	**0.002**	**-**	**90**	**0.002**	**0.031**
Layer								
Forages	Wheat	forage	10	0.009	10	10	0.009	0.134
Cereal grains	Barley	grain	100	0.003	100	90	0.002	0.036
By-products	Distiller's grain	dried	10	0.000	100	0	0.000	0.000
Roots/tubers	Turnip	roots	10	0.000	100	0	0.000	0.000
Total			**130**	**0.012**	**-**	**100**	**0.012**	**0.170**

EU								
Category	**Crop**	**Feedstuff**	**% of Diet**	**Dietary Contribution (mg/kg bw)**	**Cumulative % Diet**	**Adjusted % Diet**	**Adjusted Dietary Contribution (mg/kg bw)**	**Feed Burden (mg/kg DM)**
Turkey								
Cereal grains	Barley	grain	50	0.001	50	50	0.001	0.020
By-products	Distiller's grain	dried	10	0.000	60	10	0.000	0.004
Roots/tubers	Turnip	roots	10	0.000	70	10	0.000	0.000
Forages	Wheat	silage	0	0.000	70	0	0.000	0.000
Total			**70**	**0.002**	**-**	**70**	**0.002**	**0.023**

Scenario 2:

A summary is given in Table 6.7.2-13 and Table 6.7.2-14. In Table 6.7.2-15 and Table 6.7.2-16 the results of the total maximum and mean dietary burden calculations for cattle (beef and dairy), sheep (ram/ewe and lamb), swine (breeding and finishing) and poultry (broiler, layer, turkey) for the European Union (EU) are presented in detail.

Table 6.7.2-13: Summary: Maximum Feed burden calculation results for Europe (Reasonable Worst Case Feeding Levels (RWCFL)) – Scenario 2, all uses considered

	Beef	**Dairy**	**Ram/ewe**	**Lamb**	**Swine breeding**	**Swine finishing**	**Poultry broiler**	**Poultry layer**	**Poultry turkey**
Regions	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**
Body weight (kg)	500	650	75	40	260	100	1.7	1.9	7
Daily intake (kg DM)	12	25	2.5	1.7	6	3	0.12	0.13	0.5
Dietary Burden (mg/kg bw)	0.023	0.035	0.055	0.059	0.020	0.002	0.003	0.030	0.002
Feed Burden (mg/kg DM)	0.960	0.900	1.649	1.385	0.848	0.064	0.045	0.432	0.033

Table 6.7.2-14: Summary: Mean Feed burden calculation results for Europe (Reasonable Worst Case Feeding Levels (RWCFL)) – Scenario 2, all uses considered

	Beef	**Dairy**	**Ram/ewe**	**Lamb**	**Swine breeding**	**Swine finishing**	**Poultry broiler**	**Poultry layer**	**Poultry turkey**
Regions	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**	**EU**
Body weight (kg)	500	650	75	40	260	100	1.7	1.9	7
Daily intake (kg DM)	12	25	2.5	1.7	6	3	0.12	0.13	0.5
Dietary Burden (mg/kg bw)	0.014	0.023	0.023	0.029	0.007	0.002	0.003	0.012	0.002
Feed Burden (mg/kg DM)	0.585	0.597	0.688	0.688	0.324	0.064	0.045	0.170	0.033

Table 6.7.2-15: Total maximum dietary burden with highest contributors for commodity groups– Scenario 2

EU								
Category	**Crop**	**Feedstuff**	**% of Diet**	**Dietary Contribution (mg/kg bw)**	**Cumulative % Diet**	**Adjusted % Diet**	**Adjusted Dietary Contribution (mg/kg bw)**	**Feed Burden (mg/kg DM)**
Beef								
Forages	Corn, field	forage/silage	80	0.022	80	80	0.022	0.920
By-products	Beet, sugar	ensiled pulp	25	0.001	100	20	0.001	0.040
Roots/tubers	Swede	roots	40	0.001	100	0	0.000	0.000
Cereal grains	Barley	grain	70	0.001	100	0	0.000	0.000
Total			**215**	**0.025**	**-**	**100**	**0.023**	**0.960**
Dairy								
Forages	Wheat	forage	20	0.030	20	20	0.030	0.792
By-products	Beet, sugar	ensiled pulp	40	0.003	60	40	0.003	0.080
Roots/tubers	Swede	roots	20	0.001	80	20	0.001	0.020
Cereal grains	Barley	grain	40	0.001	100	20	0.000	0.008
Total			**120**	**0.035**	**-**	**100**	**0.035**	**0.900**
Ram/Ewe								
Forages	Wheat	forage	40	0.053	40	40	0.053	1.584
By-products	Beet, sugar	dried pulp	40	0.002	80	40	0.002	0.045
Roots/tubers	Swede	roots	30	0.001	100	20	0.001	0.020
Cereal grains	Barley	grain	40	0.001	100	0	0.000	0.000
Total			**150**	**0.056**	**-**	**100**	**0.055**	**1.649**
Lamb								
Forages	Rye	forage	40	0.056	40	40	0.056	1.320
By-products	Beet, sugar	dried pulp	40	0.002	80	40	0.002	0.045
Roots/tubers	Swede	roots	30	0.001	100	20	0.001	0.020
Cereal grains	Barley	grain	60	0.001	100	0	0.000	0.000
Total			**170**	**0.060**	**-**	**100**	**0.059**	**1.385**
Swine breeding								
Forages	Wheat	forage	20	0.018	20	20	0.018	0.792
Roots/tubers	Swede	roots	40	0.001	60	40	0.001	0.040
Cereal grains	Barley	grain	80	0.001	100	40	0.000	0.016
By-products	Beet, sugar	dried pulp	20	0.001	100	0	0.000	0.000
Total			**160**	**0.020**	**-**	**100**	**0.020**	**0.848**
Swine finishing								
Roots/tubers	Swede	roots	40	0.001	40	40	0.001	0.040
Cereal grains	Barley	grain	80	0.001	100	60	0.001	0.024
By-products	Beet, sugar	dried pulp	20	0.001	100	0	0.000	0.000
Forages	Wheat	silage	0	0.000	100	0	0.000	0.000
Total			**140**	**0.003**	**-**	**100**	**0.002**	**0.064**
Broiler								
Cereal grains	Barley	grain	70	0.002	70	70	0.002	0.028
Roots/tubers	Swede	roots	10	0.001	80	10	0.001	0.010
By-products	Potato	dried pulp	20	0.001	100	20	0.001	0.008
Forages	Wheat	silage	0	0.000	100	0	0.000	0.000
Total			**100**	**0.003**	**-**	**100**	**0.003**	**0.045**
Layer								
Forages	Wheat	forage	10	0.027	10	10	0.027	0.396
Cereal grains	Barley	grain	100	0.003	100	90	0.002	0.036
Roots/tubers	Swede	roots	10	0.001	100	0	0.000	0.000
By-products	Potato	dried pulp	15	0.000	100	0	0.000	0.000
Total			**135**	**0.031**	**-**	**100**	**0.030**	**0.432**
Turkey								
Cereal grains	Barley	grain	50	0.001	50	50	0.001	0.020
Roots/tubers	Swede	roots	10	0.001	60	10	0.001	0.010
By-products	Distiller's grain	dried	10	0.000	70	10	0.000	0.004
Forages	Wheat	silage	0	0.000	70	0	0.000	0.000
Total			**70**	**0.002**	**-**	**70**	**0.002**	**0.033**

Table 6.7.2-16: Total mean dietary burden with highest contributors for commodity groups– Scenario 2

EU								
Category	**Crop**	**Feedstuff**	**% of Diet**	**Dietary Contribution (mg/kg bw)**	**Cumulative % Diet**	**Adjusted % Diet**	**Adjusted Dietary Contribution (mg/kg bw)**	**Feed Burden (mg/kg DM)**
Beef								
Forages	Kale	leaves	20	0.012	20	20	0.012	0.489
By-products	Beet, sugar	ensiled pulp	25	0.001	45	25	0.001	0.050
Roots/tubers	Swede	roots	40	0.001	85	40	0.001	0.040
Cereal grains	Barley	grain	70	0.001	100	15	0.000	0.006
Total			**155**	**0.015**	**-**	**100**	**0.014**	**0.585**
Dairy								
Forages	Kale	leaves	20	0.019	20	20	0.019	0.489
By-products	Beet, sugar	ensiled pulp	40	0.003	60	40	0.003	0.080
Roots/tubers	Swede	roots	20	0.001	80	20	0.001	0.020
Cereal grains	Barley	grain	40	0.001	100	20	0.000	0.008
Total			**120**	**0.023**	**-**	**100**	**0.023**	**0.597**
Ram/Ewe								
Forages	Barley	forage	50	0.021	50	50	0.021	0.633
By-products	Beet, sugar	dried pulp	40	0.002	90	40	0.002	0.045
Roots/tubers	Swede	roots	30	0.001	100	10	0.000	0.010
Cereal grains	Barley	grain	40	0.001	100	0	0.000	0.000
Total			**160**	**0.024**	**-**	**100**	**0.023**	**0.688**
Lamb								
Forages	Barley	forage	50	0.027	50	50	0.027	0.633
By-products	Beet, sugar	dried pulp	40	0.002	90	40	0.002	0.045
Roots/tubers	Swede	roots	30	0.001	100	10	0.000	0.010
Cereal grains	Barley	grain	60	0.001	100	0	0.000	0.000
Total			**180**	**0.031**	**-**	**100**	**0.029**	**0.688**
Swine breeding								
Forages	Wheat	forage	20	0.006	20	20	0.006	0.268
Roots/tubers	Swede	roots	40	0.001	60	40	0.001	0.040
Cereal grains	Barley	grain	80	0.001	100	40	0.000	0.016
By-products	Beet, sugar	dried pulp	20	0.001	100	0	0.000	0.000
Total			**160**	**0.008**	**-**	**100**	**0.007**	**0.324**
Swine finishing								
Roots/tubers	Swede	roots	40	0.001	40	40	0.001	0.040
Cereal grains	Barley	grain	80	0.001	100	60	0.001	0.024
By-products	Beet, sugar	dried pulp	20	0.001	100	0	0.000	0.000
Forages	Wheat	silage	0	0.000	100	0	0.000	0.000
Total			**140**	**0.003**	**-**	**100**	**0.002**	**0.064**
Broiler								
Cereal grains	Barley	grain	70	0.002	70	70	0.002	0.028
Roots/tubers	Swede	roots	10	0.001	80	10	0.001	0.010
By-products	Potato	dried pulp	20	0.001	100	20	0.001	0.008
Forages	Wheat	silage	0	0.000	100	0	0.000	0.000
Total			**100**	**0.003**	**-**	**100**	**0.003**	**0.045**
Layer								
Forages	Wheat	forage	10	0.009	10	10	0.009	0.134
Cereal grains	Barley	grain	100	0.003	100	90	0.002	0.036
Roots/tubers	Swede	roots	10	0.001	100	0	0.000	0.000
By-products	Potato	dried pulp	15	0.000	100	0	0.000	0.000
Total			**135**	**0.013**	**-**	**100**	**0.012**	**0.170**
Turkey								
Cereal grains	Barley	grain	50	0.001	50	50	0.001	0.020
Roots/tubers	Swede	roots	10	0.001	60	10	0.001	0.010
By-products	Distiller's grain	dried	10	0.000	70	10	0.000	0.004
Forages	Wheat	silage	0	0.000	70	0	0.000	0.000
Total			70	0.002	-	70	0.002	0.033

Below it is shown which residues are to be expected in animal matrices based on the results of the feeding studies (see CA 6.4) as well as the estimation of the livestock dietary burden as shown above. All calculated livestock dietary burdens are below the lowest dose levels applied in the feeding studies. Therefore, the estimation of the expected residues in animal commodities is based on the residues found in the lowest dose groups.

Expected residues based on scenario 1 (representative uses only) are given in table Table 6.7.2-17, residues based on scenario 2 (all uses) are summarized in Table 6.7.2-18.

The results clearly show that the expected alpha-cypermethrin residues in animal matrices are well below the residue levels found in the feeding studies in the lowest dose groups for all scenarios assessed.

Table 6.7.2-17: Expected residues in animal matrices: Scenario 1, representative uses only

Matrix	Results of feeding study			Feed burden				Expected residues[1]	
	Dose level		Residue (mg/kg)	Max		Mean		Alpha-cyper-methrin (mg/kg)	
	mg/kg DM	mg/kg bw/d		mg/kg DM	mg/kg bw/d	mg/kg DM	mg/kg bw/d	Max	Mean
Bovine muscle	3.9[2]	0.14[2]	<0.05	0.823	0.020	0.521	0.012	0.011	0.007
Bovine fat	3.9	0.14	0.064	0.823	0.020	0.521	0.012	0.014	0.009
Bovine liver	3.9	0.14	<0.05	0.823	0.020	0.521	0.012	0.011	0.007
Bov. kidney	3.9	0.14	<0.05	0.823	0.020	0.521	0.012	0.011	0.007
Sheep muscle	3.9	0.14	<0.05	1.607	0.054	0.653	0.028	0.021	0.008
Sheep fat	3.9	0.14	0.064	1.607	0.054	0.653	0.028	0.026	0.011
Sheep liver	3.9	0.14	<0.05	1.607	0.054	0.653	0.028	0.021	0.008
Sheep kidney	3.9	0.14	<0.05	1.607	0.054	0.653	0.028	0.021	0.008
Swine muscle	3.9	0.14	<0.05	0.824	0.019	0.300	0.007	0.011	0.004
Swine fat	3.9	0.14	0.064	0.824	0.019	0.300	0.007	0.014	0.005
Swine liver	3.9	0.14	<0.05	0.824	0.019	0.300	0.007	0.011	0.004
Swine kidney	3.9	0.14	<0.05	0.824	0.019	0.300	0.007	0.011	0.004
Poultry meat	15[3,4]	0.965	<0.05	0.432	0.030	0.170	0.012	0.014	0.005
Poultry fat	1.6[3]	0.0987	<0.05	0.432	0.030	0.170	0.012	0.014	0.005
Poultry liver	15[3,4]	0.965	<0.05	0.432	0.030	0.170	0.012	0.014	0.005
Milk	3.9	0.14	<0.01	0.813	0.031	0.521	0.012	0.002	0.001
Poultry egg	1.6[3]	0.0987	<0.01	0.432	0.030	0.170	0.012	0.003	0.001

1 expected residues were calculated by applying the transfer factor (residue level in milk, eggs or tissue / residue level in diet) at the lowest feeding level to the dietary burden. For sheep and swine tissues, results from the cow feeding study were applied.
2 dose level calculated based on a daily dose (capsule dosing)) of 77 mg alpha-cypermethrin per animal with an estimated weight of 550 kg; dietary equivalent is based on a theoretical consumption of 20 kg dry material/animal/day
3 actual feed level
4 tissue samples from lower dose groups were not analysed because no residues above the LOQ were detected in tissue samples from the highest dose group. For calculation of expected residues the dietary burden of the lowest dose group (1.6 mg/kg DM) was used.

Table 6.7.2-18: Expected residues in animal matrices: Scenario 2, all uses

Matrix	Results of feeding study			Feed burden				Expected residues[1]	
	Dose level		Residue (mg/kg)	Max		Mean		Alpha-cyper-methrin (mg/kg)	
	mg/kg DM	mg/kg bw/d		mg/kg DM	mg/kg bw/d	mg/kg DM	mg/kg bw/d	Max	Mean
Bovine muscle	3.9[2]	0.14[2]	<0.05	0.960	0.023	0.597	0.023	0.012	0.008
Bovine fat	3.9	0.14	0.064	0.960	0.023	0.597	0.023	0.016	0.010
Bovine liver	3.9	0.14	<0.05	0.960	0.023	0.597	0.023	0.012	0.008
Bov. kidney	3.9	0.14	<0.05	0.960	0.023	0.597	0.023	0.012	0.008
Sheep muscle	3.9	0.14	<0.05	1.649	0.055	0.688	0.029	0.021	0.009
Sheep fat	3.9	0.14	0.064	1.649	0.055	0.688	0.029	0.027	0.011
Sheep liver	3.9	0.14	<0.05	1.649	0.055	0.688	0.029	0.021	0.009
Sheep kidney	3.9	0.14	<0.05	1.649	0.055	0.688	0.029	0.021	0.009
Swine muscle	3.9	0.14	<0.05	0.848	0.020	0.324	0.007	0.011	0.004
Swine fat	3.9	0.14	0.064	0.848	0.020	0.324	0.007	0.014	0.005
Swine liver	3.9	0.14	<0.05	0.848	0.020	0.324	0.007	0.011	0.004
Swine kidney	3.9	0.14	<0.05	0.848	0.020	0.324	0.007	0.011	0.004
Poultry meat	15[3,4]	0.965	<0.05	0.432	0.030	0.170	0.012	0.014	0.005
Poultry fat	1.6[3]	0.0987	<0.05	0.432	0.030	0.170	0.012	0.014	0.005
Poultry liver	15[3,4]	0.965	<0.05	0.432	0.030	0.170	0.012	0.014	0.005
Milk	3.9	0.14	<0.01	0.960	0.023	0.597	0.023	0.002	0.002
Poultry egg	1.6[3]	0.0987	<0.01	0.432	0.030	0.170	0.012	0.003	0.001

1 expected residues were calculated by applying the transfer factor (residue level in milk, eggs or tissue / residue level in diet) at the lowest feeding level to the dietary burden. For sheep and swine tissues, results from the cow feeding study were applied
2 dose level calculated based on a daily dose (capsule dosing)) of 77 mg alpha-cypermethrin per animal with an estimated weight of 550 kg; dietary equivalent is based on a theoretical consumption of 20 kg dry material/animal/day
3 actual feed level
4 tissue samples from lower dose groups were not analysed because no residues above the LOQ were detected in tissue samples from the highest dose group. For calculation of expected residues the dietary burden of the lowest dose group (1.2 mg/kg DM) was used.

Fish feed burden calculation

A fish feed burden calculation for alpha-cypermethrin based on the critical GAPs of the representative uses and all uses to be defended in future (Scenario 2) was performed using the DietaryBurdenCalculator 1.0.4 (Fraunhofer IME Schmallenberg, Germany). The model was used for carp and rainbow trout, considering the maximum reasonable balanced diet, as well as compositions by adding untreated carborhydrate concentrate, protein concentrate and fat in every combination possible.

The input values used for this calculation are summarized in the table below. Detailed information on the studies and the individual residue values used for the estimation of the livestock feed burden is given in the supplement dossier.

Table 6.7.2-19: Residue values used for calculation of the fish feed burden, Scenario 2

Crop	IFN Code	Residue value type	Residue input value (mg/kg)
Barley bran fractions	4-00-515	STMR-P	0.203[1]
Brewer's grain	5-00-516	STMR-P	0.033[2]
Cottonseed meal	5-01-617	STMR	0.01
Distiller's grain	5-00-518	STMR	0.033[2]
Linseed meal	5-02-048	STMR	0.01
Rape seed meal	5-26-093	STMR	0.01
Canola meal	5-08-136	STMR	0.01
Safflower meal	5-26-095	STMR	0.01
Corn grain	4-20-698	STMR	0.01
Cow pea seed	5-01-661	STMR	0.01
Faba bean seed		STMR	0.01
Lupin seed	5-02-707	STMR	0.01
Pea seed	5-03-600	STMR	0.01
Rice broken grains	4-03-939	STMR	0.092
Wheat grain	4-05-211	STMR	0.01

1 calculated using STMR of barley grain (0.035 mg/kg) x median TF (5.79) of pearling dust/bran from barley processing study 2007/1013068 (see MCA 6.5)

2 calculated using STMR of barley grain (0.035 mg/kg) x median TF (0.95) of spent grain from barley processing studies AL-730-061 and 2007/1013068 (see MCA 6.5)

The results presented in Table 6.7.2-20 below show that the expected alpha-cypermethrin residues in fish feed are well below the lowest dose level of 10 mg/kg administered to rainbow trout (see study CA 6.2.5/1).

Table 6.7.2-20: Summary: Fish feed burden calculation, Scenario 2

	Maximum residue burden in fish feed (mg/kg)	
	Common carp	**Rainbow trout**
Without additions	0.048	-
Without additions (MRBD)	-	-
Adding of PC	0.079	0.078
Adding of PC (MRBD)	0.079	0.055
Adding of CC	0.048	-
Adding of CC (MRBD)	-	-
Adding of fat/oil	0.048	-
Adding of fat/oil (MRBD)	-	-
Adding of PC and CC	0.079	0.078
Adding of PC and CC (MRBD)	0.079	0.055
Adding of PC and fat/oil	0.079	0.078
Adding of PC and fat/oil (MRBD)	0.079	0.056
Adding of CC and fat/oil	0.048	-
Adding of CC and fat/oil (MRBD)	-	-
Adding of PC, CC and fat/oil	0.079	0.078
Adding of PC, CC and fat/oil (MRBD)	0.079	0.056
Worst case feed burden	**0.079**	**0.078**

MRBD maximum reasonable balanced diet
PC protein concentrate (fish meal)
CC carbohydrate concentrate (starch)

In Table 6.7.2-21 it is shown which residues are to be expected in fish matrices based on the results of the fish metabolism study conducted (see CA 6.2.5) as well as the estimation of the fish dietary burden as shown above. The feed burden is well below the lowest dose levels applied in the metabolism study. Therefore, the expected residues are estimated based on the 10 mg/kg dose group by multiplying with a transfer factor (0.0079). The results are given in Table 6.7.2-21.

The results clearly show that the expected alpha-cypermethrin residues in fish matrices are below the limit of quantitation.

Table 6.7.2-21: Expected residues in fish matrices: Scenario 2

Matrix	Metabolite	[Benzyl-U-^{14}C]-BAS 310 (mg/kg)		[Cyclopropane-^{14}C]-BAS 310 I (mg/kg)	
		Residue at 10 mg/kg*	Maximum residue expected**	Residue at 10 mg/kg	Maximum residue expected**
Composite muscle	BAS 310 I	0.071	0.000561	0.060	0.000474
	Hydroxylated BAS 310 I	0.009	0.000071	0.006	0.000047
Composite skin	BAS 310 I	0.130	0.001027	0.091	0.000719
	Hydroxylated BAS 310 I	0.018	0.000142	0.012	0.000095
Liver	BAS 310 I	0.017	0.000134	0.011	0.000087
	Hydroxylated BAS 310 I	0.013	0.000103	0.013	0.000103
	Glucuronide conjugate	0.061	0.000482	0.059	0.000466

* results of fish metabolism study CA 6.2.5/1

** extrapolated by multiplying with transfer factor based on maximum dietary burden (0.0079 = 0.079 mg/kg / 10 mg/kg)

CA 6.7.3 Proposed maximum residue levels (MRLs) and justification of the acceptability of the levels proposed for imported products (import tolerance)

Plant Products

According to the proposed residue definition for MRL setting of

Alpha-cypermethrin parent

and referring to MRL derivations in chapter CA 6.7.2 it is proposed to establish EU MRLs of:

0.06 mg/kg for courgettes (group 232030)
0.08 mg/kg for oilseed rape seed (group 401060)
0.15 mg/kg for barley grain (group 500010)
0.15 mg/kg for oats grain (group 500050)
0.015 mg/kg for wheat (spelt, triticale) grain (group 500090)
0.015 mg/kg for rye grain (group 500070)

Animal products

According to the proposed residue definition for MRL setting of

Alpha-cypermethrin parent

and referring to MRL derivations in chapter CA 6.7.2 it is proposed to establish EU MRLs of:

0.05 mg/kg for ruminant and pig meat, liver and kidney
0.1 mg/kg for ruminant fat
0.05 mg/kg for pig fat
0.01 mg/kg for milk
0.01 mg/kg for eggs

CA 6.8 Proposed safety intervals

Residue trials have been conducted with applications made at the latest recommended crop growth stage with harvest taking place at the time of crop maturity following good agricultural practice.

Pre-harvest interval
In cucumber / courgette application at the growth stage BBCH 10-89 with the pre-harvest interval of 3 days (indoor) is intended. For leafy ~~cabbages~~ brassica application is possible during growth stage BBCH 10-49 with pre-harvest intervals of 3 days (outdoor). For lettuce application at the growth stage BBCH 10-49 with the pre-harvest interval of 3 days (outdoor) is intended.
For oilseed rape application is possible during growth stage BBCH 51-59 with the pre-harvest interval of 28 days (outdoor). For cereals application at the growth stage BBCH 51-83 with the pre-harvest interval of 28 days (outdoor) is intended.

Re-entry period for livestock to areas to be grazed
Because alpha-cypermethrin is not intended to be used in areas to be grazed, no re-entry period for livestock has to be defined.

Re-entry period for man to treated crops
Re-entry assessments are given for the representative uses in the supplemental product dossiers (M-CP 7.2). Re-entry is possible after the spray deposits on the crops have dried given the worker is wearing adequate work clothing.

Witholding period for animal feed stuffs
Treated cereals, oil seed rape and leafy ~~cabbage~~ brassica feed items may be used as fodder for livestock. Alpha-cypermethrin derived residues in those feed items are assessed in M-CA 6.7 by providing updated calculations of livestock dietary burdens and deriving suitable MRLs for animal products covering the intended uses. There is no additional withholding period needed for animal feeds with regard to alpha-cypermerthrin derived residues in feed items.

Waiting period between application and crop sowing or planting the crop to be protected
No waiting period is necessary since alpha-cypermethrin is not intended in pre-emergence use.

Waiting period between application and handling treated produce
This is not relevant here since a post-harvest treatment is not intended for cucumber / courgette, leafy cabbage, lettuce, oilseed rape and cereals.

Waiting period between last application and sowing or planting succeeding crops
No replant restrictions are needed; no detectable (0.01 mg/kg) residues of BAS 310 I (cis-2 isomer of cypermethrin) or any of the three other cypermethrin isomers (cis-1, trans-3, or trans-4) are expected in succeeding crops when BAS 310 I is applied at the proposed GAP. Refer to succeeding crop studies (Alphacypermethrin EU Dossier, Document M-II/Addendum, Section 6.6, 2002).

CA 6.9 Estimation of the potential and actual exposure through diet and other sources

Assessments of the potential chronic and acute dietary consumer risk due to exposure to residues of alpha-cypermethrin were performed using the EFSA model for chronic and acute risk assessment - rev. 2_0 (Model PRIMo). The EFSA model was used since it considers all the different diets in the EU and all consumer groups.
The ADI and ARfD for the active substance alpha-cypermethrin are summarized in the table below.

Table 6.9-1: Toxicological endpoints – alpha-cypermethrin

Endpoint	Value	Study	Safety factor	Reference
Acceptable Daily Intake (ADI)	0.02 mg/kg bw/d	1-year study in dogs	100	EFSA Review Report (2004), SANCO/4335/2000
Acute Reference Dose (ARfD)	0.04 mg/kg bw/d	Acute neurotoxicity study in rats	100	EFSA Review Report (2004), SANCO/4335/2000

CA 6.9.1 Acceptable Daily Intake (ADI) and Dietary Exposure Calculation

TMDI calculation

The calculation of the TMDI was performed for the representative crops cucumber / courgette, leafy brassica, oilseed rape, barley and wheat using the maximum residue levels summarised in Table 6.9.1-1. These MRLs were calculated in section M-CA 6.7 based on the residues data from section M-CA 6.3. For lettuce, oilseed rape, barley and wheat residue trials have been considered for the MRL calculations that had already been peer reviewed within the framework of the original inclusion of alpha-cypermethrin into Annex I of Directive 91/414, as described in Section M-CA 6.7. For the reviewer´s convenience, these data are summarized in the supplement information document (DocID 2014/1314800).
Beyond that, in the risk assessment also calculated MRLs, highest residues (HRs) and STMRs of residue values for crops were considered as a second scenario, which are on the one hand not foreseen in context of this submission, but are on the other hand relevant for BASF and chronic dietary risk assessment. All those residue studies were conducted according to long-term GAPs with a reduced number of applications, compared to GAPs being valid some years ago.
Calculated MRLs, HRs and STMRs are summarized in Table 6.9.1-1. Summaries of the individual studies are included as supplemental information document (DocID 2014/1314800) to this dossier. The supplement information document also includes an Appendix where all individual residue data are tabulated with their corresponding DocID, trial number and the PHI.

For animal matrices, the livestock dietary burden calculations showed that the estimated dietary burden is well below the lowest feeding level applied in the ruminant and poultry feeding studies (see M-CA 6.7). Therefore, the residue levels used to calculate the ADI contribution from animal matrices are based on the residues found in the feeding studies at the lowest dose levels.

Table 6.9.1-1: Calculated MRLs, HRs and STMRs for alpha-cypermethrin and input values for risk assessments

Code Number	Crop	EU Region	Per EU Region		N+S		Calc. MRL (mg/kg)	Input values for chronic RA			
								Scenario 1: Representative uses only		Scenario 2: all crops to be defended in future	
			HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]		(mg/kg)	comment	(mg/kg)	comment
FOOD OF PLANT ORIGIN											
151010	Table grapes	N	0.064	0.032	0.069	0.041	0.15	-	-	0.15	MRL
151020	Wine grapes	S	0.069	0.049						0.15	
152000	Strawberry	N	0.031	0.01	0.054	0.01	0.1	-	-	0.10	MRL
		S	0.028	0.01							
		Indoor	0.054	0.02							
161030	Table olives	S	0.26	0.05	-	-	0.4	-	-	0.40	MRL
211000	Potato	N	0.01	0.01	0.01	0.01	0.01	-	-	0.01	MRL
		S	0.01	0.01							
213020	Carrot	N	0.01	0.01	-	-	0.01	-	-	0.01	MRL
213030	Celeriac	Extrapolation from carrot						-	-	0.01	MRL
213040	Horseradish	Extrapolation from carrot						--	-	0.01	MRL
213070	Parsley root	Extrapolation from carrot						-	-	0.01	MRL
213080	Radishes	Extrapolation from carrot						-	-	0.01	MRL
213090	Salsify	Extrapolation from carrot						-	-	0.01	MRL
213100	Swedes	Extrapolation from carrot						-	-	0.01	MRL
213110	Turnips	Extrapolation from carrot						-	-	0.01	MRL
220010	Garlic	Extrapolation from onion						-	-	0.01	MRL
220020	Onion	N	0.01	0.01	-	-	0.01	-	-	0.01	MRL
231010	Tomato	N	0.01	0.03	0.037	0.01	0.06	-	-	0.06	MRL
		S	0.021	0.01							
		Indoor	0.037	0.016							
231020	Pepper, sweet	S	0.28	0.013	0.033	0.017	0.05[1)]	-	-	0.05	MRL
	Pepper, sweet	Indoor	0.033	0.018							
231030	Aubergines	Extrapolation from tomato						-	-	0.06	
232010	Cucumber	Indoor	0.012	0.010	0.037	0.010	0.06	-	-	0.06	MRL
		Indoor	0.37	0.014							
232030	**Courgettes**	**Extrapolation from cucumber**						**0.06**	**MRL**	**0.06**	**MRL**
233010	Melon	S	0.010	0.014	0.010	0.048	0.08	-	-	0.08	MRL
		Indoor	0.01	0.048							

Code Number	Crop	EU Region	Per EU Region		N+S		Calc. MRL (mg/kg)	Input values for chronic RA			
								Scenario 1: Representative uses only		Scenario 2: all crops to be defended in future	
			HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]		(mg/kg)	comment	(mg/kg)	comment
233020	Pumpkin	Extrapolation from melon						-	-	0.08	MRL
241010	Broccoli	N	0.029	0.016	0.047	0.018	0.1	-	-	0.10	MRL
		S	0.047	0.03							
241020	Cauliflower	N	0.085	0.01	0.085	0.01	0.15	-	-	0.15	MRL
		S	0.083	0.01							MRL
242010	Brussels sprouts	N	0.046	0.018	0.046	0.016	0.07	-	-	0.07	MRL
		S	0.029	0.012							
242020	Cabbage, head	N	0.105	0.034	0.105	0.0.014	0.2	-	-	0.20	MRL
		S	0.013	0.012							
243010	Chinese cabbage	**N**	**0.59**	**0.367**	**0.59**	**0.14**	1.5	1.5		**1.5**	**MRL**
243020	**Kale**	**S**	**0.436**	**0.075**			**1.5**	**1.5**	**MRL**	**1.5**	
251010	Lamb's lettuce	Extrapolation from lettuce							-	1.0	MRL
251020	**Lettuce**	**N**	**0.722**	**0.123**	**0.722**	**0.125**	**1.0**	**1.0**	**MRL**	**1.0**	**MRL**
		S	**0.585**	**0.195**							
251030	Scarole	Extrapolation from lettuce								1.0	MRL
251060	Rocket, Rucola	Extrapolation from lettuce								1.0	MRL
252010	Spinach	N	1.113	0.465	-	-	2.0	-	-	2.0	MRL
260010	Green beans with pods	N	0.026	0.016	0.063	0.020	0.15	-	-	0.15	MRL
		S	0.063	0.034							
260020	Beans without pods	Extrapolation from peas without pods						-	-	0.01	MRL
260030	Peas with pods	Extrapolation from beand with pods						-	-	0.15	MRL
260040	Green peas without pods	N	0.01	0.01	0.01	0.01	0.01	-	-	0.01	MRL
		S	0.01	0.01							
270010	Asparagus	Edible parts harvested before treatment					0.01	-	-	0.01	MRL
270030	Celery	N	0.30	0.22	-	-	0.7	-	-	0.70	MRL
270050	Artichoke	S	0.04	0.023	-	-	0.08	-	-	0.08	MRL
270060	Leek	N	0.071	0.044	0.105	0.044	0.2	-	-	0.20	MRL
		S	0.105	0.047							MRL
300010	Dry beans	N	0.01	0.01	0.01	0.01	0.01	-	-	0.06	MRL
		S	0.01	0.01							MRL
300030	Dry peas	N	0.01	0.01	0.042	0.01	0.06	-	-	0.06	MRL
		S	0.042	0.01							
401060	**Rape seed**	**N**	**0.01**	**0.01**	**0.06**	**0.01**	**0.08**	**0.08**	**MRL**	**0.08**	**MRL**
		S	**0.06**	**0.01**							
401080	Mustard seed	Extrapolation from rape seed						-	-	0.08	MRL
401090	Cotton seed	S	0.018	0.01	-	-	0.03	-	-	0.03	MRL
402010	Olives for oil production	Data for table olives used						-	-	0.4	MRL
500010	**Barley grain**	**N**	**0.079**	**0.033**	**0.083**	**0.035**	**0.15**	**0.15**	**-**	**0.15**	**MRL**
		S	**0.083**	**0.035**							
500030	Maize	N	0.01	0.01	0.01	0.01	0.01	-	-	0.01	MRL
		S	0.01	0.01							

Code Number	Crop	EU Region	Per EU Region		N+S		Calc. MRL (mg/kg)	Input values for chronic RA			
								Scenario 1: Representative uses only		Scenario 2: all crops to be defended in future	
			HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]		(mg/kg)	comment	(mg/kg)	comment
500050	Oats	Extrapolation from barley						**0.15**	**MRL**	0.15	MRL
500060	Rice	S	0.206	0.092	-	-	0.4	-	-	0.40	MRL
500070	Rye	Extrapolation from wheat						**0.015**	**MRL**	0.015	MRL
500090	**Wheat grain**	**N**	**0.01**	**0.01**	**0.01**	**0.01**	**0.015**	**0.015**	**MRL**	**0.015**	**MRL**
		S	**0.01**	**0.01**							
900010	Sugar beet root	N	0.228	0.03	0.228	0.01	0.4	-	-	0.4	MRL
		S	0.01	0.01							
FOOD OF ANIMAL ORIGIN-											
1011010	Swine: Meat							0.05		0.05	
1011020	Swine: Fat free of lean meat							0.05		0.05	
1011030	Swine: Liver							0.05		0.05	
1011040	Swine: Kidney							0.05		0.05	
1011050	Swine: Edible offal							0.05		0.05	
1011990	Other swine products							0.05		0.05	
1012010	Bovine: Meat							0.05		0.05	
1012020	Bovine: Fat							0.10		0.10	
1012030	Bovine: Liver							0.05		0.05	
1012040	Bovine: Kidney							0.05		0.05	
1012050	Bovine: Edible offal							0.05		0.05	
1012990	Other bovine products							0.05		0.05	
1013010	Sheep: Meat							0.05		0.05	
1013020	Sheep: Fat							0.10		0.10	
1013030	Sheep: Liver							0.05		0.05	
1013040	Sheep: Kidney							0.05		0.05	
1013050	Sheep: Edible offal							0.05		0.05	
1013990	Other sheep products							0.05		0.05	
1014010	Goat: Meat							0.05		0.05	
1014020	Goat: Fat							0.10		0.10	
1014030	Goat: Liver							0.05		0.05	
1014040	Goat: Kidney							0.05		0.05	
1014050	Goat: Edible offal							0.05		0.05	
1014990	Other goat products							0.05		0.05	
1016010	Poultry: Meat							0.05		0.05	
1016020	Poultry: Fat							0.05		0.05	
1016030	Poultry: Liver							0.05		0.05	
1016040	Poultry: Kidney							0.05		0.05	
1016050	Poultry: Edible offal							0.05		0.05	
1016990	Other poultry products							0.05		0.05	
1020010	Cattle milk							0.01		0.01	

Code Number	Crop	EU Region	Per EU Region		N+S		Calc. MRL (mg/kg)	Input values for chronic RA			
								Scenario 1: Representative uses only		Scenario 2: all crops to be defended in future	
			HR [mg/kg]	STMR [mg/kg]	HR [mg/kg]	STMR [mg/kg]		(mg/kg)	comment	(mg/kg)	comment
1020020	Sheep milk							0.01		0.01	
1020030	Goat milk							0.01		0.01	
1020040	Horse milk							0.01		0.01	
1020990	Others milk							0.01		0.01	
1030000	Bird eggs							0.01		0.01	

Data for representative uses underlined in bold
1) calculation based on outdoor use

Based on the input values as shown in the above table the following results for the chronic risk assessments are obtained:

Scenario 1 shows that considering only the representative uses as submitted in this dossier would lead to an utilisation of the ADI of 0.1 to 4.6 %. The diet with the highest TMDI is "NL child" with 4.6% of ADI. For this diet, the highest contributor is cattle milk with 1.5% of ADI. The diet with the second highest TMDI is “ES child” “with 4.2% of ADI where lettuce is the major contributor (2.1% of ADI).

Scenario 2 shows the chronic risk assessment including those crops which are to be defended in future. The ADI of 0.02 mg/kg bw/d is utilised to 1.5-49.1%. The diet with the highest TMDI is "UK toddler" with 49.1 % of ADI. For this diet, the highest contributor is sugar beet root with 45.7% of ADI. The diet with the second highest TMDI is “UK infant” with 23.2% of ADI where also sugar beet (root) is the major contributor (20.2 % of ADI).

Table 6.9.1-2: Results of chronic risk assessments

Input values	Highest exposure	
	% of ADI	Diet
Residue definition for risk assessment:	alpha-cypermethrin	
ADI:	0.02 mg/kg bw/d	
Scenario 1: Representative uses only	4.6%	NL child
Scenario 2: all crops to be defended in future	49.1%	UK toddler

According to the presented TMDI calculations a long-term intake of alpha-cypermethrin residues is unlikely to present a public health concern, even when the worst case situation is considered.

NEDI calculation

For all population groups included in the EFSA model, the use of STMR or STMRP values in the estimation of the chronic dietary consumer risk is up to this point in time not necessary since the crude overestimated TMDI of alpha-cypermethrin was well below 100% of the ADI for the representative uses courgette, kale, lettuce and cereals as well as under consideration of the contribution of all uses.

NESTI calculations

The acute dietary risk assessment was performed according to the EFSA PRIMo model, applying the calculated MRLs for the representative uses shown in Table 6.9.1-1.

An acute Reference Dose (ARfD) of 0.04 mg/kg bw was used. The results of the IESTI calculations are summarised below and in Table 6.9.1-5.

Alpha-cypermethrin

Prepare workbook for refined calculations

Undo refined calculations

Status of the active substance:		Code no.	
LOQ (mg/kg bw):	0,01	proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	**0,02**	ARfD (mg/kg bw):	**0,04**
Source of ADI:		Source of ARfD:	
Year of evaluation:		Year of evaluation:	

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI
minimum – maximum
2 49

No of diets exceeding ADI: ---

Highest calculated TMDI values in % of ADI	MS Diet	contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
49,1	UK Toddler	45,7	Sugar beet (root)	1,2	Rice	0,3	Wheat	0,3
23,2	UK Infant	20,2	Sugar beet (root)	1,3	Rice	0,2	Wheat	0,4
16,0	WHO Cluster diet B	3,8	Olives for oil production	1,8	Lettuce	1,4	Sugar beet (root)	0,5
13,1	NL child	3,7	Spinach	1,5	Milk and milk products: Cattle	1,2	Kale	1,9
12,4	FR toddler	7,1	Spinach	0,8	Beans (with pods)	0,7	Leek	0,5
11,3	UK vegetarian	7,6	Sugar beet (root)	0,8	Rice	0,7	Lettuce	0,1
11,2	UK Adult	8,0	Sugar beet (root)	0,8	Wine grapes	0,7	Rice	0,1
8,6	FR infant	4,4	Spinach	1,3	Milk and milk products: Cattle	0,6	Beans (with pods)	1,7
8,5	ES child	2,1	Lettuce	1,5	Olives for oil production	1,0	Rice	0,8
7,9	IE adult	1,3	Spinach	0,9	Wine grapes	0,9	Barley	0,5
7,5	WHO regional European diet	1,9	Lettuce	0,4	Rice	0,4	Head cabbage	0,5
7,4	ES adult	2,7	Lettuce	0,8	Olives for oil production	0,7	Spinach	0,3
7,2	DE child	2,0	Spinach	1,0	Table grapes	0,7	Milk and milk products: Cattle	1,0
7,1	WHO cluster diet E	1,2	Wine grapes	0,6	Barley	0,5	Lettuce	0,5
7,1	NL general	1,4	Spinach	0,7	Kale	0,6	Lettuce	0,5
6,4	WHO cluster diet D	1,4	Chinese cabbage	1,1	Rice	0,7	Kale	0,6
6,2	SE general population 90th percentil	1,5	Chinese cabbage	0,8	Rice	0,7	Spinach	1,0
6,2	WHO Cluster diet F	1,5	Lettuce	0,5	Chinese cabbage	0,5	Barley	0,5
6,1	FR all population	3,0	Wine grapes	0,5	Lettuce	0,4	Olives for oil production	0,2
5,4	PT General population	1,9	Wine grapes	1,6	Rice	0,5	Olives for oil production	0,4
4,7	IT adult	1,9	Lettuce	1,0	Spinach	0,4	Rice	0,0
4,0	IT kids/toddler	1,5	Lettuce	0,6	Spinach	0,5	Wheat	0,1
3,5	DK child	0,7	Lettuce	0,5	Cucumbers	0,4	Wheat	0,2
2,6	LT adult	0,4	Rice	0,4	Head cabbage	0,3	Lettuce	0,4
2,5	DK adult	1,0	Wine grapes	0,2	Rice	0,2	Wheat	0,1
1,9	FI adult	0,4	Lettuce	0,3	Chinese cabbage	0,2	Wine grapes	0,1
1,5	PL general population	0,4	Head cabbage	0,3	Tomatoes	0,2	Table grapes	0,2

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of Alpha-cypermethrin is unlikely to present a public health concern.

The commodity which had the highest calculated IESTI 1 or IESTI 2 is listed on top of this section, respectively. If IESTI is more than 90% of the ARfD/ADI, the results are reported in this sheet. However, at least the top 5 commodities are reported, even if the IESTI is lower than 90%.

The acute risk assessment showed no exceedance of the ARfD for adults for both IESTI calculations.

For children, the ARfD is exceeded for the following unprocessed commodities:

	Member State	IESTI 1 (%)	IESTI 2 (%)
Kale	NL	253.5	181.1

For processed commodities, no exceedance of the ARfD was identified.

As a refinement, the HR (0.59 mg/kg) was used instead of the MRL for calculation of the acute dietary risk assessment for kale. No exceedance of the ARfD was identified after the refinement.

Table 6.9.1-3: TMDI calculation for alpha-cypermethrin with PRIMo Model (rev 2.0) using MRLs for the representative uses submitted in this dossier (Scenario 1)

Alpha-cypermethrin			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):	0,01	proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	**0,02**	ARfD (mg/kg bw):	**0,04**
Source of ADI:		Source of ARfD:	
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum – maximum: 0 – 5

No of diets exceeding ADI: ---

Highest calculated TMDI values in % of ADI	MS Diet	contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
4,6	NL child	1,5	Milk and milk products: Cattle	1,2	Kale	0,5	Lettuce	1,5
4,2	ES child	2,1	Lettuce	0,6	Milk and milk products: Cattle	0,4	Bovine: Meat	0,7
4,1	ES adult	2,7	Lettuce	0,4	Barley	0,2	Milk and milk products: Cattle	0,3
3,9	WHO regional European diet	1,9	Lettuce	0,3	Swine: Meat	0,3	Bovine: Meat	0,3
3,7	WHO Cluster diet B	1,8	Lettuce	0,6	Wheat	0,2	Poultry: Meat	0,2
3,5	WHO Cluster diet F	1,5	Lettuce	0,5	Barley	0,3	Swine: Meat	0,2
2,7	NL general	0,7	Kale	0,6	Lettuce	0,3	Milk and milk products: Cattle	0,3
2,5	IE adult	0,9	Barley	0,4	Lettuce	0,2	Wheat	0,2
2,4	WHO cluster diet E	0,6	Barley	0,5	Lettuce	0,3	Wheat	0,2
2,3	IT adult	1,9	Lettuce	0,3	Wheat	0,1	Courgettes	
2,1	WHO cluster diet D	0,7	Kale	0,5	Wheat	0,2	Milk and milk products: Cattle	0,3
2,0	DE child	0,7	Milk and milk products: Cattle	0,3	Lettuce	0,3	Wheat	0,8
2,0	IT kids/toddler	1,5	Lettuce	0,5	Wheat	0,1	Courgettes	
1,9	FR infant	1,3	Milk and milk products: Cattle	0,2	Courgettes	0,1	Bovine: Meat	1,3
1,8	DK child	0,7	Lettuce	0,4	Wheat	0,3	Rye	0,0
1,4	SE general population 90th percentil	0,6	Milk and milk products: Cattle	0,4	Kale	0,2	Wheat	0,7
1,3	FR all population	0,5	Lettuce	0,2	Wheat	0,2	Poultry: Meat	0,1
1,2	LT adult	0,3	Lettuce	0,2	Swine: Meat	0,2	Milk and milk products: Cattle	0,2
1,1	FR toddler	0,3	Bovine: Meat	0,2	Poultry: Meat	0,2	Wheat	0,1
1,0	UK vegetarian	0,7	Lettuce	0,2	Wheat	0,0	Oats	0,0
0,8	UK Adult	0,6	Lettuce	0,1	Wheat	0,0	Barley	0,0
0,6	FI adult	0,4	Lettuce	0,1	Wheat	0,1	Oats	0,0
0,5	DK adult	0,2	Wheat	0,1	Bovine: Meat	0,1	Oats	0,0
0,5	UK Toddler	0,3	Wheat	0,1	Lettuce	0,0	Birds' eggs	0,0
0,5	UK Infant	0,2	Wheat	0,2	Oats	0,1	Birds' eggs	0,1
0,3	PT General population	0,3	Wheat	0,0	Barley	0,0	Barley	
0,1	PL general population	0,1	Lettuce	0,0	Courgettes		FRUIT (FRESH OR FROZEN)	

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of Alpha-cypermethrin is unlikely to present a public health concern.

Table 6.9.1-4: TMDI calculation for alpha-cypermethrin with PRIMo Model (rev 2.0) using MRLs for all uses to be defended in future (Scenario 2)

Alpha-cypermethrin			
Status of the active substance:		Code no.	
LOQ (mg/kg bw):	0,01	proposed LOQ:	
Toxicological end points			
ADI (mg/kg bw/day):	**0,02**	ARfD (mg/kg bw):	**0,04**
Source of ADI:		Source of ARfD:	
Year of evaluation:		Year of evaluation:	

Prepare workbook for refined calculations

Undo refined calculations

The risk assessment has been performed on the basis of the MRLs collected from Member States in April 2006. For each pesticide/commodity the highest national MRL was identified (proposed temporary MRL = pTMRL).
The pTMRLs have been submitted to EFSA in September 2006.

Chronic risk assessment

TMDI (range) in % of ADI minimum – maximum	
2	49
No of diets exceeding ADI:	---

Highest calculated TMDI values in % of ADI	MS Diet	contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	pTMRLs at LOQ (in % of ADI)
49,1	UK Toddler	45,7	Sugar beet (root)	1,2	Rice	0,3	Wheat	0,3
23,2	UK Infant	20,2	Sugar beet (root)	1,3	Rice	0,2	Wheat	0,4
16,0	WHO Cluster diet B	3,8	Olives for oil production	1,8	Lettuce	1,4	Sugar beet (root)	0,5
13,1	NL child	3,7	Spinach	1,5	Milk and milk products: Cattle	1,2	Kale	1,9
12,4	FR toddler	7,1	Spinach	0,8	Beans (with pods)	0,7	Leek	0,5
11,3	UK vegetarian	7,6	Sugar beet (root)	0,8	Rice	0,7	Lettuce	0,1
11,2	UK Adult	8,0	Sugar beet (root)	0,8	Wine grapes	0,7	Rice	0,1
8,6	FR infant	4,4	Spinach	1,3	Milk and milk products: Cattle	0,6	Beans (with pods)	1,7
8,5	ES child	2,1	Lettuce	1,5	Olives for oil production	1,0	Rice	0,8
7,9	IE adult	1,3	Spinach	0,9	Wine grapes	0,9	Barley	0,5
7,5	WHO regional European diet	1,9	Lettuce	0,4	Rice	0,4	Head cabbage	0,5
7,4	ES adult	2,7	Lettuce	0,8	Olives for oil production	0,7	Spinach	0,3
7,2	DE child	2,0	Spinach	1,0	Table grapes	0,7	Milk and milk products: Cattle	1,0
7,1	WHO cluster diet E	1,2	Wine grapes	0,6	Barley	0,5	Lettuce	0,5
7,1	NL general	1,4	Spinach	0,7	Kale	0,6	Lettuce	0,5
6,4	WHO cluster diet D	1,4	Chinese cabbage	1,1	Rice	0,7	Kale	0,6
6,2	SE general population 90th percentil	1,5	Chinese cabbage	0,8	Rice	0,7	Spinach	1,0
6,2	WHO Cluster diet F	1,5	Lettuce	0,5	Chinese cabbage	0,5	Barley	0,5
6,1	FR all population	3,0	Wine grapes	0,5	Lettuce	0,4	Olives for oil production	0,2
5,4	PT General population	1,9	Wine grapes	1,6	Rice	0,5	Olives for oil production	0,4
4,7	IT adult	1,9	Lettuce	1,0	Spinach	0,4	Rice	0,0
4,0	IT kids/toddler	1,5	Lettuce	0,6	Spinach	0,5	Wheat	0,1
3,5	DK child	0,7	Lettuce	0,5	Cucumbers	0,4	Wheat	0,2
2,6	LT adult	0,4	Rice	0,4	Head cabbage	0,3	Lettuce	0,4
2,5	DK adult	1,0	Wine grapes	0,2	Rice	0,2	Wheat	0,1
1,9	FI adult	0,4	Lettuce	0,3	Chinese cabbage	0,2	Wine grapes	0,1
1,5	PL general population	0,4	Head cabbage	0,3	Tomatoes	0,2	Table grapes	0,2

Conclusion:
The estimated Theoretical Maximum Daily Intakes (TMDI), based on pTMRLs were below the ADI.
A long-term intake of residues of Alpha-cypermethrin is unlikely to present a public health concern.

Table 6.9.1-5: IESTI calculation for the representative uses – MRLs applied

Acute risk assessment /children						Acute risk assessment / adults / general population					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**	1	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**	1		**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**	---	**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**	---		
	IESTI 1	*)	**)	**IESTI 2**	*)	**)	**IESTI 1**	*)	**)	**IESTI 2**	*)	**)
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	253,5	Kale	1,5 / 0,59	181,1	Kale	1,5 / 0,82	76,4	Kale	1,5 / -	56,8	Kale	1,5 / -
	67,3	Lettuce	1 / -	40,4	Lettuce	1 / -	27,5	Lettuce	1 / -	16,5	Lettuce	1 / -
	7,0	Courgettes	0,06 / -	5,0	Courgettes	0,06 / -	4,0	Courgettes	0,06 / -	3,0	Courgettes	0,06 / -
	3,1	Milk and milk	0,01 / -	3,1	Milk and milk	0,01 / -	2,7	Barley	0,15 / -	2,7	Barley	0,15 / -
	1,6	Bovine: Meat	0,05 / -	1,6	Bovine: Meat	0,05 / -	1,5	Poultry: Meat	0,05 / -	1,5	Poultry: Meat	0,05 / -
	No of critical MRLs (IESTI 1)	1					**No of critical MRLs (IESTI 2)**	1				
Processed commodities	**No of commodities for which ARfD/ADI is exceeded:**	---					**No of commodities for which ARfD/ADI is exceeded:**	---				
		***)						***)				
	Highest % of ARfD/ADI	Processed commodities	pTMRL/ threshold MRL (mg/kg)				Highest % of ARfD/ADI	Processed commodities	pTMRL/ threshold MRL (mg/kg)			
	0,4	Wheat flour	0,015 / -				0,2	Bread/pizza	0,015 / -			
*) The results of the IESTI calculations are reported for at least 5 commodities. If the ARfD is exceeded for more than 5 commodities, all IESTI values > 90% of ARfD are reported.												
**) pTMRL: provisional temporary MRL												
***) pTMRL: provisional temporary MRL for unprocessed commodity												
Conclusion:												
For Alpha-cypermethrin IESTI 1 and IESTI 2 were calculated for food commodities for which pTMRLs were submitted and for which consumption data are available.												
The estimated short term intake (IESTI 1) exceeded the ARfD/ADI for 1 commodities.												
Also the IESTI 2 calculation, using less conservative variability factors, resulted in exceedances of the ARfD/ADI for 1 commodities.												
For processed commodities, no exceedance of the ARfD/ADI was identified.												

Table 6.9.1-6: IESTI calculation for the representative uses – refinement applying HR for kale

Acute risk assessment /children - refined calculations						Acute risk assessment / adults / general population - refined calculations					
The acute risk assessment is based on the ARfD.											
For each commodity the calculation is based on the highest reported MS consumption per kg bw and the corresponding unit weight from the MS with the critical consumption. If no data on the unit weight was available from that MS an average European unit weight was used for the IESTI calculation.											
In the **IESTI 1** calculation, the variability factors were 10, 7 or 5 (according to JMPR manual 2002), for lettuce a variability factor of 5 was used. In the **IESTI 2** calculations, the variability factors of 10 and 7 were replaced by 5. For lettuce the calculation was performed with a variabilty factor of 3.											
Threshold MRL is the calculated residue level which would leads to an exposure equivalent to 100 % of the ARfD.											
Unprocessed commodities	**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**	---		**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**	---		**No of commodities for which ARfD/ADI is exceeded (IESTI 1):**	---		**No of commodities for which ARfD/ADI is exceeded (IESTI 2):**	---
	IESTI 1	***)**	****)**	**IESTI 2**	***)**	****)**	**IESTI 1**	***)**	****)**	**IESTI 2**	***)**	****)**
	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)	Highest % of ARfD/ADI	Commodities	pTMRL/ threshold MRL (mg/kg)
	99,7	Kale	0,59 / -	71,2	Kale	0,59 / -	30,1	Kale	0,59 / -	22,3	Kale	0,59 / -
	67,3	Lettuce	1 / -	40,4	Lettuce	1 / -	27,5	Lettuce	1 / -	16,5	Lettuce	1 / -
	7,0	Courgettes	0,06 / -	5,0	Courgettes	0,06 / -	4,0	Courgettes	0,06 / -	3,0	Courgettes	0,06 / -
	3,1	Milk and milk	0,01 / -	3,1	Milk and milk	0,01 / -	2,7	Barley	0,15 / -	2,7	Barley	0,15 / -
	1,6	Bovine: Meat	0,05 / -	1,6	Bovine: Meat	0,05 / -	1,5	Poultry: Meat	0,05 / -	1,5	Poultry: Meat	0,05 / -
	No of critical MRLs (IESTI 1)			---			**No of critical MRLs (IESTI 2)**			---		
Processed commodities	**No of commodities for which ARfD/ADI is exceeded:**	---					**No of commodities for which ARfD/ADI is exceeded:**	---				
			*****)**						*****)**			
	Highest % of ARfD/ADI	Processed commodities	pTMRL/ threshold MRL (mg/kg)				Highest % of ARfD/ADI	Processed commodities	pTMRL/ threshold MRL (mg/kg)			
	0,4	Wheat flour	0,015 / -				0,2	Bread/pizza	0,015 / -			
*) The results of the IESTI calculations are reported for at least 5 commodities. If the ARfD is exceeded for more than 5 commodities, all IESTI values > 90% of ARfD are reported. **) pTMRL: provisional temporary MRL ***) pTMRL: provisional temporary MRL for unprocessed commodity												
Conclusion: For Alpha-cypermethrin IESTI 1 and IESTI 2 were calculated for food commodities for which pTMRLs were submitted and for which consumption data are available. No exceedance of the ARfD/ADI was identified for any unprocessed commodity. For processed commodities, no exceedance of the ARfD/ADI was identified.												

Exposure calculation for metabolites

As described in detail in chapter CA 6.2 a number of alpha-cypermethrin metabolites were identified in matrices directly or indirectly related to human consumption.
To address the relevance of the alpha-cypermethrin metabolites for the human consumption, the Threshold of Toxicological Concern-Concept (TTC-Concept) was applied, which is considered by EFSA to be "the most appropriate tool for evaluating the toxicological relevance of pesticide metabolites" (Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment, EFSA Journal 2012;10(07):2799).

The following 18 compounds have to be assessed:

Metabolites
M310I001
M310I003
M310I004
M310I005
M310I006
M310I007
M310I008
M310I009
M310I010
M310I011
M310I013
M310I017
M310I018
M310I019
M310I021
M310I024
M310I025
M310I026

Exposure calculations have been performed for all metabolites mentioned above as basis for an assessment using the TTC concept.
These calculations were done using the EFSA PRIMo calculator and considering all uses to be defended in future. For each metabolite, a parent/metabolite ratio was derived from the respective metabolism studies, which was multiplied with the parent residues (STMR for the chronic risk assessment and HR for the acute risk assessment) from the supervised field trials. In the following, the calculations are summarized for the individual metabolites. The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculation are given in the supplemental information document (DocID 2014/1314800) for each metabolite.

1. M310I001

For metabolite M310I001 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.016
- ruminants muscle: 0.03
- ruminant liver: 0.78
- ruminant kidney: 4.03
- ruminant fat: 0.02
- poultry muscle: 2.63
- poultry liver: 0.01*
- eggs: 1.83

* absolute value which will not be multiplied with STMR values

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-7: Results of the chronic exposure calculations for metabolite M310I001

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (ES child)		Trigger: 0.0003 mg/kg bw/d (ES child)		Trigger: 0.0015 mg/kg bw/d (ES child)	
1st	6762	Poultry meat	56.3	Poultry meat	11.3	Poultry meat
2nd	530	Birds´ eggs	4.4	Birds´ eggs	0.9	Birds´ eggs
3rd	99.3	Swine liver	0.8	Swine liver	0.2	Swine liver
Total utilization [%]	**7835**		**65.3**		**13.1**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

2. M310I003

For metabolite M310I003 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- ruminant kidney: 1.58
- poultry liver: 0.00054*
- eggs: 0.93

* absolute value which will not be multiplied with STMR values

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-8: Results of the chronic exposure calculations for metabolite M310I003

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK infant)		Trigger: 0.0003 mg/kg bw/d (UK infant)		Trigger: 0.0015 mg/kg bw/d (UK infant)	
1st	500	Birds´ eggs	4.2	Birds´ eggs	0.8	Birds´ eggs
2nd	72.6	Bovine kidney	0.6	Bovine kidney	0.1	Bovine kidney
3rd	--	--	--	--	--	--
Total utilization [%]	**573**		**4.8**		**1.0**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

3. M310I004

For metabolite M310I004 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- ruminant liver: 0.86
- ruminant kidney: 12.95

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-9: Results of the chronic exposure calculations for metabolite M310I004

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (WHO Cluster diet B)		Trigger: 0.0003 mg/kg bw/d (WHO Cluster diet B)		Trigger: 0.0015 mg/kg bw/d (WHO Cluster diet B)	
1st	1899	Bovine kidney	15.8	Bovine kidney	3.2	Bovine kidney
2nd	126	Bovine liver	1.0	Bovine liver	0.2	Bovine liver
3rd	--	--	--	--		--
Total utilization [%]	**2025**		**16.9**		**3.4**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

4. M310I005

For metabolite M310I005 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.031

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-10: Results of the chronic exposure calculations for metabolite M310I005

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	851	sugar beet (root)	7.1	sugar beet (root)	1.4	sugar beet (root)
2nd	65.6	rice	0.5	rice	0.1	rice
3rd	48.6	wheat	0.4	wheat	0.1	wheat
Total utilization [%]	**1117**		**9.3**		**1.9**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

5. M310I006

For metabolite M310I006 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.032

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-11: Results of the chronic exposure calculations for metabolite M310I006

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	878	sugar beet (root)	7.3	sugar beet (root)	1.5	sugar beet (root)
2nd	67.8	rice	0.6	rice	0.1	rice
3rd	50.1	wheat	0.4	wheat	0.1	wheat
Total utilization [%]	**1153**		**9.6**		**1.9**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

6. M310I007

For metabolite M310I007 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.057

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-12: Results of the chronic exposure calculations for metabolite M310I007

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	1564	sugar beet (root)	13.0	sugar beet (root)	2.6	sugar beet (root)
2nd	121	rice	1.0	rice	0.2	rice
3rd	89.3	wheat	0.7	wheat	0.1	wheat
Total utilization [%]	**2053**		**17.1**		**3.4**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

7. M310I008

For metabolite M310I008 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.057

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-13: Results of the chronic exposure calculations for metabolite M310I008

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	1564	sugar beet (root)	13.0	sugar beet (root)	2.6	sugar beet (root)
2nd	121	rice	1.0	rice	0.2	rice
3rd	89.3	wheat	0.7	wheat	0.1	wheat
Total utilization [%]	**2053**		**17.1**		**3.4**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

8. M310I009

For metabolite M310I009 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.07

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-14: Results of the chronic exposure calculations for metabolite M310I009

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	1921	sugar beet (root)	16.0	sugar beet (root)	3.2	sugar beet (root)
2nd	148	rice	1.2	rice	0.2	rice
3rd	110	wheat	0.9	wheat	0.2	wheat
Total utilization [%]	**2521**		**21.0**		**4.2**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

9. M310I010

For metabolite M310I010 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.003
- ruminant liver: 0.44
- ruminant kidney: 5.06
- milk: 0.37

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-15: Results of the chronic exposure calculations for metabolite M310I010

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK infant)		Trigger: 0.0003 mg/kg bw/d (UK infant)		Trigger: 0.0015 mg/kg bw/d (UK infant)	
1st	5730	milk and cream	47.7	milk and cream	9.5	milk and cream
2nd	233	bovine kidney	1.9	bovine kidney	0.4	bovine kidney
3rd	80.9	bovine liver	0.7	bovine liver	0.1	bovine liver
Total utilization [%]	**6103**		**50.9**		**10.2**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

10. M310I011

For metabolite M310I011 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.07
- ruminant liver: 0.19
- ruminant kidney: 1.75

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-16: Results of the chronic exposure calculations for metabolite M310I011

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	1921	sugar beet (root)	16.0	sugar beet (root)	3.2	sugar beet (root)
2nd	148	rice	1.2	rice	0.2	rice
3rd	110	wheat	0.9	wheat	0.2	wheat
Total utilization [%]	**2553**		**21.3**		**4.3**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

11. M310I013

For metabolite M310I013 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.006

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-17: Results of the chronic exposure calculations for metabolite M310I013

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	165	sugar beet (root)	1.4	sugar beet (root)	0.3	sugar beet (root)
2nd	12.7	rice	0.1	rice	0.0	rice
3rd	9.4	wheat	0.1	wheat	0.0	wheat
Total utilization [%]	**216**		**1.8**		**0.4**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

12. M310I017

For metabolite M310I017 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.15

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-18: Results of the chronic exposure calculations for metabolite M310I017

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	4117	sugar beet (root)	34.3	sugar beet (root)	6.9	sugar beet (root)
2nd	318	rice	2.6	rice	0.5	rice
3rd	235	wheat	2.0	wheat	0.4	wheat
Total utilization [%]	**5402**		**45.0**		**9.0**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

13. M310I018

For metabolite M310I018 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.007

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-19: Results of the chronic exposure calculations for metabolite M310I018

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	192	sugar beet (root)	1.6	sugar beet (root)	0.3	sugar beet (root)
2nd	14.8	rice	0.1	rice	0.0	rice
3rd	11.0	wheat	0.1	wheat	0.0	wheat
Total utilization [%]	**252**		**2.1**		**0.4**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

14. M310I019

For metabolite M310I019 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- poultry liver: 2.05

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-20: Results of the chronic exposure calculations for metabolite M310I019

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (NL child)		Trigger: 0.0003 mg/kg bw/d (NL child)		Trigger: 0.0015 mg/kg bw/d (NL child)	
1st	24.0	poultry liver	0.2	poultry liver	0.0	poultry liver
2nd	--	--	--	--	--	--
3rd	--	--	--	--	--	--
Total utilization [%]	**24.0**		**0.2**		**0.0**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

15. M310I021

For metabolite M310I021 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- ruminant liver: 2.55

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-21: Results of the chronic exposure calculations for metabolite M310I021

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (IE adult)		Trigger: 0.0003 mg/kg bw/d (IE adult)		Trigger: 0.0015 mg/kg bw/d (IE adult)	
1st	1153	sheep liver	9.6	sheep liver	1.9	sheep liver
2nd	--	--	--	--	--	--
3rd	--	--	--	--	--	--
Total utilization [%]	**1153**		**9.6**		**1.9**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

16. M310I024

For metabolite M310I024 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.07

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-22: Results of the chronic exposure calculations for metabolite M310I024

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	1921	sugar beet (root)	16.0	sugar beet (root)	3.2	sugar beet (root)
2nd	148	rice	1.2	rice	0.2	rice
3rd	110	wheat	0.9	wheat	0.2	wheat
Total utilization [%]	**2521**		**21.0**		**4.2**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

17. M310I025

For metabolite M310I025 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.08

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-23: Results of the chronic exposure calculations for metabolite M310I025

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	2196	sugar beet (root)	18.3	sugar beet (root)	3.7	sugar beet (root)
2nd	169	rice	1.4	rice	0.3	rice
3rd	125	wheat	1.0	wheat	0.2	wheat
Total utilization [%]	**2881**		**24.0**		**4.8**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

18. M310I026

For metabolite M310I026 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.003

Applying the ADIs 0.0000025, 0.0003 and 0.0015 mg/kg bw/d, representing the trigger values for genotoxic potential, neurotoxic potential and Cramer class III, leads to the following results:

Table 6.9.1-24: Results of the chronic exposure calculations for metabolite M310I026

Most critical commodities	ADI utilization [%]					
	Trigger: 0.0000025 mg/kg bw/d (UK toddler)		Trigger: 0.0003 mg/kg bw/d (UK toddler)		Trigger: 0.0015 mg/kg bw/d (UK toddler)	
1st	82.3	sugar beet (root)	0.7	sugar beet (root)	0.1	sugar beet (root)
2nd	6.4	rice	0.1	rice	0.0	rice
3rd	4.7	wheat	0.0	wheat	0.0	wheat
Total utilization [%]	**108**		**0.9**		**0.2**	

The detailed input values for the chronic risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

Conclusion: Chronic risk assessments for the metabolites according to the TTC Concept

According to the TTC (Threshold of Toxicological Concern) concept for metabolites the chronic risk assessments for the 18 alpha-cypermethrin metabolites were performed.

Their potential toxicological relevance is discussed in the dossier section KCA 5.8.1. All metabolites are by weight of evidence or by direct investigation not genotoxic and not neurotoxic and thereby fall under the trigger value of Cramer Class III substances.

The calculated chronic exposure values for the compounds M310I001, M310I003, M310I004, M310I005, M310I006, M310I007, M310I008, M310I009, M310I010, M310I011, M310I013, M310I017, M310I018, M310I019, M310I021, M310I024, M310I025 and M310I026 turned out to be below 1.5 µg/kg bw/day, the trigger value of Cramer Class III substances.

Based on these data, a long-term intake of residues of these metabolites is unlikely to present a public health concern and they can be assumed to be not relevant for consumer safety.

NEDI calculations

NEDI calculations were not required as ADI utilizations for all 18 metabolites were below 100%.

Acute Reference Dose (ARfD) and Dietary Exposure Calculation

NESTI calculations

Exposure calculation for metabolites

To address the relevance of the alpha-cypermethrin metabolites for the human consumption, the Threshold of Toxicological Concern-Concept (TTC-Concept) was applied, which is considered by EFSA to be "the most appropriate tool for evaluating the toxicological relevance of pesticide metabolites" (Scientific Opinion on Evaluation of the Toxicological Relevance of Pesticide Metabolites for Dietary Risk Assessment, EFSA Journal 2012;10(07):2799).

Acute exposure was assessed for the following 18 compounds:

Metabolite
M310I001
M310I003
M310I004
M310I005
M310I006
M310I007
M310I008
M310I009
M310I010
M310I011
M310I013
M310I017
M310I018
M310I019
M310I021
M310I024
M310I025
M310I026

These calculations were done using the EFSA PRIMo calculator. For each metabolite, the parent/metabolite ratios derived from the respective metabolism studies as discussed in section 6.9.1 were used for multiplication with the parent residues (HRs) from the supervised field trials. The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculation are given in the supplemental information document (DocID 2014/1314800) for each metabolite.

1. M310I001

For metabolite M310I001 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.016
- ruminants muscle: 0.03
- ruminant liver: 0.78
- ruminant kidney: 4.03
- ruminant fat: 0.02
- poultry muscle: 2.63
- poultry liver: 0.01*
- eggs: 1.83

* absolute value which will not be multiplied with HR values

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-25: Results of the acute exposure calculations for metabolite M310I001 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	30.9	poultry meat	adults/general
2nd	29.5	poultry meat	children
3rd	20.2	scarole	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

2. M310I003

For metabolite M310I003 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- ruminant kidney: 1.58
- poultry liver: 0.00054*
- eggs: 0.93

* absolute value which will not be multiplied with HR values

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-26: Results of the acute exposure calculations for metabolite M310I003 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	5.9	bovine kidney	children
2nd	2.7	bovine kidney	adults/general
3rd	2.3	birds´ eggs swine kidney	children adults/general

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

3. M310I004

For metabolite M310I004 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- ruminant liver: 0.86
- ruminant kidney: 12.95

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-27: Results of the acute exposure calculations for metabolite M310I004 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	48.8	bovine kidney	children
2nd	22.0	bovine kidney	adults/general
3rd	18.4	swine kidney	adults/general

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

4. M310I005

For metabolite M310I005 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.031

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-28: Results of the acute exposure calculations for metabolite M310I005 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	39.1	scarole	children
2nd	24.7	kale	children
3rd	15.6	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

5. M310I006

For metabolite M310I006 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.032

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-29: Results of the acute exposure calculations for metabolite M310I006 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	40.4	scarole	children
2nd	25.5	kale	children
3rd	16.1	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

6. M310I007

For metabolite M310I007 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.057

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-30: Results of the acute exposure calculations for metabolite M310I007 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	72.0	scarole	children
2nd	45.5	kale	children
3rd	28.6	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

7. M310I008

For metabolite M310I008 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.057

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-31: Results of the acute exposure calculations for metabolite M310I008 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	72.0	scarole	children
2nd	45.5	kale	children
3rd	28.6	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

8. M310I009

For metabolite M310I009 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.07

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-32: Results of the acute exposure calculations for metabolite M310I009 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	88.4	scarole	children
2nd	55.8	kale	children
3rd	35.1	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

9. M310I010

For metabolite M310I010 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.003
- ruminant liver: 0.44
- ruminant kidney: 5.06
- milk: 0.37

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-33: Results of the acute exposure calculations for metabolite M310I010 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	19.1	bovine kidney	children
2nd	9.2	milk and milk products	children
3rd	8.6	bovine kidney	adults/general

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

10. M310I011

For metabolite M310I011 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.07
- ruminant liver: 0.19
- ruminant kidney: 1.75

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-34: Results of the acute exposure calculations for metabolite M310I011 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	88.4	scarole	children
2nd	55.8	kale	children
3rd	35.1	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

11. M310I013

For metabolite M310I013 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.006

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-35: Results of the acute exposure calculations for metabolite M310I013 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	7.6	scarole	children
2nd	4.8	kale	children
3rd	3.0	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

12. M310I017

For metabolite M310I017 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.15

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-36: Results of the acute exposure calculations for metabolite M310I017 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	189	scarole	children
2nd	120	kale	children
3rd	75.3	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

13. M310I018

For metabolite M310I018 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.007

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-37: Results of the acute exposure calculations for metabolite M310I018 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	8.8	scarole	children
2nd	5.6	kale	children
3rd	3.5	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

14. M310I019

For metabolite M310I019 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- poultry liver: 2.05

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-38: Results of the acute exposure calculations for metabolite M310I019 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	9.1	poultry liver	adults/general
2nd	--	--	--
3rd	--	--	--

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

15. M310I021

For metabolite M310I021 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- ruminant liver: 2.55

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-39: Results of the acute exposure calculations for metabolite M310I021 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	20.6	bovine liver	children
2nd	6.9	bovine liver	adults/general
3rd	2.8	swine liver	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

16. M310I024

For metabolite M310I024 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.07

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-40: Results of the acute exposure calculations for metabolite M310I024 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	88.4	scarole	children
2nd	55.8	kale	children
3rd	35.1	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

17. M310I025

For metabolite M310I025 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.08

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-41: Results of the acute exposure calculations for metabolite M310I025 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	101	scarole	children
2nd	63.8	kale	children
3rd	40.1	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

18. M310I026

For metabolite M310I026 the parent/metabolite ratios were as follows (for details see chapter 3.2 of the supplemental information document (DocID 2014/1314800)):

- all plants: 0.003

Applying the ARfD of 0.005 mg/kg bw/d, representing the trigger value for Cramer class III, leads to the following results:

Table 6.9.1-42: Results of the acute exposure calculations for metabolite M310I026 using default variability factors (IESTI 1)

Most critical commodities	Trigger: 0.005 mg/kg bw/d		
	ARfD utilization [%]	Commodity	Population
1st	3.8	scarole	children
2nd	2.4	kale	children
3rd	1.5	spinach	children

The detailed input values for the acute risk assessment and the results of the EFSA PRIMo calculations are given in in the supplemental information document (DocID 2014/1314800).

Conclusion: Acute risk assessments for the metabolites according to the TTC Concept

According to the TTC (Threshold of Toxicological Concern) concept for metabolites the acute risk assessments for the 18 alpha-cypermethrin metabolites were performed.

Their potential toxicological relevance is discussed in the dossier section KCA 5.8.1. All metabolites are by weight of evidence or by direct investigation not genotoxic and not neurotoxic and thereby fall under the trigger value of Cramer Class III substances.
The calculated maximum acute exposure values for the compounds M310I001, M310I003, M310I004, M310I005, M310I006, M310I007, M310I008, M310I009, M310I010, M310I011, M310I013, M310I018, M310I019, M310I021, M310I024 and M310I026 turned out to be below 5 µg/kg bw/day.

For metabolites M310I017 and M310I025 ARfD utilizations slightly above the trigger value were observed but by weight of evidence the toxicity data (see KCA 5.8.1) do not indicate a relevant concern for acute toxicity at the respective exposure level.
Based on these data, a short-term acute intake of residues of these metabolites is unlikely to present a public health concern and they can be assumed to be not relevant for consumer safety.

CA 6.10 Other studies

No other/special studies are deemed necessary. The studies and information provided in the previous sections are considered adequate and sufficient.

CA 6.10.1 Effect on the residue level in pollen and bee products

The objective of these studies shall be to determine the residue in pollen and bee products for human consumption resulting from residues taken up by honeybees from crops at blossom.

The representative uses of this dossier are cereals, oil seed rape, leafy brassica, lettuce and cucumber. Cereals are generally considered to be of low attractiveness to honey bees for pollen collection (although not fully excluded) and collection of nectar is not relevant (EFSA, 2013). Moreover, cereals are considered to have no melliferous capacity. Lettuce and leafy brassica vegetables are harvested before flowering, hence these crops are not relevant regarding potential exposure of bees to residues. The pollen/nectar from cucumber and courgette cultivated in greenhouses is assumed to have a negligible contribution to the total pollen/nectar foraged by honeybees for several reasons: Greenhouses represent an enclosed in-door system, often with limited direct access for honey bees and other pollinators. For that reason, often commercially bread bumble bees are specifically introduced into greenhouses to perform the pollination activity on crops such as cucurbits, tomatoes, peppers, etc., grown in greenhouses. Furthermore, when considering that the representative use on oilseed rape - which is cultivated over a much larger field area (the EU cucumber production area (greenhouse plus field) represents only 2% of the EU oil seed rape production area) - and which is treated during flowering – the oil seed rape use represents the realistic worst-case scenario with regard to the foraging by bees in view of honey production.
Due to the lack of an appropriate guidance document, but driven by the fact that oil seed rape is a fodder crop for honey bees the residue in pollen and derived bee products was evaluated. Studies were conducted to determine the effects of BAS 310 I on bees and to derive ecotoxicological endpoints. In these studies, residues of alpha-cypermethrin were determined in pollen and nectare and in honey. These studies are summarized in Document M-CP, Section 10 chapter CP 10.3.1 of this dossier. As expected from the low water solubility and the non-systemicity of alpha-cypermethrin these studies demonstrate that a transfer of residue from flowers into honey is insignificant. The consumer risk in honey consumption resulting from the uses of alpha-cypermethrin is therefore considered as negligible.

In the application, the study with the title “Determination of residues of BAS 310 55 I in nectar, pollen and flowers of winter oil seed rape after one application in Germany 2013”, Mack P., 2014, BASF DocID 2014/1000203, is listed as reference no. KCA 6.10.1/1. In deviation from this order, this study is not summarized in this section (CA 6.10.1), but in CP 10.3.1 (report CP 10.3.1.6/7).

Tier 1 Summaries of the Supervised Field Residue Trials for the Representative Crops

- **Cucumber**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	Denmark, France, Germany, The Netherlands	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
166297	Cucumber/	1. 22.01.04	medium	0.004	400	0.015	1	85	fruit	<0.01	0	Method
2004/5000720	Euphoria	2. 05.02.04-08.10.04	volume foliar				25.05.04		fruit	<0.01	3	No 567/0
Brandenburg	VC 0424	3. 25.05.04	application						fruit	<0.01	7	LOQ
Germany		28.05.04	using boom						fruit	<0.01	14	0.01 mg/kg
(ACK/08/04)		01.06.04 08.06.04	sprayer						fruit			
166297	Cucumber/	1. 22.01.04	medium	0.004	400	0.015	2	85	fruit	<0.01	0	Method
2004/5000720	Euphoria	2. 05.02.04-08.10.04	volume foliar	0.004	400	0.015	25.05.04		fruit	<0.01	3	No 567/0
Brandenburg	VC 0424	3. 25.05.04	application						fruit	<0.01	7	LOQ
Germany		28.05.04	using boom						fruit	<0.01	14	0.01 mg/kg
(ACK/08/04)		01.06.04 08.06.04	sprayer						fruit			

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	Denmark, France, Germany, The Netherlands	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
166297	Cucumber/	1. 10.06.04	medium	0.004	400	0.015	1	85	fruit	<0.01	0	Method
2004/5000720	Sheila	2. 20.06.04-27.08.04	volume foliar				17.08.04		fruit	<0.01	3	No 567/0
Limburg	VC 0424	3. 17.08.04	application						fruit	<0.01	7	LOQ
The Netherlands		20.08.04	using boom						fruit	<0.01	13	0.01 mg/kg
(AGR/10/04)		24.08.04 30.08.04	sprayer						fruit			
166297	Cucumber/	1. 10.06.04	medium	0.004	400	0.015	2	85	fruit	<0.01	0	Method
2004/5000720	Sheila	2. 20.06.04-27.08.04	volume foliar	0.004	400	0.015	17.08.04		fruit	<0.01	3	No 567/0
Limburg	VC 0424	3. 17.08.04	application						fruit	<0.01	7	LOQ
The Netherlands		20.08.04	using boom						fruit	<0.01	13	0.01 mg/kg
(AGR/10/04)		24.08.04 30.08.04	sprayer						fruit			
166297	Cucumber/	1. 15.05.04	medium	0.004	400	0.015	1	77	fruit	0.023	0	Method
2004/5000720	Naomi	2. 01.07.04-30.07.04	volume foliar				15.07.04		fruit	<0.01	4	No 567/0
S. Jutland	VC 0424	3. 15.07.04	application						fruit	<0.01	7	LOQ
Denmark		19.07.04	using boom						fruit	<0.01	13	0.01 mg/kg
(ALB/07/04)		22.07.04 28.07.04	sprayer						fruit			
166297	Cucumber/	1. 15.05.04	medium	0.004	400	0.015	2	77	fruit	0.030	0	Method
2004/5000720	Naomi	2. 01.07.04-30.07.04	volume foliar	0.004	400	0.015	15.07.04		fruit	0.012	4	No 567/0
S. Jutland	VC 0424	3. 15.07.04	application						fruit	<0.01	7	LOQ
Denmark		19.07.04	using boom						fruit	<0.01	13	0.01 mg/kg
(ALB/07/04)		22.07.04 28.07.04	sprayer						fruit			

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	Denmark, France, Germany, The Netherlands	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
166297	Cucumber/	1. 10.05.04	medium	0.004	400	0.015	1	73	fruit	<0.01	0	Method
2004/5000720	Solverde	2. 20.06.04-30.06.04	volume foliar				23.06.04		fruit	<0.01	2	No 567/0
Alsace	VC 0424	3. 23.06.04	application						fruit	<0.01	8	LOQ
France North		25.06.04	using boom						fruit	<0.01	14	0.01 mg/kg
(FAN/10/04)		01.07.04 07.07.04	sprayer						fruit			
166297	Cucumber/	1. 10.05.04	medium	0.004	400	0.015	2	73	fruit	<0.01	0	Method
2004/5000720	Solverde	2. 20.06.04-30.06.04	volume foliar	0.004	400	0.015	23.06.04		fruit	<0.01	2	No 567/0
Alsace	VC 0424	3. 23.06.04	application						fruit	<0.01	8	LOQ
France North		25.06.04	using boom						fruit	<0.01	14	0.01 mg/kg
(FAN/10/04)		01.07.04 07.07.04	sprayer						fruit			

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

ESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	Belgium, Denmark, Germany	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
227122	Cucumber/	1. 22.06.05	SOLO	0.0100	400	0.04	1	69	fruit	<0.01	0	Method
2006/1036934	Avonis	2. 01.07.05	knapsack				08.08.05		fruit	<0.01	3	No 567/0
Großrudestedter Str. 3-4, 99195 Alperstedt, Thuringia	VC 0424	3. 10.07.05	sprayer						fruit	<0.01	7	LOQ
Germany									fruit	<0.01	15	0.01 mg/kg
(AT-05/007-1)												
227122	Cucumber/	1. 07.07.05	RAM3-10	0.0100	400	0.04	1	87-89	fruit	0.020	0	Method
2006/1036934	Loretta	2. . 15.08.-20.08.05					06.09.05		fruit	0.014	3	No 567/0
Rue Dominique Seret, 34 6210 Villers-Perwin, Wagnelée (Hainaut)	VC 0424	3. . not reported							fruit	<0.01	7	LOQ
Belgium									fruit	<0.01	14	0.01 mg/kg
(G022-05 I-B)												
227122	Cucumber/	1. 13.05.05	spray lance #126,	0.0100	400	0.04	1	81	fruit	0.014	0	Method
2006/1036934	Naomi	2. 10.05.-01.06.05	air assisted				10.06.05		fruit	0.020	3	No 567/0
Tuevej 39, 7000 Fredericia Jylland	VC 0424	3. 15.06.-31.07.05	Knapsack						fruit	0.013	7	LOQ
Denmark			precision						fruit	<0.01	14	0.01 mg/kg
(ALB/190507-01)			sprayer with 1 nozzle on lance									

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

Cucumber

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
166297	Cucumber/	1. 17.08.04	medium	0.004	400	0.015	1	73	fruit	<0.01	0	Method
2004/5000720	Suxo	2. 12.09.04-30.09.04	volume foliar				30.09.04		fruit	<0.01	4	No 567/0
Andalucia	VC 0424	3. 30.09.04	application						fruit	<0.01	7	LOQ
Spain		04.10.04	using boom						fruit	<0.01	13	0.01 mg/kg
(ALO/17/04)		07.10.04 13.10.04	sprayer									
166297	Cucumber/	1. 17.08.04	medium	0.004	400	0.015	2	73	fruit	<0.01	0	Method
2004/5000720	Suxo	2. 12.09.04-30.09.04	volume foliar	0.004	400	0.015	30.09.04		fruit	<0.01	4	No 567/0
Andalucia	VC 0424	3. 30.09.04	application						fruit	<0.01	7	LOQ
Spain		04.10.04	using boom						fruit	<0.01	13	0.01 mg/kg
(ALO/17/04)		07.10.04 13.10.04	sprayer									
166297	Cucumber/	1. 21.06.04	medium	0.004	400	0.015	1	81	fruit	<0.01	0	Method
2004/5000720	Rawa	2. 05.07.04-22.07.04	volume foliar				19.07.04		fruit	<0.01	3	No 567/0
Rhone-Alpes	VC 0424	3. 19.07.04	application						fruit	<0.01	7	LOQ
France South		22.07.04	using boom						fruit	<0.01	14	0.01 mg/kg
(FBD/11/04)		26.07.04 02.08.04	sprayer									

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
166297	Cucumber/	1. 21.06.04	medium	0.004	400	0.015	2	81	fruit	0.015	0	Method
2004/5000720	Rawa	2. 05.07.04-22.07.04	volume foliar	0.004	400	0.015	19.07.04		fruit	<0.01	3	No 567/0
Rhone-Alpes	VC 0424	3. 19.07.04	application						fruit	<0.01	7	LOQ
France South		22.07.04	using boom						fruit	<0.01	14	0.01 mg/kg
(FBD/11/04)		26.07.04 02.08.04	sprayer									
166297	Cucumber/	1. 22.07.04	medium	0.004	400	0.015	1	81	fruit	0.026	0	Method
2004/5000720	Palmera	2. 15.08.04-NA	volume foliar				31.08.04		fruit	<0.01	4	No 567/0
N. Greece-Macedonia	VC 0424	3. 31.08.04	application						fruit	<0.01	8	LOQ
Greece		04.09.04	using boom						fruit	<0.01	14	0.01 mg/kg
(GRE/12/04)		08.09.04 14.09.04	sprayer									
166297	Cucumber/	1. 22.07.04	medium	0.004	400	0.015	2	81	fruit	0.014	0	Method
2004/5000720	Palmera	2. 15.08.04-NA	volume foliar	0.004	400	0.015	31.08.04		fruit	<0.01	4	No 567/0
N. Greece-Macedonia	VC 0424	3. 31.08.04	application						fruit	<0.01	8	LOQ
Greece		04.09.04	using ~~boom~~						fruit	<0.01	14	0.01 mg/kg
(GRE/12/04)		08.09.04 14.09.04	~~Sprayer~~ atomizer									
166297	Cucumber/	1. 10.06.04	medium	0.004	400	0.015	1	79	fruit	<0.01*	0	Method
2004/5000720	Hiyield	2. 30.08.04-14.09.04	volume foliar				14.09.04		fruit	<0.01*	3	No 567/0
Piemonte	VC 0424	3. 14.09.04	application						fruit	<0.01*	7	LOQ
Italy		17.09.04	using ~~boom~~						fruit	<0.01*	14	0.01 mg/kg
(ITA/09/04)		21.09.04 28.09.04	~~Sprayer~~ mistblower									

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
166297	Cucumber/	1. 10.06.04	medium	0.004	400	0.015	2	79	fruit	<0.01	0	Method
2004/5000720	Hiyield	2. 30.08.04-14.09.04	volume foliar	0.004	400	0.015	14.09.04		fruit	<0.01	3	No 567/0
Piemonte	VC 0424	3. 14.09.04	application						fruit	<0.01	7	LOQ
Italy		17.09.04	using boom						fruit	<0.01	14	0.01 mg/kg
(ITA/09/04)		21.09.04 28.09.04	sprayer									

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
227122	Cucumber/	1. 03.06.05	Pulvexper,	0.0100	400	0.04	1	71	fruit	0.027	0	Method
2006/1036934	Defens	2. 15.06.05	spray boom with				08.07.05		fruit	0.012	3	No 567/0
Chemin Croix Rouge		11.07.05	nozzle							0.012	7	
84140 Montfavet	VC 0424	3.							fruit			LOQ
Provence France									fruit	<0.01	14	0.01 mg/kg
(05 I CL FR P32)												
227122	Cucumber/	1. 10.05.05	Pulvexper	0.0100	400	0.04	1	76	fruit	0.043	0	Method
2006/1036934	Columbia	2. 15.06.05*	type,				18.07.05		fruit	0.031	3	No 567/0
Route Les Paluds, 13940 Mollèges, Provence	VC 0424	3. 15.06.-31.08.05	lance sprayer						fruit	0.014	7	LOQ
France		*continuous flowering	fitted with a						fruit	0.011	14	0.01 mg/kg
(05 I CL FR P36)			single nozzle									
227122	Cucumber/	1. 10.04.05	hand held	0.0100	400	0.04	1	88	fruit	0.087	0	Method
2006/1036934	Jumbo	2. 10.05.-25.05.05	sprayer				23.05.05		fruit	0.037	3	No 567/0
Via Pioppa 81, 44030 Pontegradella, Ferrara	VC 0424	3. 23.05.-06.06.05	(GZ SIP I 015						fruit	0.011	7	LOQ
Italy									fruit	<0.01	14	0.01 mg/kg
(IR05BASL11LG01)												

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cucumber (fruiting vegetables, cucurbits edible peel)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
227122	Cucumber/	1. 16.03.05	Schachtner air	0.0100	400	0.04	1	75	fruit	<0.01	0	Method
2006/1036934	Suso	2. 18.04.05	compressed				27.05.05		fruit	<0.01	3	No 567/0
Finca La Dehesilla, 41710 Utrera, Andalucia, Sevilla	VC 0424	3. 03.05.-10.06.05	boom sprayer						fruit	<0.01	7	LOQ
Spain									fruit	<0.01	14	0.01 mg/kg
(05ES087R)												
227122	Cucumber/	1. 01.06.05	Fox-Motori	0.0100	400	0.04	1	81	fruit	0.036	0	Method
2006/1036934	Luberon	2. 25.06.-Aug. 05	motorized				30.06.05		fruit	0.014	3	No 567/0
Profitis		01.07 -Sep. 05	knapsack							<0.01	7	
Thessaloniki	VC 0424	3.							fruit			LOQ
Central Macedonia												
Greece			sprayer with						fruit	<0.01	14	0.01 mg/kg
(05RF044)			lance									

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

Leafy brassica

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Leafy brassica (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Total cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
181963	Curly kale/	1. 15.07.05	foliar	0.003	400	0.0125	1	48-49	Foliage	0.154	0	Method
2006/1026862	Winterbox	2. -	application				04.11.05		Foliage	0.227	3	No 567/0
Ippenstedter Straße 16 30982 Pattensen, Jeinsen	VL 0480	3. 04.11. -18.11.05	with						Foliage	0.171	7	LOQ
Germany (AF/8815/BA/1)			boom						Foliage	0.083	14	0.01 mg/kg
181963	Curly kale/	1. 15.07.05	foliar	0.003	400	0.0125	2	48-49	Foliage	0.241	0	Method
2006/1026862	Winterbox	2. -	application	0.003	400	0.0125	04.11.05		Foliage	0.170	3	No 567/0
Ippenstedter Straße 16 30982 Pattensen, Jeinsen	VL 0480	3. 04.11. -18.11.05	with						Foliage	0.084	7	LOQ
Germany (AF/8815/BA/1)			boom						Foliage	0.152	14	0.01 mg/kg
181963	Curly kale/	1. 19.06.06	foliar	0.003	400	0.0125	1	48	Foliage	0.159	0	Method
2006/1026862	Winetou	2. -	application				27.11.06		Foliage	0.203	3	No 567/0
Asmall, Scarisbruck, Ormskirk, Lancashire, L40 8JL	VL 0480	3. 27.11 -11.12.06	with						Foliage	0.237	7	LOQ
The United Kingdom (AF/8815/BA/5)			boom						Foliage	0.116	14	0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Leafy brassica (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Total cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
181963	Curly kale/	1. 19.06.06	foliar	0.003	400	0.0125	2	48	Foliage	0.241	0	Method
2006/1026862	Winetou	2. -	application	0.003	400	0.0125	27.11.2006		Foliage	0.348	3	No 567/0
Asmall, Scarisbruck, Ormskirk, Lancashire, L40 8JL	VL 0480	3. 27.11 -11.12.06	with						Foliage	0.383	7	LOQ
The United Kingdom			boom						Foliage	0.245	14	0.01 mg/kg
(AF/8815/BA/5)												
227098	Leafy cabbage/	1. 19.06.06	foliar spray	0.003	400	0.0125	1	49	Leaves	0.26	0	Method
2007/1013342	Reflex	2. -	using boom				21.09.06		Leaves	0.26	3	No 567/0
Warmeriville Champagne-Ardennes	VL 0480	3. 05.10.06	sprayer						Leaves	0.22	8	LOQ
France									Leaves	0.17	14	0.01 mg/kg
(A/NF/I/06/115)												
227098	Leafy cabbage/	1. 19.06.06	foliar spray	0.003	419	0.0125	2	49	Leaves	0.47	0	Method
2007/1013342	Reflex	2. -	using boom	0.003	399	0.0125	21.09.06		Leaves	0.59	3	No 567/0
Warmeriville Champagne-Ardennes	VL 0480	3. 05.10.06	sprayer						Leaves	0.35	8	LOQ
France									Leaves	0.24	14	0.01 mg/kg
(A/NF/I/06/115)												
227098	Leafy cabbage/	1. 03.08.06	foliar spray	0.003	415	0.0125	1	48-49	Leaves	0.26	0	Method
2007/1013342	Winnetou	2. -	using boom				05.10.06		Leaves	0.18	4	No 567/0
NV Meterik, Limburg	VL 0480	3. 20.10.06	sprayer						Leaves	0.15	7	LOQ
The Netherlands									Leaves	0.074	15	0.01 mg/kg
(A/NL/I/06/116)												

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Leafy brassica (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Total cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
227098	Leafy cabbage/	1. 03.08.06	foliar spray	0.003	396	0.0125	2	48-49	Leaves	0.48	0	Method
2007/1013342	Winnetou	2. -	using boom	0.003	402	0.0125	05.10.06		Leaves	0.35	4	No 567/0
NV Meterik, Limburg	VL 0480	3. 20.10.06	sprayer						Leaves	0.21	7	LOQ
The Netherlands									Leaves	0.19	15	0.01 mg/kg
(A/NL/I/06/116)												

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Kale (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	AK Lück	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH**	**Portion Analysed**	**Residues (mg/kg)**	**DALA [2] (days)**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
-													DFG S19
2007/1035745	Kale	1.	31.07.06	Spraying	0.00208	600	0.0125	2	16-17	leaves	0.24	0	
Münster	Kobold	2.	-		0.00208	600	0.0125	05.10.06		leaves	0.07	7	LOQ = 0.01
48147	VL 0480	3.	from							leaves	0.09	10	mg/kg
Gemany (N)			November							leaves	0.08	14	
RU-I-05 06 NW BN 1/1			2006										
-													DFG S19
2007/1035745	Kale	1.	31.07.06	Spraying	0.00208	600	0.0125	2	18-19	leaves	0.62	0	
Münster	Kobold	2.	-		0.00208	600	0.0125	11.10.06		leaves	0.20	7	LOQ = 0.01
48147	VL 0480	3.	from							leaves	0.24	10	mg/kg
Gemany (N)			November							leaves	0.19	14	
RU-I-05 06 NW BN 1/2			2006										
-													DFG S19
2007/1035745	Kale	1.	05.07.06	Spraying	0.00208	600	0.0125	2	39	leaves	0.24	7	
Köln	Winterbor	2.	-		0.00208	600	0.0125	12.10.06		leaves	0.17	14	LOQ = 0.01
50765	VL 0480	3.	from										mg/kg
Gemany (N)	,		October										
RU-I-05 06 NW BN 1/4			2006										
-													DFG S19
2007/1035745	Kale	1.	08.06.06	Spraying	0.00139	900	0.0125	2	48-49	leaves	0.54	0	
Oberbessingen	Winnetou	2.	-		0.00139	900	0.0125	30.08.06		leaves	0.26	7	LOQ = 0.01
35423	VL 0480	3.	30.08.06							leaves	0.16	10	mg/kg
Gemany (N)										leaves	0.05	14	
RU-I-05 06 HE WE 1/1													

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Kale (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	AK Lück	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	SC (BAS 310 41 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
-													DFG S19
2014/1083417	Curly kale	1.	02.07.07		0.0021	600	0.0125	2	47-49	leaves	0.18	0	
Butzbach	Winnetou	2.	-					24.09.07		leaves	0.05	7	LOQ = 0.01
35510	VL 0480	3.	from 01.10.07	Spraying						leaves	0.08	10	mg/kg
Gemany (N)										leaves	0.06	14	
RU-I-03 07 HE WE 2/1													
-													DFG S19
			25.05.07 (sowing)										
2014/1083417	Curly kale	1.	27.07.07 (planting)		0.0021	600	0.0125	2	47	leaves	0.34	0	
Köln	Winterboer	2.	-	Spraying				26.09.07		leaves	0.06	7	LOQ = 0.01
50765	VL 0480	3.	from							leaves	0.07	10	mg/kg
Gemany (N)			December							leaves	0.03	14	
RU-I-03 07 NW BN 2/1			2007										
-													DFG S19
			25.05.07 (sowing)										
2014/1083417	Curly kale	1.	02.07.07 (planting)		0.0021	600	0.0125	2	47	leaves	0.38	0	
Köln	Winterboer	2.	-	Spraying				02.10.07		leaves	0.23	7	LOQ = 0.01
50765	VL 0480	3.	from							leaves	0.19	10	mg/kg
Gemany (N)			December							leaves	0.26	14	
RU-I-03 07 NW BN 2/2			2007										
-													DFG S19
2014/1083417	Curly kale	1.	18.07.07 (sowing) 29.08.07		0.0021	600	0.0125	2	44	leaves	0.58	0	
Bonn	Westländer	2.	-					18.10.07		leaves	0.34	7	LOQ = 0.01
53229	VL 0480	3.	from	Spraying						leaves	0.34	10	mg/kg
Gemany (N)			December							leaves	0.25	14	
RU-I-03 07 NW BN 2/3			2007										

1) at last treatment
2) days after last application

Leafy brassica

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Leafy brassica (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0] kg a.s./hL**	**Water (L/ha)**	**kg a.s./ha**	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
181963	Curly kale/	1. 05.09.05	foliar	0.006	400	0.025	1	47-48	Foliage	0.517	0	Method
2006/1026862	Black Toscano	2. n.a.	application				02.12.05		Foliage	0.436	3	No 567/0
Az Agr Cammelli, Viuzzo di Fagna 2, 50142 Firenze	VL 0480	3. 02.12 - 16.12.05	with boom						Foliage	0.263	7	LOQ
Italy									Foliage	0.243	14	0.01 mg/kg
(AF/8815/BA/3)												
181963	Curly kale/	1. 05.09.05	foliar	0.006	400	0.025	1	48	Foliage	0.349	0	Method
2006/1026862	Vates	2. n.a.	application				08.11.05		Foliage	0.278	3	No 567/0
Nea Magnisia, Thessaloniki, Central Macedonia, GR-57008	VL 0480	3. 08.11 - 22.11.05	with boom						Foliage	0.257	7	LOQ
Greece									Foliage	0.137	14	0.01 mg/kg
(AF/8815/BA/4)												
227098	Leafy cabbage/	1. Seeding 14.06.06, transplant. 11.07.0(	foliar	0.006	403	0.025	1	47	Leaves	0.066	0	Method
2007/1013342	Castelard	2. -	spray				12.09.06		Leaves	0.046	3	No 567/0
Chateaurenard, Bouches-de-Rhone	xxx	3. 26.09.06	using						Leaves	0.011	7	LOQ
France			boom						Leaves	<0.01	14	0.01 mg/kg
(A/SF/I/06/117)			sprayer									

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Leafy brassica (brassica leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation (common name and content)**	-
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
227098	Leafy cabbage/	1. 12.10.06	foliar	0.006	397	0.025	1	47	Leaves	0.13	0	Method
2007/1013342	Manoko F-1 (Chinese cabbage)	2. -	spray				29.11.06		Leaves	0.056	3	No 567/0
Almussafes, Valencia	VL 0467	3. 13.12.06	using						Leaves	0.056	6	LOQ
Spain			boom						Leaves	0.011	14	0.01 mg/kg
(A/SP/I/06/118)			sprayer									

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**BAS 310 55 I**
Crop/crop group:	**Chinese cabbage (Leafy brassica)**	**Producer of commercial product**	**BASF SE**
Responsible body for reporting (name, address)	**BASF SE**	**Indoor/Glasshouse/Outdoor**	**Outdoor**
Country	**Germany**	**Other active substance in the formulation**	**-**
Content of active substance (g/kg or g/L)	**50 g/L**	**(common name and content)**	
Formulation (e.g. WP)	**ME (BAS 310 55 I)**	**Residues calculated as:**	**BAS 310 I**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2013/1416285	Chinese	1. 13.08.13	Plot 2	0.006	200	0.0125	24.09.13		head with wrapper leaves	0.13	0	Method:
S13-00427-01 / L130046	Cabbage /	2. na	broadcast	0.006	200	0.0125	01.10.13	47-48	head with wrapper leaves	0.086	3	BASF method No. 567/0
82000 Montauban	Kaboko	3. 15.10.13	foliar						head with wrapper leaves	0.041	7	
France	VL 0467		application						head with wrapper leaves	0.027	14	
BASF DocID 2013/1416285	Chinese	1. 23.08.13	Plot 2	0.006	200	0.0125	26.09.13		head with wrapper leaves	0.27	0	Method:
S13-00427-02 / L130047	Cabbage /	2. na	broadcast	0.006	200	0.0125	04.10.13	43-45	head with wrapper leaves	0.11	3	BASF method No. 567/0
57007 Thessaloniki	Guest Star	3. 17.10.13	foliar						head with wrapper leaves	0.036	7	
Greece	VL 0467		application						head with wrapper leaves	<0.01	13	
BASF DocID 2013/1416285	Chinese	1. 02.05.13	Plot 2	0.006	200	0.0125	02.07.13		head with wrapper leaves	0.10	0	Method:
S13-00427-03 / L130048	Cabbage /	2. na	broadcast	0.006	200	0.0125	09.07.13	47	head with wrapper leaves	0.064	3	BASF method No. 567/0
40057 Granarolo	Paranlo	3. 22.07.13	foliar						head with wrapper leaves	0.029	6	
Italy	VL 0467		application						head with wrapper leaves	0.013	13	
BASF DocID 2013/1416285	Chinese	1. 21.02.13	Plot 2	0.006	200	0.0125	24.04.13		head with wrapper leaves	0.093	0	Method:
S13-00427-04 / L130049	Cabbage /	2. na	broadcast	0.006	200	0.0125	30.04.13	47	head with wrapper leaves	0.019	2	BASF method No. 567/0
46810 Enguera	Manoko (Bejo)	3. 06.05.13	foliar						head with wrapper leaves	0.013	6	
Spain	VL 0467		application									

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

Lettuce

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182014 2006/1026855 St Lambert de Levees 49400 France (AF/8817/BA/1)	Lettuce, head/ Estelle VL 0482	1. 26.09.05 2. n.a. 3. 22.11-06.12.05	foliar application with boom	0.003	400	0.0125	1 22.11.05	45-47	Head Head Head Head	0.421 0.382 0.243 0.194	0 2 7 14	Method No 567/0 LOQ 0.01 mg/kg
182014 2006/1026855 St Lambert de Levees 49400 France (AF/8817/BA/1)	Lettuce, head/ Estelle VL 0482	1. 26.09.05 2. n.a. 3. 22.11-06.12.05	foliar application with boom	0.003 0.003	400 400	0.0125 0.0125	2 22.11.05	45-47	Head Head Head Head	0.654 0.722 0.491 0.412	0 2 7 14	Method No 567/0 LOQ 0.01 mg/kg
182014 2006/1026855 Shepshed, Leicestershire, LE12 9EJ The United Kingdom (AF/8817/BA/2)	Lettuce, head/ Tamburo VL 0482	1. 28.06.05 2. n.a. 3. 29.07-12.08.05	foliar application with boom	0.003	400	0.0125	1 29.07.05	47	Head Head Head Head	0.295 0.244 0.064 0.025	0 3 7 14	Method No 567/0 LOQ 0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182014	Lettuce, head/	1. 28.06.05	foliar	0.003	400	0.0125	2	47	Head	0.401	0	Method
2006/1026855	Tamburo	2. n.a.	application	0.003	400	0.0125	29.07.05		Head	0.236	3	No 567/0
Shepshed, Leicestershire, LE12 9EJ	VL 0482	3. 29.07 - 12.08.05	with boom						Head	0.086	7	LOQ
The United Kingdom									Head	0.047	14	0.01 mg/kg
(AF/8817/BA/2)												
182014	Lettuce, head/	1. 11.07.05	foliar	0.003	400	0.0125	1	45	Head	0.282	0	Method
2006/1026855	Rheinea	2. n.a.	application				18.07.05		Head	0.117	3	No 567/0
Bedelaarspad, 5976 Kronenberg, Limburg	VL 0482	3. 18.07 - 01.08.05	with boom						Head	0.042	7	LOQ
The Netherlands									Head	<0.01	14	0.01 mg/kg
(AF/8817/BA/3)												
182014	Lettuce, head/	1. 11.07.05	foliar	0.003	400	0.0125	2	45	Head	0.442	0	Method
2006/1026855	Rheinea	2. n.a.	application	0.003	400	0.0125	18.07.05		Head	0.125	3	No 567/0
Bedelaarspad, 5976 Kronenberg, Limburg	VL 0482	3. 18.07 - 01.08.05	with boom						Head	0.052	7	LOQ
The Netherlands									Head	<0.01	14	0.01 mg/kg
(AF/8817/BA/3)												
182014	Lettuce, head/	1. 18.08.05	foliar	0.003	400	0.0125	1	48-49	Head	0.399	0	Method
2006/1026855	Nobilan	2. n.a.	application				07.10.05		Head	0.165	3	No 567/0
Hundepfuhlstr, 30966 Hemmingen/ Arnum,	VL 0482	3. 07.10 - 21.10.05	with boom						Head	0.167	7	LOQ
Germany									Head	0.112	14	0.01 mg/kg
(AF/8817/BA/4)												

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182014	Lettuce, head/	1. 18.08.05	foliar	0.003	400	0.0125	2	48-49	Head	0.315	0	Method
2006/1026855	Nobilan	2. n.a.	application	0.003	400	0.0125	07.10.05		Head	0.221	3	No 567/0
Hundepfuhlstr, 30966 Hemmingen/ Arnum, Germany	VL 0482	3. 07.10 - 21.10.05	with boom						Head	0.176	7	LOQ
(AF/8817/BA/4)									Head	0.085	14	0.01 mg/kg
182062	Lettuce, head/	1. 10.05.06	foliar	0.003	400	0.0125	1	47-48	Head	0.131	0	Method
2007/1007938	Jambis	2. n.a.	application				19.06.06		Head	0.036	3	No 567/0
St Lambert des Levées, 49400, Maine-et-Loire, France	VL 0482	3. 19.06 - 03.07.06	with boom						Head	0.018	7	LOQ
(AF/10503/BA/1)									Head	<0.01	14	0.01 mg/kg
182062	Lettuce, head/	1. 10.05.06	foliar	0.003	400	0.0125	2	47-48	Head	0.193	0	Method
2007/1007938	Jambis	2. n.a.	application	0.003	400	0.0125	19.06.06		Head	0.072	3	No 567/0
St Lambert des Levées, 49400, Maine-et-Loire	VL 0482	3. 19.06 - 03.07.06	with boom						Head	0.018	7	LOQ
France (AF/10503/BA/1)									Head	<0.01	14	0.01 mg/kg
182062	Lettuce, head/	1. 12.07.06	foliar	0.003	400	0.0125	1	45	Head	0.088	0	Method
2007/1007938	Igoma	2. n.a.	application				21.08.06		Head	<0.01	3	No 567/0
Haskayne, Preston, Lancashire	VL 0482	3. 21.08 - 04.09.06	with boom						Head	<0.01	7	LOQ
The United Kingdom (AF/10503/BA/2)									Head	<0.01	14	0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182062	Lettuce, head/	1. 12.07.06	foliar	0.003	400	0.0125	2	45	Head	0.134	0	Method
2007/1007938	Igoma	2. n.a.	application	0.003	400	0.0125	21.08.06		Head	<0.01	3	No 567/0
Haskayne, Preston, Lancashire	VL 0482	3. 21.08 - 04.09.06	with boom						Head	<0.01	7	LOQ
The United Kingdom									Head	<0.01	14	0.01 mg/kg
(AF/10503/BA/2)												
227104	Lettuce, head/	1. 02.08.06	foliar	0.003	381	0.012	1	45	Head	0.38	0	Method
2007/1008496	Lucan	2. -	spray using				13.09.06		Head	0.13	3	No 567/0
Sedan (Champagne-Ardennes)	VL 0482	3. 26.09.06	boom						Head	0.080	7	LOQ
France			sprayer						Head	0.025	13	0.01 mg/kg
(A/NF/I/06/106)												
227104	Lettuce, head/	1. 02.08.06	foliar	0.003	403	0.013	2	45	Head	0.35	0	Method
2007/1008496	Lucan	2. -	spray using	0.003	379	0.012	13.09.06		Head	0.27	3	No 567/0
Sedan (Champagne-Ardennes)	VL 0482	3. 26.09.06	boom						Head	0.12	7	LOQ
France			sprayer						Head	0.039	13	0.01 mg/kg
(A/NF/I/06/106)												
227104	Lettuce, head/	1. 13.06.06	foliar	0.003	337	0.011	1	45	Head	0.024	0	Method
2007/1008496	mixture of	2. -	spray using				20.07.06		Head	0.014	2	No 567/0
Pegau (Sachsen)	Dynnmite and	3. 03.08.06	boom						Head	<0.01	7	LOQ
Germany	Dolly		sprayer						Head	<0.01	14	0.01 mg/kg
(A/GE/I/06/107)	VL 0482											
227104	Lettuce, head/	1. 13.06.06	foliar	0.003	413	0.013	2	45	Head	0.030	0	Method
2007/1008496	mixture of	2. -	spray using	0.003	340	0.011	20.07.06		Head	0.021	2	No 567/0
Pegau (Sachsen)	Dynnmite and	3. 03.08.06	boom						Head	<0.01	7	LOQ
Germany	Dolly		sprayer						Head	<0.01	14	0.01 mg/kg
(A/GE/I/06/107)	VL 0482											

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
227104	Lettuce, head/	1. 08.05.06	foliar	0.003	388	0.012	1	47	Head	0.13	0	Method
2007/1008496	Matilda	2. -	spray using				27.06.06		Head	0.096	3	No 567/0
Middelfart (Fyn)	VL 0482	3. 11.07.06	boom						Head	0.027	7	LOQ
Denmark			sprayer						Head	<0.01	14	0.01 mg/kg
(A/DK/I/06/108)												
227104	Lettuce, head/	1. 08.05.06	foliar	0.003	392	0.012	2	47	Head	0.13	0	Method
2007/1008496	Matilda	2. -	spray using	0.003	398	0.012	27.06.06		Head	0.12	3	No 567/0
Middelfart (Fyn)	VL 0482	3. 11.07.06	boom						Head	0.042	7	LOQ
Denmark			sprayer						Head	0.010	14	0.01 mg/kg
(A/DK/I/06/108)												
227104	Lettuce, head/	1. 19.04.06	foliar	0.003	417	0.013	1	45	Head	0.14	0	Method
2007/1008496	Pinochio	2. -	spray using				26.06.06		Head	0.041	3	No 567/0
Badsey (Gloucestershire)	VL 0482	3. 10.07.06	boom						Head	0.027	7	LOQ
The United Kingdom			sprayer						Head	0.010	14	0.01 mg/kg
(A/UK/I/06/109)												
227104	Lettuce, head/	1. 19.04.06	foliar	0.003	365	0.011	2	45	Head	0.13	0	Method
2007/1008496	Pinochio	2. -	spray using	0.003	410	0.013	26.06.06		Head	0.055	3	No 567/0
Badsey (Gloucestershire)	VL 0482	3. 10.07.06	boom						Head	0.055	7	LOQ
The United Kingdom			sprayer						Head	0.013	14	0.01 mg/kg
(A/UK/I/06/109)												

0) actual application rates varied by 10% at most
1) days after last application
2) at last treatment

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	WG (BAS 310 08 I)	**Residues calculated as:**	Total cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182062	Lettuce, head/	1. 10.05.06	foliar	0.003	400	0.0125	1	47-48	Head	0.024	0	Method
2007/1007938	Jambis	2. n.a.	application				19.06.06		Head	0.028	3	No 567/0
St Lambert des Levées, 49400, Maine-et-Loire, France	VL 0482	3. 19.06 - 03.07.06	with boom						Head	0.019	7	LOQ
(AF/10503/BA/1)									Head	<0.01	14	0.01 mg/kg
182062	Lettuce, head/	1. 10.05.06	foliar	0.003	400	0.0125	2	47-48	Head	0.090	0	Method
2007/1007938	Jambis	2. n.a.	application	0.003	400	0.0125	19.06.06		Head	0.057	3	No 567/0
St Lambert des Levées, 49400, Maine-et-Loire, France	VL 0482	3. 19.06 - 03.07.06	with boom						Head	0.022	7	LOQ
(AF/10503/BA/1)									Head	<0.01	14	0.01 mg/kg
182062	Lettuce, head/	1. 12.07.06	foliar	0.003	400	0.0125	1	45	Head	0.049	0	Method
2007/1007938	Igoma	2. n.a.	application				21.08.06		Head	<0.01	3	No 567/0
Haskayne, Preston, Lancashire	VL 0482	3. 21.08 - 04.09.06	with boom						Head	<0.01	7	LOQ
The United Kingdom (AF/10503/BA/2)									Head	<0.01	14	0.01 mg/kg
182062	Lettuce, head/	1. 12.07.06	foliar	0.003	400	0.0125	2	45	Head	0.131	0	Method
2007/1007938	Igoma	2. n.a.	application	0.003	400	0.0125	21.08.06		Head	<0.01	3	No 567/0
Haskayne, Preston, Lancashire	VL 0482	3. 21.08 - 04.09.06	with boom						Head	<0.01	7	LOQ
The United Kingdom (AF/10503/BA/2)									Head	<0.01	14	0.01 mg/kg

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

Residue studies reported in supplement information

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany, The Netherlands	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 51 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090703 Germany - 67245 Lamsheim (L080177)	Lettuce (Tizian)	1. 2008 2. 21.10.2008 3. n. a.	Gloria pneumatic sprayer, installed on an unicycle	0.0038	400	0.0150	2 07.10.2008	48	head head head head	0.66 0.35 0.33 0.10	0 3 7 13	BASF method No. 567/0
DocID 2009/1090703 9541 XL, Vlagtwedde Netherlands (L080178)	Lettuce (Santoro)	1. 14.08.2008 2. 06.10.2008 3. n. a.	Plotsprayer (Agrotop)	0.0038	400	0.0150	2 29.09.2008	47	head head head head	0.29 0.10 0.10 0.06	0 3 7 15	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany, The Netherlands	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090703 Germany - 67245 Lamsheim (L080177)	Lettuce (Tizian)	1. 2008 2. 21.10.2008 3. n. a.	Gloria pneumatic sprayer, installed on an unicycle	0.0038	400	0.0150	2 07.10.2008	48	head head head head	0.36 0.22 0.15 0.12	0 3 7 13	BASF method No. 567/0
DocID 2009/1090703 9541 XL, Vlagtwedde Netherlands (L080178)	Lettuce (Santoro)	1. 14.08.2008 2. 06.10.2008 3. n. a.	Plotsprayer (Agrotop)	0.0038	400	0.0150	2 29.09.2008	47	head head head head	0.39 0.14 0.13 0.06	0 3 7 15	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	Fastac EC	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID AL-726-012 France - Machecoul, Pays de la Loire (MA/44/05/1)	Lambs Lettuce (Verte de Cambrai)	1. n. a. 2. n. a. 3. 13.10.1992	foliar spray	0.001	1000	0.010	2 06.10.1992			0.294	7	use of plastic tunnel 8 days after sowing
DocID AL-726-012 France - Machecoul, Pays de la Loire (MA/44/05/2)	Lambs Lettuce (Verte de Cambrai)	1. n. a. 2. n. a. 3. 13.10.1992	foliar spray	0.001	1000	0.010	2 06.10.1992			0.283	7	use of plastic tunnel 8 days after sowing

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 03 I SC	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/1006469 Germany - 16818 Wustrau (ACK/03/03)	Lettuce (Nadine)	1. 12.05.2003 2. n. a. 3. 14.-18.06.2003	foliar spray	0.005	200	0.009	1 10.06.2003	45	Heads Heads Heads	0.189 0.099 <0.05	0 3 7	BASF Method No. 546/0; Residues of BAS 310 03 I
DocID 2004/1006469 Germany - 47638 Straelen (AGR/03/03)	Lettuce (Jesina)	1. 02.05.2003 2. n. a. 3. 05.-18.06.2003	foliar spray	0.005	200	0.009	1 03.06.2003	48	Heads Heads Heads	0.441 0.068 0.054	0 3 7	BASF Method No. 546/0; Residues of BAS 310 03 I
DocID 2004/1006469 Germany - 69121 Handschuhsheim (DU2/03/03)	Lettuce (Ponchito)	1. 08.05.2003 2. n. a. 3. 27.06.2003	foliar spray	0.005	200	0.009	1 23.06.2003	48	Heads Heads Heads	0.073 <0.05 0.060	0 3 7	BASF Method No. 546/0; Residues of BAS 310 03 I
DocID 2004/1006469 Germany - 67376 Harthausen (DU4/03/03)	Lettuce (Nadine)	1. 15.05.2003 2. n. a. 3. 27.06.2003	foliar spray	0.005	200	0.009	1 23.06.2003	49	Heads Heads Heads	0.232 0.149 0.063	0 3 7	BASF Method No. 546/0; Residues of BAS 310 03 I

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 QC I	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/1006469 Germany - 16818 Wustrau (ACK/03/03)	Lettuce (Nadine)	1. 12.05.2003 2. n. a. 3. 14.-18.06.2003	foliar spray	0.005	200	0.009	1 10.06.2003	45	Heads Heads Heads	0.174 0.108 <0.05	0 3 7	BASF Method No. 546/0; Residues of BAS 310 QCI
DocID 2004/1006469 Germany - 47638 Straelen (AGR/03/03)	Lettuce (Jesina)	1. 02.05.2003 2. n. a. 3. 05.-18.06.2003	foliar spray	0.005	200	0.009	1 03.06.2003	48	Heads Heads Heads	0.287 0.091 0.054	0 3 7	BASF Method No. 546/0; Residues of BAS 310 QCI
DocID 2004/1006469 Germany - 69121 Handschuhsheim (DU2/03/03)	Lettuce (Ponchito)	1. 08.05.2003 2. n. a. 3. 27.06.2003	foliar spray	0.005	200	0.009	1 23.06.2003	48	Heads Heads Heads	0.155 0.058 <0.05	0 3 7	BASF Method No. 546/0; Residues of BAS 310 QCI
DocID 2004/1006469 Germany - 67376 Harthausen (DU4/03/03)	Lettuce (Nadine)	1. 15.05.2003 2. n. a. 3. 27.06.2003	foliar spray	0.005	200	0.009	1 23.06.2003	49	Heads Heads Heads	0.105 0.104 0.082	0 3 7	BASF Method No. 546/0; Residues of BAS 310 QCI

n. a. not available

Lettuce

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182014 2006/1026855 31790 St Jory France (AF/8817/BA/5)	Lettuce, head/ Sagesse VL 0482	1. 16.08.05 2. n.a. 3. 09.09 - 23.09.05	foliar application with boom	0.006	400	0.025	1 09.09.05	41-43	Head Head Head Head	0.448 0.103 0.067 0.013	0 3 7 14	Method No 567/0 LOQ 0.01 mg/kg
182014 2006/1026855 Granarolo, Bologna, 40057 Italy (AF/8817/BA/6)	Lettuce, head/ Gentilina VL 0482	1. 11.07.05 2. n.a. 3. 02.08 - 16.08.05	foliar application with boom	0.006	400	0.025	1 02.08.05	45-46	Head Head Head Head	0.737 0.305 0.065 0.021	0 3 8 14	Method No 567/0 LOQ 0.01 mg/kg
182014 2006/1026855 El Viso del Alcor, 41520, Spain (AF/8817/BA/7)	Lettuce, head/ Filipus VL 0482	1. 10.06.05 2. n.a. 3. 15.07 - 29.07.05	foliar application with boom	0.006	400	0.025	1 15.07.05	47	Head Head Head Head	0.299 0.210 0.062 0.022	0 3 7 14	Method No 567/0 LOQ 0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
182014	Lettuce, head/	1. 08.09.05	foliar	0.006	400	0.025	1	47	Head	1.161* (1.354/0.968)	0	Method
2006/1026855	Atraxion	2. n.a.	application				31.10.05		Head	0.585* (0.535/0.635)	3	No 567/0
Thessaloniki, Central Macedonia, GR-57200,	VL 0482	3. 31.10 - 15.11.05	with boom						Head	0.520* (0.534/0.482/ 0.543)	7	LOQ
Greece									Head	0.112* (0.08/0.145)	15	0.01 mg/kg
(AF/8817/BA/8)												
182062	Lettuce, head/	1. 22.04.06	foliar	0.006	400	0.025	1	35-37	Head	0.424	0	Method
2007/1007938	Filipo	2. n.a.	application				26.05.06		Head	0.139	3	No 567/0
El Viso del Alcor, Sevilla, Andalusia	VL 0482	3. 26.05 - 09.06.06	with boom						Head	0.106	6	LOQ
Spain									Head	<0.01	14	0.01 mg/kg
(AF/10503/BA/3)												
182062	Lettuce, head/	1. 01.03.06	foliar	0.006	400	0.025	1	46-47	Head	0.512	0	Method
2007/1007938	Gentilina	2. n.a.	application				26.04.06		Head	0.387	2	No 567/0
Granarolo, Bologna, Emilia-Romagna	VL 0482	3. 26.04 - 10.05.06	with boom						Head	0.065	7	LOQ
Italy									Head	<0.01	14	0.01 mg/kg
(AF/10503/BA/4)												
227104	Lettuce, head/	1. 27.07.06	foliar	0.006	417	0.026	1	47	Head	0.36	0	Method
2007/1008496	Soleilan	2. -	spray using				31.08.06		Head	0.32	3	No 567/0
Noves (Bouches de Rhone)	VL 0482	3. 14.09.06	boom						Head	0.10	7	LOQ
France			sprayer						Head	0.021	14	0.01 mg/kg
(A/SF/I/06/110)												

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
227104	Lettuce, head/	1. 05.07.06	foliar	0.006	372	0.023	1	45	Head	0.24	0	Method
2007/1008496	Filippus	2. -	spray using				26.07.06		Head	0.11	3	No 567/0
Torrent (Valencia)	VL 0482	3. 08.08.06	knapsack						Head	0.038	8	LOQ
Spain			sprayer						Head	0.011	13	0.01 mg/kg
(A/SP/I/06/111)												
227104	Lettuce, head/	1. 18.09.2006	foliar	0.006	410	0.026	1	30-35	Head	1.1	0	Method
2007/1008496	Carinos	2. -	spray using				17.10.06		Head	0.41	3	No 567/0
Cassibile (Siracusa - Sicily)	VL 0482	3. 31.10.2006	knapsack						Head	0.12	7	LOQ
Italy			sprayer						Head	0.031	14	0.01 mg/kg
(A/IT/I/06/112)												
227104	Lettuce, head/	1. 26.05.06	foliar	0.006	405	0.025	1	44	Head	0.33	0	Method
2007/1008496	Aberam	2. -	spray using				29.06.06		Head	<0.01	4	No 567/0
Nea Magnisia (Central Macedonia	VL 0482	3. 13.07.06	boom						Head	<0.01	8	LOQ
Greece			sprayer						Head	<0.01	14	0.01 mg/kg
(A/GR/I/06/113)												

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	WG (BAS 310 08 I)	**Residues calculated as:**	Total cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
182062	Lettuce, head/	1. 22.04.06	foliar	0.006	400	0.025	1	35-37	Head	0.228	0	Method
2007/1007938	Filipo	2. n.a.	application				26.05.06		Head	0.195	3	No 567/0
El Viso del Alcor, Sevilla, Andalusia	VL 0482	3. 26.05 - 09.06.06	with boom						Head	0.131	6	LOQ
Spain									Head	0.013	14	0.01 mg/kg
(AF/10503/BA/3)												
182062	Lettuce, head/	1. 01.03.06	foliar	0.006	400	0.025	1	46-47	Head	0.475	0	Method
2007/1007938	Gentilina	2. n.a.	application				26.04.06		Head	0.289	2	No 567/0
Granarolo, Bologna, Emilia-Romagna	VL 0482	3. 26.04 - 10.05.06	with boom						Head	0.035	7	LOQ
Italy									Head	<0.01	14	0.01 mg/kg
(AF/10503/BA/4)												

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety** (a)	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest** (b)	**Method of Treatment** (c)	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date** (d)	**Growth Stage (BBCH)[1]** (e)	**Portion Analysed** (a)	**Residues (mg/kg)**	**DALA (days)[2]** (f)	**Remarks** (g)
408662	Lettuce,	1. 05.07.13	broadcast	0.006	200	0.0125	2	45-47	Head	0.44	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	30.07.13		Head	0.42	1	No 567/0
82000 Montauban, Tarn-et-Garonne	Kirina	3. 06.08.13	application						Head	0.24	3	LOQ
France	VL 0483								Head	0.089	7	0.01 mg/kg
(L130050)												
408662	Lettuce,	1. 22.04.13	broadcast	0.006	200	0.0125	2	47-49	Head	0.13	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	29.05.13		Head	0.082	1	No 567/0
57008 Nea Magnisia, Thessaloniki Central Macedonia	Parris Island	3. 30.05. -05.06.13	application						Head	0.075	2	LOQ
Greece	VL 0483								Head	0.015	8	0.01 mg/kg
(L130051)												
408662	Lettuce,	1. 02.04.13	broadcast	0.006	200	0.0125	2	47-49	Head	0.10	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	24.06.13		Head	0.084	1	No 567/0
40057 San Marino di Bentivoglio, Provincia di Bologna	Foglia di Quercia	3. 10.07.13	application						Head	0.032	3	LOQ
Italy	VL 0483								Head	0.012	7	0.01 mg/kg
(L130052)												
408662	Lettuce,	1. 05.03.13	broadcast	0.006	200	0.0125	2	49	Head	0.38	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	13.05.13		Head	0.23	1	No 567/0
41140 Conil de la Frontera, Andalucia	Flavius	3. 20.05.13	application						Head	0.21	3	LOQ
Spain	VL 0483								Head	0.094	7	0.01 mg/kg
(L130053)												

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety** (a)	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest** (b)	**Method of Treatment** (c)	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date** (d)	**Growth Stage (BBCH)[1]** (e)	**Portion Analysed** (a)	**Residues (mg/kg)**	**DALA (days)[2]** (f)	**Remarks** (g)
408662	Lettuce,	1. 06.08.13	broadcast	0.006	200	0.0125	2	47	Head	0.12	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	09.09.13		Head	0.17	1	No 567/0
82290 Meauzac, Tarn-et-Garonne	Ukulele	3. 16.09.13	application						Head	0.049	3	LOQ
France	VL 0483								Head	0.042	7	0.01 mg/kg
(L130054)												
408662	Lettuce,	1. 27.05.13	broadcast	0.006	200	0.0125	2	47	Head	0.23	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	25.06.13		Head	0.19	1	No 567/0
57007 Chalkidona, Thessaloniki, Central Macedonia	Magister F1	3. 25.06.13-10.07.13	application						Head	0.11	3	LOQ
Greece	VL 0483								Head	0.031	7	0.01 mg/kg
(L130055)												
408662	Lettuce,	1. 23.08.13	broadcast	0.006	200	0.0125	2	48	Head	0.063	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	22.10.13		Head	0.058	1	No 567/0
40055 Castellano, Provincia di Bologna	Analena	3. 28.10.13	application						Head	0.037	2	LOQ
Italy	VL 0483								Head	0.014	6	0.01 mg/kg
(L130056)												

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce (leafy vegetables)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation (common name and content)**	-
Content of active substance (g/kg or g/L)	50 g/L		
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety (a)**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest (b)**	**Method of Treatment (c)**	**Application Rate per Treatment[0] kg a.s./hL**	**Water (L/ha)**	**kg a.s./ha**	**No of Treatm. and Last Date (d)**	**Growth Stage (BBCH)[1] (e)**	**Portion Analysed (a)**	**Residues (mg/kg)**	**DALA (days)[2] (f)**	**Remarks (g)**
408662	Lettuce,	1. 12.04.13	broadcast	0.006	200	0.0125	2	49	Head	0.19	0	Method
2014/1140312	open leaf /	2. -	foliar	0.006	200	0.0125	14.05.13		Head	0.18	1	No 567/0
41520 El Viso del Alcor, Andalucia	Filipo	3. 21.05.13	application						Head	0.091	3	LOQ
Spain	VL 0483								Head	0.054	7	0.01 mg/kg
(L130057)												

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

Residue studies reported in supplement information

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**-**
Crop/crop group:	**Lettuce/leafy vegetables**	**Producer of commercial product**	**67056 Ludwigshafen, Germany**
Responsible body for reporting (name, address)	**BASF SE, 67056 Ludwigshafen**	**Indoor/Glasshouse/Outdoor**	**outdoor**
Country	**France**	**Other active substance in the formulation**	**none**
Content of active substance (g/kg or g/L)	**100 g/L**	**(common name and content)**	
Formulation (e.g. WP)	**BAS 310 I (EC)**	**Residues calculated as:**	**Alpha-Cypermethrin**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-726-004 BEGR.82.035 France - France 33 – Eysines (W/FR/E 81/446)	Lettuce (Capitan)	1. 09.09.81 2. - 3. Sampling 23.10.-02.11.81; near harvest (one week)	spraying	0.0009	1000	0.009	1 23.10.81	near harvest	lettuce lettuce lettuce	0.31 0.21 0.12	0 3 10	Method SAMS 233-1
AL-726-004 BEGR.82.035 France - France 33 – Eysines (W/FR/E 81/446)	Lettuce (Capitan)	1. 09.09.81 2. - 3. Sampling 23.10.-02.11.81; near harvest (one week)		0.0015	1000	0.015	1 23.10.81	near harvest	lettuce lettuce lettuce	0.41 0.28 0.17	0 3 10	Method SAMS 233-1

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Italy	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 51 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090703 France - St. Avit St. Nazaire (L080179)	Lettuce (Forlina)	1. 18.08.2008 2. 29.09.2008 3. n. a.	Agrotop Plot sprayer	0.0075	400	0.0300	1 17.09.2008	41	head head head head	0.42 0.20 0.11 0.07	0 2 6 13	BASF method No. 567/0
DocID 2009/1090703 Italy - 40127 Viadagola - Granarolo (L080180)	Lettuce (Noisette)	1. 29.05.2008 2. 08.07.-15.07.2008 3. n. a.	Echo SHR 150 SI	0.0075	400	0.0300	1 01.07.2008	47	head head head head	0.56 0.17 0.07 0.02	0 3 7 14	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Lettuce/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Italy	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090703 France - St. Avit St. Nazaire (L080179)	Lettuce (Forlina)	1. 18.08.2008 2. 29.09.2008 3. n. a.	Agrotop Plot sprayer	0.0075	400	0.0300	1 17.09.2008	41	head head head head	0.46 0.19 0.15 0.09	0 2 6 13	BASF method No. 567/0
DocID 2009/1090703 Italy - 40127 Viadagola - Granarolo (L080180)	Lettuce (Noisette)	1. 29.05.2008 2. 08.07.-15.07.2008 3. n. a.	Echo SHR 150 SI	0.0075	400	0.0300	1 01.07.2008	47	head head head head	0.40 0.21 0.15 0.03	0 3 7 14	BASF method No. 567/0

n. a. not available

Residue studies reported in supplement information

Glasshouse

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**-**
Crop/crop group:	**Lettuce/leafy vegetables**	**Producer of commercial product**	**67056 Ludwigshafen, Germany**
Responsible body for reporting (name, address)	**BASF SE, 67056 Ludwigshafen**	**Indoor/Glasshouse/Outdoor**	**indoor**
Country	**Belgium, Denmark, France, Germany, Greece, Italy, Spain**	**Other active substance in the formulation**	**none**
Content of active substance (g/kg or g/L)	**100 g/L**	**(common name and content)**	
Formulation (e.g. WP)	**BAS 310 40 I (EC)**	**Residues calculated as:**	**Alpha-Cypermethrin**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1037555 25 Rue Motte 37700 Saint Pierre des Corps, Centre France (05 I CL FR P33)	VL 0482 Mathilda	1. 23.09.2005 2. N/A 3. 12.11.2005	Pulvexper, spray boom with nozzles	0.0100	400	0.0400	1 25.10.2005	41	head head head head	0.93 1.10 0.68 0.43	0 3 6 14	BASF method No. 567/0
2006/1037555 Luchstraße 16833 Fehrbellin Brandenburg Germany (AC/05/044)	VL 0482 Ponchito	1. 12.08.2005 2. N/A 3. 15.09.2005	custom built bicycle mounted boom sprayer	0.0100	400	0.0400	1 08.09.2005	44	head head head head	1.05 0.21 0.09 0.02	0 4 7 13	BASF method No. 567/0
2006/1037555 3454 Rummen Limburg Belgium (AGR/52/05)	VL 0482 Omega	1. 24.05.2005 2. N/A 3. 24.06.2005	VCR portable knapsack boom sprayer	0.0100	400	0.0400	1 16.06.2005	47	head head head head	1.04 0.44 0.21 0.05	0 3 8 14	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**-**
Crop/crop group:	**Lettuce/leafy vegetables**	**Producer of commercial product**	**67056 Ludwigshafen, Germany**
Responsible body for reporting (name, address)	**BASF SE, 67056 Ludwigshafen**	**Indoor/Glasshouse/Outdoor**	**indoor**
Country	**Belgium, Denmark, France, Germany, Greece, Italy, Spain**	**Other active substance in the formulation**	**none**
Content of active substance (g/kg or g/L)	**100 g/L**	**(common name and content)**	
Formulation (e.g. WP)	**BAS 310 40 I (EC)**	**Residues calculated as:**	**Alpha-Cypermethrin**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1037555 Tuevej 39 7000 Fredericia Denmark (ALB/190509-01)	VL 0482 Hawaii	1. 20.05.2005 2. N/A 3. 20.06.-30.06.2005	spray boom #112, air assisted Knapsack precision sprayer with 2 m boom	0.0100	400	0.0400	1 17.06.2005	44	head head head head	0.61 0.43 0.27 0.18	0 3 7 14	BASF method No. 567/0
2006/1037555 Quartier Les Ferrages 13550 Noves Provence France (05 I CL FR P37)	VL 0482 Boreal	1. 06.10.2005 2. N/A 3. 05.12.2005	Pulvexper spray boom with nozzles	0.0100	400	0.0400	1 29.11.2005	47	head head head head	0.93 0.77 0.68 0.45	0 3 7 14	BASF method No. 567/0
2006/1037555 Via Giare, 10 45020 Lusia (RO) Rovigo Italy (IR05BASI11LG01)	VL 0482 Limax	1. 10.06.2005 2. N/A 3. 29.06.-13.07.2005	hand held sprayer (GZ SIP 02)	0.0100	400	0.0400	1 29.06.2005	47	head head head head	0.95 0.85 0.30 0.15	0 3 7 14	BASF method No. 567/0
2006/1037555 Finca La Dehesilla 41710 Utrera Andalucia Sevilla Spain (05ES/084R)	VL 0482 Filipus	1. 23.08.2005 2. N/A 3. 21.10.-25.10.2005	Schachtner air compressed boom sprayer	0.0100	400	0.0400	1 07.10.2005	47	head head head head	0.80 0.56 0.30 0.20	0 3 7 13	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**-**
Crop/crop group:	**Lettuce/leafy vegetables**	**Producer of commercial product**	**67056 Ludwigshafen, Germany**
Responsible body for reporting (name, address)	**BASF SE, 67056 Ludwigshafen**	**Indoor/Glasshouse/Outdoor**	**indoor**
Country	**Belgium, Denmark, France, Germany, Greece, Italy, Spain**	**Other active substance in the formulation**	**none**
Content of active substance (g/kg or g/L)	**100 g/L**	**(common name and content)**	
Formulation (e.g. WP)	**BAS 310 40 I (EC)**	**Residues calculated as:**	**Alpha-Cypermethrin**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1037555 Profitis Thessaloniki Central Macedonia GR 57200 Greece (05RF045)	VL 0482 Atraxion	1. 01.08.2005 2. N/A 3. 10.10.-30.10.2005	Azo pressurized gas sprayer with 6-nozzle boom	0.0100	400	0.0400	1 12.10.2005	47	head head head head	1.65 0.77 0.57 0.15	0 2 7 13	BASF method No. 567/0

- **Oilseed rape**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation (common name and content)**	none
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
311992													BASF
2008/1019999	Oilseed rape /	1.	22.08.06	foliar	0.004	300	0.0105	2	79-80	whole plant w/o root	0.159	0	method no.
Aulnay-la-Riviere	Adriana	2.	-	appl.	0.004	300	0.0105	04.06.07		seed	<0.01	14	567/0 and
Loiret,	SO 0088	3.	04.06-	with						seed	<0.01	21	567/1
45390			02.07.07	AUK						seed	<0.01	28	LOQ = 0.01
France				plot									mg/kg
AF/12151/BA/2				sprayer									
311992													BASF
2008/1019999	Oilseed rape /	1.	20.08.06	foliar	0.004	300	0.0105	2	79-80	whole plant w/o root	0.109	0	method no.
Roinvilliers,	Robuste	2.	-	appl.	0.004	300	0.0105	04.06.07		whole pods	0.098	14	567/0 and
Essonne,	SO 0088	3.	04.06-	with						seed	<0.01	21	567/1
91150			02.07.07	AUK						seed	<0.01	28	LOQ = 0.01
France				plot									mg/kg
AF/12151/BA/3				sprayer									

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
319697													BASF
2009/1090702	Oilseed rape /	1.	27.08.07	Plot-	0.005	300	0.015	2	87	whole plant (no roots)	0.17	0	method no.
Kirchstrasse 9	Elektra	2.	09.04.-	sprayer	0.005	300	0.015	11.07.08		seed	<0.01	13	567/0
26169 Altenoythe	SO 0495		05.05.08							seed	<0.01	20	
		3.	24.07.08							seed	<0.01	27	LOQ = 0.01
Germany													mg/kg
L080185													
319697													BASF
2009/1090702	Oilseed rape /	1.	07.09.07	Boom-	0.005	300	0.015	2	79	whole plant (no roots)	0.16	0	method no.
2 rue du Général Patton	Campo	2.	05.05.-	sprayer	0.005	300	0.015	18.06.08		pods with seed	0.03	15	567/0
	SO 0495		20.05.08							seed	0.01	22	
51220 Courcy		3.	18.07.08							seed	<0.01	27	LOQ = 0.01
France													mg/kg
L080186													

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67117 Limburgerhof	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 51 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
319697 2009/1090702 Kirchstrasse 9 26169 Altenoythe Germany L080185	Oilseed rape / Elektra SO 0495	1. 2. 3.	27.08.07 09.04.- 05.05.08 24.07.08	Plot- sprayer	0.005 0.005	300 300	0.015 0.015	2 11.07.08	87	whole plant (no roots) seed seed seed	0.20 <0.01 <0.01 <0.01	0 13 20 27	BASF method no. 567/0 LOQ = 0.01 mg/kg
319697 2009/1090702 2 rue du Général Patton 51220 Courcy France L080186	Oilseed rape / Campo SO 0495	1. 2. 3.	07.09.07 05.05.- 20.05.08 18.07.08	Boom- Sprayer	0.005	300	0.015	2 18.06.08	79	whole plant (no roots) pods with seed seed seed	0.26 0.05 <0.01 <0.01	0 15 22 27	BASF method no. 567/0 LOQ = 0.01 mg/kg

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408612	Oilseed rape/	1. 06.04.12	foliar	0.00625	200	0.0125	2	87	whole plant*	0.49	0	Method
2013/1037957	Heros	2. 20.07.-30.07.12	application	0.00625	200	0.0125	15.08.12		seed	<0.01	14	No 567/0
47574 Goch-Kessel	SO 0495	3. 05.09.12							rest of plant*	0.42	14	LOQ
Germany									seed	<0.01	21	0.01 mg/kg
(L120460)									rest of plant*	0.22	21	
									seed	<0.01	29	
									rest of plant*	0.16	29	
408612	Oilseed rape/	1. 03.09.11	foliar	0.00625	200	0.0125	2	79	whole plant*	0.19	0	Method
2013/1037957	DK Exquisite	2. 05.05.-20.05.12	application	0.00625	200	0.0125	19.06.12		rest of plant*	0.063	14	No 567/0
3470 Kortenaken	SO 0495	3. 10.07.12							pods with seeds	0.025	14	LOQ
Belgium									rest of plant*	0.059	21	0.01 mg/kg
(L120461)									pods with seeds	0.039	21	
									seed	<0.01	29	
									rest of plant*	0.11	29	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408616	Oilseed rape/	1. 04.09.12	Plot 2	0.006	200	0.0125	24.06.13	79-81	whole plant*	0.20	0	Method
2013/1416283	Marquis	2. 25.04.13-20.05.13	broadcast	0.006	228	0.0142	01.07.13		rest of plant*	0.10	14	No 567/0
16356 Blumberg	SO 0495	3. 30.07.13	foliar						pods with seeds	0.057	14	LOQ
Germany			application						seed	<0.01	22	0.01 mg/kg
(S13-00443-01 / L130038)									rest of plant*	0.10	22	
									seed	<0.01	29	
									rest of plant*	0.14	29	
408616	Oilseed rape/	1. 03.09.12	Plot 2	0.006	200	0.0125	24.06.13	80	whole plant*	0.31	0	Method
2013/1416283	DK Esquisite	2. 04.05.13-25.05.13	broadcast	0.006	200	0.0125	01.07.13		rest of plant*	0.15	14	No 567/0
1450 Cortil Noirmont	SO 0495	3. 29.07.13	foliar						pods with seeds	0.13	14	LOQ
Belgium			application						seed	<0.01	21	0.01 mg/kg
(S13-00443-02/ L130039)									rest of plant*	0.056	21	
									seed	<0.01	28	
									rest of plant*	0.066	28	
408616	Oilseed rape/	1. 05.09.12	Plot 2	0.006	200	0.0125	02.07.13	78	whole plant*	0.24	0	Method
2013/1416283	Safran	2. 01.05.13-15.06.13	broadcast	0.006	200	0.0125	09.07.13		rest of plant*	0.14	14	No 567/0
91150 Mespuits	SO 0495	3. 06.08.13	foliar						pods with seeds	0.16	14	LOQ
France			application						seed	<0.01	21	0.01 mg/kg
(S13-00443-03/ L130040)									rest of plant*	0.13	21	
									seed	<0.01	28	
									rest of plant*	0.27	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408616	Oilseed rape/	1. 19.04.13	Plot 2	0.006	200	0.0125	25.07.13	83	whole plant*	0.21	0	Method
2013/1416283	msl559c x ccc621205	2. nr	broadcast	0.006	200	0.0125	01.08.13		rest of plant*	0.055	13	No 567/0
6615 AJ Leur	SO 0495	3. Aug 13	foliar						pods with seeds	0.14	13	LOQ
The Netherlands			application						rest of plant*	<0.01	20	0.01 mg/kg
(S13-00443-04/ L130041)									pods with seeds	0.12	20	
									seed	<0.01	27	
									rest of plant*	0.13	27	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

Residue studies reported in supplement information

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	United Kingdom	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (WL85871)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-750-004 SBGR.84.014 Castle Howard Farms United Kingdom Trial no. not reported	Oilseed rape /Jet Neuf	1. 2. 3.	not reported not reported 31.07.83 (sampling)	spray	0.01	200	0.02	1 31.05.83	Not reported	Seed	<0.01	61	Method No. SAMS 351-1
BASF RDI No. AL-750-004 SBGR.84.014 Stratford Davies United Kingdom (WROSR 3)	Oilseed rape /Jet Neuf	1. 2. 3.	not reported not reported 26.07.83 (sampling)	spray	0.01	200	0.02	1 18.05.83	Not reported	Seed	<0.01	69	Method No. SAMS 351-1
BASF RDI No. AL-750-004 SBGR.84.014 S.Smart, United Kingdom (83/307)	Oilseed rape /Jet Neuf	1. 2. 3.	not reported not reported 29.07.83 (sampling)	spray	0.01	200	0.02	1 17.05.83	Not reported	Seed	<0.01	73	Method No. SAMS 351-1
BASF RDI No. AL-750-004 SBGR.84.014 P. Gordon United Kingdom (83/306)	Oilseed rape /Jet Neuf	1. 2. 3.	not reported not reported 25.07.83 (sampling)	spray	0.01	200	0.02	1 16.05.83	Not reported	Seed	<0.01	70	Method No. SAMS 351-1

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	United Kingdom	**Other active substance in the formulation (common name and content)**	none
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (WL85871)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-750-004 SBGR.84.014 J.Kidston United Kingdom (83/305)	Oilseed rape /Jet Neuf	1. 2. 3.	not reported not reported 30.07.83 (sampling)	spray	0.01	200	0.02	1 16.05.83	Not reported	Seed	<0.01	75	Method No. SAMS 351-1

1) at last treatment
2) days after last application

Oilseed rape

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Italy, Spain	**Other active substance in the formulation (common name and content)**	none
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
311992 2008/1019999 Montauben Tarn-et-Garonne, 82000 France AF/12151/BA/6	Oilseed rape / Corail SO 0088	1. 2. 3.	02.09.06 - 19.06.- 16.07.07	foliar appl. with AUK plot sprayer	0.004 0.004	300 300	0.0105 0.0105	2 19.06.07	89	whole plant* seed seed seed	0.327 <0.01 <0.01 <0.01	0 13 21 27	BASF method no. 567/0 and 567/1 LOQ = 0.01 mg/kg
311992 2008/1019999 Sistels, Tarn-et-Garonne 82340 France AF/12151/BA/7	Oilseed rape / Standy SO 0088	1. 2. 3.	20.09.06 - 18.06.- 16.07.07	foliar appl. with AUK plot sprayer	0.004 0.004	300 300	0.0105 0.0105	2 18.06.07	87	whole plant* seed seed seed	0.390 0.012 <0.01 <0.01	0 14 21 28	BASF method no. 567/0 and 567/1 LOQ = 0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
319697 2009/1090702 Fondo Carbonara Via Coronella 120 44028 Coronella, Ferrara Italy L080187	Oilseed rape / Fantasio SO 0495	1. 2. 3.	28.02.08 18.05.- 04.06.08 14.07.- 21.07.08	Honda WJR 2525	0.010	300	0.030	1 23.06.08	75	whole plant*) seed seed seed	0.54 0.01 <0.01 0.01	0 14 21 28	BASF method no. 567/0 LOQ = 0.01 mg/kg
319697 2009/1090702 SGS Espanola de Control S.A. 29780 Nerja, Malaga Spain L080188	Oilseed rape / Kabel SO 0495	1. 2. 3.	30.04.08 24.06.- 23.09.08 26.09.08	Boom- Sprayer	0.010	300	0.030	1 26.08.08	65	whole plant* seed seed seed	0.12 <0.01 <0.01 <0.01	0 14 21 28	BASF method no. 567/0 LOQ = 0.01 mg/kg

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 51 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
319697 2009/1090702 Fondo Carbonara Via Coronella 120 44028 Coronella, Ferrara Italy L080187	Oilseed rape / Fantasio SO 0495	1. 2. 3.	28.02.08 18.05.- 04.06.08 14.07.- 21.07.08	Honda WJR 2525	0.010	300	0.030	1 23.06.08	75	whole plant* seed seed seed	0.41 0.02 0.01 <0.01	0 14 21 28	BASF method no. 567/0 LOQ = 0.01 mg/kg
319697 2009/1090702 SGS Espanola de Control S.A. 29780 Nerja, Malaga Spain L080188	Oilseed rape / Kabel SO 0495	1. 2. 3.	30.04.08 24.06.- 23.09.08 26.09.08	Boom- Sprayer	0.010	300	0.030	1 26.08.08	65	whole plant* seed seed seed	0.33 0.01 <0.01 0.01	0 14 21 28	BASF method no. 567/0 LOQ = 0.01 mg/kg

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Oilseed rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408612	Oilseed rape/	1. 25.08.11	foliar	0.00625	200	0.0125	2	80	whole plant*	0.17	0	Method
2013/1037957	CSZ8992 + ES Alias	2. 10.04.-20.05.12	application	0.00625	200	0.0125	12.06.12		rest of plant*	0.087	13	No 567/0
17380 Torxè	SO 0495	3. 02.07.12							pods with seeds	0.037	13	LOQ
France									seed	<0.01	20	0.01 mg/kg
(L120462)									rest of plant*	0.093	20	
									seed	<0.01	28	
									rest of plant*	0.067	28	
408612	Oilseed rape/	1. 25.08.11	foliar	0.0125	200	0.025	1	80	whole plant*	0.25	0	Method
2013/1037957	CSZ8992 + ES Alias	2. 10.04.-20.05.12	application				12.06.12		rest of plant*	0.14	13	No 567/0
17380 Torxè	SO 0495	3. 02.07.12							pods with seeds	0.064	13	LOQ
France									seed	<0.01	20	0.01 mg/kg
(L120462)									rest of plant*	0.096	20	
									seed	<0.01	28	
									rest of plant*	0.061	28	
408612	Oilseed rape/	1. 30.11.11	foliar	0.00625	200	0.0125	2	77/79	whole plant*	0.28	0	Method
2013/1037957	PR46W31	2. 10.04.-30.04.12	application	0.00625	200	0.0125	21.05.12		seed	<0.01	14	No 567/0
57003 Nea Mesimvria, Thessaloniki	SO 0495	3. 11.06.12							rest of plant*	0.033	14	LOQ
Greece									seed	<0.01	21	0.01 mg/kg
(L120463)									rest of plant*	0.011	21	
									seed	<0.01	28	
									rest of plant*	0.018	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Oilseed rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	-
Content of active substance (g/kg or g/L)	50 g/L		
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408612	Oilseed rape/	1. 30.11.11	foliar	0.0125	200	0.025	1	77/79	whole plant*	0.44	0	Method
2013/1037957	PR46W31	2. 10.04.-30.04.12	application				21.05.12		seed	<0.01	14	No 567/0
57003 Nea Mesimvria, Thessaloniki	SO 0495	3. 11.06.12							rest of plant*	0.013	14	LOQ
Greece									seed	<0.01	21	0.01 mg/kg
(L120463)									rest of plant*	0.010	21	
									seed	<0.01	28	
									rest of plant*	<0.01	28	
408612	Oilseed rape/	1. 15.09.11	foliar	0.00625	200	0.0125	2	80	whole plant*	0.20	0	Method
2013/1037957	PR44W29	2. 15.04.-15.05.12	application	0.00625	200	0.0125	31.05.12		rest of plant*	0.10	13	No 567/0
20096 Pioltello	SO 0495	3. 21.06.12							pods with seeds	0.033	13	LOQ
Italy									seed	<0.01	21	0.01 mg/kg
(L120464)									rest of plant*	0.090	21	
									seed	<0.01	28	
									rest of plant*	0.19	28	
408612	Oilseed rape/	1. 15.09.11	foliar	0.0125	200	0.025	1	80	whole plant*	0.30	0	Method
2013/1037957	PR44W29	2. 15.04.-15.05.12	application				31.05.12		rest of plant*	0.12	13	No 567/0
20096 Pioltello	SO 0495	3. 21.06.12							pods with seeds	0.028	13	LOQ
Italy									seed	<0.01	21	0.01 mg/kg
(L120464)									rest of plant*	0.14	21	
									seed	<0.01	28	
									rest of plant*	0.22	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Oilseed rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	-
Content of active substance (g/kg or g/L)	50 g/L		
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408612	Oilseed rape/	1. 07.03.12	foliar	0.00625	200	0.0125	2	79	whole plant*	0.17	0	Method
2013/1037957	Kabel	2. 03.05.-03.06.12	application	0.00625	200	0.0125	11.06.12		rest of plant*	0.082	14	No 567/0
41710 Utrera	SO 0495	3. 03.07.12							pods with seeds	0.014	14	LOQ
Spain									seed	<0.01	22	0.01 mg/kg
(L120465)									rest of plant*	0.12	22	
									seed	<0.01	29	
									rest of plant*	0.13	29	
408612	Oilseed rape/	1. 07.03.12	foliar	0.0125	200	0.025	1	79	whole plant*	0.28	0	Method
2013/1037957	Kabel	2. 03.05.-03.06.12	application				11.06.12		rest of plant*	0.13	14	No 567/0
41710 Utrera	SO 0495	3. 03.07.12							pods with seeds	0.021	14	LOQ
Spain									seed	<0.01	22	0.01 mg/kg
(L120465)									rest of plant*	0.17	22	
									seed	<0.01	29	
									rest of plant*	0.17	29	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Oilseed rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408616	Oilseed rape/	1. 03.09.12	Plot 2	0.006	200	0.0125	03.06.13	80-81	whole plant*	0.29	0	Method
2013/1416283	Anderson	2. 05.04.13-25.04.13	broadcast	0.006	200	0.0125	10.06.13		rest of plant*	0.078	14	No 567/0
82290 Tarn et Garonne	SO 0495	3. 09.07.13	foliar						pods with seeds	0.14	14	LOQ
France			application						seed	<0.01	22	0.01 mg/kg
(S13-00443-05/ L130042)									rest of plant*	0.038	22	
									seed	<0.01	29	
									rest of plant*	0.040	29	
408616	Oilseed rape/	1. 10.10.12	Plot 2	0.006	200	0.0125	01.05.13	75	whole plant*	0.27	0	Method
2013/1416283	Nelson	2. 20.03.13-15.04.13	broadcast	0.006	200	0.0125	08. 05.13		rest of plant*	0.015	14	No 567/0
62100 Paralimnio	SO 0495	3. 20.05.13-10.06.13	foliar						pods with seeds	<0.01	14	LOQ
Greece			application						rest of plant*	0.019	22	0.01 mg/kg
(S13-00443-06/ L130043)									pods with seeds	0.036	22	
									seed	0.015	29	
									rest of plant*	0.092	29	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Oilseed rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408616	Oilseed rape/	1. 10.10.12	Plot 2	0.006	200	0.0125	13.05.13	80	whole plant*	0.28	0	Method
2013/1416283	Bagira	2. Apr 13 - May 13	broadcast	0.006	200	0.0125	20.05.13		rest of plant*	0.067	15	No 567/0
44019 Gualdo	SO 0495	3. 18.06.13	foliar						pods with seeds	0.038	15	LOQ
Italy			application						rest of plant*	0.047	21	0.01 mg/kg
(S13-00443-07/ L130044)									pods with seeds	0.056	21	
									seed	0.012	29	
									rest of plant*	0.084	29	
408616	Oilseed rape/	1. 26.11.12	Plot 2	0.006	200	0.0125	13.06.13	78-79	whole plant*	0.25	0	Method
2013/1416283	Frilola	2. nr	broadcast	0.006	200	0.0125	20.06.13		seed	<0.01	14	No 567/0
50637 Remolinos	SO 0495	3. 18.07.13	foliar						rest of plant*	0.057	14	LOQ
Spain			application						seed	<0.01	21	0.01 mg/kg
(S13-00443-08/ L130045)									rest of plant*	0.13	21	
									seed	<0.01	28	
									rest of plant*	0.060	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

Residue studies reported in supplement information

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (Fastac)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-750-021 BEGR.93.012 01-Massieux France South (W/FR/R/92/244)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)	spray	0.0025	400	0.01	1 04.05.92	63	Seed	<0.01	64	Method No. SAMS 351-2
BASF RDI No. AL-750-021 BEGR.93.012 01-Massieux France South (W/FR/R/92/244)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)	spray	0.005	400	0.02	1 04.05.92	63	Seed	<0.01	64	Method No. SAMS 351-2
BASF RDI No. AL-750-021 BEGR.93.012 69-Genay France South (W/FR/R/92/246)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)	spray	0.0025	400	0.01	1 23.04.92	-	Seed	<0.01	75	Method No. SAMS 351-2
BASF RDI No. AL-750-021 BEGR.93.012 69-Genay France South (W/FR/R/92/246)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)	spray	0.005	400	0.02	1 23.04.92	-	Seed	<0.01	75	Method No. SAMS 351-2

1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	PVP (SF07537, Fastac)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-750-021 BEGR.93.012 01-Massieux France South (W/FR/R/92/244)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)				0.01	1 04.05.92	63	Seed	<0.01	64	Method No. SAMS 351-2
BASF RDI No. AL-750-021 BEGR.93.012 01-Massieux France South (W/FR/R/92/244)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)				0.02	1 04.05.92	63	Seed	<0.01	64	Method No. SAMS 351-2
BASF RDI No. AL-750-021 BEGR.93.012 69-Genay France South (W/FR/R/92/246)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)				0.01	1 23.04.92	-	Seed	<0.01	75	Method No. SAMS 351-2
BASF RDI No. AL-750-021 BEGR.93.012 69-Genay France South (W/FR/R/92/246)	Winter oilseed rape/ Samourai	1. 2. 3.	Not reported Not reported 07.07.92 (sampling)				0.02	1 23.04.92	-	Seed	<0.01	75	Method No. SAMS 351-2

1) at last treatment
2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Oilseed Rape (oilseeds)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF AG, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/kg	**(common name and content)**	
Formulation (e.g. WP)	WG (BAS 310 08 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
2002/1004087 E-41410 Carmona Andalucia Spain (ALO/10/01)	Winter oilseed rape /Kabel	1. 2. 3.	01.11.2000 10.-30.03.01 17-18.05.01	Medium volume foliar application using boom sprayer	0.003	300	0.01	1 05.04.01	77	plants plants minus pods pods seed	0.11 0.11 0.11 <0.05	0 29 29 42	Method No. RLA 12594.02V (989/0)
2002/1004087 82170 Pompignan Midi-Pyrenees France South (FTL/07/01)	Winter oilseed rape /Cheyenne	1. 2. 3.	02.09.2000 02.04.-11.05.01 22.-29.06.01	Medium volume foliar application using boom sprayer	0.003	315	0.01	1 09.05.01	69	plants plants minus pods pods seed seed seed	0.07 <0.05 <0.05 <0.05 <0.05 <0.05	0 29 29 43 50 57	Method No. RLA 12594.02V (989/0)
2002/1004087 E-41410 Carmona Andalucia Spain (ALO/10/01)	Winter oilseed rape /Kabel	1. 2. 3.	01.11.2000 10.-30.03.01 17-18.05.01	Medium volume foliar application using boom sprayer	0.003 0.003	308 303	0.01 0.01	23.03.01 05.04.01	77	plants plants minus pods pods seed	0.28 0.24 0.36 0.06	0 29 29 42	Method No. RLA 12594.02V (989/0)
2002/1004087 82170 Pompignan Midi-Pyrenees France South (FTL/07/01)	Winter oilseed rape /Cheyenne	1. 2. 3.	02.09.2000 02.04.-11.05.01 22.-29.06.01	Medium volume foliar application using boom sprayer	0.003 0.003	353 297	0.012 0.010	2 09.05.01	69	plants plants minus pods pods seed seed seed	0.06 <0.05 <0.05 <0.05 <0.05 <0.05	0 29 29 43 50 57	Method No. RLA 12594.02V (989/0)

1) at last treatment
2) days after last application

Barley

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, The United Kingdom	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408610	Winter barley/	1. 03.10.11	foliar	0.00625	200	0.0125	2	81	whole plant*	0.32	0	Method
2013/1388974	Pelican	2. N/A	with boom	0.00625	200	0.0125	28.06.12		ears	0.18	13	No 567/0
21737, Wischhafen, Niedersachsen,	GC 0640	3. 27.07.12	sprayer						rest of plant*	0.21	13	LOQ
Germany									ears	0.18	20	0.01 mg/kg
(L120444)									rest of plant*	0.19	20	
									grain	0.035	29	
									straw	0.25	29	
408610	Spring barley/	1. 20.03.12	foliar	0.00781	196	0.0153	2	79	whole plant*	0.57	0	Method
2013/1388974	Westminster	2. N/A	with boom	0.00625	200	0.0125	26.07.12		ears	0.33	13	No 567/0
Arnold, Nottingham,	GC 0640	3. 24.08.12	sprayer						rest of plant*	0.45	13	LOQ
The United Kingdom									grain	0.029	20	0.01 mg/kg
(L120445)									straw	0.44	20	
									grain	0.053	29	
									straw	0.49	29	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, The United Kingdom	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408610	Winter barley/	1. 28.09.11	foliar	0.00625	200	0.0125	2	76	whole plant*	0.34	0	Method
2013/1388974	Naomi	2. N/A	with boom	0.00625	200	0.0125	12.06.12		ears	0.16	14	No 567/0
6662 PK Elst,	GC 0640	3. 10.7.12	sprayer						rest of plant*	0.14	14	LOQ
The Netherlands									ears	0.15	21	0.01 mg/kg
(L120446)									rest of plant*	0.20	21	
									grain	0.032	28	
									straw	0.26	28	
408610	Barley/	1. 02.03.12	foliar	0.00625	200	0.0125	2	83	whole plant*	0.37	0	Method
2013/1388974	Sebastian	2. 10.-29.06.12	with boom	0.00625	200	0.0125	27.06.12		ears	0.099	14	No 567/0
45300 Audeville,	GC 0640	3. 25.07.12	sprayer						rest of plant*	0.34	14	LOQ
France									ears	0.10	21	0.01 mg/kg
(L120447)									rest of plant*	0.39	21	
									grain	0.023	21	
									straw	0.41	21	
									ears	0.12	28	
									rest of plant*	0.40	28	
									grain	0.031	28	
									straw	0.47	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, Germany, The Netherlands	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2013/1416284		1. 19.09.12	Plot T	0.006	200	0.0125	19.06.13		Whole plant*	0.31	0	
S13-00441-01 / L130026	Barley /	2. 25/05/13-30/05/13	broadcast	0.006	200	0.0125	26.06.13	83	Ears	0.048	14	
16321 Bernau	Semper	3. 23.07.13	foliar						Rest of plant*	0.38	14	Method: BASF method No. 567/0
Germany	GC 0640		application						Grain	0.027	20	LOQ 0.01 mg/kg
									Straw	0.37	20	
									Grain	0.020	27	
									Straw	0.35	27	
BASF DocID 2013/1416284	Barley /	1. 02.10.12	Plot T	0.006	200	0.0125	18.06.13		Whole Plant*	0.51	0	
S13-00441-03 / L130028	Malabar	2. nr	broadcast	0.006	200	0.0125	25.06.13	77	Ears	0.29	14	Method:
6662 PK Elst	GC 0640	3. 18.07.13	foliar						Rest Of Plant*	0.44	14	BASF method No. 567/0
The Netherlands			application						Grain	0.077	22	LOQ 0.01 mg/kg
									Straw	0.48	22	
BASF DocID 2013/1416284	Barley /	1. 28.12.12	Plot T	0.006	200	0.0125	24.06.13		Whole Plant*	0.26	0	
S13-00441-04 / L130029	Meridian	2. 22/05/13-10/06/13	broadcast	0.006	200	0.0125	01.07.13	85	Grain	0.075	14	
5140 Sombreffe	GC 0640	3. 22/07/13-29/07/13	foliar						Straw	0.37	14	Method:
Belgium			application						Grain	0.11	21	BASF method No. 567/0
									Straw	0.43	21	LOQ 0.01 mg/kg
									Grain	0.079	28	
									Straw	0.41	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	The United Kingdom	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2013/1416284	Barley /	1. 21/10/12	Plot T	0.004	213	0.0080	08/07/13		Whole plant*	0.13	0	Method: BASF method No. 567/0
S13-00441-02 / L130027	Saffron	2. nr	broadcast	0.003	228	0.0057	15/07/13	75-77	Ears	0.019	15	LOQ 0.01 mg/kg
WV5 7HT Trysull	GC 0640	3. 02/08/13	foliar						Rest of plant*	0.34	15	Trial S13-00441-02 / L130027: The actual application rates for plot 2, applications A1 and A2, were -36% and -54% of the nominal rate due to the incorrect amount of chemical weighed out
UK			application						Grain	0.026	18	
									Straw	0.35	18	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	The United Kingdom	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)**	**Portion Analysed**	**Residues (mg/kg)***	**DALA[1] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2014/1173599		1.	foliar	0.006	200	12.5	2		Whole plant*	0.18	0	
S14-00678 / L140092	Barley /	2.	application	0.006	200	12.5			Ears	0.16	14±1	
The United Kingdom	GC 0640	3.							Rest of plant*	0.21	14±1	Method: BASF method No. 567/0
									Grain	0.05	21±1	
									Straw	0.23	21±1	
									Grain	0.03	28±1	
									Straw	0.20	28±1	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

- **Barley**

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408610	Winter barley/	1. 20.10.11	foliar	0.00625	200	0.0125	2	75	whole plant*	0.27	0	Method
2013/1388974	Baracca	2. 20.04.12-10.05.12	with boom	0.00625	200	0.0125	23.05.12		ears	0.31	14	No 567/0
40023, Castel Guelfo, Bologna, Italy	GC 0640	3. 18.06.12	sprayer						rest of plant*	0.25	14	LOQ
									ears	0.29	20	0.01 mg/kg
(L120448)									rest of plant*	0.37	20	
									grain	0.050	26	
									straw	0.40	26	
408610	Winter barley/	1. 20.10.11	foliar	0.0125	200	0.025	1	75	whole plant*	0.36	0	Method
2013/1388974	Baracca	2. 20.04.12-10.05.12	with boom				23.05.12		ears	0.14	14	No 567/0
40023, Castel Guelfo, Bologna, Italy	GC 0640	3. 18.06.12	sprayer						rest of plant*	0.35	14	LOQ
									ears	0.13	20	0.01 mg/kg
(L120448)									rest of plant*	0.031	20	
									grain	0.040	26	
									straw	0.29	26	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408610	Winter barley/	1. 16.11.11	foliar	0.00625	200	0.0125	2	85	whole plant*	0.51	0	Method
2013/1388974	Loverde	2. Apr-May 2012	with boom	0.00625	224	0.014	15.06.12		grain	0.052	14	No 567/0
Castelmayran, 82210 Tarn-et-Garonne, France	GC 0640	3. 06.07.12	sprayer						straw	0.43	14	LOQ
(L120450)									grain	0.066	21	0.01 mg/kg
									straw	0.24	21	
408610	Winter barley/	1. 16.11.11	foliar	0.01248	238	0.0297	1	85	whole plant*	0.32	0	Method
2013/1388974	Loverde	2. Apr-May 2012	with boom				15.06.12		grain	0.099	14	No 567/0
Castelmayran, 82210 Tarn-et-Garonne, France	GC 0640	3. 06.07.12	sprayer						straw	0.32	14	LOQ
(L120450)									grain	0.061	21	0.01 mg/kg
									straw	0.20	21	
408610	Winter barley/	1. 20.12.11	foliar	0.00625	200	0.0125	2	73	whole plant*	0.51	0	Method
2013/1388974	Arta	2. 15.-22.05.12	with boom	0.00625	200	0.0125	07.06.12		ears	0.19	14	No 567/0
Florina, West Macedonia, GR-53200, Greece	GC 0640	3. 05.07.12	sprayer						rest of plant*	0.22	14	LOQ
(L120451)									ears	0.12	21	0.01 mg/kg
									rest of plant*	0.23	21	
									grain	0.026	28	
									straw	0.21	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408610	Winter barley	1. 20.12.11	foliar	0.0125	200	0.025	1	73	whole plant*	0.61	0	Method
2013/1388974	Arta	2. 15.-22.05.12	with boom				07.06.12		ears	0.23	14	No 567/0
Florina, West Macedonia, GR-53200,	GC 0640	3. 05.07.12	sprayer						rest of plant*	0.28	14	LOQ
Greece									ears	0.21	21	0.01 mg/kg
(L120451)									rest of plant*	0.39	21	
									grain	0.043	28	
									straw	0.33	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	BAS 310 55 I
Crop/crop group:	Barley (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) [1] at last treatment or date**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2013/1416281 S13-00446-01 / L130003 Aragon Spain	Winter barley / Unia GC 0640	1. 20.12.12 2. N/A 3. 15.07.13	Foliar with boom sprayer	0.00625 0.00625	200 200	0.0125 0.0125	17.06.13 24.06.13	79-83	whole plant* ears rest of plant* grain straw grain straw	0.39 0.38 0.38 0.067 0.52 0.083 0.43	0 14 14 21* 21* 28* 28*	Method: BASF method No. 567/0 LOQ 0.01 mg/kg No residues above the LOQ were found in any of the untreated specimens * BBCH 89 N/A – not applicable
BASF DocID 2013/1416281 S13-00446-01 / L130003 Aragon Spain	Winter barley / Unia GC 0640	1. 20.12.12 2. N/A 3. 15.07.13	Foliar with boom sprayer	0.0125	200	0.025	24.06.13	79-83	whole plant* ears rest of plant* grain straw grain straw	0.34 0.42 0.41 0.12 0.57 0.092 0.46	0 14 14 21* 21* 28* 28*	Method: BASF method No. 567/0 LOQ 0.01 mg/kg No residues above the LOQ were found in any of the untreated specimens * BBCH 89 N/A – not applicable

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)***	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2013/1416284	Barley /	1. 13.12.12	Plot T	0.006	200	0.0125	17.05.13		Whole plant no roots	0.78	0	
S13-00441-05 / L130030	Atomo	2. Apr 13 - May 13	broadcast	0.006	200	0.0125	24.05.13	85	Ears	0.15	13	Method: BASF method No. 567/0 LOQ 0.01 mg/kg
40064 Castel S. Pietro	GC 0640	3. 20.06.13	foliar						Rest of plant without roots	0.55	13	
Italy			application						Grain	0.034	20	
									Straw	0.92	20	
BASF DocID 2013/1416284	Barley /	1. 23.12.12	Plot T	0.006	200	0.0125	20.05.13		Whole Plant No Roots	0.25	0	
S13-00441-06 / L130031	Meseta	2. nr	broadcast	0.006	200	0.0125	30.05.13	73-75	Ears	0.18	13	
50180 Utebo	GC 0640	3. 25.06.13	foliar						Rest of plant without roots	0.16	13	Method: BASF method No. 567/0 LOQ 0.01 mg/kg
Spain			application						Ears	0.20	20	
									Rest of plant without roots	0.20	20	
									Grain	0.079	26	
									Straw	0.47	26	
BASF DocID 2013/1416284	Barley /	1. 08/11/12	Plot T	0.006	200	0.0125	04/06/13		Whole plant no roots	0.41	0	
S13-00441-07 / L130032	Ketos	2. 01/05/13-15/05/13	broadcast	0.006	200	0.0125	11/06/13	83-85	Grain	0.023	14	
82700 Bourret	GC 0640	3. 09/07/13	foliar						Straw	0.61	14	Method: BASF method No. 567/0 LOQ 0.01 mg/kg
France			application						Grain	0.051	22	
									Straw	0.46	22	
									Grain	0.035	28	
									Straw	0.68	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	**-**
Crop/crop group:	Barley (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)***	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
BASF DocID 2013/1416284	Barley /	1. 09/11/12	Plot T	0.006	200	0.0125	23/04/13		Whole plant no roots	0.49	0	
S13-00441-08 / L130033	Mutso	2. 08/04/13-18/04/13	broadcast	0.006	200	0.0125	29/04/13	73	Ears	0.23	14	
58300 Galatades	GC 0640	3. 30/05/13-05/06/13	foliar						Rest of plant without roots	0.39	14	
Greece			application						Ears	0.15	21	Method: BASF method No. 567/0 LOQ 0.01 mg/kg
									Rest of plant without roots	0.45	21	
									Ears	0.14	28	
									Rest of plant without roots	0.38	28	
									Grain	0.024	36	
									Straw	0.40	36	

Residue studies reported in supplement information

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Barley (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	PVP	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/238)	Summer barley/Delta	1. 2. 3.	04.03.92 - 22.07.92	spray	0.0015	1000	0.015	1 04.06.92	no information	grain straw	<0.01 0.05	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/238)	Summer barley/Delta	1. 2. 3.	04.03.92 - 22.07.92	Spray	0.003	1000	0.03	1 04.06.92	no information	grain straw	<0.01 0.10	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Barley (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/238)	Summer barley/Delta	1. 2. 3.	04.03.92 - 22.07.92	spray	0.0015	1000	0.015	1 04.06.92	no information	grain straw	<0.01 0.08	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/238)	Summer barley/Delta	1. 2. 3.	04.03.92 - 22.07.92	spray	0.003	1000	0.03	1 04.06.92	no information	grain straw	<0.01 0.08	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR

1) at last treatment

2) days after last application

- **Wheat**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, Denmark, Germany, The Netherlands	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.29	0	BASF
2008/1002701	Wheat /	1.	28.09.06		0.005	298	0.015	2	73	ears	0.06	28	method no.
5500 Middelfart	Opus	2.	30.05.-	Foliar spray	0.005	290	0.015	28.06.07		rest of plant	0.30	28	567/0
(Fyn)	A 020/GC 0654		05.06.07	using boom						grain	<0.01	35	
Denmark (N)		3.	09.08.07	sprayer						straw	0.28	35	LOQ
L070422										grain	<0.01	42	0.01 mg/kg
										straw	0.33	42	
253702										plant without roots	0.25	0	BASF
2008/1002701	Wheat /	1.	27.09.06		0.005	306	0.015	2	75	ears	0.26	28	method no.
08459 Neukirchen	Bussard	2.	29.05.-	Foliar spray	0.005	296	0.015	19.06.07		rest of plant	0.46	28	567/0
(Sachsen)	A 020/GC 0654		10.06.07	using boom						grain	<0.01	36	
Germany (N)		3.	30.07.07	sprayer						straw	0.48	36	LOQ
L070423										grain	0.01	41	0.01 mg/kg
										straw	0.56	41	
253702										plant without roots	0.22	0	BASF
2008/1002701	Wheat /	1.	18.12.06		0.005	309	0.015	2	71	grain	<0.01	27	method no.
6662 Elst	Limes	2.	n.a.	Foliar spray	0.005	306	0.015	22.06.07		straw	0.46	27	567/0
(Gelderland)	A 020/GC 0654	3.	02.08.07	using boom						grain	<0.01	34	
The Netherlands (N)				sprayer						straw	0.28	34	LOQ
L070424										grain	<0.01	41	0.01 mg/kg
										straw	0.24	41	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, Denmark, Germany, The Netherlands	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.42	0	BASF
2008/1002701	Wheat /	1.	08.11.06		0.005	297	0.015	2	71	grain	<0.01	27	method no.
3400 Wange	Einstein	2.	24.05.-	Foliar spray	0.005	294	0.015	19.06.07		straw	0.26	27	567/0
(Landen)	A 020/GC 0654		30.05.07	using boom						grain	<0.01	34	
Belgium (N)		3.	31.07.07	sprayer						straw	0.56	34	LOQ
L070425										grain	<0.01	42	0.01 mg/kg
										straw	0.96	42	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, Denmark, Germany, The Netherlands	**Other active substance in the formulation**	Acetamiprid (100 g/L)
Content of active substance (g/kg or g/L)	25 g/L	**(common name and content)**	
Formulation (e.g. WP)	SL (BAS 370 00 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.38	0	BASF
2008/1002701	Wheat /	1.	28.09.06		0.004	302	0.013	2	73	ears	0.06	28	method no.
5500 Middelfart	Opus	2.	30.05.-	Foliar spray using boom sprayer	0.004	292	0.012	28.06.07		rest of plant	0.34	28	567/0
(Fyn)	A 020/GC 0654		05.06.07							grain	<0.01	35	
Denmark (N)		3.	09.08.07							straw	0.36	35	LOQ
L070422										grain	<0.01	42	0.01 mg/kg
										straw	0.23	42	
253702										plant without roots	0.37	0	BASF
2008/1002701	Wheat /	1.	27.09.06		0.004	303	0.013	2	75	ears	0.27	28	method no.
08459 Neukirchen	Bussard	2.	29.05.-	Foliar spray using boom sprayer	0.004	302	0.013	19.06.07		rest of plant	0.73	28	567/0
(Sachsen)	A 020/GC 0654		10.06.07							grain	<0.01	36	
Germany (N)		3.	30.07.07							straw	0.73	36	LOQ
L070423										grain	<0.01	41	0.01 mg/kg
										straw	0.70	41	
253702										plant without roots	0.18	0	BASF
2008/1002701	Wheat /	1.	18.12.06		0.004	284	0.012	2	71	grain	<0.01	27	method no.
6662 Elst	Limes	2.	n.a.	Foliar spray using boom sprayer	0.004	317	0.013	22.06.07		straw	0.51	27	567/0
(Gelderland)	A 020/GC 0654	3.	02.08.07							grain	<0.01	34	
The Netherlands (N)										straw	0.28	34	LOQ
L070424										grain	<0.01	41	0.01 mg/kg
										straw	0.25	41	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, Denmark, Germany, The Netherlands	**Other active substance in the formulation**	Acetamiprid (100 g/L)
Content of active substance (g/kg or g/L)	25 g/L	**(common name and content)**	
Formulation (e.g. WP)	SL (BAS 370 00 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.59	0	BASF
2008/1002701	Wheat /	1.	08.11.06		0.004	312	0.013	2	71	grain	<0.01	27	method no.
3400 Wange	Einstein	2.	24.05.-	Foliar spray using boom sprayer	0.004	295	0.012	19.06.07		straw	0.16	27	567/0
(Landen)	A 020/GC 0654		30.05.07							grain	0.01	34	
Belgium (N)		3.	31.07.07							straw	0.48	34	LOQ
L070425										grain	<0.01	42	0.01 mg/kg
										straw	0.82	42	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The United Kingdom	**Other active substance in the formulation (common name and content)**	-
Content of active substance (g/kg or g/L)	50 g/L		
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment[0] kg a.s./hL	Water (L/ha)	kg a.s./ha	No of Treatm. and Last Date	Growth Stage (BBCH)[1]	Portion Analysed	Residues (mg/kg)	DALA[2] (days)	Remarks
408613	Winter wheat/	1. 08.10.11	foliar with	0.00625	200	0.0125	2	83	whole plant*	0.46	0	Method
2014/1028112	Tabasco	2. N/A	boom	0.00625	200	0.0125	24.07.12		ears	0.24	13	No 567/0
21739 Dollern	GC 0654	3. 21.08.12	sprayer						rest of plant*	0.35	13	LOQ
Germany									grain	0.013	20	0.01 mg/kg
(L120452)									straw	0.71	20	
									grain	<0.01	28	
									straw	0.53	28	
408613	Spring wheat/	1. 05.02.12	foliar with	0.00625	200	0.0125	2	77-79	whole plant*	0.86	0	Method
2014/1028112	AC Barrie	2. N/A	boom	0.00625	200	0.0125	01.08.12		ears	0.34	13	No 567/0
Castlemorton, Worcestershire WR13 6BL,	GC 0654	3. 30.08.12	sprayer						rest of plant*	0.44	13	LOQ
The United Kingdom									ears	0.41	22	0.01 mg/kg
(L120453)									rest of plant*	0.38	22	
									grain	<0.01	29	
									straw	0.29	29	
408613	Spring wheat/	1. 20.03.12	foliar with	0.00625	200	0.0125	2	69-71	whole plant*	0.60	0	Method
2014/1028112	Granary	2. N/A	boom	0.00625	200	0.0125	01.08.12		ears	0.12	15	No 567/0
Withington, Andoversford, GL54 4BL	GC 0654	3. 05.10.12	sprayer						rest of plant*	0.21	15	LOQ
The United Kingdom									ears	0.077	22	0.01 mg/kg
(L120454)									rest of plant*	0.21	22	
									ears	0.047	29	
									rest of plant*	0.13	29	
									grain	<0.01	65	
									straw	0.181	65	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The United Kingdom	**Other active substance in the formulation (common name and content)**	-
Content of active substance (g/kg or g/L)	50 g/L		
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment[0] kg a.s./hL	Water (L/ha)	kg a.s./ha	No of Treatm. and Last Date	Growth Stage (BBCH)[1]	Portion Analysed	Residues (mg/kg)	DALA[2] (days)	Remarks
408613	Winter wheat/	1. 02.11.11	foliar with	0.00625	200	0.0125	2	75	whole plant*	0.30	0	Method
2014/1028112	Gabriel	2. 21.-28.05.12	boom	0.00625	200	0.0125	14.06.12		ears	0.088	14	No 567/0
45300 Reigneville, Yèvre-la-Ville	GC 0654	3. 19.07.12	sprayer						rest of plant*	0.25	14	LOQ
France									ears	0.078	21	0.01 mg/kg
(L120455)									rest of plant*	0.25	21	
									grain	<0.01	21	
									straw	0.26	21	
									ears	0.12	28	
									rest of plant*	0.33	28	
									grain	<0.01	28	
									straw	0.40	28	
									grain	<0.01	35	
									straw	0.35	35	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	BAS 310 55 I
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)at last treatment or date [1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408665	Winter wheat	1. 01.11.12	Plot 2	0.006	200	0.0125	11.07.13	75	Whole plant no roots	0.37	0	
BASF DocID 2013/1416282	Tabasco	2. 28.06.-05.07.13	broadcast	0.006	200	0.0125	18.07.13		Ears	0.026	14	
S13-00445-01 / L130034	GC0654	3. 14.08.13	foliar						Rest of plant without roots	0.25	14	Method: BASF method No. 567/0 LOQ 0.01 mg/kg
21739 Dollern			application						Ears	0.082	20	
Germany									Rest of plant without roots	0.23	20	
									Grain	<0.01	27	
									Straw	0.36	27	
408665	Winter wheat	1. 31.10.12	Plot 2	0.006	200	0.0125	26.06.13	77	Whole plant no roots	0.35	0	
BASF DocID 2013/1416282	Courtot	2. 08.06.13-15.06.13	broadcast	0.006	200	0.0125	04.07.13		Ears	0.095	14	
S13-00445-02 / L130035	GC0654	3. 01.08.13	foliar						Rest of plant without roots	0.34	14	Method: BASF method No. 567/0 LOQ 0.01 mg/kg
45300 Rouvres Saint Jean			application						Grain	<0.01	21	
France									Straw	0.38	21	
									Grain	<0.01	28	
									Straw	0.40	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

Residue studies reported in supplement information

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-001 BEGR.83.061 27 – Breuilpont France North (S/FR/E83/878)	Winter wheat/Caton	1. 2. 3.	10.10. 82 N/A 22.07. 83	spray	0.0035	500	0.0175	1 06.06.83	stage 10.52	grain straw	<0.01 0.17	46 46	Method no. SAMS 351-1

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/l	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-031 BEGR.93.013 D-7775 Bermatingen Germany (BE61)	Summer wheat/Star	1. 2. 3.	09.03.92 N/A 06.08.92	spray	0.0038	400	0.0150	1 03.07.92	75	grain straw	<0.01 0.01	34 34	Method no. SAMS 351-02 residues of CF05835; other chemical applied: Tristar

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	150 g/kg	**(common name and content)**	
Formulation (e.g. WP)	PVP	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-031 BEGR.93.013 D-7775 Bermatingen Germany (BE61)	Summer wheat/Star	1. 2. 3.	09.03.92 N/A 06.08.92	spray	0.0038	400	0.0150	1 03.07.92	75	grain straw	<0.01 0.03	34 34	Method no. SAMS 351-02 residues of CF05835; other chemical applied: Tristar

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-003 BEGR.85.004 27 – Le Plessis Hebert France North (S/FR/E84/890)	Winter wheat/Caton	1. 2. 3.	no information	no information	no information	no information	0.015	1 06.06.84	no information	grain grain straw straw straw straw straw	<0.01 <0.01 0.58 0.25 0.04 0.06 0.07	28 42 0 7 14 28 42	Method no. SAMS 233-1
BASF RDI No. AL-730-003 BEGR.85.004 27 – Le Plessis Hebert France North (S/FR/E84/891)	Winter wheat/Caton	1. 2. 3.	no information	no information	no information	no information	0.015	1 05.07.84	no information	grain grain grain straw straw straw straw	<0.01 <0.01 <0.01 0.54 0.40 0.36 0.29	7 13 20 0 7 13 20	Method no. SAMS 233-1
BASF RDI No. AL-730-003 BEGR.85.004 27 – Le Plessis Hebert France North (S/FR/E84/892)	Winter wheat/Rivoli	1. 2. 3.	no information	no information	no information	no information	0.015	1 14.06.84	no information	grain grain grain straw straw straw straw straw straw	<0.01 <0.01 <0.01 0.38 0.17 0.21 0.19 0.13 0.10	28 42 63 0 7 14 28 42 63	Method no. SAMS 233-1

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-003 BEGR.85.004 27 – Le Plessis Hebert France North (S/FR/E84/893)	Winter wheat/Rivoli	1. 2. 3.	no information	no information	no information	no information	0.015	1 05.07.84	no information	grain grain grain grain grain grain straw straw straw straw straw straw	<0.01 <0.01 <0.01 <0.01 <0.01 <0.01 0.50 0.28 0.18 0.14 0.12 0.10	0 7 14 21 28 42 0 7 14 21 28 42	Method no. SAMS 233-1

1) at last treatment

2) days after last application

Wheat

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation (common name and content)**	None
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.53	0	BASF
2008/1002701	Wheat /	1.	05.11.06		0.005	303	0.015	2	83	ears	0.17	28	method no.
13200 Arles	Dakter	2.	23.04.-	Foliar spray	0.005	308	0.015	31.05.07		rest of plant	0.52	28	567/0
(PACA)	A 020/GC 0654		01.05.07	using boom						grain	<0.01	35	
France (S)		3.	12.07.07	sprayer						straw	0.81	35	LOQ
L070426										grain	<0.01	42	0.01 mg/kg
										straw	0.83	42	
253702										plant without roots	0.69	0	BASF
2008/1002701	Wheat /	1.	20.11.06		0.005	300	0.015	2	71	grain	<0.01	27	method no.
58300 Galatades	Eva	2.	25.04.-	Foliar spray	0.005	301	0.015	04.05.07		straw	0.57	27	567/0
(Central Macedonia)	A 020/GC 0654		10.05.07	using boom						grain	<0.01	34	
Greece (S)		3.	15.06.07	sprayer						straw	0.56	34	LOQ
L070427										grain	<0.01	42	0.01 mg/kg
										straw	0.62	42	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC (BAS 310 40 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.38	0	BASF
2008/1002701	Wheat /	1.	Nov. 06		0.005	312	0.016	2	71-75	grain	<0.01	28	method no.
Regalbuto	Arcancelo	2.	April-May	Foliar spray	0.005	293	0.015	11.05.07		straw	1.04	28	567/0
(Enna)	A 020/GC 0654	3.	21.06.07	using boom						grain	<0.01	35	
Italy (S)				sprayer						straw	0.70	35	LOQ
L070428										grain	<0.01	41	0.01 mg/kg
										straw	0.77	41	
253702										plant without roots	0.28	0	BASF
2008/1002701	Wheat /	1.	10.01.07		0.005	306	0.014	2	69	ears	0.04	27	method no.
Albacete	Galera	2.	mid May-	Foliar spray	0.005	318	0.015	01.06.07		rest of plant	0.09	27	567/0
(Albacete)	A 020/GC 0654		end May	using boom						grain	<0.01	36	
Spain (S)		3.	13.07.07	sprayer						straw	0.16	36	LOQ
L070429										grain	<0.01	42	0.01 mg/kg
										straw	0.15	42	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	Acetamiprid (100 g/L)
Content of active substance (g/kg or g/L)	25 g/L	**(common name and content)**	
Formulation (e.g. WP)	SL (BAS 370 00 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.82	0	BASF
2008/1002701	Wheat /	1.	05.11.06		0.004	303	0.013	2	83	ears	0.31	28	method no.
13200 Arles	Dakter	2.	23.04.-	Foliar spray	0.004	314	0.013	31.05.07		rest of plant	0.99	28	567/0
(PACA)	A 020/GC 0654		01.05.07	using boom sprayer						grain	<0.01	35	
France (S)		3.	12.07.07							straw	0.80	35	LOQ
L070426										grain	<0.01	42	0.01 mg/kg
										straw	0.50	42	
253702										plant without roots	0.54	0	BASF
2008/1002701	Wheat /	1.	20.11.06		0.004	300	0.012	2	71	grain	<0.01	27	method no.
58300 Galatades	Eva	2.	25.04.-	Foliar spray	0.004	306	0.013	04.05.07		straw	0.49	27	567/0
(Central Macedonia)	A 020/GC 0654		10.05.07	using boom sprayer						grain	<0.01	34	
Greece (S)		3.	15.06.07							straw	0.58	34	LOQ
L070427										grain	<0.01	42	0.01 mg/kg
										straw	0.82	42	
253702										plant without roots	0.58	0	BASF
2008/1002701	Wheat /	1.	Nov. 06		0.004	313	0.013	2	71-75	grain	<0.01	28	method no.
Regalbuto	Arcancelo	2.	April-May	Foliar spray	0.004	307	0.013	11.05.07		straw	1.04	28	567/0
(Enna)	A 020/GC 0654	3.	21.06.07	using boom sprayer						grain	<0.01	35	
Italy (S)										straw	0.66	35	LOQ
L070428										grain	<0.01	41	0.01 mg/kg
										straw	0.79	41	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	Acetamiprid (100 g/L)
Content of active substance (g/kg or g/L)	25 g/L	**(common name and content)**	
Formulation (e.g. WP)	SL (BAS 370 00 I)	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (Trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / Planting Flowering Harvest**	**Method of Treatment**	**Application Rate Per Treatment**			**No of Treat-ment(s) and Last Date**	**Growth stage at Last Treat. or Date BBCH[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**				**BAS 310 I**		
253702										plant without roots	0.25	0	BASF
2008/1002701	Wheat /	1.	10.01.07		0.004	296	0.012	2	69	ears	0.04	27	method no.
Albacete	Galera	2.	mid May-	Foliar spray	0.004	307	0.013	01.06.07		rest of plant	0.15	27	567/0
(Albacete)	A 020/GC 0654		end May	using boom sprayer						grain	<0.01	36	
Spain (S)		3.	13.07.07							straw	0.20	36	LOQ
L070429										grain	<0.01	42	0.01 mg/kg
										straw	0.15	42	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]** kg a.s./hL	Water (L/ha)	kg a.s./ha	**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
408613	Winter wheat/	1. 04.11.11	foliar with	0.00625	200	0.0125	2	83-85	whole plant*	0.30	0	Method
2014/1028112	Imgeuio	2. 12.-25.05.12	boom	0.00625	200	0.0125	14.06.12		grain	<0.01	14	No 567/0
82700 La Barraqué, Cordes-Tolosannes, Tarn-et-Garonne,	GC 0654	3. 12.07.12	sprayer						straw	0.35	14	LOQ
France									grain	<0.01	21	0.01 mg/kg
(L120456)									straw	0.56	21	
									grain	<0.01	28	
									straw	0.54	28	
408613	Winter wheat/	1. 04.11.11	foliar with	0.0125	200	0.025	1	83-85	whole plant*	0.44	0	Method
2014/1028112	Imgeuio	2. 12.-25.05.12	boom				14.06.12		grain	<0.01	14	No 567/0
82700 La Barraqué, Cordes-Tolosannes, Tarn-et-Garonne,	GC 0654	3. 12.07.12	sprayer						straw	0.45	14	LOQ
France									grain	<0.01	21	0.01 mg/kg
(L120456)									straw	0.84	21	
									grain	<0.01	28	
									straw	0.74	28	
408613	Spring wheat/	1. 30.03.12	foliar with	0.00625	200	0.0125	2	69-71/73	whole plant*	0.61	0	Method
2014/1028112	Simeto	2. 25.05.-05.06.12	boom	0.00625	200	0.0125	07.06.12		ears	0.12	14	No 567/0
Florina, West Macedonia, GR-53200,	GC 0654	3. 05.07.12	sprayer						rest of plant*	0.25	14	LOQ
Greece									ears	0.088	21	0.01 mg/kg
(L120457)									rest of plant*	0.19	21	
									grain	<0.01	28	
									straw	0.33	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408613	Spring wheat/	1. 30.03.12	foliar with	0.0125	200	0.025	1	69-71/73	whole plant*	0.89	0	Method
2014/1028112	Simeto	2. 25.05.-05.06.12	boom				07.06.12		ears	0.18	14	No 567/0
Florina, West Macedonia, GR-53200,	GC 0654	3. 05.07.12	sprayer						rest of plant*	0.26	14	LOQ
Greece									ears	0.15	21	0.01 mg/kg
(L120457)									rest of plant*	0.32	21	
									grain	<0.01	28	
									straw	0.38	28	
408613	Winter wheat/	1. 17.12.11	foliar with	0.00625	200	0.0125	2	69-71	whole plant*	0.17	0	Method
2014/1028112	Dylan	2. 26.04.-20.05.12	boom	0.00625	200	0.0125	24.05.12		ears	0.088	14	No 567/0
40057 Granarolo dell'Emilia, Bologna,	GC 0654	3. 29.06.12	sprayer						rest of plant*	0.11	14	LOQ
Italy									ears	0.069	21	0.01 mg/kg
(L120458)									rest of plant*	0.13	21	
									ears	0.05	28	
									rest of plant*	0.11	28	
									grain	<0.01	36	
									straw	0.18	36	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408613	Winter wheat/	1. 17.12.11	foliar with	0.0125	200	0.025	1	69-71	whole plant*	0.50	0	Method
2014/1028112	Dylan	2. 26.04.-20.05.12	boom				24.05.12		ears	0.098	14	No 567/0
40057 Granarolo dell'Emilia, Bologna,	GC 0654	3. 29.06.12	sprayer						rest of plant*	0.12	14	LOQ
Italy									ears	0.11	21	0.01 mg/kg
(L120458)									rest of plant*	0.11	21	
									ears	0.064	28	
									rest of plant*	0.12	28	
									grain	<0.01	36	
									straw	0.26	36	
408613	Winter wheat/	1. 15.11.12	foliar with	0.00625	200	0.0125	2	75-77	whole plant*	0.45	0	Method
2014/1028112	Bastide	2. N/A	boom	0.00625	200	0.0125	17.07.12		ears	0.40	15	No 567/0
50369, Lechón, Zaragoza,	GC 0654	3. 14.08.12	sprayer						rest of plant*	0.17	15	LOQ
Spain									ears	0.27	22	0.01 mg/kg
(L120459)									rest of plant*	0.32	22	
									grain	<0.01	22	
									straw	0.38	22	
									ears	0.24	28	
									rest of plant*	0.20	28	
									grain	<0.01	28	
									straw	0.35	28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Wheat (cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	Alpha-cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment[0]**			**No of Treatm. and Last Date**	**Growth Stage (BBCH)[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA[2] (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408613	Winter wheat/	1. 15.11.12	foliar with	0.0125	200	0.025	1	75-77	whole plant*	0.49	0	Method
2014/1028112	Bastide	2. N/A	boom				17.07.12		ears	0.32	15	No 567/0
50369, Lechón, Zaragoza,	GC 0654	3. 14.08.12	sprayer						rest of plant*	0.27	15	LOQ
Spain									ears	0.27	22	0.01 mg/kg
(L120459)									rest of plant*	0.27	22	
									grain	<0.01	22	
									straw	0.46	22	
									ears	0.21	28	
									rest of plant*	0.19	28	
									grain	<0.01	28	
									straw	0.36	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	BAS 310 55 I
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Italy, Spain	**Other active substance in the formulation**	-
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	ME (BAS 310 55 I)	**Residues calculated as:**	BAS 310 I

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment or date[1]**	**Portion Analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
408665	Wheat	1. 15.10.12	Plot 2	0.006	200	0.0125	21.05.13	83	Whole plant no roots	0.47	0	
BASF DocID 2013/1416282	Blasco	2. May 13	broadcast	0.006	200	0.0125	28.05.13		Ears	0.048	14	
S13-00445-03 / L130036	GC0654	3. 24.06.13	foliar						Rest of plant without roots	0.40	14	Method:
40057 Granarolo			application						Grain	<0.01	21	BASF method No. 567/0
Italy									Straw	0.35	21	LOQ 0.01 mg/kg
									Grain	<0.01	27	
									Straw	0.37	27	
408665	Wheat	1. 01.12.12	Plot 2	0.006	200	0.0125	17.06.13	81-87	Whole plant no roots	0.56	0	
BASF DocID 2013/1416282	Burgos	2. nr	broadcast	0.006	200	0.0125	24.06.13		Grain	<0.01	14	
S13-00445-04 / L130037	GC0654	3. 22.07.13	foliar						Straw	0.32	14	Method:
50059 Montañana			application						Grain	<0.01	21	BASF method No. 567/0
Spain									Straw	0.32	21	LOQ 0.01 mg/kg
									Grain	<0.01	28	
									Straw	0.47	28	

0) actual application rates varied by 10% at most
1) at last treatment
2) days after last application
* without roots

Residue studies reported in supplement information

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-001 BEGR.83.061 01 – Nievroz France South (S/FR/E83/220)	Winter wheat/Talent	1. 2. 3.	10.11. 82 N/A 15.07. 83	spray	0.0035	500	0.0175	1 07.06.83	stage 10.51	grain straw	<0.01 0.44	39 39	Method no. SAMS 351-1
BASF RDI No. AL-730-001 BEGR.83.061 24 – Cherval France South (S/FR/E83/419)	Winter wheat/Hardi	1. 2. 3.	05.11. 82 N/A 22.07. 83	spray	0.0035	500	0.0175	1 09.06.83	stage 10.2	grain straw	<0.01 0.29	42 42	Method no. SAMS 351-1

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-003 BEGR.85.004 69 – Taponas France South (S/FE/E84/217)	Winter wheat/Talent	1. 2. 3.	no information	no information	no information	no information	0.015	1 05.06.84	no information	grain grain grain grain straw straw straw straw straw straw	<0.01 <0.01 <0.01 <0.01 0.66 0.23 0.09 0.08 0.12 0.12	14 28 42 70 0 7 14 28 42 70	Method no. SAMS 233-1
BASF RDI No. AL-730-003 BEGR.85.004 69 – Drace France South (S/FR/E84/218)	Winter wheat/Talent	1. 2. 3.	no information	no information	no information	no information	0.015	1 26.06.84	no information	grain grain grain grain grain grain straw straw straw straw straw straw	<0.01 <0.01 <0.01 <0.01 <0.01 <0.01 0.16 0.12 0.09 0.05 0.06 0.13	0 7 14 21 28 63 0 7 14 21 28 63	Method no. SAMS 233-1

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-003 BEGR.85.004 33 – Salleboeuf France South (S/FR/E84/347)	Winter wheat/Top	1. 2. 3.	no information	no information	no information	no information	0.015	1 25.05.84	no information	grain grain grain straw straw straw straw straw straw	<0.01 <0.01 <0.01 1.1 0.62 0.18 0.16 0.09 0.03	28 42 70 0 7 14 28 42 70	Method no. SAMS 233-1
BASF RDI No. AL-730-003 BEGR.85.004 33 – Salleboeuf France South (S/FR/E84/348)	Winter wheat/Top	1. 2. 3.	no information	no information	no information	no information	0.015	1 22.06.84	no information	grain grain grain grain grain straw straw straw straw straw straw	<0.01 <0.01 <0.01 <0.01 <0.01 0.15 0.07 0.05 0.04 0.03 0.06	7 14 21 28 42 0 7 14 21 28 42	Method no. SAMS 233-1

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/239)	Summer wheat/Pinkal	1. 2. 3.	04.03.92 - 22.07.92	spray	0.0015	1000	0.015	1 04.06.92	no information	grain straw	<0.01 0.02	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/239)	Summer wheat/Pinkal	1. 2. 3.	04.03.92 - 22.07.92	spray	0.003i	1000	0.03	1 04.06.92	no information	grain straw	<0.01 0.08	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	PVP	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/239)	Summer wheat/Pinkal	1. 2. 3.	04.03.92 - 22.07.92	spray	0.0015	1000	0.015	1 04.06.92	no information	grain straw	<0.01 0.05	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR
BASF RDI No. AL-730-029 BEGR.93.009 69 – Quincieux France South (W/FR/E/92/239)	Summer wheat/Pinkal	1. 2. 3.	04.03.92 - 22.07.92	spray	0.003	1000	0.03	1 04.06.92	no information	grain straw	<0.01 0.06	49 49	Method no. SAMS 351-02 other chemical applied: Ioniz VR

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-030 BEGR.93.010 38 – Montalieu France South (W/FR/E/92/209)	Winter wheat/Darius	1. 2. 3.	08.11.91 N/A 20.07.92	spray	0.0038	400	0.015	1 01.06.92	68	grain straw	<0.01 0.09	49 49	Method no. SAMS 351-02 other chemical applied: Foctar
BASF RDI No. AL-730-030 BEGR.93.010 38 – Montalieu France South (W/FR/E/92/209)	Winter wheat/Darius	1. 2. 3.	08.11.91 N/A 20.07.92	spray	0.0075	400	0.03	1 01.06.92	68	grain straw	<0.01 0.30	49 49	Method no. SAMS 351-02 other chemical applied: Foctar

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	PVP	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-030 BEGR.93.010 38 – Montalieu France South (W/FR/E/92/209)	Winter wheat/Darius	1. 2. 3.	08.11.91 N/A 20.07.92	spray	0.0038	400	0.015	1 01.06.92	68	grain straw	<0.01 0.06	49 49	Method no. SAMS 351-02 other chemical applied: Foctar
BASF RDI No. AL-730-030 BEGR.93.010 38 – Montalieu France South (W/FR/E/92/209)	Winter wheat/Darius	1. 2. 3.	08.11.91 N/A 20.07.92	spray	0.0075	400	0.03	1 01.06.92	68	grain straw	<0.01 0.19	49 49	Method no. SAMS 351-02 other chemical applied: Foctar

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	EC	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-030 BEGR.93.010 01 – Parcay France South (W/FR/E/92/869)	Winter wheat/Artaban	1. 2. 3.	02.11.91 N/A 17.07.92	spray	0.0038	400	0.015	1 09.06.92	74	grain straw	<0.01 0.75	39 39	Method no. SAMS 351-02
BASF RDI No. AL-730-030 BEGR.93.010 01 – Parcay France South (W/FR/E/92/869)	Winter wheat/Artaban	1. 2. 3.	02.11.91 N/A 17.07.92	spary	0.0075	400	0.03	1 09.06.92	74	grain straw	<0.01 2.00	39 39	Method no. SAMS 351-02

1) at last treatment

2) days after last application

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	BAS 310 I	**Commercial Product (name)**	
Crop/crop group:	Wheat (Cereals)	**Producer of commercial product**	BASF SE
Responsible body for reporting (name, address)	BASF SE	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	None
Content of active substance (g/kg or g/L)	50 g/kg	**(common name and content)**	
Formulation (e.g. WP)	PVP	**Residues calculated as:**	Alpha-cypermethrin, BAS 310 I

1	2	3		4	5			6	7	8	9	10	11
Report-No. Location (trial no.)	**Commodity/ Variety**	**1. 2. 3.**	**Date of Sowing / planting Flowering Harvest**	**Method of treatment**	**Application rate per treatment**			**No of treatment(s) and last date**	**Growth stage at last treat. or date BBCH[1]**	**Portion analysed**	**Residues (mg/kg)**	**DALA (days)[2]**	**Remarks**
					kg a.s./hL	**Water L/ha**	**kg a.s./ha**				**BAS 310 I**		
BASF RDI No. AL-730-030 BEGR.93.010 01 – Parcay France South (W/FR/E/92/869)	Winter wheat/Artaban	1. 2. 3.	02.11.91 N/A 17.07.92	spray	0.0038	400	0.015	1 09.06.92	74	grain straw	<0.01 0.60	39 39	Method no. SAMS 351-02
BASF RDI No. AL-730-030 BEGR.93.010 01 – Parcay France South (W/FR/E/92/869)	Winter wheat/Artaban	1. 2. 3.	02.11.91 N/A 17.07.92	spray	0.0075	400	0.03	1 09.06.92	74	grain straw	<0.01 1.60	39 39	Method no. SAMS 351-02

1) at last treatment

2) days after last application

Tier 1 Summaries of the Supervised Field Residue Trials for Supplemental Information

- **Grape**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation (common name and content)**	none
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of : 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment			No of Treatm. and Last Date	Growth stage at last treatment	Portion Analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
DocID 2005/1006474 France - 67550 Rosheim Alsace (FAN/19/03)	Wine grape (Chardonnay)	1. 01.04.1995 2. 03.-15.06.2013 3. 10.-18.09.2003	medium volume foliar application using mistblower	0.0019	800	0.015	1 03.09.2004	89	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 3 7 14	BASF method No. 546/0
							2 03.09.2004	89	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 3 7 14	
DocID 2005/1006474 France - 67117 Handschuheim Alsace (FAN/20/03)	Wine grape (Pinot Noir)	1. n. a. 2. 10.-26.06.2003 3. 15.09.2003	medium volume foliar application using mistblower	0.0019	800	0.015	1 03.09.2004	85	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 3 7 14	BASF method No. 546/0
							2 03.09.2004	85	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of : 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment			No of Treatm. and Last Date	Growth stage at last treatment	Portion Analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
DocID 2005/1006474 France - 49540 Martigné-Briand Pays de la Loire (FBM/14/03)	Wine grape (Grolleau)	1. 10.03.1950 2. 11.-20.06.2003 3. 15.09.2003	medium volume foliar application using mistblower	0.0019	800	0.015	1 11.09.2003	85	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 4 7 14	BASF method No. 546/0
							2 11.09.2003	85	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 4 7 14	
DocID 2005/1006474 France - 49190 Saint Aubin de Luigné Pays de la Loire (FBM/15/03)	Wine grape (Grolleau)	1. 12.03.1963 2. 11.-20.06.2003 3. 17.09.2003	medium volume foliar application using mistblower	0.0019	800	0.015	1 11.09.2003	85	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 4 7 14	BASF method No. 546/0
							2 11.09.2003	85	fruit fruit fruit fruit	<0.05 <0.05 <0.05 <0.05	0 4 7 14	

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007591 Germany - 74193 Stetten (DU2/06/04)	Wine grape (Spaet-burgunder)	1. 01.10.1995 2. 04.-21.06.2004 3. 18.10.04	medium volume foliar application using mistblower	0.0019	800	0.015	1 28.09.2004	85	fruit fruit fruit fruit	0.013 0.011 0.011 <0.01	0 3 7 14	BASF method No. 567/0
							2 28.09.2004	85	fruit fruit fruit fruit	0.027 0.026 0.030 0.024	0 3 7 14	
DocID 2005/1007591 Germany - 76831 Eschbach (DU4/06/04)	Wine grape (Gewuerz-traminer)	1. n. a. 2. 10.-16.06.2004 3. 20.10.2004	medium volume foliar application using mistblower	0.0019	800	0.015	1 28.09.2004	89	fruit fruit fruit fruit	0.013 0.046 <0.01 0.011	0 3 7 14	BASF method No. 567/0
							2 28.09.2004	89	fruit fruit fruit fruit	0.045 0.010 0.031 0.029	0 3 7 14	
DocID 2005/1007591 France - 67117 Handschuheim, Alsace (FAN/12/04)	Wine grape (Auxerrois)	1. 01.04.1993 2. 12.-26.06.2004 3. 15.09.2004	medium volume foliar application using mistblower	0.0019	800	0.015	1 06.09.2004	85	fruit fruit fruit fruit	<0.01 <0.01 0.010 <0.01	0 3 7 14	BASF method No. 567/0
							2 06.09.2004	85	fruit fruit fruit fruit	0.018 0.015 0.018 0.013	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007591 France - 49540 Martigné-Briand, Pays de la Loire (FBM/06/04)	Wine grape (Chenin)	1. 05.03.1993 2. 01.-05.07.2004 3. 04.10.2004	medium volume foliar application using mistblower	0.0019	800	0.015	1 28.09.2004	85	fruit fruit fruit fruit	0.021 0.012 <0.01 0.014	0 3 7 14	BASF method No. 567/0
							2 28.09.2004	85	fruit fruit fruit fruit	0.014 0.019 0.019 0.020	0 3 7 14	

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 55 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2013/1066508 Germany - 55437 Ockenheim (L110429)	Wine grape (Johanniter)	1. 2008 2. 26.05.-06.06.2011 3. 12.09.2011	Knapsack sprayer, Solo airblast	0.0016	800	0.0125	1 30.08.2011	89	fruit fruit fruit fruit	0.06 0.04 0.04 0.03	0 3 7 15	BASF method No. 567/0
DocID 2013/1066508 France - 72340 Ruillè sur Loire Pays de la Loire (L110430)	Wine grape (Gamay)	1. 01.01.1979 2. 03.06.-09.06.2011 3. 19.09.2011	Atomizer, Stihl	0.0016	800	0.0125	1 06.09.2011	87	fruit fruit fruit fruit	0.05 0.05 0.04 0.04	0 3 7 13	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2013/1066508 Germany - 55437 Ockenheim (L110429)	Wine grape (Johanniter)	1. 2008 2. 26.05.-06.06.2011 3. 12.09.2011	Knapsack sprayer, SOLO airblast	0.0016	800	0.0125	1 30.08.2011	89	fruit fruit fruit fruit	0.03 0.02 0.03 0.02	0 3 7 15	BASF method No. 567/0
DocID 2013/1066508 France - 72340 Ruillè sur Loire Pays de la Loire (L110430)	Wine grape (Gamay)	1. 01.01.1979 2. 03.06.-09.06.2011 3. 19.09.2011	Atomizer, Stihl	0.0016	800	0.0125	1 06.09.2011	87	fruit fruit fruit fruit	0.06 0.03 0.05 0.04	0 3 7 13	BASF method No. 567/0
DocID 2006/1026853 France - 71260 Collongette, Lugny (AF/8830/BA/1)	Wine grape (Chardonnay)	1. 1998 2. n.a. 3. 07.09. - 21.09.2005	foliar application with mist blower	0.00125	1000	0.0125	1 07.09.2005	85	bunches bunches bunches bunches	0.049 0.033 0.031 0.033	0 2 7 14	BASF method No. 567/0
							2 07.09.2005	85	bunches bunches bunches bunches	0.082 0.077 0.064 0.063	0 2 7 14	
DocID 2006/1026853 France - 49320 La Ropeliere, St Jean des Mauvrets (AF/8830/BA/2)	Wine grape (Grolleau)	1. 1945 2. n.a. 3. 13.09. - 27.09.2005	foliar application with mist blower	0.00125	1000	0.0125	1 13.09.2005	85-89	bunches bunches bunches bunches	0.019 0.026 0.021 0.015	0 3 7 14	BASF method No. 567/0
							2 13.09.2005	85-89	bunches bunches bunches bunches	0.026 0.028 0.033 0.013	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026853 Germany - 69181 Leimen (AF/8830/BA/3)	Wine grape (Riesling)	1. 1994 2. n.a. 3. 06.09. - 20.09.2005	foliar application with mist blower	0.00125	1000	0.0125	1 06.09.2005	85	bunches bunches bunches bunches	<0.01 <0.01 <0.01 0.013	0 3 7 14	BASF method No. 567/0
							2 06.09.2005	85	bunches bunches bunches bunches	0.016 <0.01 0.028 0.037	0 3 7 14	
DocID 2006/1026853 Germany - 69231 Rauenberg (AF/8830/BA/4)	Wine grape (Müller-Thurgau)	1. 1983 2. n.a. 3. 30.08. - 13.09.2005	foliar application with mist blower	0.00125	1000	0.0125	1 30.08.2005	83	bunches bunches bunches bunches	0.033 0.019 0.030 0.026	0 3 7 14	BASF method No. 567/0
							2 30.08.2005	83	bunches bunches bunches bunches	0.049 0.037 0.043 0.039	0 3 7 14	
DocID 2007/1008492 France - 227080 Ay, Champagne-Ardennes (A/NF/I/06/127)	Wine grape (Chardonnay)	1. 1987 2. 10-18.06.2006 3. 27.09.2006	Foliar spray using atomizer	0.001	911	0.011	1 12.09.2006	85	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 3 6 15	BASF method No. 567/0
				0.001 0.001	1094 1036	0.014 0.013	2 12.09.2006	85	fruits fruits fruits fruits	0.018 0.016 0.012 0.012	0 3 6 15	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008492 France - Reims, Champagne-Ardennes (A/NF/I/06/128)	Wine grape (Chardonnay)	1. 1985 2. 14-21.06.2006 3. 13.09.2006	Foliar spray using atomizer	0.001	1061	0.013	1 30.08.2006	79	fruits fruits fruits fruits	0.012 0.010 0.011 <0.01	0 4 8 14	BASF method No. 567/0
				0.001 0.001	918 1061	0.011 0.013	2 30.08.2006	79	fruits fruits fruits fruits	0.017 0.025 0.019 0.010	0 4 8 14	
DocID 2007/1008492 Germany - Geinsheim, Rheinland-Pfalz (A/GE/I/06/129)	Wine grape (Mueller-Thurgau)	1. 1977 2. 15-30.06.2006 3. 18.09.2006	Foliar spray using motorised knapsack sprayer	0.001	969	0.013	1 04.09.2006	85	fruits fruits fruits fruits	0.012 <0.01 <0.01 0.016	0 3 7 14	BASF method No. 567/0
				0.001 0.001	979 984	0.013 0.013	2 04.09.2006	85	fruits fruits fruits fruits	0.016 0.014 0.020 0.021	0 3 7 14	
DocID 2007/1008492 Germany - Kirchheim, Rheinland-Pfalz (A/GE/I/06/130)	Wine grape (Phoenix)	1. 2001 2. 18-25.06.2006 3. 18.09.2006	Foliar spray using motorised knapsack sprayer	0.001	1141	0.014	1 04.09.2006	87	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
				0.001 0.001	1002 992	0.012 0.012	2 04.09.2006	87	fruits fruits fruits fruits	0.013 0.010 0.014 0.028	0 3 7 14	

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1004977 Spain - 41710 Utrera (Sevilla) Palmera 16 Andalucia (ALO/15/03)	Red wine grape (Cardenal)	1. 15.02.1987 2. 05.05.-17.05.2003 3. 17.07.-21.07.2003	Maruyama lance spraying	0.0019	800	0.015	2 07.07.2003	85	bunches bunches bunches bunches	<0.05 <0.05 <0.05 <0.05	0 3 7 14	BASF method No. 546/0
DocID 2005/1004977 Spain - 41720 Los Palacios (Sevilla) Enrique Granados 26, Andalucia (ALO/24/03)	White wine grape (Airen)	1. 15.02.1996 2. 30.04.-10.05.2003 3. 11.08.-12.08.2003	Maruyama lance spraying	0.0019	800	0.015	2 28.07.2003	85	bunches bunches bunches bunches	<0.05 <0.05 <0.05 <0.05	0 3 7 14	BASF method No. 546/0
DocID 2005/1004977 Italy - 15059 Monleale Fraz. Ville, 9 Piedmont (ITA/10/03)	Wine grape (Croatina)	1. n. a. 2. 01.06.-15.06.2003 3. 10.09.-18.09.2003	mistblower solo	0.0019	800	0.015	2 05.09.2013	85	bunches bunches bunches bunches	<0.05 <0.05 <0.05 <0.05	0 3 6 14	BASF method No. 546/0
DocID 2005/1004977 Italy - 15057 Tortona Strada Communale 7 Piedmont (ITA/11/03)	Wine grape (Barbera)	1. n. a. 2. 01.06.-15.06.2003 3. 10.09.-20.09.2003	mistblower solo	0.0019	800	0.015	2 05.09.2013	85	bunches bunches bunches bunches	<0.05 <0.05 <0.05 <0.05	0 3 6 14	BASF method No. 546/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007589 Greece - 59200 Naousa (GRE/03/04)	Wine grape (Xinomavro)	1. n. a. 2. 25.05.-05.08.2004 3. 01.-10.10.2004	medium volume foliar application using mistblower	0.0019	800	0.015	2 16.09.2004	79	fruit fruit fruit fruit	0.079 0.027 0.048 0.045	0 2 7 13	BASF method No. 567/0
DocID 2005/1007589 Greece - 59200 Marina (GRE/04/04)	Wine grape (Xinomavro)	1. n. a. 2. 25.05.-03.06.2005 3. 01.-10.10.2004	medium volume foliar application using mistblower	0.0019	800	0.015	2 16.09.2004	81	fruit fruit fruit fruit	0.075 0.054 0.051 0.043	0 2 7 13	BASF method No. 567/0
DocID 2005/1007589 France - 26600 Pont de l´Isère Rhone-Alpes (FBD/01/04)	Wine grape (Syrah)	1. 01.02.1992 2. 14.-29.05.2004 3. 11.-18.09.2004	medium volume foliar application using mistblower	0.0019	800	0.015	2 03.09.2004	85	fruit fruit fruit fruit	0.090 0.084 0.055 0.069	0 3 6 14	BASF method No. 567/0
DocID 2005/1007589 France - 31620 Fronton Midi-Pyreneés (FTL/01/04)	Wine grape (Negrette)	1. 20.05.1979 2. 14.-20.06.2004 3. 15.-25.09.2004	medium volume foliar application using mistblower	0.0019	800	0.015	2 07.09.2004	85	fruit fruit fruit fruit	0.084 0.034 0.036 0.061	0 3 7 14	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 55 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2013/1066508 Spain - 29700 Vélez-Málaga (L110431)	Wine grape (Moscatel)	1. 02.1997 2. 12.05.-01.06.2011 3. 24.08.2011	Atomizer Stihl SR 420	0.0016	800	0.0125	1 25.08.2011	89	fruit fruit fruit fruit	0.01 0.02 0.02 <0.01	0 2 6 13	BASF method No. 567/0
DocID 2013/1066508 Greece - GR 60100 Central Macedonia (L110432)	Wine grape (Muscat)	1. 1993 2. 25.05.-10.06.2011 3. 28.08.-15.09.2011	Knapsack sprayer, A20	0.0016	800	0.0125	1 23.08.2011	87	fruit fruit fruit fruit	0.04 0.04 0.02 0.02	0 2 8 15	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2013/1066508 Spain - 29700 Vélez-Málaga (L110431)	Wine grape (Moscatel)	1. 02.1997 2. 12.05.-01.06.2011 3. 24.08.2011	Atomizer Stihl SR 420	0.0016	800	0.0125	1 17.08.2011	88	fruit fruit fruit fruit	0.04 0.02 0.03 0.02	0 3 7 14	BASF method No. 567/0
DocID 2013/1066508 Greece - GR 60100 Central Macedonia (L110432)	Wine grape (Muscat)	1. 1993 2. 25.05.-10.06.2011 3. 28.08.-15.09.2011	Knapsack sprayer, A20	0.0016	800	0.0125	1 23.08.2011	87	fruit fruit fruit fruit	0.04 0.03 0.02 0.01	0 2 8 15	BASF method No. 567/0
DocID 2006/1026853 France - 82100 Les Barthes Labastide du Temple Ropeliere (AF/8830/BA/5)	Wine grape (Gamay)	1. 1984 2. n.a. 3. 08.09. - 22.09.2005	foliar application with mist blower	0.00125	1000	0.0125	1 08.09.2005	89	bunches bunches bunches bunches	0.035 0.044 0.020 0.027	0 4 8 14	BASF method No. 567/0
							2 08.09.2005	89	bunches bunches bunches bunches	0.071 0.036 0.034 0.051	0 4 8 14	
DocID 2006/1026853 Italy - 40024 Bologna, Castel S. Pietro (AF/8830/BA/6)	Wine grape (Montuni)	1. 1993 2. n.a. 3. 29.08. - 12.09.2005	foliar application with mist blower	0.00125	1000	0.0125	1 29.08.2005	88	bunches bunches bunches bunches	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
							2 29.08.2005	88	bunches bunches bunches bunches	0.020 <0.01 <0.01 <0.01	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026853 Spain - Muela Alta S/N, Malejan (AF/8830/BA/7)	Wine grape (Macabeo)	1. 06.02.2000 2. n.a. 3. 16.09. - 30.09.2005	foliar spray	0.00125	1000	0.0125	1 16.09.2005	85	bunches bunches bunches bunches	0.021 0.039 0.013 0.019	0 3 7 14	BASF method No. 567/0
							2 16.09.2005	85	bunches bunches bunches bunches	0.035 0.027 0.045 0.020	0 3 7 14	
DocID 2006/1026853 Greece - Anchialos, 57011 Thessaloniki (AF/8830/BA/8)	Wine grape (Roditis)	1. 1987 2. n.a. 3. 25.08. - 08.09.2005	foliar spray	0.00125	1000	0.0125	1 25.08.2005	85	bunches bunches bunches bunches	<0.01 0.019 0.012 <0.01	0 3 7 14	BASF method No. 567/0
							2 25.08.2005	85	bunches bunches bunches bunches	0.030 0.035 <0.01 <0.01	0 3 7 14	
DocID 2007/1008492 France - Mazan, Vaucluse (A/SF/I/06/131)	Wine grape (Grenache)	1. 1936 2. 15-25.05.2006 3. 18.09.2006	Foliar spray using atomizer	0.001	986	0.012	1 04.09.2006	85	fruits fruits fruits fruits	0.014 0.014 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
				0.001 0.001	977 1039	0.012 0.013	2 04.09.2006	85	fruits fruits fruits fruits	0.029 0.015 0.015 0.011	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Grapes/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008492 Greece - Kato Milea (A/GR/I/06/132)	Wine grape (Muscat)	1. 1991 2. 30.05.-10.06.2006 3. 21.09.2006	Foliar spray using knapsack sprayer	0.001	1011	0.013	1 08.09.2006	87	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF method No. 567/0
				0.001 0.001	853 1021	0.012 0.013	2 08.09.2006	87	fruits fruits fruits fruits	0.022 0.011 0.012 <0.01	0 3 7 13	
DocID 2007/1008492 Spain - Pobla del Duc, Valencia (A/SP/I/06/133)	Wine grape (Tempranillo)	1. 1998 2. May 2006 3. 07.09.2006	Foliar spray using motorised knapsack sprayer	0.001	1011	0.013	1 25.08.2006	85-87	fruits fruits fruits fruits	0.043 0.026 0.043 0.023	0 3 7 13	BASF method No. 567/0
				0.001 0.001	1001 1005	0.013 0.013	2 25.08.2006	85-87	fruits fruits fruits fruits	0.035 0.029 0.028 0.039	0 3 7 13	
DocID 2007/1008492 Italy - Costigliole D'Asti, Piemont (A/IT/I/06/134)	Wine grape (Barbera)	1. 1971 2. 30.05.-11.06.2006 3. 17.10.2006	Foliar spray using electric knapsack sprayer	0.001	1032	0.013	1 04.10.2006	88	fruits fruits fruits fruits	0.029 0.016 0.014 0.011	0 4 6 13	BASF method No. 567/0
				0.001 0.001	1016 972	0.013 0.012	2 04.10.2006	88	fruits fruits fruits fruits	0.043 0.035 0.030 0.025	0 4 6 13	

• Strawberry

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Strawberries/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008489 Belgium - Nivelles, Brabant (A/BE/I/05/88)	Strawberry (Elsanta)	1. n. a. 2. n. a. 3. 23.08.2005	foliar spray	0.004	310	0.013	1 16.08.2005	89	fruits fruits fruits fruits	<0.01 0.010 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
				0.004 0.004	308 308	0.013 0.013	2 16.08.2005	89	fruits fruits fruits fruits	0.012 0.012 <0.01 <0.01	0 1 3 7	
DocID 2007/1008489 Germany - Ladenburg, Baden-Wuerrtemberg (A/GE/I/05/81)	Strawberry (Chandler)	1. n. a. 2. May - June 3. 22.06.2005	foliar spray	0.004	280	0.012	1 15.06.2005	87-89	fruits fruits fruits fruits	<0.02 <0.01 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
				0.004 0.004	285 290	0.012 0.012	2 15.06.2005	87-89	fruits fruits fruits fruits	0.01 <0.01 <0.01 <0.01	0 1 3 7	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Strawberries/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008489 France - Fontaines-en-Sologne (Centre) (A/NF/I/05/80)	Strawberry (Cirrafine)	1. Oct 2004 2. 12.05.2005 3. 18.07.2005	foliar spray	0.004	300	0.012	1 12.07.2005	89	fruits fruits fruits fruits	0.032 0.029 0.022 0.017	0 1 3 6	BASF method No. 567/0
				0.004 0.004	302 301	0.012 0.012	2 12.07.2005	89	fruits fruits fruits fruits	0.049 0.042 0.031 0.020	0 1 3 6	
DocID 2007/1008489 UK - Ledbury, Herefordshire (A/UK/I/05/83)	Strawberry (Florence)	1. 2003 2. 23.05.-21.06.2005 3. 04.07.2005	foliar spray	0.004	293	0.012	1 27.06.2005	75-81	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
				0.004 0.004	293 313	0.012 0.013	2 27.06.2005	75-81	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 3 7	
DocID 2007/1007935 Belgium - Nivelles, Brabant (A/BE/I/05/78)	Strawberry (Darselect)	1. 20.06.2005 2. n.a. 3. 14.09.2005	foliar spray	0.0133	296	0.039	1 06.09.2005	85-87	fruits fruits fruits	0.0472 0.0400 0.0234	0 3 8	BASF method No. 567/0
DocID 2007/1007935 Germany - Ladenburg, Baden-Wuerrtemberg (A/GE/I/05/72)	Strawberry (Avanta)	1. n.a. 2. April 3. 11.05.2005	foliar spray	0.0133	280	0.037	1 04.05.2005	85-87	fruits fruits fruits	<0.01 <0.01 <0.01	0 3 7	BASF method No. 567/0
DocID 2007/1007935 Germany - Schmilau, Schleswig-Holstein (A/GE/I/05/73)	Strawberry (Rosella)	1. n.a. 2. May 3. 25.05.2005	foliar spray	0.0133	290	0.039	1 18.05.2005	85-89	fruits fruits fruits	0.0121 <0.01 <0.01	0 3 7	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Strawberries/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007935 France - Fontaines-en-Sologne (Centre) (A/NF/I/05/70)	Strawberry (Cirrafine)	1. Oct 2003 2. 10.04.-10.07.2005 3. 10.06.2005	foliar spray	0.0133	320	0.043	1 03.06.2005	89	fruits fruits fruits	0.0563 0.0541 0.0410	0 3 7	BASF method No. 567/0
DocID 2007/1008493 France - Vraux, Marne (A/NF/I/06/88)	Strawberry (Florence)	1. 26.05.2004 2. 05.05.-01.06.2006 3. 23.06.2006	foliar spray	0.004	285	0.012	1 16.06.2006	85	fruits fruits fruits fruits	0.011 <0.01 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
				0.004 0.004	326 292	0.014 0.012	2 16.06.2006	85	fruits fruits fruits fruits	0.018 0.018 <0.01 <0.01	0 1 3 7	
DocID 2007/1008493 Germany - Schmilau, Schleswig-Holstein (A/GE/I/06/89)	Strawberry (Florence)	1. May 2005 2. 12.06.-02.07.2006 3. 08.07.2006	foliar spray	0.004	314	0.013	1 30.06.2006	86	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 2 8	BASF method No. 567/0
				0.004 0.004	298 310	0.012 0.013	2 30.06.2006	86	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 2 8	
DocID 2007/1008493 Netherlands - AK Nymegen, Gelderland (A/NL/I/06/90)	Strawberry (Elsanta)	1. May 2006 2. Jul 2006 3. 15.08.2006	foliar spray	0.004	307	0.013	1 07.08.2006	87	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 4 8	BASF method No. 567/0
				0.004 0.004	315 301	0.013 0.013	2 07.08.2006	87	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 4 8	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Strawberries/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008493 UK - Harvington Evesham, Worcestershire (A/UK/I/06/91)	Strawberry (Pegasus)	1. Okt 2003 2. early May - 3. 18.05.2006 27.06.2006	foliar spray	0.004	347	0.014	1 20.06.2006	85	fruits fruits fruits fruits	0.012 0.012 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
				0.004 0.004	317 327	0.013 0.014	2 20.06.2006	85	fruits fruits fruits fruits	0.020 0.027 0.018 0.023	0 1 3 7	

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Strawberries/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece. Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008489 Italy - Gavi Ligure, Piemont (A/IT/I/05/86)	Strawberry (Granada)	1. Jul 2004 2. May 2005 3. 19.06.2005	foliar spray	0.006	384	0.024	1 12.06.2005	86-87	fruits fruits fruits fruits	0.015 <0.01 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
DocID 2007/1008489 Italy - S. Stefano Roero, Piemont (A/IT/I/05/87)	Strawberry (Grande)	1. Aug 2004 2. Apr 2005 3. 15.06.2005	foliar spray	0.006	452	0.028	1 08.06.2005	85-86	fruits fruits fruits fruits	0.038 0.028 n.a. 0.022	0 1 3 7	BASF method No. 567/0
DocID 2007/1008489 France - Feugarolles, Lot and Garonne (A/SF/I/05/84)	Strawberry (Cariguette)	1. 25.08.2004 2. 25.02-10.06.2005 3. 14.06.2005	foliar spray	0.006	409	0.026	1 07.06.2005	89	fruits fruits fruits fruits	0.012 0.011 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
DocID 2007/1008489 Spain - Quatretonda, Valencia (A/SP/I/05/85)	Strawberry (Plantafrigo)	1. 20.08.2004 2. Jan 2005 3. 10.05.2005	foliar spray	0.006	420	0.026	1 03.05.2005	87	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
DocID 2007/1007935 Greece - Svoronos (A/GR/I/05/77)	Strawberry (Aroma)	1. 20.08.2004 2. 05.05.-10.10.2005 3. 26.08.2005	foliar spray	0.0133	296	0.039	1 19.08.2005	87	fruits fruits fruits	0.0601 0.0484 0.0348	0 3 7	BASF method No. 567/0
DocID 2007/1007935 Italy - S. Stefano Roero, Piemont (A/IT/I/05/76)	Strawberry (Alba)	1. 20.07.2004 2. Mar 2005 3. 23.05.2005	foliar spray	0.0133	340	0.045	1 16.05.2005	84-86	fruits fruits fruits	0.0367 0.0289 0.0149	0 3 7	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Strawberries/Berries and small fruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece. Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007935 France - Feugarolles, Lot and Garonne (A/SF/I/05/71)	Strawberry (Dark Select)	1. 16.08.2004 2. 20.02.-10.06.2005 3. 14.06.2005	foliar spray	0.0133	298	0.040	1 07.06.2005	87	fruits fruits fruits	<0.01 <0.01 <0.01	0 3 7	BASF method No. 567/0
DocID 2007/1007935 Spain - Lloc Nou Del Fenollet, Valencia (A/SP/I/05/75)	Strawberry (Ventana)	1. 24.10.2004 2. Jan 2005 3. 13.05.2005	foliar spray	0.0133	300	0.040	1 06.05.2005	87	fruits fruits fruits	0.0134 <0.01 <0.01	0 3 7	BASF method No. 567/0
DocID 2007/1008493 France - Lansargue, Herault (A/SF/I/06/92)	Strawberry (Cleret)	1. 25.08.2005 2. 03.03.-09.06.2006 3. 06.06.2006	foliar spray	0.006	423	0.026	1 30.05.2006	89	fruits fruits fruits fruits	0.015 0.030 0.018 <0.01	0 1 3 7	BASF method No. 567/0
DocID 2007/1008493 Spain - Quatretonda, Valencia (A/SP/I/06/93)	Strawberry (Camarosa)	1. 13.10.2005 2. 25.-30.01.2006 3. 24.05.2006	foliar spray	0.006	399	0.025	1 16.05.2006	87	fruits fruits fruits fruits	0.026 0.017 0.017 0.015	0 1 3 8	BASF method No. 567/0
DocID 2007/1008493 Italy - S. Stefano Roero, Piemont (A/IT/I/06/94)	Strawberry (Marmohede)	1. Jul 2005 2. 10.04.-11.05.2006 3. 17.06.2006	foliar spray	0.006	430	0.027	1 10.06.2006	86	fruits fruits fruits fruits	0.016 0.019 <0.01 <0.01	0 1 3 7	BASF method No. 567/0
DocID 2007/1008493 Greece - Svoronos, Pierin (A/GR/I/06/95)	Strawberry (Aroma)	1. 20.04.2006 2. 20.04.-20.08.2006 3. 08.06.2006	foliar spray	0.006	402	0.025	1 02.06.2006	85	fruits fruits fruits fruits	<0.01 <0.01 <0.01 <0.01	0 1 3 6	BASF method No. 567/0

n. a. not available

• Olive

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Olives/oilfruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1004975 Spain - 41500 Alcalá de Guadaira (ALO/16/03)	Olive (Manzanilla)	1. 15.02.1985 2. 25.04.-10.05.2003 3. 12.09.-20.09.2003	High volume foliar application using mistblower sprayer	0.0015	1000	0.015	1 29.08.2003	6[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 3 6 14	BASF method No. 546/0
				0.0015 0.0015	1000 1000	0.015 0.015	2 29.08.2003	6[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 3 6 14	
DocID 2005/1004975 Spain - 41500 Arahal (ALO/17/03)	Olive (Manzanilla)	1. 15.03.99 2. 23.04.03-08.05.03 3. 12.09.-15.09.03	High volume foliar application using mistblower sprayer	0.0015	1000	0.015	1 29.08.2003	6[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 3 6 14	BASF method No. 546/0
				0.0015 0.0015	1000 1000	0.015 0.015	2 29.08.2003	6[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 3 6 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Olives/oilfruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1004975 Greece - 60063 Leptokaria (GRE/11/03)	Olive (Koroneiki)	1. n. a. 2. n. a. 3. 30.10.-15.11.03	High volume foliar application using mistblower sprayer	0.0015	1000	0.015	1 17.10.2003	7[1]	fruits fruits fruits fruits	<0.05 <0.05 0.066 <0.05	0 3 7 14	BASF method No. 546/0
				0.0015 0.0015	1000 1000	0.015 0.015	2 17.10.2003	7[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 3 7 14	
DocID 2005/1004975 Greece - 59100 Veria, Asomata (GRE/12/03)	Olive (Amfisis)	1. 01.12.1997 2. n. a. 3. 22.10.-10.11.2003	High volume foliar application using mistblower sprayer	0.0015	1000	0.015	1 10.10.2003	8[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 4 8 15	BASF method No. 546/0
				0.0015 0.0015	1000 1000	0.015 0.015	2 10.10.2003	8[1]	fruits fruits fruits fruits	<0.05 <0.05 <0.05 <0.05	0 4 8 15	

[1]dbh: days before harvest
n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Olives/oilfruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007582 Spain - 41500 Alcalá de G. Sevilla (ALO/23/04)	Olive (Manzanilla)	1. 1985 2. 20.04.-05.05.2004 3. 10.09.-15.09.2004	foliar application with lance	0.0015	1000	0.015	1 27.08.2004	78	fruits fruits fruits fruits	0.01 0.02 0.02 0.02	0 3 7 13	BASF method No. 546/0
				0.0015	1000	0.015	2 27.08.2004	78	fruits fruits fruits fruits	0.04 0.04 0.04 0.04	0 3 7 13	
DocID 2005/1007582 Spain - 41500 Arahal (ALO/24/04)	Olive (Manzanilla)	1. 1999 2. 20.04.-05.05.2004 3. 10.09.-15.09.2004	foliar application with lance	0.0015	1000	0.015	1 27.08.2004	78	fruits fruits fruits fruits	0.09 0.10 0.09 0.06	0 3 7 13	BASF method No. 546/0
				0.0015	1000	0.015	2 27.08.2004	78	fruits fruits fruits fruits	0.16 0.08 0.09 0.07	0 3 7 13	
DocID 2005/1007582 Greece - 59100 Imathia, Veria (GRE/14/04)	Olive (Halkidikis)	1. 1986 2. 20.05.-02.06.2004 3. 01.10.-10.11.2004	foliar application with lance	0.0015	1000	0.015	1 20.09.2004	79	fruits fruits fruits fruits	0.02 0.02 0.05 0.01	0 3 7 14	BASF method No. 546/0
				0.0015	1000	0.015	2 20.09.2004	79	fruits fruits fruits fruits	0.02 0.04 0.04 0.02	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Olives/oilfruits	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007582 Greece - 59100 Imathia, Veria (GRE/15/04)	Olive (Halkidikis)	1. 1983 2. 20.05.-02.06.2004 3. 01.10.-10.11.2004	foliar application with lance	0.0015	1000	0.015	1 29.09.2004	79	fruits fruits fruits fruits	0.02 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 546/0
				0.0015	1000	0.015	2 29.09.2004	79	fruits fruits fruits fruits	0.03 <0.01 <0.01 <0.01	0 3 7 14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**-**
Crop/crop group:	**Olives/oilfruits**	**Producer of commercial product**	**67056 Ludwigshafen, Germany**
Responsible body for reporting (name, address)	**BASF SE, 67056 Ludwigshafen**	**Indoor/Glasshouse/Outdoor**	**Outdoor**
Country	**Italy, Spain**	**Other active substance in the formulation**	**none**
Content of active substance (g/kg or g/L)	**100 g/L**	**(common name and content)**	
Formulation (e.g. WP)	**BAS 310 40 I (EC)**	**Residues calculated as:**	**Alpha-Cypermethrin (BAS 310 I)**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2012/1157548 Spain –Seville (L110427)	Olive FT0305 (Picual)	1. 1990 2. 05.11. 3. 11.11.	Backpack power sprayer	0.0015 0.0015	1000 1000	0.015 0.015	31.10.11 07.11.11	85-88	fruits fruits fruits fruits	0.16 0.17 0.14 0.12	0 3 7 14	BASF method No. 567/0
DocID 2012/1157548 Italy – Palagiano (L110428)	Olive FT0305 (Cima di Melfi)	1. 10.03.78 2. - 3 14.11.11	Knapsack sprayer	0.0015 0.0015	1000 1000	0.015 0.015	31.10.11 07.11.11	88	fruits fruits fruits fruits	0.12 0.10 0.19 0.13	0 3 7 14	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	**Alpha-cypermethrin**	**Commercial Product (name)**	**-**
Crop/crop group:	**Olives/oilfruits**	**Producer of commercial product**	**67056 Ludwigshafen, Germany**
Responsible body for reporting (name, address)	**BASF SE, 67056 Ludwigshafen**	**Indoor/Glasshouse/Outdoor**	**Outdoor**
Country	**Italy, Spain**	**Other active substance in the formulation (common name and content)**	**none**
Content of active substance (g/kg or g/L)	**50 g/L**		
Formulation (e.g. WP)	**BAS 310 55 I (ME)**	**Residues calculated as:**	**Alpha-Cypermethrin (BAS 310 I)**

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of : 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment kg a.s./hL	Water (L/ha)	kg a.s./ha	No of Treatm. and Last Date	Growth stage at last treatment	Portion Analysed	Residues (mg/kg)	PHI (days)	Remarks
DocID 2012/1157548 Spain –Seville (L110427)	Olive FT0305 (Picual)	1. 1990 2. 05.11. 3. 11.11.	Backpack power sprayer	0.0015 0.0015	1000 1000	0.015 0.015	31.10.11 07.11.11	85-88	fruits fruits fruits fruits	0.15 0.10 0.08 0.06	0 3 7 14	BASF method No. 567/0
DocID 2012/1157548 Spain –Seville (L110427)	Olive FT0305 (Picual)	1. 1990 2. 05.11. 3. 11.11.	Backpack power sprayer	0.030 0.030	30 30	0.009 0.009	31.10.11 07.11.11	85-88	fruits fruits fruits fruits	0.08 0.07 0.07 0.04	0 3 7 14	BASF method No. 567/0 Plot was treated with a mixture of the test item BAS 310 55 I and a commercial bait applied at a concentration of 2%.
DocID 2012/1157548 Italy – Palagiano (L110428)	Olive FT0305 (Cima di Melfi)	1. 10.03.78 2. - 3 14.11.11	Knapsack sprayer	0.0015 0.0015	1000 1000	0.015 0.015	31.10.11 07.11.11	88	fruits fruits fruits fruits	0.03 0.08 0.13 0.09	0 3 7 14	BASF method No. 567/0
DocID 2012/1157548 Italy – Palagiano (L110428)	Olive FT0305 (Cima di Melfi)	1. 10.03.78 2. - 3 14.11.11	Knapsack sprayer	0.030 0.030	30 30	0.009 0.009	31.10.11 07.11.11	88	fruits fruits fruits fruits	0.47 0.05 0.26 0.21	0 3 7 14	BASF method No. 567/0 Plot was treated with a mixture of the test item BAS 310 55 I and a commercial bait applied at a concentration of 2%.

- **Potato**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, The Netherlands	**Other active substance in the formulation (common name and content)**	Acetamiprid (100 g/L)
Content of active substance (g/kg or g/L)	25 g/L		
Formulation (e.g. WP)	BAS 370 00 I (SL)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2008/1002704 France - 80340 Herleville (Somme) (L070446)	Potato (Agria)	1. 11.04.2007 2. 18.06.2007-20.07.2007 3. 03.09.2007	foliar spraying	0.003 0.003	385 386	0.012 0.012	2 13.08.2007	48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 6 14 21	BASF method No. 567/0
DocID 2008/1002704 Netherlands - 6687 LC Angeren (Gelderland) (L070447)	Potato (Agria)	1. 11.04.2007 2. n. a. 3. 24.08.2007	foliar spraying	0.003 0.003	405 415	0.013 0.013	2 02.08.2007	44	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 8 14 22	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Denmark, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2008/1002704 France - 80340 Herleville (Somme) (L070446)	Potato (Agria)	1. 11.04.2007 2. 18.06.2007-20.07.2007 3. 03.09.2007	foliar spraying	0.003 0.003	388 394	0.012 0.012	2 13.08.2007	48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 6 14 21	BASF method No. 567/0
DocID 2008/1002704 Netherlands - 6687 LC Angeren (Gelderland) (L070447)	Potato (Agria)	1. 11.04.2007 2. n. a. 3. 24.08.2007	foliar spraying	0.003 0.003	392 402	0.012 0.013	2 02.08.2007	44	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 8 14 22	BASF method No. 567/0
DocID 2012/1157550 Germany - Goch-Nierswalde (L110423)	Potato (Bintje)	1. 18.04.2000 2. 14.07.2000 3. 08.09.2000	Portable knapsack boom sprayer	0.0075 0.0075	200 200	0.015 0.015	2 26.08.2011	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 13 20	BASF method No. 567/0
DocID 2012/1157550 UK - Croughton (L110424)	Potato (Cara)	1. 16.06.2000 2. n. a. 3. 29.09.2000	Boom sprayer	0.0075 0.0075	200 200	0.015 0.015	2 29.09.2011	47-48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 15 20	BASF method No. 567/0
DocID 2006/1026846 UK - Barn Farm, Cranebrook Lane, Hilton, Lichfield, Staffordshire WS14 0EZ (AF/8831/BA/1)	Potato (Wilja)	1. 21.04.2005 2. n. a. 3. 02.09. - 23.09.2005	foliar spray	0.0030	400	0.013	1 02.09.2005	48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (Wilja)	1. 21.04.2005 2. n. a. 3. 02.09. - 23.09.2005	foliar spray	0.0030	400	0.013	2 02.09.2005	48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Denmark, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026846 France - Le Quart Mallet, Uchizy 71700 (AF/8831/BA/2)	Potato (Synthomas)	1. 09.05.2005 2. n. a. 3. 03.08. - 24.08.2005	foliar spray	0.0030	400	0.013	1 03.08.2005	46	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (Synthomas)	1. 09.05.2005 2. n. a. 3. 03.08. - 24.08.2005	foliar spray	0.0030	400	0.013	2 03.08.2005	46	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2006/1026846 Germany - Waldstr. 5, 69256 Mauer (AF/8831/BA/3)	Potato (Agria)	1. 11.05.2005 2. n. a. 3. 23.08. - 13.09.2005	foliar spray	0.0030	400	0.125	1 23.08.2005	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (Agria)	1. 11.05.2005 2. n. a. 3. 23.08. - 13.09.2005	foliar spray	0.0030	400	0.125	2 23.08.2005	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2006/1026846 The Netherlands - Reethsestraat, 6662 PK Elst, Geiderland (AF/8831/BA/4)	Potato (Agria)	1. 20.05.2005 2. n. a. 3. 24.08. - 14.09.2005	foliar spray	0.0030	400	0.125	1 24.08.2005	69	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (Agria)	1. 20.05.2005 2. n. a. 3. 24.08. - 14.09.2005	foliar spray	0.0030	400	0.125	2 24.08.2005	69	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Denmark, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007945 France - 1932 Route d'Orlèans, St Hilaire St Mesmin, 45160 Loiret (AF/10497/BA/1)	Potato (Mona Lisa)	1. 25.04.2006 2. n. a. 3. 19.07. - 09.08.2006	foliar spray	0.0030	400	0.013	1 19.07.2006	47	tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (Mona Lisa)	1. 25.04.2006 2. n. a. 3. 19.07. - 09.08.2006	foliar spray	0.0030	400	0.013	2 19.07.2006	47	tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2007/1007945 Denmark - Agrolab A/S, Rojleskovvej 18, Middelfart, Fyn (AF/10497/BA/2)	Potato (Hamlet)	1. 13.05.2006 2. 16. - 26.07.2006 3. 26.07.2006 - 16.08.2006	foliar spray	0.0030	400	0.013	1 26.07.2006	45	tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (Hamlet)	1. 13.05.2006 2. 16. - 26.07.2006 3. 26.07.2006 - 16.08.2006	foliar spray	0.0030	400	0.013	2 26.07.2006	45	tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2007/1007945 Germany - Peiner Weg 60, 31303 Burgdorf, Lower Saxony (AF/10497/BA/3)	Potato (Bernadette)	1. 20.04.2006 2. n. a. 3. 07. - 28.07.2006	foliar spray	0.0030	400	0.013	1 07.07.2006	65-69	tubers	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
	Potato (Bernadette)	1. 20.04.2006 2. n. a. 3. 07. - 28.07.2006	foliar spray	0.0030	400	0.013	2 07.07.2006	65-69	tubers	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Denmark, France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007945 UK - 29 Ryecroft Road, Hemmington, Derby, Derbyshire, DE74 2RE (AF/10497/BA/9)	Potato (King Edward)	1. 05.06.2006 2. n. a. 3. 19.09. - 11.10.2006	foliar spray	0.0030	400	0.013	1 19.09.2006	71	tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
	Potato (King Edward)	1. 05.06.2006 2. n. a. 3. 19.09. - 11.10.2006	foliar spray	0.0030	400	0.013	2 19.09.2006	71	tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 55 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2012/1157550 Germany - Goch-Nierswalde (L110423)	Potato (Bintje)	1. 18.04.2000 2. 14.07.2000 3. 08.09.2000	Portable knapsack boom sprayer	0.0075 0.0075	200 200	0.015 0.015	2 26.08.2011	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 13 20	BASF method No. 567/0
DocID 2012/1157550 UK - Croughton (L110424)	Potato (Cara)	1. 16.06.2000 2. n. a. 3. 29.09.2000	Boom sprayer	0.0075 0.0075	200 200	0.015 0.015	2 29.09.2011	47-48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 15 20	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID AL-721-049 Netherlands - Ottersum, Zansteeg 18, 6595 Limburg (AGR/27/01)	Potato (Cilena)	1. 20.04.2001 2. 22.06. - 06.06.2001 3. 10. - 11.09.2001	foliar spray	0.0050	308	0.015	1 07.08.2001	43	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 20 28 34	BASF method RLA 12513.03V
	Potato (Cilena)	1. 20.04.2001 2. 22.06. - 06.07.2001 3. 10. - 11.09.2001	foliar spray	0.005 0.005	298 296	0.015 0.015	2 07.08.2001	43	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 20 28 34	BASF method RLA 12513.03V
DocID AL-721-049 Germany - 67376 Herthausen, Schulstraße 5, Rheinland-Pfalz (DU4/10/01)	Potato (Solana)	1. 03.06.2001 2. n. a. 3. 02.10.2001	foliar spray	0.0050	308	0.015	1 13.08.2001	41	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 8 21 28 35	BASF method RLA 12513.03V
	Potato (Solana)	1. 03.06.2001 2. n. a. 3. 02.10.2001	foliar spray	0.005 0.005	300 300	0.015 0.015	2 27.08.2001	33	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V
DocID AL-721-049 France - 72800 Thoree les Pins, le Point du Jour (FBM/06/01)	Potato (Nicolas)	1. 05.05.2001 2. 16. - 20.07.2001 3. 21.09. - 04.10.2001	foliar spray	0.005	329	0.017	1 28.08.2001	91	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V
	Potato (Nicolas)	1. 05.05.2001 2. 16. - 20.07.2001 3. 21.09. - 04.10.2001	foliar spray	0.005 0.005	344 303	0.017 0.015	2 28.08.2001	91	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID AL-721-049 UK - Akeman Street Farm, Combe Oxfordshire (OAT/08/01)	Potato (Wilja)	1. 23.05.2001 2. 15. - 25.06.2001 3. 25. - 26.09.2001	foliar spray	0.0050	280	0.014	1 17.08.2001	79	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 6 20 28 35	BASF method RLA 12513.03V
	Potato (Wilja)	1. 23.05.2001 2. 15. - 25.06.2001 3. 25. - 26.09.2001	foliar spray	0.005 0.005	295 301	0.014 0.015	2 17.08.2001	79	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 6 20 28 35	BASF method RLA 12513.03V

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Spain	**Other active substance in the formulation**	Acetamiprid (100 g/L)
Content of active substance (g/kg or g/L)	25 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 370 00 I (SL)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2008/1002704 Spain - 46470 Albal (Valencia) (L070448)	Potato (Condor)	1. 10.03.2007 2. n. a. 3. 08.06.2007	foliar spraying	0.003 0.003	405 392	0.013 0.012	2 18.05.2007	65	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 8 15 21	BASF method No. 567/0
DocID 2008/1002704 France - 84800 Lagnes (Provence) (L070449)	Potato (Mona Lisa)	1. 15.03.2007 2. 08.06.-29.06.2007 3. 18.07.2007	foliar spraying	0.003 0.003	402 423	0.013 0.013	2 27.06.2007	68	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2008/1002704 Spain - 46470 Albal (Valencia) (L070448)	Potato (Condor)	1. 10.03.2007 2. n. a. 3. 08.06.2007	foliar spraying	0.003 0.003	419 409	0.013 0.013	2 18.05.2007	65	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 8 15 21	BASF method No. 567/0
DocID 2008/1002704 France - 84800 Lagnes (Provence) (L070449)	Potato (Mona Lisa)	1. 15.03.2007 2. 08.06.-29.06.2007 3. 18.07.2007	foliar spraying	0.003 0.003	408 421	0.013 0.013	2 27.06.2007	68	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2012/1157550 Italy - Ginosa (L110425)	Potato (Spunta)	1. 30.08.2000 2. n. a. 3. 15.12.2000	Boom sprayer compressed air pump	0.0075 0.0075	200 200	0.015 0.015	2 01.12.2011	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 8 14 21	BASF method No. 567/0
DocID 2012/1157550 Spain - Seville (L110426)	Potato (Liseta)	1. 29.08.2000 2. n. a. 3. 15.12.2000	Backpack power sprayer	0.0075 0.0075	200 200	0.015 0.015	2 01.12.2011	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2005/1007592 Greece (N) - 50100 Kozani, Macedonia (GRE/02/04)	Potato (Agria)	1. 28.03.2004 2. 01.06. - 18.07.2004 3. 20.08. - 15.09.2004	foliar spray	0.0050	300	0.015	1 28.07.2004	49	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 3 8 14	BASF method No. 567/0
	Potato (Agria)	1. 28.03.2004 2. 01.06. - 18.07.2004 3. 20.08. - 15.09.2004	foliar spray	0.0050	300	0.015	2 28.07.2004	49	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 3 8 14	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007592 Italy - 27050 Casei, Gerola, Pavia (ITA/01/04)	Potato (Agata)	1. 20.03.12004 2. 30.04. - 15.05.2004 3. 07. - 10.08.2004	foliar spray	0.0050	300	0.015	1 27.07.2004	49	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
	Potato (Agata)	1. 20.03.12004 2. 30.04. - 15.05.2004 3. 07. - 10.08.2004	foliar spray	0.0050	300	0.015	2 27.07.2004	49	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
DocID 2006/1026846 France - Route de Monciar de Quercy, Montauban 82000 (AF/8831/BA/5)	Potato (Agatha)	1. 08.2005 2. n. a. 3. 07.10. - 28.10.2005	foliar spray	0.0060	400	0.025	1 07.10.2005	47	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2006/1026846 Italy - Via Olmo, 60 - Budrio, Bologna 40054 (AF/8831/BA/6)	Potato (Agata)	1. 03.05.2005 2. n. a. 3. 29.06. - 19.07.2005	foliar spray	0.0060	400	0.025	1 29.06.2005	45-46	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2006/1026846 Spain - Replazeto, S/N, Villarreal 50490 (AF/8831/BA/7)	Potato (Agria)	1. 08.04.2005 2. n. a. 3. 29.08. - 20.09.2005	foliar spray	0.0060	400	0.025	1 29.08.2005	46	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0
DocID 2006/1026846 Greece - Zoodochos Pygi, Kozani, Central Macedonia, GR-50100 (AF/8831/BA/8)	Potato (Sprunta)	1. 13.04.2005 2. n. a. 3. 01.08. - 22.08.2005	foliar spray	0.0060	400	0.025	1 01.08.2005	42	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007945 France - 7 Chemin de Coudoumac, St Jory, 31790 Haute-Garonne (AF/10497/BA/5)	Potato (Mona Lisa)	1. 30.03.2006 2. n. a. 3. 04. - 25.07.2006	foliar spray	0.0030	400	0.013	1 04.07.2006	46	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
DocID 2007/1007945 Spain - C/Justica de Aragon 11, Alcalá de Moncayo, Aragon 50591 (AF/10497/BA/6)	Potato (Agria)	1. 08.04.2006 2. n. a. 3. 08. - 29.08.2006	foliar spray	0.0060	400	0.025	1 08.08.2006	81-83	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 8 14 21	BASF method No. 567/0
DocID 2007/1007945 Italy - Via Olmo 60, Budrio (BO), Emilie-Romagna 40054 (AF/10497/BA/7)	Potato (Almera)	1. 04.04.2006 2. n. a. 3. 14.07. - 04.08.2006	foliar spray	0.0060	400	0.025	1 14.07.2006	46-48	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 6 14 21	BASF method No. 567/0
DocID 2007/1007945 Greece - Nea Magnisia, Thessaloniki, Central Macedonia, GR-57008 (AF/10497/BA/8)	Potato (Agria)	1. 26.03.2006 2. n. a. 3. 16.06. - 07.07.2006	foliar spray	0.0030	400	0.013	1 16.06.2006	649-664	tubers tubers tubers tubers	<0.01 <0.01 <0.01 <0.01	0 7 13 21	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 55 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2012/1157550 Italy - Ginosa (L110425)	Potato (Spunta)	1. 30.08.2000 2. n. a. 3. 15.12.2000	Boom sprayer compressed air pump	0.0075 0.0075	200 200	0.015 0.015	2 01.12.2011	47	tuber tuber tuber tuber	<0.01 <0.01 <0.01 <0.01	0 8 14 21	BASF method No. 567/0
DocID 2012/1157550 Spain - Seville (L110426)	Potato (Liseta)	1. 29.08.2000 2. n. a. 3. 15.12.2000	Backpack power sprayer	0.0075 0.0075	200 200	0.015 0.015	2 01.12.2011	47	tuber tuber tuber tuber	<0.01 <0.01 <0.01 <0.01	0 7 14 21	BASF method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID AL-721-049 Spain - Virgen del Castilio 31, E-41710 Sevilla, Andalucia (ALO/39/01)	Potato (Spunta)	1. 02.03.2001 2. 07. - 08.06.2001 3. 07. - 08.06.2001	foliar spray	0.0050	308	0.015	1 03.05.2001	48	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V
	Potato (Spunta)	1. 02.03.2001 2. 07. - 08.06.2001 3. 07. - 08.06.2001	foliar spray	0.005 0.005	308 302	0.015 0.015	1 03.05.2001	48	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V
DocID AL-721-049 France - 26380 Perins, Le plan Rhone-Alpes (FBD/05/01)	Potato (Mana)	1. 04.06.2001 2. 22.08.2001 3. 22.08.2001	foliar spray	0.0050	311	0.016	1 24.07.2001	32	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 8 22 28 35	BASF method RLA 12513.03V
	Potato (Mana)	1. 04.06.2001 2. 22.08.2001 3. 22.08.2001	foliar spray	0.005 0.005	310 320	0.015 0.016	2 24.07.2001	32	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 8 22 28 35	BASF method RLA 12513.03V
DocID AL-721-049 Italy - 27050 Casei Gerola, Strada Voghera, Pavia (ITA/07/01)	Potato (Monalisa)	1. 20.03.2001 2. 01. - 25.06.2001 3. 08. - 18.08.2001	foliar spray	0.0050	300	0.015	1 13.07.2001	85	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 8 22 29 36	BASF method RLA 12513.03V
	Potato (Monalisa)	1. 20.03.2001 2. 01. - 25.06.2001 3. 08. - 18.08.2001	foliar spray	0.005 0.005	302 340	0.015 0.017	1 13.07.2001	85	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 8 22 29 36	BASF method RLA 12513.03V

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Potato/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID AL-721-049 Greece - Edessa, Nisi, Macedonia (HEL/01/01)	Potato (Spunta)	1. 28.05.2001 2. n. a. 3. n. a.	foliar spray	0.0070	204	0.015	1 28.09.2001	89	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V
	Potato (Spunta)	1. 28.05.2001 2. n. a. 3. n. a.	foliar spray	0.007 0.008	214 194	0.016 0.015	2 28.09.2001	89	tubers tubers tubers tubers tubers	<0.05 <0.05 <0.05 <0.05 <0.05	0 7 21 28 35	BASF method RLA 12513.03V

n. a. not available

• Carrot

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Carrot/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	Fastac SC	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1035294 Germany - 67435 Neustadt a. d. W. RU-I-13 04 RP NW 2/1	Carrot (Nandera)	1. 13.05.2004 2. n. a. 3. 27.07.-10.08.2004	spraying	0.002 0.002	596 578	0.0124 0.0120	2 27.07.2004	14 days before harvest	root root root root	0.01/0.02 <0.01 <0.01 <0.01	0 3 7 14	Method DFG S19
DocID 2005/1035294 Germany - 67435 Neustadt a. d. W. RU-I-13 04 RP NW 2/2	Carrot (Nandera)	1. 13.05.2004 2. n. a. 3. 03.08.-17.08.2004	spraying	0.002 0.002	596 613	0.0124 0.0128	2 03.08.2004	14 days before harvest	root root root root	0.01 <0.01 <0.01 <0.01	0 3 7 14	Method DFG S19
DocID 2005/1035294 Germany - 67435 Neustadt a. d. W. RU-I-13 04 RP NW 2/3	Carrot (Napoli)	1. 17.06.2004 2. n. a. 3. 13.09.2004	spraying	0.003 0.003	416 400	0.0130 0.0125	2 30.08.2004	405/407	root root root root	0.01 <0.01 <0.01 <0.01	0 3 7 14	Method DFG S19

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Carrot/root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	Fastac SC Super Contact	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1029533 Germany - Bonn RU-I-06 05 NW BN 2/1	Carrot (Dordogne)	1. 20.06.2005 2. n. a. 3. September 2005	spraying	0.002 0.002	600 600	0.0125 0.0125	2 27.09.2005	n. a.	root root root root	<0.005 <0.005 <0.005 <0.005	0 3 7 14	SAA/C/PSM 023
DocID 2006/1029533 Germany - Bonn RU-I-06 05 NWBN 2/2	Carrot (Yukon)	1. 20.06.2005 2. n. a. 3. 11.-18.10.2005	spraying	0.002 0.002	600 600	0.0125 0.0125	2 04.10.2005	n. a.	root root	<0.005 <0.005	7 14	SAA/C/PSM 023
DocID 2006/1029533 Germany - Kiel RU-I-06 05 SHKI 2/1	Carrot (Naploi)	1. 26.04.2005 2. n. a. 3. 05.08.2005	spraying	0.003 0.003	400 400	0.0125 0.0125	2 29.07.2005	n. a.	root	<0.005	7	SAA/C/PSM 023
DocID 2006/1029533 Germany - Kiel RU-I-06 05 SHKI 2/2	Carrot (Bolero)	1. 26.04.2005 2. n. a. 3. 05.08.2005	spraying	0.003 0.003	400 400	0.0125 0.0125	2 29.07.2005	n. a.	root	<0.005	7	SAA/C/PSM 023
DocID 2006/1029533 Germany - Freising RU-I-06 05 BYFS 2/2	Carrot (Tino)	1. 18.06.2005 2. n. a. 3. 21.09.2005	spraying	0.002 0.002	600 600	0.0125 0.0125	2 19.09.2005	77	root root	<0.005 <0.005	7 14	SAA/C/PSM 023
DocID 2006/1029533 Germany - Freising RU-I-06 05 BYFS 2/1	Carrot (Tino)	1. 18.06.2005 2. n. a. 3. 21.09.2005	spraying	0.002 0.002	600 600	0.0125 0.0125	2 14.09.2005	76	root root	<0.005 <0.005	7 14	SAA/C/PSM 023

• Onion

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Onion/Bulb vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, UK	**Other active substance in the formulation (common name and content)**	none
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026854 UK - Southery, Downham Market, Norfolk PE 38 0NL (AF/8827/BA/1)	Bulb onion (Hyskin)	1. 14.03.2005 2. n. a. 3. 08.08. - 22.08.2005	foliar spray with boom	0.003	400	0.0125	1 08.08.2005	47	bulbs	<0.01 0.012 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 08.08.2005	47	bulbs	0.017 0.014 0.010 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026854 France - 45300 Rouvres St Jean (AF/8827/BA/2)	Bulb onion (Summit)	1. 21.03.2005 2. n. a. 3. 05.08. - 19.08.2005	foliar spray with boom	0.003	400	0.0125	1 05.08.2005	47-48	bulbs	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 05.08.2005	47-48	bulbs	0.013 <0.01 0.010 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026854 France - 45390 Aulnay la Riviere, Loiret (AF/8827/BA/3)	Bulb onion (Summit)	1. 05.04.2005 2. n. a. 3. 05.08. - 19.08.2005	foliar spray with boom	0.003	400	0.0125	1 05.08.2005	47-48	bulbs	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 05.08.2005	47-48	bulbs	0.016 <0.01	0 3	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Onion/Bulb vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, UK	**Other active substance in the formulation (common name and content)**	none
Content of active substance (g/kg or g/L)	100 g/L		
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
										<0.01 <0.01	7 14	
DocID 2006/1026854 Netherlands - 6678 PB Oosterhout, Gelderland (AF/8827/BA/4)	Bulb onion (Hyfort)	1. 23.03.2005 2. n. a. 3. 18.08. - 01.09.2005	foliar spray with boom	0.003	400	0.0125	1 18.08.2005	47	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 18.08.2005	47	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2007/1008499 France - Bignicourt, Champagne-Ardennes (A/NF/I/06/150)	Onion (Barito)	1. 20.03.2006 2. n. a. 3. 06.09.2006	foliar spray with boom	0.003	397	0.012	1 22.08.2006	47	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 3 8 15	BASF Method No. 567/0
				0.003 0.003	420 408	0.013 0.013	2 22.08.2006	45 47	bulbs bulbs bulbs bulbs	0.014 <0.01 <0.01 <0.01	0 3 8 15	BASF Method No. 567/0
DocID 2007/1008499 Germany - Schmilau, Schleswig-Holstein (A/GE/I/06/151)	Onion (Stuttgarter Riesen)	1. 03.06.2006 2. n. a. 3. 27.08.2006	foliar spray with boom	0.003	427	0.013	1 13.08.2006	44	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	423 427	0.013 0.013	2 13.08.2006	43-44 44	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Onion/Bulb vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany, The Netherlands, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008499 Germany - Crossen, Sachsen (A/GE/I/06/152)	Onion (Sturon)	1. 06.04.2006 2. n. a. 3. 07.09.2006	foliar spray with boom	0.003	373	0.012	1 24.08.2006	44-45	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
				0.003 0.003	418 405	0.013 0.013	2 24.08.2006	44 44-45	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2007/1008499 Netherlands - NB Elst, Gelderland (A/NL/I/06/153)	Onion (Donna)	1. 24.03.2006 2. n. a. 3. 24.08.2006	foliar spray with boom	0.003	388	0.012	1 09.08.2006	46-47	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 3 7 15	BASF Method No. 567/0
				0.003 0.003	408 405	0.013 0.013	2 09.08.2006	44-45 46-47	bulbs bulbs bulbs bulbs	<0.01 <0.01 <0.01 <0.01	0 3 7 15	BASF Method No. 567/0

n. a. not available

• Tomato

Glasshouse/Indoor

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	DE, DK, ES, FR, GR, IT, NL	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/5000719 Germany - Brandenburg (ACK/07/04)	Tomato (Swift)	1. 24.02.2004 2. 08.03.-10.08.2004 3. 25.05.-08.06.2004	foliar spray	0.004	400	0.015	1 25.05.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 25.05.2004	85	fruit fruit fruit fruit	0.012 0.012 0.010 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2004/5000719 Netherlands - Limburg (AGR/09/04)	Tomato (Cedrico)	1. 08.01.2004 2. 26.01.-30.11.2004 3. 12.-25.10.2004	foliar spray	0.004	400	0.015	1 12.10.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 12.10.2004	85	fruit fruit fruit fruit	0.017 0.018 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
DocID 2004/5000719 Denmark - S. Jutland (ALB/06/04) [1]	Tomato (Aromata)	1. 05.01.2004 2. 01.03.-30.10.2004 3. 10.-24.06.2004	foliar spray	0.004	400	0.015	1 10.06.2004	83	fruit fruit fruit fruit	<0.01 <0.01 0.019 <0.01	0 4 7 14	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 10.06.2004	83	fruit fruit fruit fruit	0.019 <0.01 0.013 0.011	0 4 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	DE, DK, ES, FR, GR, IT, NL	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/5000719 Spain - Andalucia (ALO/16/04)	Tomato (Antilla)	1. 17.08.2004 2. 15.09.-27.10.2004 3. 03.-16.11.2004	foliar spray	0.004	400	0.015	1 03.11.2004	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 03.11.2004	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
DocID 2004/5000719 France - Pays de la Loire (FBM/05/04)	Tomato (Petula)	1. 17.04.2004 2. 20.06.-12.07.2004 3. 16.-30.07.2004	foliar spray	0.004	400	0.015	1 16.07.2004	83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 16.07.2004	83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2004/5000719 France - Rhone-Alpes (FTL/12/04) [1]	Tomato (Brenda)	1. 02.06.2004 2. 26.-31.07.2004 3. 08.-21.09.2004	foliar spray	0.004	400	0.015	1 16.09.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 2 6 13	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 16.09.2004	85	fruit fruit fruit fruit	0.011 <0.01 <0.01 0.015	0 2 6 13	BASF Method No. 567/0
DocID 2004/5000719 France - Pays de la Loire (FBM/05/04)	Tomato (Petula)	1. 17.04.2004 2. 20.06.-12.07.2004 3. 16.-30.07.2004	foliar spray	0.004	400	0.015	1 16.07.2004	83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 16.07.2004	83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	DE, DK, ES, FR, GR, IT, NL	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/5000719 Greece - Macedonia (GRE/11/04)	Tomato (Alma)	1. 22.07.2004 2. 14.08.2004-n. a. 3. 17.09.-01.10.2004	foliar spray	0.004	400	0.015	1 17.09.2004	76	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 17.09.2004	76	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2004/5000719 Italy - Torino (ITA/08/04) [1]	Tomato (Seni)	1. 05.07.2004 2. 12.-30.08.2004 3. 13.-27.09.2004	foliar spray	0.004	400	0.015	1 13.09.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004 0.004	400 400	0.015 0.015	2 13.09.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

n. a. not available

[1]Samples from trials ALB/06/04, FTL12/04, ITA08/04 thawed during delays in customs, but arrived cold in the laboratory. This indicates that they have not been without dry ice for long and were in good condition for analysis.

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruitsing vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Indoor
Country	BE, DE, ES, FR, GR, IT	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007934 France - St. Genouph, Tourraine (A/NF/I/05/50)	Tomato (Sympathic)	1. 21.03.2005 2. 01.04.-30.06.2005 3. 04.07.2005	foliar spray	0.010	390	0.039	1 20.06.2005	81	fruit fruit fruit fruit	<0.01 0.010 0.011 0.016	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007934 Belgium - Villers-Perwin, Hainaut (A/BE/I/05/54)	Tomato (Paola)	1. 10.05.2005 2. n. a. 3. 09.08.2005	foliar spray	0.010	423	0.042	1 26.07.2005	71-85	fruit fruit fruit fruit	<0.01 0.015 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007934 Germany - Hassloch-Meckenheim (A/GE/I/05/52)	Tomato (Pipo)	1. end May 2005 2. mid June 2005 3. 27.07.2005	foliar spray	0.010	370	0.037	1 13.07.2005	85-88	fruit fruit fruit fruit	0.019 0.025 <0.01 0.013	0 2 7 14	BASF Method No. 567/0
DocID 2007/1007934 Germany - Engelbrechtsche Wildnis, Schleswig-Holstein (A/GE/I/05/53)	Tomato (Alma)	1. May 2005 2. mid June 2005 3. 16.08.2005	foliar spray	0.010	415	0.042	1 01.08.2005	85-87	fruit fruit fruit fruit	0.012 0.016 <0.01 <0.01	0 3 7 15	BASF Method No. 567/0
DocID 2007/1007934 France - St. Remy de Provence, Bouches du Rhone (A/SF/I/05/51)	Tomato (Brenda)	1. 18.05.2005 2. 20.06.-20.08.2005 3. 02.08.2005	foliar spray	0.010	416	0.042	1 19.07.2005	74-81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007934 Spain - Benifayo, Valencia (A/SP/I/05/55)	Tomato (Marmande RAF)	1. 15.02.2005 2. March 3. 21.06.2005	foliar spray	0.010	405	0.040	1 07.06.2005	85	fruit fruit fruit fruit	0.030 0.024 0.015 0.014	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruitsing vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Indoor
Country	BE, DE, ES, FR, GR, IT	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007934 Greece - Profitis, Thessaloniki (A/GR/I/05/57)	Tomato (Alma)	1. 01.05.2005 2. 10.07.-30.08.2005 3. 22.08.2005	foliar spray	0.010	386	0.038	1 29.07.2005	84	fruit fruit fruit fruit	<0.01 <0.01 0.014 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007934 Italy - Motta di Costigliole, Loc. Reimoneirini, Piemont (A/IT/I/05/56)	Tomato (Cuor di bue, hybrid)	1. April 2005 2. starting - 12.05.2005 3. 25.07.2005	foliar spray	0.010	410	0.041	1 11.07.2005	85	fruit fruit fruit fruit	0.029 0.037 0.024 0.032	0 3 7 14	BASF Method No. 567/0

n. a. not available

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090704 Germany - 67245 Lambsheim (L080173)	Tomato (Vanessa)	1. 11.05.2008 2. End of May 2008 3. 18.07.2008	Gloria pneumatic sprayer, installed on an unicycle	0.0038	400	0.0150	2 06.08.2008	82	fruits fruits fruits fruits	0.02 0.03 0.02 0.02	0 2 8 14	BASF Method No. 567/0
DocID 2009/1090704 France - 49650, Allonnes (L080174)	Tomato (Topkapi)	1. 12.06.2008 2. 12.07.-18.08.2008 3. 15.09.-24.09.2008	Sprayer with booms (AGROTOP) TRS mesu 06007	0.0038	400	0.0150	2 08.09.2008	89	fruits fruits fruits fruits	0.02 <0.01 0.01 <0.01	0 3 8 14	BASF Method No. 567/0
DocID 2007/1008488 France - Dame Marie les Bois (A/NF/I/05/60)	Tomato (Joker)	1. 13.06.2005 2. 01.07.-15.08.2005 3. 12.09.2005	foliar spray	0.003	405	0.013	1 29.08.2005	82	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	407 408	0.013 0.013	2 29.08.2005	82	fruit fruit fruit fruit	0.013 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008488 France - Saint Martin des Bois (A/NF/I/05/61)	Tomato (Hector)	1. 05.06.2005 2. 01.07.-15.08.2005 3. 23.09.2005	foliar spray	0.003	405	0.013	1 09.09.2005	84	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	427 403	0.013 0.013	2 09.09.2005	84	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008488 Germany - Lambsheim (A/GE/I/05/62)	Tomato (Vanessa)	1. May 2. June 3. 07.09.2005	foliar spray	0.003	425	0.013	1 24.08.2005	81-85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	410 375	0.013 0.013	2 24.08.2005	81-85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008488 Belgium - Villers-Perwin, Hainaut (A/BE/I/05/63)	Tomato (Felicie)	1. 02.06.2005 2. n. a. 3. 20.09.2005	foliar spray	0.003	406	0.013	1 06.09.2005	69-83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	400 419	0.013 0.013	2 06.09.2005	69-83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007937 France - Allonnes, 49650 Maine-et-Loire (AF/10504/BA/1)	Tomato (Topkapi)	1. 15.06.2006 2. n. a. 3. 25.08.-08.09.2006	foliar spray	0.003	400	0.0125	1 25.08.2006	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 25.08.2006	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007937 Germany - Ringstr. 46, 67245 Lambsheim (AF/10504/BA/2)	Tomato (Vanessa)	1. 12.05.2006 2. n. a. 3. 26.07.-08.08.2006	foliar spray	0.003	400	0.0125	1 26.07.2006	73	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 2 7 13	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
				0.003	400	0.0125	2	73	fruit	0.011	0	BASF Method No.
				0.003	400	0.0125	26.07.2006		fruit	<0.01	2	567/0
									fruit	<0.01	7	
									fruit	<0.01	13	
DocID 2007/1008494 France - Warmeriville, Marne (A/NF/I/06/97)	Tomato (Medina)	1. 06.06.2006 2. 25.06.-30.07.2006 3. 06.09.2006	foliar spray	0.003	387	0.012	1 23.08.2006	83	fruit	<0.01	0	BASF Method No.
									fruit	<0.01	4	567/0
									fruit	<0.01	7	
									fruit	<0.01	14	
				0.003	389	0.012	2	83	fruit	<0.01	0	BASF Method No.
				0.003	423	0.013	23.08.2006		fruit	<0.01	4	567/0
									fruit	<0.01	7	
									fruit	<0.01	14	
DocID 2007/1008494 Germany - Schleswig, Schleswig-Holstein (A/GE/I/06/98)	Tomato (Harzfeuer)	1. 02.06.2006 2. 02.06.-01.08.2006 3. 12.08.2006	foliar spray	0.003	431	0.014	1 29.07.2006	84	fruit	<0.01	0	BASF Method No.
									fruit	<0.01	3	567/0
									fruit	<0.01	8	
									fruit	<0.01	14	
				0.003	440	0.014	2	84	fruit	<0.01	0	BASF Method No.
				0.003	394	0.012	29.07.2006		fruit	0.0101	3	567/0
									fruit	<0.01	8	
									fruit	<0.01	14	
DocID 2007/1008494 Germany - Lambsheim, Rheinland-Pfalz (A/GE/I/06/99)	Tomato (Vanessa)	1. 12.03.-13.05.2006 2. n. a. 3. 01.08.2006	foliar spray	0.003	400	0.013	1 18.07.2006	79	fruit	<0.01	0	BASF Method No.
									fruit	<0.01	3	567/0
									fruit	<0.01	7	
									fruit	<0.01	14	
				0.003	409	0.013	2	79	fruit	0.011	0	BASF Method No.
				0.003	382	0.012	18.07.2006		fruit	<0.01	3	567/0
									fruit	<0.01	7	
									fruit	<0.01	14	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Belgium, France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008494 Germany - Kirchheim, Rheinland-Pfalz (A/GE/I/06/100)	Tomato (Tombolino St. Pierre Fiaschetto Mix)	1. 15.03.-26.05.2006 2. n. a. 3. 17.08.2006	foliar spray	0.003	392	0.012	1 03.08.2006	80	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	417 417	0.013 0.013	2 03.08.2006	80	fruit fruit fruit fruit	0.0121 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/kg	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007937 France - Allonnes, 49650 Maine-et-Loire (AF/10504/BA/1)	Tomato (Topkapi)	1. 15.06.2006 2. n. a. 3. 25.08.-08.09.2006	foliar spray	0.003	400	0.0125	1 25.08.2006	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 25.08.2006	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007937 Germany - Ringstr. 46, 67245 Lambsheim (AF/10504/BA/2)	Tomato (Vanessa)	1. 12.05.2006 2. n. a. 3. 26.07.-08.08.2006	foliar spray	0.003	400	0.0125	1 26.07.2006	73	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 2 7 13	BASF Method No. 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 26.07.2006	73	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 2 7 13	BASF Method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 51 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090704 Germany - 67245 Lambsheim (L080173)	Tomato (Vanessa)	1. 11.05.2008 2. End of May 2008 3. 18.07.2008	Gloria pneumatic sprayer, installed on an unicycle	0.0038	400	0.0150	2 06.08.2008	82	fruits fruits fruits fruits	0.03 0.03 0.02 0.02	0 2 8 14	BASF Method No. 567/0
DocID 2009/1090704 France - 49650, Allonnes (L080174)	Tomato (Topkapi)	1. 12.06.2008 2. 12.07.-18.08.2008 3. 15.09.-24.09.2008	Sprayer with booms (AGROTOP) TRS mesu 06007	0.0038	400	0.0150	2 08.09.2008	89	fruits fruits fruits fruits	0.01 <0.01 0.01 0.01	0 3 8 14	BASF Method No. 567/0

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090704 Italy - 44023 Lagosanta Ferrara (L080175)	Tomato (Asterix)	1. 08.05.2008 2. 10.06-23.06.2008 3. 18.08.-25.08.2008	Echo SHR 150 SI	0.0075	400	0.0300	1 11.08.2008	85	fruit fruit fruit fruit	0.03 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2009/1090704 Spain - Union del Llano Zafarraya, Granada (L080176)	Tomato (Brillante)	1. 20.05.2008 2. 10.07.2008 3. 21.08.2008	Stihl Atomizer	0.0075	400	0.0300	1 21.08.2008	71	fruit fruit fruit fruit	0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008488 France - Boisseron, Languedoc Roussillon (A/SF/I/05/64)	Tomato (All R 50)	1. 19.05.2005 2. 17.07.-28.08.2005 3. 30.09.2005	foliar spray	0.006	449	0.028	1 16.09.2005	81	fruit fruit fruit fruit	<0.01 <0.01 0.012 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008488 France - Caumont, Midi Pyrenees (A/SF/I/05/65)	Tomato (Coudoulet)	1. 10.05.2005 2. 01.-31.07.2005 3. 19.08.2005	foliar spray	0.006	400	0.025	1 05.08.2005	87	fruit fruit fruit fruit	0.015 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008488 Spain - Turis, Valencia (A/SP/I/05/66)	Tomato (Rio Grande)	1. 14.06.2005 2. 15.07.2005 3. 12.09.2005	foliar spray	0.006	400	0.025	1 29.08.2005	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008488 Italy - Costigliole d'Asti, Piemont (A/IT/I/05/67)	Tomato (Rio Grande)	1. 14.05.2005 2. 10.07.2005 3. 01.09.2005	foliar spray	0.006	430	0.027	1 17.08.2005	89	fruit fruit fruit fruit	<0.01 <0.01 0.021 <0.01	0 4 7 15	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007937 Spain - 50669 Santa Engracia (AF/10504/BA/3)	Tomato (Tina)	1. 05.05.2006 2. n. a. 3. 11.-25.09.2006	foliar spray	0.006	400	0.025	1 11.09.2006	85	fruit fruit fruit fruit	<0.01 0.013 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2007/1007937 France - Castelmayran, Tarn-et-Garronne (AF/10504/BA/4)	Tomato (Leader)	1. 10.05.2006 2. n. a. 3. 26.07.-09.08.2006	foliar spray	0.006	400	0.025	1 26.07.2006	81-85	fruit fruit fruit fruit	0.018 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2007/1008494 France - Grillo, Vaucluse (A/SF/I/06/101)	Tomato (Perfectil)	1. 28.04.-05.05.2006 2. 07.07.-01.08.2006 3. 25.08.2006	foliar spray	0.006	396	0.025	1 11.08.2006	81	fruit fruit fruit fruit	0.0122 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008494 Greece - Profodis, Thessaloniki (A/GR/I/06/102)	Tomato (Alma)	1. 20.06.2006 2. 15.06.-10.08.2006 3. 01.08.2006	foliar spray	0.006	391	0.024	1 18.07.2006	88	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 4 8 14	BASF Method No. 567/0
DocID 2007/1008494 Spain - Benicario, Castellon (A/SP/I/06/103)	Tomato (Bodar)	1. march 2. April - end of cycle 3. 28.07.2006	foliar spray	0.006	400	0.025	1 14.07.2006	73	fruit fruit fruit fruit	0.021 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008494 Italy - Costigliole d'Asti, Piemont (A/IT/I/06/104)	Tomato (Rio Grande)	1. 14.05.2006 2. 17.06.-11.08.2006 3. 26.08.2006	foliar spray	0.006	398	0.025	1 12.08.2006	81	fruit fruit fruit fruit	0.0129 0.0187 0.0126 <0.01	0 2 6 14	BASF Method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/kg	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007937 Spain - 50669 Santa Engracia (AF/10504/BA/3)	Tomato (Tina)	1. 05.05.2006 2. n. a. 3. 11.-25.09.2006	foliar spray	0.006	400	0.025	1 11.09.2006	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2007/1007937 France - Castelmayran, Tarn-et-Garronne (AF/10504/BA/4)	Tomato (Leader)	1. 10.05.2006 2. n. a. 3. 26.07.-09.08.2006	foliar spray	0.006	400	0.025	1 26.07.2006	81-85	fruit fruit fruit fruit	0.016 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Tomato/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 51 I (ME)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2009/1090704 Italy - 44023 Lagosanta Ferrara (L080175)	Tomato (Asterix)	1. 08.05.2008 2. 10.06-23.06.2008 3. 18.08.-25.08.2008	Echo SHR 150 SI	0.0075	400	0.0300	1 11.08.2008	85	fruit fruit fruit fruit	0.02 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2009/1090704 Spain - Union del Llano Zafarraya, Granada (L080176)	Tomato (Brillante)	1. 20.05.2008 2. 10.07.2008 3. 21.08.2008	Stihl Atomizer	0.0075	400	0.0300	1 21.08.2008	71	fruit fruit fruit fruit	0.02 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

• Sweet pepper

Glasshouse

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sweet pepper/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	Belgium, France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1036933 France - Chemin Madeleines, 84800 Isle sur la Sorgue (05 I CL FR P35)	Pepper (Hannibal)	1. 16.03.2005 2. 10.05.2005 - end of trial 3. n. .	foliar spray	0.010	400	0.040	1 26.07.2005	73	fruit	0.016 0.016 0.015 0.011	0 3 7 14	BASF Method No. 567/0
DocID 2006/1036933 Belgium - Rue Dominique Seret 34, 6210 Villers-Pervin Wagnelee (G023-05 I)	Pepper (Dazzle)	1. 10.05.2005 2. 20.-25.06.2005 3. 25.-30.09.2005	foliar spray	0.010	400	0.040	1 20.09.2005	87-89	fruit	0.019 0.028 0.029 0.033	0 3 7 14	BASF Method No. 567/0
DocID 2006/1036933 France - Chemin d'Arles, 13870 Rognonas (05 I Cl FR P39)	Pepper (Galileo)	1. 25.03.2005 2. 15.05.-Aug 2005 3. June-Sept 2005	foliar spray	0.010	400	0.040	1 08.08.2005	72	fruit	0.016 0.017 <0.01 0.013	0 3 7 14	BASF Method No. 567/0
DocID 2006/1036933 Italy - Via Foggia 110, 70056 Molfetta, Bari (IR05BASL51PL01)	Pepper (Quadrato d'Asti)	1. 10.05.2005 2. 10.06.-30.07.2005 3. 02.07.-25.08.2005	foliar spray	0.010	400	0.040	1 27.07.2005	74	fruit	0.044 0.011 0.018 0.013	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sweet pepper/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	Belgium, France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1036933 Spain - Finca La Dehesilla, 41710 Utrera, Sevilla (05ES086R)	Pepper (Italico)	1. 06.04.2005 2. 08.06.2005 3. 10.07.-01.09.2005	foliar spray	0.010	400	0.040	1 01.07.2005	75	fruit	0.031 0.027 0.029 0.023	0 3 7 13	BASF Method No. 567/0
DocID 2006/1036933 Greece - Profitis, 57200 Thessaloniki (05RF047)	Pepper (Staboli)	1. 20.05.2005 2. June-Aug 2005 3. July-Sept 2005	foliar spray	0.010	400	0.040	1 08.07.2005	81	fruit	0.049 0.016 0.016 0.016	0 3 7 14	BASF Method No. 567/0

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sweet pepper/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008497 France - Chateaurenard, Bouche de Rohne (A/SF/I/06/162)	Sweet pepper (Galilo)	1. 10.04.2006 2. 19.05.-17.07.2006 3. 17.07.2006	foliar spray	0.006	402	0.025	1 03.06.2006	81	fruit fruit fruit fruit	0.065 0.011 <0.01 <0.01	0 4 8 14	BASF Method No. 567/0
DocID 2007/1008497 Italy - Costigliole d'Asti, Piemont (A/IT/I/06/163)	Sweet pepper (Quadrato d'Asti)	1. 17.05.2006 2. 27.07.-30.08.2006 3. 11.09.2006	foliar spray	0.006	404	0.025	1 29.08.2006	81	fruit fruit fruit fruit	0.032 0.015 <0.01 <0.01	0 3 8 13	BASF Method No. 567/0
DocID 2007/1008497 Spain - Almussafes, Valencia (A/SP/I/06/164)	Sweet pepper (Stilo)	1. 10.04.2006 2. Mid May - Mid 3. June 2006 13.07.2006	foliar spray	0.006	395	0.025	1 30.06.2006	72	fruit fruit fruit fruit	0.033 0.025 0.024 0.012	0 3 8 13	BASF Method No. 567/0
DocID 2007/1008497 Greece - Profitis, 57200 Thessaloniki (A/GR/I/06/165)	Sweet pepper (Laser F1)	1. 20.05.2006 2. 10.06.-10.08.2006 3. 24.07.2006	foliar spray	0.006	398	0.025	1 11.07.2006	87	fruit fruit fruit fruit	0.019 0.030 0.017 <0.01	0 3 7 13	BASF Method No. 567/0
DocID 2006/1026860 France - Labarthe, 82220 Tarn-et-garonne (AF/8820/BA/1)	Pepper (Albi)	1. 04.05.2005 2. n. a. 3. 28.06.-12.07.2005	foliar spray	0.006	400	0.025	1 28.06.2005	68	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026860 Spain - El Viso Del Alcor, 415200 Sevilla (AF/8820/BA/2)	Pepper (Negrillo)	1. 15.04.2005 2. n. a. 3. 20.06.-04.07.2005	foliar spray	0.006	400	0.025	1 20.06.2005	71	fruit fruit fruit fruit	0.011 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sweet pepper/fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026860 Italy - Castenaso, 40055 Bologna (AF/8820/BA/3)	Pepper (Senior)	1. 30.03.2005 2. n. a. 3. 13.-26.09.2005	foliar spray	0.006	400	0.025	1 13.09.2005	81-82	fruit fruit fruit fruit	0.018 0.024 0.028 <0.01	0 2 6 13	BASF Method No. 567/0
DocID 2006/1026860 Greece - 57200 Thesalloniki (AF/8820/BA/4)	Pepper (Florinis)	1. 20.05.2005 2. n. a. 3. 30.07.-13.08.2005	foliar spray	0.006	400	0.025	1 30.07.2005	87	fruit fruit fruit fruit	0.032 0.026 0.014 0.015	0 3 7 14	BASF Method No. 567/0

• Melon

Glasshouse

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Melone/Cucurbit fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	DK, ES, Fr, IT, NL,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/5000721 Netherlands - Limburg (AGR/11/04)	Melon (Lunabel)	1. 01.06.2004 2. 30.06.-28.07.2004 3. 01.-15.09.2004	foliar spray	0.004	400	0.015	1 01.09.2004	87	fruit fruit fruit fruit	0.013 <0.01 0.010 0.011	0 2 7 14	BASF Method No. 567/0
				0.004	400	0.015	2 01.09.2004	87	fruit fruit fruit fruit	0.014 0.010 0.019 0.012	0 2 7 14	BASF Method No. 567/0
DocID 2004/5000721 Denmark - S. Jutland (ALB/08/04)	Melon (Aroma)	1. 08.05.2004 2. 10.-20.07.2004 3. 27.07.-09.08.2004	foliar spray	0.004	400	0.015	1 27.07.2004	79	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
				0.004	400	0.015	2 27.07.2004	79	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Melone/Cucurbit fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	DK, ES, Fr, IT, NL,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/5000721 Spain - Andalucia (ALO/18/04)	Melon (Maxdimon)	1. 17.08.2004 2. 12.09.-20.10.2004 3. 26.10.-09.11.2004	foliar spray	0.004	400	0.015	1 26.10.2004	79	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004	400	0.015	2 26.10.2004	79	fruit fruit fruit fruit	0.011 0.011 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2004/5000721 France - Rhone-Alpes (FBD/12/04)	Melon (Nagaro)	1. 21.06.2004 2. 24.07.-15.08.2004 3. 19.08.-02.09.2004	foliar spray	0.004	400	0.015	1 19.08.2004	83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
				0.004	400	0.015	2 19.08.2004	83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2004/5000721 France - Languedoc, Roussillon (FTL/13/04)	Melon (Panchito)	1. 15.02.2004 2. 20.-25.05.2004 3. 08.-22.06.2004	foliar spray	0.004	400	0.015	1 08.06.2004	77	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
				0.004	400	0.015	2 08.06.2004	77	fruit fruit fruit fruit	0.011 0.011 0.010 0.013	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Melone/Cucurbit fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Glasshouse
Country	DK, ES, Fr, IT, NL,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/5000721 Italy - Piemonte (ITA/10/04)	Melon (Macigno)	1. 05.03.2004 2. 28.04.-25.05.2004 3. 07.-22.06.2004	foliar spray	0.004	400	0.015	1 07.06.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 15	BASF Method No. 567/0
				0.004	400	0.015	2 07.06.2004	85	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 15	BASF Method No. 567/0

Note: eight trials initiated, two trials (France North and Greece) cancelled due to poor crop growth.

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Melone/Cucurbit fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Belgium, Denmark, France, Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1024607 France - SCA Rouge Gorge du Thouet, Charteau, Taize, 79100 Noize (AF/8816/BA/1)	Melon (Anasta)	1. 13.06.2005 2. n.a. 3. 19.08.-02.09.2005	foliar spray	0.006	400	0.025	1 19.08.2005	74	whole fruit whole fruit whole fruit whole fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0 Amendment: DocID 2007/1011007
DocID 2006/1024607 France - SCA Rouge Gorge du Thouet, Charteau, Taize, 79100 Noize (AF/8816/BA/2)	Melon (Anasta)	1. 17.06.2005 2. n.a. 3. 22.08.-05.09.2005	foliar spray	0.006	400	0.025	1 22.08.2005	73	whole fruit whole fruit whole fruit whole fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0 Amendment: DocID 2007/1011007
DocID 2006/1037507 Belgium - 3454 Rummen, Limburg (AGR/54/05)	Melon (Delta F1)	1. 19.05.2005 2. 19.06.-19.07.2005 3. 06.09.2005	foliar spray	0.010	400	0.040	1 30.08.2005	87	fruit fruit fruit fruit	0.058 0.057 0.022 0.030	0 3 7 13	BASF Method No. 567/0
DocID 2006/1037507 Germany - 47574 Goch-Hülm (AGR/53/05)	Melon (Delta F1)	1. 20.05.2005 2. 20.06.-20.07.2005 3. 06.09.2005	foliar spray	0.010	400	0.040	1 29.08.2005	87	fruit fruit fruit fruit	0.047 0.045 0.029 0.022	0 3 8 14	BASF Method No. 567/0
DocID 2006/1037507 Denmark - Borkopskovvej 114A, 7080 Borkop (ALB/190508-01)	Melon (Aroma)	1. 29.05.2005 2. 20.06.-05.07.2005 3. 20.-30.07.2005	foliar spray	0.010	400	0.040	1 13.07.2005	75	fruit fruit fruit fruit	0.012 0.010 <0.01 <0.01	0 2 7 14	BASF Method No. 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Melone/Cucurbit fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1024607 Spain - 21450 Villamanrique de la Condesa (AF/8816/BA/3)	Melon (Lusitano)	1. 08.03.2005 2. n.a. 3. 03.-17.06.2005	foliar spray	0.006	400	0.025	1 03.06.2005	72	whole fruit whole fruit whole fruit whole fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0 Amendment: DocID 2007/1011007
DocID 2006/1024607 Italy - Via Nuove 44, Funo, 40050 Bologna (AF/8816/BA/4)	Melon (C5)	1. 03.06.2005 2. n.a. 3. 20.07.-14.08.2005	foliar spray	0.006	400	0.025	1 20.07.2005	85	whole fruit whole fruit whole fruit	<0.01 <0.01 <0.01 <0.01	0 2 7 14	BASF Method No. 567/0 Amendment: DocID 2007/1011007
DocID 2007/1007940 France - 82290 Meauzac (AF/10490/BA/1)	Melon (Cezanne)	1. 05.05.2006 2. n.a. 3. 11.-25.07.2006	foliar spray	0.006	400	0.025	1 11.07.2006	81	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 2 7 14	BASF Method No. 567/0
DocID 2007/1007940 Greece - Platanos, 59032 Imathia (AF/10490/BA/2)	Melon (Masada F1)	1. 10.07.2006 2. n.a. 3. 31.08.-14.09.2006	foliar spray	0.006	400	0.025	1 31.08.2006	88	fruit fruit fruit fruit	0.014 0.014 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007940 Spain - Los Palacios y Villafranca (AF/10490/BA/3)	Melon (Piel de Sapo, Nicolas)	1. 08.03.2006 2. n.a. 3. 09.-23.06.2006	foliar spray	0.006	400	0.025	1 09.06.2006	71-83	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 6 14	BASF Method No. 567/0
DocID 2007/1007940 Italy - 40054 Budrio (AF/10490/BA/4)	Melon (Tamaris)	1. 20.05.2006 2. n.a. 3. 27.07.-10.08.2006	foliar spray	0.006	400	0.025	1 27.07.2006	87-88	fruit fruit fruit fruit	0.014 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Melone/Cucurbit fruiting vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1037507 France - Route d'Alleins, 13560 Senas (05 I CL FR P34)	Melon (Anasta)	1. 11.04.2005 2. 05.-20.05.2005 3. 21.06.2005	foliar spray	0.010	400	0.040	1 14.06.2005	71	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
DocID 2006/1037507 France - Route d'Alleins, 13560 Senas (05 I CL FR P38)	Melon (Luna Star)	1. 27.03.2005 2. 01.-15.05.2005 3. 15.06.2005	foliar spray	0.010	400	0.040	1 08.06.2005	71	fruit fruit fruit fruit	<0.01 0.012 <0.01 <0.01	0 3 7 13	BASF Method No. 567/0
DocID 2006/1037507 Italy - Via Cacciatori, 40014 Palata Pepoli, Bologna (IR05BASG61LG01)	Melon (Creso)	1. 05.03.2005 2. 01.-15.06.2005 3. 10.-25.06.2005	foliar spray	0.010	400	0.040	1 10.06.2005	87	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2006/1037507 Spain - 41710 Utrera, Sevilla (05ES/085R)	Melon (Maxdimon)	1. 22.08.2005 2. 13.10.2005 3. 02.-03.12.2005	foliar spray	0.010	400	0.040	1 18.11.2005	79	fruit fruit fruit fruit	0.074 0.070 0.048 0.045	0 3 7 13	BASF Method No. 567/0
DocID 2006/1037507 Greece - Profitis, Thessaloniki (05RF046)	Melon (Gali F1)	1. 07.05.2005 2. 10.06.-05.08.2005 3. 20.07.-30.08.2005	foliar spray	0.010	400	0.040	1 30.07.2005	84	fruit fruit fruit fruit	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

n. a. not available

• Broccoli

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Broccoli/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026863 Germany - Deichstr. 7, 67227 Frankenthal (AF/8814/BA/1)	Broccoli (Ovation)	1. 05.05.2005 2. n. a. 3. 27.06.-11.07.2005	foliar spray with boom	0.003	400	0.0125	1 27.06.2005	55	Inflorescence Inflorescence Inflorescence Inflorescence	0.163 0.048 <0.01 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 27.06.2005	55	Inflorescence Inflorescence Inflorescence Inflorescence	0.259 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026863 Netherlands - 4756 SB Kruisland, Brabant (AF/8814/BA/2)	Broccoli (Volta)	1. 20.07.2005 2. n. a. 3. 20.09.-04.10.2005	foliar spray with boom	0.003	400	0.0125	1 20.09.2005	44	Inflorescence Inflorescence Inflorescence Inflorescence	0.044 0.030 0.015 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 20.09.2005	44	Inflorescence Inflorescence Inflorescence Inflorescence	0.074 0.039 0.015 <0.01	0 3 7 14	BASF Method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Broccoli/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026863 UK - Westgate Farm, Hesketh Bank, Lancashire PR4 6XS (AF/8814/BA/3)	Broccoli (Chevalier)	1. 27.07.2005 2. n. a. 3. 10.-24.10.2005	foliar spray with boom	0.003	400	0.0125	1 10.10.2005	44	Inflorescence Inflorescence Inflorescence Inflorescence	0.056 0.033 0.016 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 10.10.2005	44	Inflorescence Inflorescence Inflorescence Inflorescence	0.048 0.015 <0.01 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026863 France - 1 Rue des Petits Champs, 49730 Varennes-sur-Loire (AF/8814/BA/4)	Broccoli (Marathon)	1. 15.06.2005 2. n. a. 3. 07.-21.10.2005	foliar spray with boom	0.003	400	0.0125	1 07.10.2005	45	Inflorescence Inflorescence Inflorescence Inflorescence	0.035 0.018 0.013 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 07.10.2005	45	Inflorescence Inflorescence Inflorescence Inflorescence	0.069 0.034 0.018 <0.01	0 3 7 14	BASF Method 567/0
DocID 2007/1013274 France - Sedan, Champagne-Ardennes (A/NF/I/06/120)	Broccoli (Monterey)	1. 26.06.2006 2. n. a. 3. 26.09.2006	foliar spray with boom	0.003	427	0.013	1 13.09.2006	45	Inflorescence Inflorescence Inflorescence Inflorescence	0.036 0.016 0.010 <0.01	0 3 7 13	BASF Method 567/0
				0.003 0.003	435 395	0.014 0.012	2 13.09.2006	39 45	Inflorescence Inflorescence Inflorescence Inflorescence	0.050 0.017 0.011 <0.01	0 3 7 13	BASF Method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Broccoli/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1013274 Germany - Erfurt, Thüringen (A/GE/I/06/121)	Broccoli (Ironman)	1. 11.07.2006 2. n. a. 3. 08.11.2006	foliar spray with boom	0.003	400	0.013	1 25.10.2006	49	Inflorescence Inflorescence Inflorescence Inflorescence	0.032 0.030 0.017 0.017	0 3 6 14	BASF Method 567/0
				0.003 0.003	400 400	0.012 0.013	2 25.10.2006	48-49 49	Inflorescence Inflorescence Inflorescence Inflorescence	0.052 0.035 0.029 0.018	0 3 6 14	BASF Method 567/0
DocID 2007/1013274 Denmark - Middelfart, Fyn (A/DK/I/06/122)	Broccoli (Marathon)	1. 09.05.2006 2. n. a. 3. 26.07.2006	foliar spray with boom	0.003	420	0.013	1 12.07.2006	43	Inflorescence Inflorescence Inflorescence Inflorescence	0.053 0.030 0.014 <0.01	0 2 7 14	BASF Method 567/0
				0.003 0.003	395 407	0.012 0.013	2 12.07.2006	43 46	Inflorescence Inflorescence Inflorescence Inflorescence	0.052 0.032 0.017 <0.01	0 2 7 14	BASF Method 567/0
DocID 2007/1013274 UK - Chipping Campden, Gloucestershire (A/UK/I/06/123)	Broccoli (Marathon)	1. 01.04.2006 2. 19.08.2006 3. 19.08.2006	foliar spray with boom	0.003	422	0.013	1 04.08.2006	48	Inflorescence Inflorescence Inflorescence Inflorescence	0.044 0.032 0.025 <0.01	0 3 7 15	BASF Method 567/0
				0.003 0.003	396 409	0.012 0.013	2 04.08.2006	47 48	Inflorescence Inflorescence Inflorescence Inflorescence	0.050 0.036 0.018 <0.01	0 3 7 15	BASF Method 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Broccoli/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026863 France - 82170 Canals (AF/8814/BA/5)	Broccoli (Chevalier)	1. 13.04.2005 2. n. a. 3. 06.-20.06.2005	foliar spray with boom	0.006	400	0.025	1 06.06.2005	47	Inflorescence Inflorescence Inflorescence Inflorescence	0.070 0.028 0.017 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026863 Italy - Via Cadriano 60/2, 40057 Granarolo (AF/8814/BA/6)	Broccoli (Eyron)	1. 05.08.2005 2. n. a. 3. 17.-31.10.2005	foliar spray with boom	0.006	400	0.025	1 17.10.2005	46-47	Inflorescence Inflorescence Inflorescence Inflorescence	0.065 0.047 0.027 0.011	0 3 7 14	BASF Method 567/0
DocID 2007/1013274 Greece - Chalkidona, Thessaloniki (A/GR/I/06/124)	Broccoli (Marathon)	1. 10.07.2006 2. 01.-25.10.2006 3. 17.10.2006	foliar spray with boom	0.006	397	0.025	1 03.10.2006	47-48	Inflorescence Inflorescence Inflorescence Inflorescence	0.056 0.032 0.014 <0.01	0 3 7 14	BASF Method 567/0
DocID 2007/1013274 Spain - Nava Campana, Albacete, Castilla la Mancha (A/SP/I/06/125)	Broccoli (Monaco)	1. end Jul 2006 2. end Oct 2006 3. 06.11.2006	foliar spray with motorized boom	0.006	428	0.027	1 24.10.2006	49	Inflorescence Inflorescence Inflorescence Inflorescence	0.050 0.026 0.013 <0.01	0 4 7 13	BASF Method 567/0

n. a. not available

• Cauliflower

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026864 France - 49320 Coutures (AF/8813/BA/1)	Cauliflower (Optimist)	1. 20.07.2005 2. n. a. 3. 11.10.-25.10.2005	foliar application with boom	0.003	400	0.0125	1 11.10.2005	41-43	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
				0.003	400	0.0125	2 11.10.2005	41-43	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
DocID 2006/1026864 UK - Mullensgrove Farm, Marston, Curdworh, Warwickshire (AF/8813/BA/2)	Cauliflower (white rock)	1. 14.07.2005 2. n. a. 3. 16.09.-30.09.05	foliar application with boom	0.003	400	0.0125	1 16.09.05	43	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
				0.003	400	0.0125	2 16.08.05	43	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026864 Germany - Pattensen/Jeinsen (AF/8813/BA/3)	Cauliflower (freedom)	1. 08.07.2005 2. n. a. 3. 30.08.-14.10.2005	foliar application with boom	0.003	357	0.0111	1 30.09.2005	43-47	Inflorescence Inflorescence Inflorescence Inflorescence	0.1231 0.0416 0.0531 0.0112	0 3 7 14	BASF method No. 567/0
				0.003	400	0.0125	2 30.09.2005	43-47	Inflorescence Inflorescence Inflorescence Inflorescence	0.2007 0.0750 0.0852 0.0271	0 3 7 14	BASF method No. 567/0
DocID 2006/1026864 The Netherlands - 3281 L W Numansdorp Zuid, Holland (AF/8813/BA/4)	Cauliflower (fremont)	1. 07.06.2005 2. n. a. 3. 16.08.-30.08.2005	foliar application with boom	0.003	400	0.0125	1 16.08.2005	43-45	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
				0.003	400	0.0125	2 16.08.2005	43-45	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
DocID 2007/1007936 Germany - 67071 Ludwigshafen (AF/10502/BA/1)	Cauliflower (Gregor)	1. 25.06.2006 2. n. a. 3. 13.09.-27.09.2006	foliar application with boom sprayer	0.003	400	0.0125	1 13.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 2 7 14	BASF method No. 567/0
				0.003	400	0.0125	2 13.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 2 7 14	BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007936 UK - Marshalls, Butterwick, Boston, Lincolshire PE20 3SS (AF/10502/BA/2)	Cauliflower (Correll)	1. 17.07.2006 2. n. a. 3. 23.09.-07.10.06	foliar application with boom sprayer	0.003	400	0.0125	1 23.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 4 6 14	BASF method No. 567/0
				0.003	400	0.0125	2 23.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 4 6 14	BASF method No. 567/0
DocID 2007/1008495 France - Sedan, Champagne-Ardennes (A/NF/I/06/136)	Cauliflower (Cartier)	1. 20.06.2006 2. n. a. 3. 21.10.2006	foliar spray	0.003	406	0.013	1 08.10.2006	47	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 6 13	BASF Method No. 567/0
				0.003 0.003	383 394	0.012 0.012	2 08.10.2006	45 47	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 6 13	BASF Method No. 567/0
DocID 2007/1008495 UK - Chipping Campden, Gloucestershire (A/UK/I/06/137)	Cauliflower (Fremont)	1. 01.04.2006 2. n. a. 3. 30.09.2006	foliar spray	0.003	405	0.013	1 17.09.2006	42	Inflorescence Inflorescence Inflorescence Inflorescence	0.022 0.025 <0.01 <0.01	0 3 8 13	BASF Method No. 567/0
				0.003 0.003	385 393	0.012 0.012	2 17.09.2006	41 42	Inflorescence Inflorescence Inflorescence Inflorescence	0.056 0.017 <0.01 <0.01	0 3 8 13	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, DK, FR, NL, UK,	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1008495 Netherlands - LB Angeren, Gelderland (A/NL/I/06/138)	Cauliflower (Fremont)	1. 14.07.2006 2. n. a. 3. 10.10.2006	foliar spray	0.003	387	0.012	1 26.09.2006	45-46	Inflorescence Inflorescence Inflorescence Inflorescence	0.028 0.017 0.013 <0.01	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	396 405	0.012 0.013	2 26.09.2006	43-44 45-46	Inflorescence Inflorescence Inflorescence Inflorescence	0.036 0.017 0.010 <0.01	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008495 Denmark - Middelfart, Fyn (A/DK/I/06/139)	Cauliflower (Fremont)	1. 10.05.2006 2. n. a. 3. 15.08.2006	foliar spray	0.003	387	0.012	1 01.08.2006	45	Inflorescence Inflorescence Inflorescence Inflorescence	0.028 0.032 0.012 <0.01	0 3 8 14	BASF Method No. 567/0
				0.003 0.003	402 398	0.013 0.012	2 01.08.2006	43 45	Inflorescence Inflorescence Inflorescence Inflorescence	0.048 0.024 0.011 <0.01	0 3 8 14	BASF Method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007936 Germany - 67071 Ludwigshafen (AF/10502/BA/1)	Cauliflower (Gregor)	1. 25.06.2006 2. n. a. 3. 13.09.-27.09.2006	foliar application with boom sprayer	0.003	400	0.0125	1 13.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 2 7 14	BASF method No. 567/0
				0.003	400	0.0125	2 13.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 2 7 14	BASF method No. 567/0
DocID 2007/1007936 UK - Marshalls, Butterwick, Boston, Lincolshire PE20 3SS (AF/10502/BA/2)	Cauliflower (Correll)	1. 17.07.2006 2. n. a. 3. 23.09.-07.10.06	foliar application with boom sprayer	0.003	400	0.0125	1 23.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 4 6 14	BASF method No. 567/0
				0.003	400	0.0125	2 23.09.2006	43-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 4 6 14	BASF method No. 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026864 France - 31330 Merville (AF/8813/BA/5)	Cauliflower (Fremont)	1. 14.07.05 2. n. a. 3. 30.09.-14.10.05	foliar application with boom	0.006	400	0.025	1 30.09.2005	43-47	Inflorescence Inflorescence Inflorescence Inflorescence	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
DocID 2006/1026864 Italy - 40057 Viadagola (AF/8813/BA/6)	Cauliflower (Emeraude)	1. 05.08.2005 2. n. a. 3. 08.11.-22.11.05	foliar application with boom	0.006	400	0.025	1 08.11.2005	43-46	Inflorescence Inflorescence Inflorescence Inflorescence	0.0144 <0.01 <0.01 <0.01	0 3 7 14	BASF method No. 567/0
DocID 2007/1007936 Italy - 40054 Bologna (AF/1052/BA/3)	Cauliflower (Dunia)	1. 27.07.2006 2. n. a. 3. 20.09.-03.10.06	foliar application with boom sprayer	0.006	400	0.025	1 20.09.2006	45-47	Heads Heads Heads Heads	0.160 0.050 <0.01 <0.01	0 2 8 13	BASF method No. 567/0
DocID 2007/1007936 France - 31330 St. Caprais (AF/10502/BA/4)	Cauliflower (Kintore)	1. 01.07.2006 2. n. a. 3. 18.09.-02.10.06	foliar application with boom sprayer	0.006	400	0.025	1 18.09.2006	45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 8 14	BASF method No. 567/0
DocID 2007/1008495 Spain - Benicarlo, Castellon (A/SP/I/06/140)	Cauliflower (Flamenco)	1. 28.07.2006 2. n. a. 3. 02.11.2006	foliar spray	0.006	432	0.027	1 19.10.2006	46	Inflorescence Inflorescence Inflorescence Inflorescence	0.016 <0.01 <0.01 <0.01	0 4 7 14	BASF Method No. 567/0
DocID 2007/1008495 Greece - Chalkidona, Thessaloniki (A/GR/I/06/141)	Cauliflower (Siria)	1. 10.07.2006 2. 01.-25.10.2006 3. 19.10.2006	foliar spray	0.006	398	0.025	1 06.10.2006	47-48	Inflorescence Inflorescence Inflorescence Inflorescence	0.014 <0.01 <0.01 <0.01	0 4 7 13	BASF Method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cauliflower/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France,Italy	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	150 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 08 I (WG)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007936 Italy - 40054 Bologna (AF/1052/BA/3)	Cauliflower (Dunia)	1. 27.07.2006 2. n. a. 3. 20.09.-03.10.06	foliar application with boom sprayer	0.006	400	0.025	1 20.09.2006	45-47	Heads Heads Heads Heads	0.130 0.083 0.014 <0.01	0 2 8 13	BASF method No. 567/0
DocID 2007/1007936 France - 31330 St. Caprais (AF/10502/BA/4)	Cauliflower (Kintore)	1. 01.07.2006 2. n. a. 3. 18.09.-02.10.06	foliar application with boom sprayer	0.006	400	0.025	1 18.09.2006	45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 8 14	BASF method No. 567/0

n. a. not available

• Brussels sprouts

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Brussel sprouts/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026859 UK - Mullensgrove farm, Marston, Curdworth, Warwickshire (AF/8818/BA/1)	Brussels sprouts (Romulus)	1. 08.06.2005 2. n. a. 3. 09.-23.11.2005	foliar spray with boom	0.003	400	0.0125	1 09.11.2005	43-45	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 09.11.2005	43-45	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 0.014 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026859 France - Le Pot Dore, 49650 Allonnes (AF/8818/BA/2)	Brussels sprouts (Cirius)	1. 21.09.2005 2. n.a. 3. 26.01.-09.02.2006	foliar spray with boom	0.003	400	0.0125	1 26.01.2006	43-45	Sprouts Sprouts Sprouts Sprouts	0.011 <0.01 <0.01 <0.01	0 4 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 26.01.2006	43-45	Sprouts Sprouts Sprouts Sprouts	0.017 0.013 0.017 0.014	0 4 7 14	BASF Method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Brussel sprouts/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026859 Germany - Hauptstr. 100, 67365 Schwegenheim (AF/8818/BA/3)	Brussels sprouts (Esperal)	1. 16.06.2005 2. n. a. 3. 22.11.-06.12.2005	foliar spray with boom	0.003	400	0.0125	1 22.11.2005	76	Sprouts Sprouts Sprouts Sprouts	0.051 0.055 0.046 0.033	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 22.11.2005	76	Sprouts Sprouts Sprouts Sprouts	0.018 0.016 0.016 0.014	0 3 7 14	BASF Method 567/0
DocID 2006/1026859 Netherlands - Teeuwenhofweg, 5809 EJ Leunen, Limburg (AF/8818/BA/4)	Brussels sprouts (Cumulus)	1. 09.-10.05.2005 2. n. a. 3. 04.-18.11.2005	foliar spray with boom	0.003	400	0.0125	1 04.11.2005	45-47	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 04.11.2005	45-47	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0
DocID 2007/1007943 France - 19 Chemin St. Remy, 45570 Dampierre en Burly (AF/10499/BA/1)	Brussels sprouts (Diablo)	1. 04.07.2006 2. n. a. 3. 05.-19.12.2006	foliar spray with boom	0.003	400	0.0125	1 05.12.2006	47-49	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 0.01 0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 05.12.2006	47-49	Sprouts Sprouts Sprouts Sprouts	0.03 0.02 0.03 0.02	0 3 7 14	BASF Method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Brussel sprouts/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007943 Belgium - 50 Rue de Thuin, 6534 Gozee (AF/10499/BA/2)	Brussels sprouts (Millenium)	1. 05.06.2006 2. n. a. 3. 23.10.-06.11.2006	foliar spray with boom	0.003	400	0.0125	1 23.10.2006	47	Sprouts Sprouts Sprouts Sprouts	0.01 0.01 <0.01 <0.01	0 4 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 23.10.2006	47	Sprouts Sprouts Sprouts Sprouts	0.02 0.02 0.02 0.01	0 4 7 14	BASF Method 567/0
DocID 2007/1007943 Germany - Hauptstr. 100, 67365 Schwegenheim (AF/10499/BA/3)	Brussels sprouts (F1 Esperal, F1 Lunet)	1. n. a. 2. n. a. 3. 22.11.-06.12.2006	foliar spray with boom	0.003	400	0.0125	1 22.11.2006	77	Sprouts Sprouts Sprouts Sprouts	0.02 0.02 0.01 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 22.11.2006	77	Sprouts Sprouts Sprouts Sprouts	0.04 0.02 0.02 0.02	0 3 7 14	BASF Method 567/0
DocID 2007/1007943 UK - Taylors Farm, Marsh Road, Banks, Lancashire PR9 8DX (AF/10499/BA/4)	Brussels sprouts (Hellemus)	1. n. a. 2. n. a. 3. 24.11.-07.12.2006	foliar spray with boom	0.003	400	0.0125	1 24.11.2006	48-49	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 <0.01 <0.01	0 3 6 13	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 24.11.2006	48-49	Sprouts Sprouts Sprouts Sprouts	<0.01 0.02 0.01 <0.01	0 3 6 13	BASF Method 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Brussel sprouts/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026859 Spain - 50641 Boquineni (AF/8818/BA/5)	Brussels sprouts (Jade Cross)	1. 19.08.2005 2. n. a. 3. 27.01.-10.02.2006	foliar spray with boom	0.006	400	0.025	1 27.01.2006	46	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 <0.01 <0.01	0 4 7 14	BASF Method 567/0
DocID 2006/1026859 Greece - Chalkidona, 57007 Thessaloniki (AF/8818/BA/6)	Brussels sprouts (Ikaros)	1. 23.07.2005 2. n.a. 3. 01.-15.11.2005	foliar spray with boom	0.006	400	0.025	1 01.11.2005	47	Sprouts Sprouts Sprouts Sprouts	0.016 0.013 0.011 <0.01	0 3 7 14	BASF Method 567/0
DocID 2007/1007943 France - 13 Av. Gascogne, 31700 Mondonville (AF/10499/BA/5)	Brussels sprouts (Dominator)	1. 01.07.2006 2. n. a. 3 05.-19.12.2006	foliar spray with boom	0.006	400	0.025	1 05.12.2006	47	Sprouts Sprouts Sprouts Sprouts	0.02 0.03 0.02 0.02	0 4 7 14	BASF Method 567/0
DocID 2007/1007943 Italy - Via Cadriano 60/2, Granarolo, 40057 Bologna (AF/10499/BA/6)	Brussels sprouts (Grosso di Cassano)	1. 25.07.2006 2. n. a. 3. 21.11-05.12.2006	foliar spray with boom	0.006	400	0.025	1 21.11.2006	47	Sprouts Sprouts Sprouts Sprouts	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0

n. a. not available

• Head Cabbage

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Head Cabbage/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Denmark, Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/1006470 Germany - 16833 Lentzke, Brandenburg (ACK/14/03)	Red Cabbage (Rodon F1)	1. 13.06.2003 2. n. a. 3. 21-22.10.2001	foliar spray	0.0025	600	0.015	1 08.10.2003	48	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 2 7 14	BASF Method 546/0
				0.0025 0.0025	600 600	0.015 0.015	2 08.10.2003	48	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 2 7 14	BASF Method 546/0
DocID 2004/1006470 Denmark - 5500 Middelfart, Fuenen (ALB/12/03)	Savoy cabbage (Mila)	1. 12.05.2003 2. n. a. 3. 25.08.-15.09.2003	foliar spray	0.0025	600	0.015	1 20.08.2003	48	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 2 7 14	BASF Method 546/0
				0.0025 0.0025	600 600	0.015 0.015	2 20.08.2003	48	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 2 7 14	BASF Method 546/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Head Cabbage/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Denmark, Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 11 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2004/1006470 France - 67203 Oberschaeffolsheim, Alsace (FAN/21/03)	White cabbage (Zerlina)	1. 19.05.2003 2. n. a. 3. 25.09-31.10.2003	foliar spray	0.0025	600	0.015	1 19.09.2003	47	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 3 7 14	BASF Method 546/0
				0.0025 0.0025	600 600	0.015 0.015	2 19.09.2003	47	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 3 7 14	BASF Method 546/0
DocID 2004/1006470 UK - OX27 9AS Bicester, Oxfordshire (OAT/18/03)	Red cabbage (Rona)	1. 13.05.2003 2. n. a. 3. 08.-09.09.2003	foliar spray	0.0025	600	0.015	1 02.09.2003	48	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 3 6 13	BASF Method 546/0
				0.0025 0.0025	600 600	0.015 0.015	2 02.09.2003	48	Heads Heads Heads Heads	<0.05 <0.05 <0.05 <0.05	0 3 6 13	BASF Method 546/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Head Cabbage/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Belgium, France, Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026852 UK - Banks, Lancashire, PR9 8DX (AF/8829/BA/1)	Head cabbage (Summer greens)	1. 15.07.2005 2. n. a. 3. 11.-25.10.2005	foliar spray with boom	0.003	400	0.0125	1 11.10.2005	43	Heads Heads Heads Heads	0.069 0.035 0.012 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 11.10.2005	43	Heads Heads Heads Heads	0.088 0.055 0.018 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026852 France - 49650 Allonnes (AF/8829/BA/2)	Head cabbage (Spitfire)	1. 07.09.2005 2. n. a. 3. 29.11.-13.12.2005	foliar spray with boom	0.003	400	0.0125	1 29.11.2005	47	Heads Heads Heads Heads	<0.01 0.012 <0.01 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 29.11.2005	47	Heads Heads Heads Heads	0.017 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026852 Germany - 30982 Pattensen Jeinsen (AF/8829/BA/4)	Head cabbage (Nula)	1. 15.07.2005 2. n. a. 3. 04.-18.11.2005	foliar spray with boom	0.003	400	0.0125	1 04.11.2005	48	Heads Heads Heads Heads	0.084 0.082 0.062 0.072	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 04.11.2005	48	Heads Heads Heads Heads	0.157 0.152 0.104 0.097	0 3 7 14	BASF Method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Head Cabbage/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Belgium, France, Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026852 UK - DE73 1DD Kings Newton, Derbyshire (AF/8829/BA/7)	Head cabbage (Wirosa F1)	1. 03.07.2005 2. n. a. 3. 29.11.-13.12.2005	foliar spray with boom	0.003	400	0.0125	1 29.11.2005	47	Heads Heads Heads Heads	0.015 0.030 <0.01 <0.01	0 3 7 14	BASF Method 567/0
				0.003 0.003	400 400	0.0125 0.0125	2 29.11.2005	47	Heads Heads Heads Heads	0.025 0.424 0.014 <0.01	0 3 7 14	BASF Method 567/0
DocID 2007/1013148 France - Sedan, Champagne-Ardennes (A/NF/I/06/143)	Head cabbage (Guisor)	1. 17.06.2006 2. not during trial 3. 02.10.2006	foliar spray with boom	0.003	400	0.012	1 18.09.2006	47	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 4 8 14	HPLC and LC-MS/MS Method LOQ = 0.01
				0.003 0.003	384 374	0.012 0.012	2 18.09.2006	47	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 4 8 14	HPLC and LC-MS/MS Method LOQ = 0.01
DocID 2007/1013148 Belgium - Lille, Antwerpen (A/BE/I/06/144)	Head cabbage (Galaxy)	1. 01.06.2006 2. not during trial 3. 09.10.2006	foliar spray with boom	0.003	400	0.013	1 25.09.2006	44-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01
				0.003 0.003	407 398	0.013 0.012	2 25.09.2006	43-44 44-45	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Head Cabbage/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Belgium, France, Germany, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1013148 UK - Chipping Campden, Gloucestershire (A/UK/I/06/145)	Head cabbage (Stonehead)	1. 01.04.2006 2. not during trial 3. 10.07.2006	foliar spray with boom	0.003	404	0.013	1 26.06.2006	45	Heads Heads Heads Heads	0.067 0.077 0.051 0.030	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01
				0.003 0.003	423 423	0.013 0.013	2 26.06.2006	43 45	Heads Heads Heads Heads	0.160 0.180 0.100 0.032	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01
DocID 2007/1013148 Germany - Pegau, Sachsen (A/GE/I/06/146)	Head cabbage (Kilatou)	1. 10.05.2006 2. not during trial 3. 02.10.2006	foliar spray with boom	0.003	411	0.013	1 18.09.2006	48	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01
				0.003 0.003	429 411	0.013 0.013	2 18.09.2006	48 48	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Head Cabbage/Brassica vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026852 France - 82170 Canals (AF/8829/BA/5)	Head cabbage (Castello)	1. 20.06.2005 2. n. a. 3. 16.-30.08.2005	foliar spray with boom	0.006	400	0.025	1 16.08.2005	47	Heads Heads Heads Heads	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method 567/0
DocID 2006/1026852 Italy - 40057 Viadagola (AF/8829/BA/6)	Head cabbage (Primero)	1. 05.08.2005 2. n. a. 3. 17.-31.10.2005	foliar spray with boom	0.006	400	0.025	1 17.10.2005	47-48	Heads Heads Heads Heads	0.015 0.013 <0.01 <0.01	0 3 7 14	BASF Method 567/0
DocID 2007/1013148 France - Chateaurenard, Bouche de Rhone (A/SF/I/06/147)	Head cabbage (Clarissa HF1)	1. 11.07.2006 2. not during trial 3. 27.09.2006	foliar spray with boom	0.006	417	0.026	1 13.09.2006	48	Heads Heads Heads Heads	0.045 0.010 <0.01 <0.01	0 3 7 14	HPLC and LC-MS/MS Method LOQ = 0.01
DocID 2007/1013148 Spain - Almussafes, Valencia (A/SP/I/06/148)	Head cabbage (Melissa)	1. 25.05.2006 2. not during trial 3. 08.08.2006	foliar spray with knapsack sprayer	0.006	392	0.024	1 26.07.2006	46	Heads Heads Heads Heads	0.010 0.010 0.013 <0.01	0 3 6 13	HPLC and LC-MS/MS Method LOQ = 0.01

n. a. not available

• Spinach

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Spinach/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Germany, The Netherlands	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007944 France - 49400 St. Lambert des Levees (AF/10498/BA/1)	Spinach (Laska)	1. 12.09.2006 2. n. a. 3. 21.11.-06.12.2006	foliar spray	0.003	400	0.0125	1 21.11.2006	49	Foliage Foliage Foliage Foliage	1.10 0.72 0.55 0.21	0 3 7 15	BASF Method No. 567/0
				0.003	400	0.0125	2 21.11.2006	49	Foliage Foliage Foliage Foliage	1.90 1.30 0.89 0.39	0 3 7 15	BASF Method No. 567/0
DocID 2007/1007944 Germany - Ippenstedterstr. 16, 0982 Patternsen-Jeinsen (AF/10498/BA/2)	Spinach (Elan)	1. 09.08.2006 2. n. a. 3. 13.-26.09.2006	foliar spray	0.003	400	0.0125	1 13.09.2006	35	Foliage Foliage Foliage Foliage	0.55 0.37 0.19 0.07	0 2 7 13	BASF Method No. 567/0
				0.003	400	0.0125	2 13.09.2006	35	Foliage Foliage Foliage Foliage	0.61 0.33 0.19 0.09	0 2 7 13	BASF Method No. 567/0
DocID 2006/1026849 The Netherlands - Kreupeleweg 49, 3286	Spinach (Monza)	1. 19.07.2005 2. n. a. 3. 16.-30.08.2006	foliar spray	0.003	400	0.0125	1 16.08.2005	13	Foliage Foliage Foliage	0.698 0.220	0 3	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Spinach/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Germany, The Netherlands	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
bb Klaaswaal (AF/8819/BA/1)									Foliage	0.179 0.037	7 14	
				0.003	400	0.0125	2 16.08.2005	13	Foliage Foliage Foliage Foliage	0.950 0.394 0.135 0.033	0 3 7 14	BASF Method No. 567/0
DocID 2008/1037135 France - 91160 Saulx les Chartreux (AF/10498/BA/6)	Spinach (Boeing)	1. 20.09.06 2. n. a. 3. 08.12.-22.12.2006	foliar application with boom	0.003	400	0.0125	1 08.12.2006	47-49	Foliage Foliage Foliage Foliage	0.69 0.81 0.71 0.84	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 08.12.2006	47-49	Foliage Foliage Foliage Foliage	1.1 1.4 1.1 0.96	0 3 7 14	BASF Method No. 567/0

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Spinach/leafy vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	Fastac SC Super Contact	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1035745 Germany - Dresden-Pilnitz (RU-I-18 06 SN DD 1/1)	Spinach (Puma F1)	1. 08.05.2006 2. n. a. 3. 22.06.2006	spraying	0.003 0.003	400 400	0.0125 0.0125	2 15.06.2006	BBCH 14-16	foliage	0.46	7	Method DFG S19
DocID 2007/1035745 Germany - 15328 Hathenow, Germany (RU-I-18 06 BB FO 1/1 (RF0906))	Spinach (Stamm 1146)	1. 07.04.2006 2. n. a. 3. 06.06.2006	spraying	0.004 0.004	293.4 306.6	0.0122 0.0128	2 30.05.2006	BBCH 37	foliage	0.47	7	Method DFG S19
DocID 2007/1035745 Germany - 48147 Münster (RU-I-18 06 NW BN 1/1)	Spinach (Rhino RZ)	1. 22.06.2006 2. n. a. 3. Aug 2006	spraying	0.002 0.002	600 600	0.0125 0.0125	2 01.08.2006	BBCH 17-18	foliage	0.36	7	Method DFG S19
DocID 2007/1035745 Germany - 53229 Bonn (RU-I-18 06 NW BN 1/2)	Spinach (Paso)	1. 08.08.2006 2. n. a. 3. September 2006	spraying	0.002 0.002	600 600	00125 0.0125	2 15.09.2006	BBCH 43-45	foliage	0.48	7	Method DFG S19

n. a. not available

• Green beans

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026857 UK - Woodfield Farm, Pershore, Birmingham WR10 3AG (AF/8825/BA/1)	Green beans (Paulista)	1. 15.05.2005 2. n. a. 3. 21.07.-04.08.2005	foliar spray	0.003	400	0.0125	1 21.07.2005	69-79	Beans with pods	0.052 0.028 0.021 0.011	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.519 0.249 0.138 0.034	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 21.07.2005	69-79	Beans with pods	0.048 0.027 0.026 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.394 0.132 0.087 0.055	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026857 France - Rue Gabriel Jeanton, 71700	Green bean (Booster)	1. 28.05.2005 2. n. a. 3. 19.07.-02.08.2005	foliar spray	0.003	400	0.0125	1 19.07.2005	77	Beans with pods	0.019 0.013 0.011 <0.01	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
Lacrost (AF/8825/BA/2)									Remaining plant	0.284 0.237 0.067 0.041	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 19.07.2005	77	Beans with pods	0.025 0.016 0.011 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.373 0.184 0.144 0.077	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026857 Germany - Biedensandstr. 32, 68623 Lampertheim (AF/8825/BA/3)	Green bean (Sigma)	1. 18.04.2005 2. n. a. 3. 22.07.-05.08.2005	foliar spray	0.003	400	0.0125	1 22.07.2005	78	Beans with pods	0.016 0.011 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.861 0.192 0.034 0.032	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 22.07.2005	78	Beans with pods	0.025 0.020 <0.01 0.014	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.816 0.253 0.077 0.092	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026857 Netherlands - Leembaan, 6595 MH Ottersum, Limburg (AF/8825/BA/4)	Green bean (Mention)	1. 08.06.2005 2. n. a. 3. 19.08.-02.09.2005	foliar spray	0.003	400	0.0125	1 19.08.2005	76-77	Beans with pods	0.017 0.014 0.031 0.020	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.341 0.131 0.128 0.071	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 19.08.2005	76-77	Beans with pods	0.023 0.031 0.015 0.014	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.339 0.305 0.177 0.107	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007950 France - Les Serres de Moulin a Vent, 49680 Le Moulin a Vent (AF/10492/BA/1)	Green bean (Angers)	1. 01.06.2006 2. n. a. 3. 28.07.-11.08.2006	foliar spray	0.003	400	0.0125	1 28.07.2006	78	Beans with pods	<0.01 <0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.42 0.08 0.07 0.04	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 28.07.2006	78	Beans with pods	<0.01 0.01 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.54 0.12 0.09 0.06	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007950 Netherlands - Zelder 1, 6595 NW Ottersum, Limburg (AF/10492/BA/2)	Green bean (Minidor)	1. 14.06.2006 2. n. a. 3. 22.08.-05.09.2006	foliar spray	0.003	400	0.0125	1 22.08.2006	76	Beans with pods	<0.01 0.01 0.01 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.25 0.15 0.08 0.08	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 22.08.2006	76	Beans with pods	0.02 0.01 0.02 0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.33 0.20 0.17 0.13	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007950 Belgium - Rue de la Chapelle, 96210 Villers-Perwin (AF/10492/BA/3)	Green bean (Polder)	1. 11.05.2006 2. n. a. 3. 25.07.-08.08.2006	foliar spray	0.003	400	0.0125	1 25.07.2006	77	Beans with pods	0.01 0.01 0.01 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.39 0.20 0.08 0.05	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 25.07.2006	77	Beans with pods	0.03 0.02 0.02 0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.36 0.34 0.13 0.07	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007950 UK - Woodfield Farm, Pershore, Birmingham WR10 3AG (AF/10492/BA/4)	Green bean (Paulifta)	1. n. a. 2. n. a. 3. 07.-21.09.2006	foliar spray	0.003	400	0.0125	1 07.09.2006	77-79	Beans with pods	0.02 0.02 0.01 <0.01	0 4 8 14	BASF Method No. 567/0
									Remaining plant	0.25 0.20 0.1 <0.01	0 4 8 14	BASF Method No. 567/0
				0.003	400	0.0125	2 07.09.2006	77-79	Beans with pods	0.02 0.02 0.02 <0.01	0 4 8 14	BASF Method No. 567/0
									Remaining plant	0.52 0.31 0.12 0.07	0 4 8 14	BASF Method No. 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026857 France - Les Saignes, 69610 Montromant (AF/8825/BA/5)	Green bean (Adagio)	1. 09.05.2005 2. n. a. 3. 26.07.-09.08.2005	foliar spray	0.006	400	0.025	1 26.07.2005	77	Beans with pods	0.047 0.028 0.017 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.504 0.256 0.105 0.050	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026857 Italy - Viabastia 6609, Lugo, 48022 Emilia Romagna (AF/8825/BA/6)	Green bean (Avalon)	1. 04.05.2005 2. n. a. 3. 05.-19.07.2005	foliar spray	0.006	400	0.025	1 05.07.2005	71-75	Beans with pods	0.054 0.063 0.033 0.021	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.888 0.796 0.305 0.160	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026857 Spain - Valtierra, 31320 Milagro (AF/8825/BA/7)	Green bean (Antea)	1. 20.07.2005 2. n. a. 3. 26.09.-10.10.2005	foliar spray	0.006	400	0.025	1 26.09.2005	72-73	Beans with pods	0.042 0.020 0.013 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.856 0.455 0.273 0.139	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026857 Greece - 56224 Thessaloniki (AF/8825/BA/8)	Green bean (Plati)	1. 15.08.2005 2. n. a. 3. 05.-19.09.2005	foliar spray	0.006	400	0.025	1 05.09.2005	78	Beans with pods	0.023 0.013 0.018 0.010	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.921 0.594 0.364 0.186	0 3 7 14	BASF Method No. 567/0
DocID 2007/1007950 France - 26 Rue Neuve, 31330 St. Caprais (AF/10492/BA/5)	Green bean (Booster)	1. 10.07.2006 2. n. a. 3. 05.-19.09.2006	foliar spray	0.006	400	0.025	1 05.09.2006	77-79	Beans with pods	0.06 0.05 <0.01 <0.01	0 3 8 14	BASF Method No. 567/0
									Remaining plant	1.50 0.98 0.04 0.02	0 3 8 14	BASF Method No. 567/0
DocID 2007/1007950 Spain - C/neuva 8, Melida, 31382 Navarra (AF/10492/BA/6)	Green bean (Aneto)	1. 28.07.2006 2. n. a. 3. 22.09.-06.10.2006	foliar spray	0.006	400	0.025	1 22.09.2006	77	Beans with pods	0.02 0.03 0.02 0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.91 0.63 0.43 0.24	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green beans/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007950 Italy - Az. Agr. Rilli, Via Boncellino 57, 48012 Bagnacavallo (RA) (AF/10492/BA/8)	Green bean (Masai)	1. 28.07.2006 2. n. a. 3. 25.09.-09.10.2006	foliar spray	0.006	400	0.025	1 25.09.2006	85-86	Beans with pods	0.02 0.04 0.02 <0.01	0 3 7 14	BASF Method No. 567/0
									Remaining plant	0.98 0.20 0.07 0.05	0 3 7 14	BASF Method No. 567/0

n. a. not available

• Green peas

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DK, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026856 France, Le Moulin Avent Vivy 49680 (AF/8826/BA/2)	Pea (Utrio)	1. 04.07.2005 2. n. a. 3. 30.08. - 13.09.2005	foliar spray	0.003	400	0.0125	1 30.08.2005	77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.07 <0.01 0.05 <0.01 0.06 <0.01 0.05	0 0 3 3 7 7 14 14	BASF Method No. 567/0
	Pea (Utrio)	1. 04.07.2005 2. n. a. 3. 30.08. - 13.09.2005	foliar spray	0.003	400	0.0125	2 30.08.2005	77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.10 <0.01 0.06 <0.01 0.08 <0.01 0.05	0 0 3 3 7 7 14 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DK, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026856 Germany - Hermannshof/Schafbruckenweg, 67126 Hochdorf-Assenheim (AF/8826/BA/3)	Pea (Maxigold)	1. 14.04.2005 2. n. a. 3. 21.06. - 05.07.2005	foliar spray	0.003	400	0.0125	1 21.06.2005	76	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.06 <0.01 0.04 <0.01 0.02 <0.01 0.01	0 0 3 3 7 7 14 14	BASF Method No. 567/0
	Pea (Maxigold)	1. 14.04.2005 2. n. a. 3. 21.06. - 05.07.2005	foliar spray	0.003	400	0.0125	2 21.06.2005	76	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.07 <0.01 0.06 <0.01 0.03 <0.01 0.01	0 0 3 3 7 7 14 14	BASF Method No. 567/0
DocID 2006/1026856 The Netherlands - Stuwweg, 6598 Heijen, Limburg (AF/8826/BA/4)	Pea (Arlette)	1. 20.05.2005 2. n. a. 3. 03. - 17.08.2005	foliar spray	0.003	400	0.0125	1 03.08.2005	78-79	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.83 <0.01 0.24 <0.01 0.25 <0.01 0.19	0 0 3 3 7 7 14 14	BASF Method No. 567/0
	Pea (Arlette)	1. 20.05.2005 2. n. a. 3. 03. - 17.08.2005	foliar spray	0.003	400	0.0125	2 03.08.2005	78-79	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.86 <0.01 0.35 <0.01 0.23 <0.01 0.34	0 0 3 3 7 7 14 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DK, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026856 UK - Bishop Burton, Yorkshire HU17 8QA (AF/8826/BA/7)	Pea (Wavarex)	1. 03.05.2005 2. n. a. 3. 27.07. - 10.08.2005	foliar spray	0.003	400	0.0125	1 27.07.2005	76-77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.35 <0.01 0.06 <0.01 0.10 <0.01 0.10	0 0 3 3 7 7 14 14	BASF Method No. 567/0
	Pea (Wavarex)	1. 03.05.2005 2. n. a. 3. 27.07. - 10.08.2005	foliar spray	0.003	400	0.0125	2 27.07.2005	76-77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.42 <0.01 0.21 <0.01 0.30 <0.01 0.29	0 0 3 3 7 7 14 14	BASF Method No. 567/0
DocID 2007/1007951 France - Clery St Andre, 45370 (AF/10491/BA/1)	Pea (Merveille de Kelvedon)	1. 12.04.2006 2. n. a. 3. 19.06. - 03.07.2006	foliar spray	0.003	400	0.0125	1 19.06.2006	77-79	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.159 <0.01 0.037 <0.01 0.047 <0.01 0.033	0 0 3 3 7 7 14 14	BASF Method No. 567/0
	Pea (Merveille de Kelvedon)	1. 12.04.2006 2. n. a. 3. 19.06. - 03.07.2006	foliar spray	0.003	400	0.0125	2 19.06.2006	77-79	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.225 <0.01 0.057 <0.01 0.104 <0.01 0.068	0 0 3 3 7 7 14 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DK, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007951 Denmark - Rojleskovvej 18, DK-5500 Middelfart (AF/10491/BA/2)	Pea (Progress No. 9)	1. 28.04.2006 2. n. a. 3. 12. - 26.07.2006	foliar spray	0.003	400	0.0125	1 12.07.2006	77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.514 <0.01 0.361 <0.01 0.227 <0.01 0.281	0 0 2 2 7 7 14 14	BASF Method No. 567/0
	Pea (Progress No. 9)	1. 28.04.2006 2. n. a. 3. 12. - 26.07.2006	foliar spray	0.003	400	0.0125	2 12.07.2006	77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.333 <0.01 0.430 <0.01 0.200 <0.01 0.166	0 0 2 2 7 7 14 14	BASF Method No. 567/0
DocID 2007/1007951 UK - Holbeach Hurn, Lincolnshire P§12 8LR (AF/10491/BA/3)	Pea (Geisha)	1. 07.06.2006 2. n. a. 3. 31.07. - 14.08.2006	foliar spray	0.003	400	0.0125	1 31.07.2006	77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.254 <0.01 0.087 <0.01 0.062 <0.01 0.066	0 0 3 3 7 7 14 14	BASF Method No. 567/0
	Pea (Geisha)	1. 07.06.2006 2. n. a. 3. 31.07. - 14.08.2006	foliar spray	0.003	400	0.0125	2 31.07.2006	77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.275 <0.01 0.190 <0.01 0.199 <0.01 0.226	0 0 3 3 7 7 14 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DK, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007951 Germany - Vor den Hofen, 31303, Burgdorf-Hulptingsen, Lower Saxony (AF/10491/BA/4)	Pea (Santana)	1. 10.08.2006 2. n. a. 3. 26.10. - 09.11.2006	foliar spray	0.003	400	0.0125	1 26.10.2006	71	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.289 <0.01 0.266 <0.01 0.185 <0.01 0.180	0 0 4 4 7 7 14 14	BASF Method No. 567/0
	Pea (Santana)	1. 10.08.2006 2. n. a. 3. 26.10. - 09.11.2006	foliar spray	0.003	400	0.0125	2 26.10.2006	71	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.636 <0.01 0.230 <0.01 0.346 <0.01 0.259	0 0 4 4 7 7 14 14	BASF Method No. 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026856 Italy - San Benedetto, S. Pietro in Casale, 40018 Bologna (AF/8826/BA/5)	Pea (Atlas)	1. 25.03.2005 2. n. a. 3. 20.06. - 04.07.2005	foliar spray	0.0060	400	0.025	1 20.06.2005	77-79	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.24 <0.01 0.27 <0.01 0.09 <0.01 0.10	0 0 3 3 7 7 14 14	BASF Method No. 567/0
DocID 2006/1026856 Greece - Komaras, Evros, Thrace, GR-68200 (AF/8826/BA/6)	Pea (Ambass adeur)	1. 10.04.2005 2. n. a. 3. 24.06. - 08.07.2005	foliar spray	0.0060	400	0.025	1 24.06.2005	74	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.57 <0.01 0.20 <0.01 0.28 <0.01 0.26	0 0 3 3 7 7 14 14	BASF Method No. 567/0
DocID 2007/1007951 France - St Nicolas de la Grave 82210.000 (AF/10491/BA/5)	Pea (Milan)	1. 14.02.2006 2. n. a. 3. 22.05. - 05.06.2006	foliar spray	0.0060	400	0.025	1 22.05.2006	69-71	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 0.721 <0.01 0.165 <0.01 0.068 <0.01 0.140	0 0 2 2 7 7 14 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Green Peas/legume vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007951 Spain - Castillo Pompien Monflorite Lascasas 22196 (AF/8826/BA/6)	Pea (Meteor)	1. 17.01.2006 2. n. a. 3. 01. - 15.06.2006	foliar spray	0.0060	400	0.025	1 01.06.2006	76-77	peas rest of plant peas rest of plant peas rest of plant peas rest of plant	<0.01 1.023 <0.01 0.431 <0.01 0.297 <0.01 0.071	0 0 4 4 7 7 14 14	BASF Method No. 567/0

n. a. not available

• Celery

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Celery/stem vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Germany	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (Fastac SC super contact)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1029533 Germany -Schifferstadt (RU-I-16 05 RP NW 2/1)	Celery (Tango)	1. 04.05.2005 2. n. a. 3. 08.08.2005	spraying	0.002 0.002	630 590	0.012 0.013	2 01.08.2005	n. a.	stem stem stem stem stem	0.2120 < LOD 0.1443 0.1360 0.0658	0 7 7 10 14	BASF Method No. 567/0
DocID 2006/1029533 Germany -Schifferstadt (RU-I-16 05 RP NW 2/2)	Celery (Tango)	1. 04.05.2005 2. n. a. 3. 01.08.2005	spraying	0.002 0.002	640 630	0.013 0.013	2 25.07.2005	n. a.	stem stem stem	0.3381 01822 0.1212 0.1045	0 7 10 14	BASF Method No. 567/0
DocID 2007/1035745 Germany - 67105 Schifferstadt (RU-I-17 06 RP NW 1/1)	Celery (Tango)	1. 27.04.2006 2. n. a. 3. 25.07.2006	spraying	0.003 0.003	424 432	0.013 0.014	2 18.07.2006	n. a.	stem stem	0.30 0.17	7 14	Method DFG S 19
DocID 2007/1035745 Germany - 67105 Schifferstadt (RU-I-17 06 RP NW 1/2)	Celery (Tango)	1. 21.06.2006 2. n. a. 3. 12.09.2006	spraying	0.003 0.002	438 640	0.014 0.013	2 05.09.2006	n. a.	stem stem	0.23 0.13	7 14	Method DFG S 19

n. a. not available, LOD= 0.001 and 0.0036 mg/kg

• Artichoke

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Artichoke/stem vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026845 Spain, 21002, Finca la Hincosa, Camino Bollullos, Bollullos del Candado (AF/8828/BA/1)	Artichoke (Chata de Tudela)	1. 20.08.2005 2. n. a. 3. 13. - 27.12.2005	foliar spray	0.0060	400	0.025	1 13.12.2005	51	flower head flower head flower head flower head	0.038 0.019 0.021 0.012	0 3 7 14	analytical method used: BASF method 576/0
DocID 2006/1026845 Italy, 57020, Ag Agr Pistolesi, Podere Pappasous, Via Aurelia, Toscana (AF/8828/BA/2)	Artichoke (Violetto Toscano)	1. 04.2003 2. n. a. 3. 29.04. - 13.05.2005	foliar spray	0.0060	400	0.025	1 29.04.2005	47	flower head flower head flower head flower head	0..06 0.06 0.04 0.014	0 3 7 14	analytical method used: BASF method 576/0
DocID 2007/1007948 France - 31330 Garosses, St Caprais, Darles (AF/10494/BA/1)	Artichoke (Macau)	1. 2004 2. n. a. 3. 11. - 25.05.2006	foliar spray	0.0060	400	0.025	1 11.05.2006	47	flower head flower head flower head flower head	0.04 0.03 0.02 0.01	0 3 7 13	analytical mehod used: BASF method 567/0
DocID 2007/1007948 Greece - Central Greece, GR-19007 Attiki, Kato Souli (AF/10494/BA/2)	Wild artichoke (variety unknown)	1. 08.2005 2. n. a. 3. 05. - 19.05.2006	foliar spray	0.0060	400	0.025	1 05.05.2006	47-49	flower head flower head flower head flower head	0.04 0.04 0.03 <0.01	0 3 7 14	analytical mehod used: BASF method 567/0

n. a. not available

- **Leek**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Leek/stem vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026850 UK - Ravenshead, Nottinghamshire NG5 8PB (AF/8821/BA/1)	Leek (Shelton)	1. 07.05.2005 2. n. a. 3. 04.10-18.10.2005	foliar spray with boom	0.003	400	0.0125	1 04.10.2005	43	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.183 0.078 0.047 0.055	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 04.10.2005	43	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.144 0.082 0.056 0.059	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026850 France - Dampierre en Burly 45730 (AF/8821/BA/2)	Leek (Ventura)	1. 12.07.2005 2. n. a. 3. 27.09.-11.10.2005	foliar spray with boom	0.003	400	0.0125	1 27.09.2005	47-49	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.059 0.052 0.037 0.017	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 27.09.2005	47-49	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.083 0.070 0.053 0.034	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026850 Germany - Hinter dem Munchhof, 67125 Dannstadt (AF/8821/BA/3)	Leek (Teuton)	1. 27.07.2005 2. n. a. 3. 26.04.-10.05.2006	foliar spray with boom	0.003	400	0.0125	1 26.04.2005	49	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.056 0.055 0.046 0.033	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 26.04.2005	49	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.084 0.069 0.071 0.064	0 3 7 14	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Leek/stem vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026850 Netherlands - Ploegstraat, 5375 KR Reek, Brabant (AF/8821/BA/4)	Leek (Kenton)	1. 10.07.2005 2. n. a. 3. 04.10.-18.10.2005	foliar spray with boom	0.003	400	0.0125	1 04.10.2005	44	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.026 0.026 0.014 0.014	0 3 7 14	BASF Method No. 567/0
				0.003	400	0.0125	2 04.10.2005	44	Plant w/o root Plant w/o root Plant w/o root Plant w/o root	0.055 0.031 0.033 0.021	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008498 France - Mardeuil, Marne (A/NF/I/06/155)	Leek (Pasteur)	1. 07.06.2006 2. n. a. 3. 13.09.2006	foliar spray with boom	0.003	438	0.0125	1 31.08.2006	45	Whole plant Whole plant Whole plant Whole plant	0.026 0.024 0.016 <0.01	0 3 7 13	BASF Method No. 567/0
				0.003 0.003	406 408	0.0125 0.0125	2 31.08.2006	45 45	Whole plant Whole plant Whole plant Whole plant	0.032 0.029 0.018 0.010	0 3 7 13	BASF Method No. 567/0
DocID 2007/1008498 UK - Pershore, Worcestershire (A/UK/I/06/156)	Leek (Pancho)	1. 22.03.2006 2. n. a. 3. 26.09.2006	foliar spray with boom	0.003	425	0.0125	1 11.09.2006	45	Whole plant Whole plant Whole plant Whole plant	0.063 0.052 0.037 <0.01	0 4 7 15	BASF Method No. 567/0
				0.003 0.003	405 416	0.0125 0.0125	2 11.09.2006	44 45	Whole plant Whole plant Whole plant Whole plant	0.130 0.078 0.048 <0.01	0 4 7 15	BASF Method No. 567/0
DocID 2007/1008498 Belgium - Sint-Kateljine-Waver, Antwerpen (A/BE/I/06/157)	Leek (Ashton)	1. 23.06.2006 2. n. a. 3. 09.10.2006	foliar spray with boom	0.003	397	0.012	1 25.09.2006	44-45	Whole plant Whole plant Whole plant Whole plant	0.021 0.029 0.020 0.015	0 3 7 14	BASF Method No. 567/0
				0.003 0.003	387 390	0.012 0.012	2 25.09.2006	43-44 44-45	Whole plant Whole plant Whole plant	0.048 0.038	0 3	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Leek/stem vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	BE, DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
									Whole plant	0.039 0.026	7 14	
DocID 2007/1008498 Germany - Erfurt, Thueringen (A/GE/I06/158)	Leek (Kenton)	1. 02.05.2006 2. n. a. 3. 13.11.2006	foliar spray with boom	0.003	424	0.013	1 30.10.2006	48	Whole plant Whole plant Whole plant Whole plant	0.018 0.025 0.023 0.017	0 2 7 14	BASF Method No. 567/0
				0.003 0.003	431 424	0.014 0.013	2 30.10.2006	48 48	Whole plant Whole plant Whole plant Whole plant	0.042 0.036 0.032 0.032	0 2 7 14	BASF Method No. 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Leek/stem vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026850 France - Seilh 31840 (AF/8821/BA/5)	Leek (Pegasus)	1. 19.07.05 2. n. a. 3. 10.10.-24.10.05	foliar spray with boom	0.006	400	0.0250	1 10.10.2005	47	Whole plants without roots	0.093 0.062 0.045 0.030	0 3 7 14	BASF Method No. 567/0
DocID 2006/1026850 Italy - Lusia, Rovigo 45020 (AF/8821/BA/6)	Leek (Armor)	1. 20.05.05 2. n. a. 3. 21.09.-06.10.05	foliar spray with boom	0.006	400	0.0250	1 21.09.2005	47	Whole plants without roots	0.149 0.106 0.105 0.054	0 2 7 15	BASF Method No. 567/0
DocID 2007/1008498 Spain - Masanasa, Valencia (A/SP/I/06/159)	Leek (Stal)	1. 01.06.2006 2. n. a. 3. 13.10.2006	foliar spray with knapsack sprayer	0.006	420	0.027	1 29.09.2006	45	Whole plant	0.042 0.017 0.011 0.014	0 3 7 14	BASF Method No. 567/0
DocID 2007/1008498 France - Noves, Bouches de Rhone (A/SF/I/06/160)	Leek (Selecta)	1. 06.03.06/05.07.06 2. n. a. 3. 25.09.2006	foliar spray with boom	0.006	412	0.026	1 11.09.2006	47	Whole plant	0.140 0.032 <0.01 <0.01	0 3 7 14	BASF Method No. 567/0

n. a. not available

• Pulses

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026858 UK - Smithy Farm, Main Road, Morley, Ikeston, DE7 6DF Derbyshire (AF/8824/BA/1)	Dry pea (Kobleackie)	1. 15.03.2005 2. n. a. 3. 15.07. - 12.08.2005	foliar spray	0.003	400	0.0125	1 15.07.2005	75-77	seed straw seed straw seed straw seed straw	<0.01 0.190 <0.01 0.122 <0.01 0.251 <0.01 0.274	0 0 14 14 21 21 28 28	BASF Method No. 567/0
	Dry pea (Kobleackie)	1. 15.03.2005 2. n. a. 3. 15.07. - 12.08.2005	foliar spray	0.003	400	0.0125	2 15.07.2005	75-77	seed straw seed straw seed straw seed straw	<0.01 0.550 <0.01 0.439 <0.01 0.524 <0.01 0.519	0 0 14 14 21 21 28 28	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026858 France - 10 Rue d'Enjanville, Rouvres St Jean, 45300 (AF/8824/BA/2)	Dry pea (Hardy)	1. 19.03.2005 2. n. a. 3. 21.06. - 19.07.2005	foliar spray	0.003	400	0.0125	1 21.06.2005	79	seed straw seed straw seed straw seed straw	<0.01 0.121 <0.01 0.119 <0.01 0.237 <0.01 0.203	0 0 14 14 21 21 28 28	BASF Method No. 567/0
	Dry pea (Hardy)	1. 19.03.2005 2. n. a. 3. 21.06. - 19.07.2005	foliar spray	0.003	400	0.0125	2 15.07.2005	79	seed straw seed straw seed straw seed straw	<0.01 0.209 <0.01 0.299 <0.01 0.365 <0.01 0.362	0 0 14 14 21 21 28 28	BASF Method No. 567/0
DocID 2006/1026858 UK - Peter Avey, Chapel Lane, Aslockton, Nottingham NG13 9AR (AF/8824/BA/5)	Dry bean (Clipper)	1. n. a. 2. n. a. 3. 09.07. - 26.08.2005	foliar spray	0.003	400	0.0125	1 29.07.2005	83	seed straw seed straw seed straw seed straw	<0.01 0.167 <0.01 0.310 <0.01 0.158 <0.01 0.089	0 0 14 14 21 21 28 28	BASF Method No. 567/0
	Dry bean (Clipper)	1. n. a. 2. n. a. 3. 09.07. - 26.08.2005	foliar spray	0.003	400	0.0125	2 29.07.2005	83	seed straw seed straw seed straw seed straw	<0.01 0.278 <0.01 0.295 <0.01 0.396 <0.01 0.109	0 0 14 14 21 21 28 28	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026858 France - Mavry, St Georges de layon, 49700 (AF/8824/BA/6)	Dry bean (Castel)	1. 15.11.2005 2. n. a. 3. 27.06. - 25.07.2005	foliar spray	0.003	400	0.0125	1 27.06.2005	86	seed straw seed straw seed straw seed straw	<0.01 0.436 <0.01 0.205 <0.01 0.236 <0.01 0.210	0 0 14 14 21 21 28 28	BASF Method No. 567/0
	Dry bean (Castel)	1. 15.11.2005 2. n. a. 3. 27.06. - 25.07.2005	foliar spray	0.003	400	0.0125	2 27.06.2005	86	seed straw seed straw seed straw seed straw	<0.01 0.574 <0.01 0.459 <0.01 0.453 <0.01 0.593	0 0 14 14 21 21 28 28	BASF Method No. 567/0
DocID 2007/1007949 France - 1 Rue d'Enzanville, 45300 Rouevres-St Jean (AF/10493/BA/1)	Dry pea (Arthur)	1. 20.03.2006 2. n. a. 3. 29.06. - 27.07.2006	foliar spray	0.003	400	0.0125	1 29.06.2006	85-87	seed straw seed straw seed straw seed straw	<0.01 0.127 <0.01 0.246 <0.01 0.577 <0.01 0.295	0 0 14 14 21 21 28 28	analytical method used: BASF method 567/0
	Dry pea (Arthur)	1. 20.03.2006 2. n. a. 3. 29.06. - 27.07.2006	foliar spray	0.003	400	0.0125	2 29.06.2006	85-87	seed straw seed straw seed straw seed straw	<0.01 0.305 <0.01 0.249 <0.01 1.007 <0.01 0.665	0 0 14 14 21 21 28 28	analytical method used: BASF method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007949 Germany - Vor den Hofen, 31303 Burhdorf-Hulptsingen, Lower Saxony (AF/10493/BA/2)	Dry pea (Santana)	1. 10.08.2006 2. n. a. 3. 27.10. - 24.11.2006	foliar spray	0.003	400	0.0125	1 27.10.2006	71	seed straw seed straw seed straw seed straw	<0.01 0.345 <0.01 0.244 <0.01 0.249 <0.01 0.212	0 0 13 13 20 20 28 28	analytical method used: BASF method 567/0
	Dry pea (Santana)	1. 10.08.2006 2. n. a. 3. 27.10. - 24.11.2006	foliar spray	0.003	400	0.0125	2 27.10.2006	71	seed straw seed straw seed straw seed straw	<0.01 0.389 <0.01 0.376 <0.01 0.268 <0.01 0.332	0 0 13 13 20 20 28 28	analytical method used: BASF method 567/0
DocID 2007/1007949 The Netherlands - Lingestraat, 6662 NN Elst, Gelderland (AF/10493/BA/6)	Dry bean (Cebeco)	1. 21.03.2006 2. n. a. 3. 11.07. - 08.08.2006	foliar spray	0.003	400	0.0125	1 11.07.2006	73	seed straw seed straw seed straw seed straw	<0.01 0.488 <0.01 0.107 <0.01 0.077 <0.01 0.050	0 0 14 14 22 22 28 28	analytical method used: BASF method 567/0
	Dry bean (Cebeco)	1. 21.03.2006 2. n. a. 3. 11.07. - 08.08.2006	foliar spray	0.003	400	0.0125	2 11.07.2006	73	seed straw seed straw seed straw seed straw	<0.01 0.638 <0.01 0.167 <0.01 0.075 <0.01 0.101	0 0 14 14 22 22 28 28	analytical method used: BASF method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	DE, FR, NL, UK	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007949 France - Gaec du Pre de Chene, Boce 49150 (AF/10493/BA/9)	Dry bean (Irena)	1. 10.11.2005 2. n. a. 3. 29.06. - 27.07.2006	foliar spray	0.003	400	0.0125	1 29.06.2006	81	seed straw seed straw seed straw seed straw	<0.01 0.584 <0.01 0.079 <0.01 0.107 <0.01 0.084	0 0 14 14 21 21 28 28	analytical method used: BASF method 567/0
	Dry bean (Irena)	1. 10.11.2005 2. n. a. 3. 29.06. - 27.07.2006	foliar spray	0.003	400	0.0125	2 29.06.2006	81	seed straw seed straw seed straw seed straw	<0.01 0.536 <0.01 0.095 <0.01 0.182 <0.01 0.203	0 0 14 14 21 21 28 28	analytical method used: BASF method 567/0

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026858 Spain - Rey de los Moros, Cortes, 31530 (AF/8824/BA/3)	Dry pea (Ideal)	1. 09.03.2005 2. n. a. 3. 01. - 22.06.2005	foliar spray	0.006	400	0.025	1 01.06.2005	73-74	seed straw seed straw seed straw	<0.01 0.230 <0.01 0.323 <0.01 0.673	0 0 14 14 21 21	BASF Method No. 567/0
DocID 2006/1026858 Italy - Az Agr Cantaglia, Via Ponticelli 2, Pepola di Malalbergo 40058 (AF/8824/BA/4)	Dry pea (Coralio)	1. 03.04.2005 2. n. a. 3. 22.06. - 20.07.2005	foliar spray	0.006	400	0.025	1 22.06.2005	87-89	seed straw seed straw seed straw seed straw	<0.01 1.213 <0.01 0.154 0.022 0.477 0.040 0.355	0 0 14 14 21 21 28 28	BASF Method No. 567/0
DocID 2006/1026858 Spain - Rey de los Moros, Cortes, 31530 (AF/8824/BA/7)	Dry bean (Agua Dulce)	1. 10.10.2004 2. n. a. 3. 01.06. - 29.06.2005	foliar spray	0.006	400	0.025	1 01.06.2005	79	seed straw seed straw seed straw seed straw	<0.01 0.490 <0.01 0.095 <0.01 0.077 <0.01 0.092	0 0 14 14 21 21 28 28	BASF Method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026858 Italy - Az Agr Cantaglia, Via Ponticelli 2, Pepola di Malalbergo 40058 (AF/8824/BA/8)	Dry bean (Polo)	1. 12.04.2005 2. n. a. 3. 13.06. - 11.07.2005	foliar spray	0.006	400	0.025	1 13.06.2005	87	seed straw seed straw seed straw seed straw	<0.01 0.760 <0.01 0.289 <0.01 0.153 <0.01 0.265	0 0 14 14 21 21 28 28	BASF Method No. 567/0
DocID 2007/1007949 France - St Martin, Castelsarrasin, 82100 (AF/10493/BA/3)	Dry pea (Baceaua)	1. 15.01.2006 2. n. a. 3. 06. - 27.06.2006	foliar spray	0.006	400	0.025	1 06.06.2006	87	seed straw seed straw seed straw	<0.01 1.503 <0.01 0.849 <0.01 0.997	0 0 13 13 21 21	analytical method used: BASF method 567/0
DocID 2007/1007949 Greece - Apollonia, Thessaloniki, GR-57015 Central Macedonia (AF/10493/BA/4)	Dry pea (Lotus)	1. 10.04.2006 2. n. a. 3. 26.05. - 22.06.2006	foliar spray	0.006	400	0.025	1 26.05.2006	87-89	seed straw seed straw seed straw seed straw	<0.01 1.097 <0.01 0.411 <0.01 0.290 <0.01 0.505	0 0 14 14 21 21 27 27	analytical method used: BASF method 567/0
DocID 2007/1007949 Spain - Plaza Major, Cortes, 31530 (AF/10493/BA/8)	Dry bean (Reina Blanca)	1. 25.10.2005 2. n. a. 3. 01. - 29.06.2006	foliar spray	0.006	400	0.025	1 01.06.2006	93	seed straw seed straw seed straw seed straw	<0.01 0.731 <0.01 0.424 <0.01 0.245 <0.01 0.261	0 0 14 14 21 21 28 28	analytical method used: BASF method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	dry beans and peas/Pulses	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007949 France - La Guille, 11150 Villasavary (AF/10493/BA/11)	Dry bean (Linex)	1. 27.05.2006 2. n. a. 3. 02. - 30.08.2006	foliar spray	0.006	400	0.025	2 02.08.2006	79	seed straw seed straw seed straw seed straw	<0.01 1.077 <0.01 0.107 <0.01 0.057 <0.01 0.045	0 0 14 14 21 21 28 28	analytical method used: BASF method 567/0

n. a. not available

• Cotton

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cotton/Oilseeds	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007583 Spain - E-41729 Trajano, Sevilla (ALO/06/04)	Cotton (Hermes)	1. 06.05.2004 2. 20.07. - 20.08.2004 3. 15. & 16.10.2004	foliar spray	0.0050	300	0.01500	1 29.09.2004	85	seeds plants seeds plants seeds plants	<0.01 0.37 <0.01 0.21 <0.01 0.05	0 0 6 6 14 14	analytical method used: BASF method 567/0
	Cotton (Hermes)	1. 06.05.2004 2. 20.07. - 3. 20.08.2004 15. & 16.10.2004	foliar spray	0.0050	300	0.01500	2 29.09.2004	85	seeds plants seeds plants seeds plants	<0.01 0.49 <0.01 0.32 <0.01 0.05	0 0 6 6 14 14	analytical method used: BASF method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cotton/Oilseeds	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 41 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2005/1007583 Greece - G-59100 Imathia, Veria (GRE/01/04)	Cotton (Sandra)	1. 15.05.2004 2. 17.06. - 15.08.2004 3. 20.10. - 15.11.2004	foliar spray	0.0050	300	0.01500	1 18.10.2004	86	seeds plants seeds plants seeds plants	0.02 0.06 <0.01 0.06 <0.01 0.07	0 0 6 6 14 14	analytical method used: BASF method 567/0
	Cotton (Sandra)	1. 15.05.2004 2. 17.06. - 15.08.2004 3. 10.20. - 15.11.2004	foliar spray	0.0050	300	0.01500	2 18.10.2004	86	seeds plants seeds plants seeds plants	0.04 0.09 <0.01 0.11 0.01 0.2	0 0 6 6 14 14	analytical method used: BASF method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cotton/Oilseeds	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026847 Spain - 41740 Lebrija, Seville (AF/8822/BA/1)	Cotton (Flroa)	1. 20.03.2005 2. n. a. 3. 26.09. - 10.10.2005	foliar spray	0.0060	200	0.02500	1 29.06.2005	85	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	0.011 0.704 0.01 0.203 <0.01 0.196 <0.01 0..078	0 0 3 3 7 7 14 14	analytical method used: BASF method 567/0
DocID 2006/1026847 Spain - 41100 Coria del Rio, La Vega de Coria, Seville (AF/8822/BA/2)	Cotton (Pandora)	1. 15.03.2005 2. n. a. 3. 27.09. - 10.10.2005	foliar spray	0.0060	200	0.02500	1 27.09.2005	86	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	0.011 0.781 <0.01 0.279 <0.01 0.455 0.018 0.194	0 0 2 2 7 7 13 13	analytical method used: BASF method 567/0
DocID 2006/1026847 Spain - 41820 Seville, La Base, Dos Hermanas (AF/8822/BA/3)	Cotton (Flora)	1. 27.03.2005 2. n. a. 3. 04. - 18.10.2005	foliar spray	0.0060	200	0.02500	1 04.10.2005	85	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	<0.01 0.274 <0.01 0.382 <0.01 0.153 <0.01 0.143	0 0 3 3 6 6 14 14	analytical method used: BASF method 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cotton/Oilseeds	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2006/1026847 Gefyra, Thessaloniki, Central Macedonia (AF/8822/BA/4)	Cotton (Velos)	1. 19.05.2005 2. n. a. 3. 07. - 21.10.2005	foliar spray	0.0060	200	0.02500	1 07.10.2005	87	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	0.013 0.986 <0.01 0.359 <0.01 0.548 <0.01 0.187	0 0 3 3 7 7 14 14	analytical method used: BASF method 567/0
DocID 2007/1007947 Greece, GR-59032, Platanos , Imathia, Central Macedonia (AF/10495/BA/1)	Cotton (Ermis)	1. 28.05.2006 2. n. a. 3. 13. - 26.10.2006	foliar spray	0.0125	200	0.02500	1 13.10.2006	86-87	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	<0.01 0.84 <0.01 0.61 <0.01 0.38 <0.01 0.2	0 0 3 3 7 7 13 13	analytical method used: BASF method No. 567/0
DocID 2007/1007947 Greece, GR-57011 - Gefyra, Thessaloniki, Central Macedonia (AF/10495/BA/2)	Cotton (Ermis)	1. 20.05.2006 2. n. a. 3. 13. - 26.10.2006	foliar spray	0.0125	200	0.02500	1 13.10.2006	86-87	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	<0.01 0.57 <0.01 0.94 <0.01 0.34 <0.01 0.38	0 0 3 3 7 7 13 13	analytical method used: BASF method No. 567/0

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Cotton/Oilseeds	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	outdoor
Country	Greece, Spain	**Other active substance in the formulation**	none
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth stage at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
DocID 2007/1007947 Spain - 41220 Sevilla, Burguillos, Finca El Vergel (AF/10495/BA/3)	Cotton (Viky)	1. 04.2006 2. n. a. 3. 26.09. - 10.10.2006	foliar spray	0.0125	200	0.02500	1 26.09.2006	85-86	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	0.02 1.1 <0.01 0.75 <0.01 0.48 <0.01 0.38	0 0 3 3 7 7 13 13	analytical method used: BASF method No. 567/0
DocID 2007/1007947 Spain - 11640 Cadiz, C/Guadalete, 16, Coto de Bornos (Bornos) (AF/10495/BA/4)	Cotton (Celia)	1. 18.04.2006 2. n. a. 3. 25.08. - 09.10.2006	foliar spray	0.0125	200	0.02500	1 25.08.2006	86	seeds rest of plant seeds rest of plant seeds rest of plant seeds rest of plant	<0.01 0.58 <0.01 0.34 <0.01 0.21 <0.01 0.15	0 0 3 3 7 7 14 14	analytical method used: BASF method No. 567/0

n. a. not available

- ***Maize***

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	WL85871 (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-730-013 France (N) - 28 - Chateaudun (W/FR/E81/883)	maize (Fontenac)	1. 20.04.1981 2. n. a. 3. 19.10.1981	spray	n. a.	n. a.	0.0300	1 16.07.1981	30-39? ("height 1.3 m")	plant grain	<0.01 <0.01	14	analytical method used: SAM 233-1 LOQ 0.01 mg/kg
	maize (Fontenac)	1. 20.04.1981 2. n. a. 3. 19.10.1981	spray	n. a.	n. a.	0.0500	1 16.07.1981	30-39? ("height 1.3 m")	plant grain	<0.01 <0.01	14	

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	- (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-730-058 Germany - Bensheim (R31-86)	maize (Limac)	1. 02.05.1986 2. n. a. 3. 07.10.1986	spray	0.0044	400	0.0175	1 06.08.1986	n. a.	whole plant whole plant whole plant whole plant cob straw	0.4 0.25 0.12 0.07 <0.01 0.05	0 7 14 21 61 61	analytical method used: SAMS 383-1 LOQ 0.01 mg/kg
AL-730-057 Germany - Groß-Gerau (R32-86)	maize (Limac)	1. 04.05.1986 2. n. a. 3. 01.10.1986	spray	0.0044	400	0.0175	1 05.08.1986	n. a.	whole plant whole plant whole plant whole plant cob straw	0.52 0.16 0.33 0.1 <0.01 0.08	0 7 14 21 57 57	analytical method used: SAMS 383-1 LOQ 0.01 mg/kg
AL-730-059 Germany - Seligenstadt (R33-86)	maize (Limac)	1. 04.05.1986 2. n. a. 3. 03.10.1986	spray	0.004	400	0.018	1 05.08.1986	n. a.	whole plant whole plant whole plant whole plant cob straw	0.14 0.1 0.18 0.14 <0.01 0.08	0 7 14 21 59 59	analytical method used: SAMS 383-1 LOQ 0.01 mg/kg
AL-730-060 Germany - Bad Segeberg (R34-86)	maize (Limac)	1. 30.04.1986 2. n. a. 3. 27.09.1986	spray	0.0044	400	0.0175	1 30.07.1986	n. a.	whole plant whole plant whole plant whole plant cob straw	1.19 0.86 0.32 0.15 <0.01 0.23	0 7 14 21 59 59	analytical method used: SAMS 383-1 LOQ 0.01 mg/kg

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	Fastac (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-730-062 Germany - Bensheim (R94-87)	maize (Limac)	1. 04.05.1987 2 n. a. 3. 08.10.1987	spray	0.0044	400	0.0175	1 06.08.1987	57	whole plant whole plant whole plant whole plant grain	0.02 0.01 0.01 0.02 <0.01	0 7 14 21 63	analytical method used: SAMS 351-1 LOQ 0.01 mg/kg
AL-730-063 Germany - Groß-Gerau (R95-87)	maize (Limac)	1. 06.05.1987 2. n. a. 3. 30.09.1987	spray	0.0044	400	0.0175	1 05.08.1987	57	whole plant whole plant whole plant whole plant grain	0.83 0.2 0.29 0.29 <0.01	0 7 14 21 56	analytical method used: SAMS 351-1 LOQ 0.01 mg/kg
AL-730-064 Germany - Billingsdorf/Freising (R96-87)	maize (Tau)	1. 24.04.1987 2. n. a. 3. 09.10.1987	spray	0.0044	400	0.0175	1 10.08.1987	57	whole plant whole plant whole plant whole plant grain	1 0.38 0.11 0.18 <0.01	0 7 14 21 60	analytical method used: SAMS 351-1 LOQ 0.01 mg/kg
AL-730-065 Germany - Seligenstadt (R97-87)	maize (Limac)	1. 02.05.1987 2. n. a. 3. 14.10.1987	spray	0.0044	400	0.0175	1 06.08.1987	57	whole plant whole plant whole plant whole plant grain	0.174 0.69 0.46 0.34 <0.01	0 7 14 21 69	analytical method used: SAMS 351-1 LOQ 0.01 mg/kg

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Germany	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2009/1125196 Germany - 21709 Burweg (L090227)	GC 0645 (Bredao)	1. 22.04.2009 2. n. a. 3. 25.09.-27.10.2009	boom spray	0.0075	201-232	0.0151 - 0.0174	2 25.09.09	85	whole plant cobs w/o husks rest of plant with husks (w/o roots) cobs w/o husks rest of plant with husks (w/o roots) grain rest of plant w/o root grain rest of plant w/o root	0.290 nd 0.287 nd 0.137 nd 0.134 nd 0.134	0 6 6 13 13 20 20 32 32	analytical method used: 567/0 LOQ 0.01 mg/kg
2009/1125196 France (N) - 91150 Ezerville (L090228)	GC 0645 (Zidarte)	1. 03.04.2009 2. n. a. 3. 16.09.-07.10.2009	boom spray	0.0075	208-210	0.0156 - 0.0158	2 16.09.09	87	whole plant grain rest of plant w/o roots grain rest of plant w/o roots grain rest of plant w/o root	0.267 nd 0.189 nd 0.214 nd 0.182	0 7 7 14 14 21 21	analytical method used: 567/0 LOQ 0.01 mg/kg

n. a. not available
nd not detectable

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	WL85871 (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-730-013 France (S) - 01 - Loyettes (W/FR/E81/221)	maize (Stella)	1. 12.05.1981 2. n. a. 3. 04.09. (silage) and 09.10.1981 (grain)	spray	0.0006	480	0.0030	1 01.07.1981	30-39? ("height 85 cm")	plant grain	<0.01 <0.01	9 14	analytical method used: SAM 233-1 LOQ 0.01 mg/kg
	maize (Stella)	1. 12.05.1981 2. n. a. 3. 04.09. (silage) and 09.10.1981 (grain)	spray	0.0010	480	0.0050	1 01.07.1981	30-39? ("height 85 cm")	plant grain	<0.01 <0.01	9 14	

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	50 g/L	**(common name and content)**	none
Formulation (e.g. WP)	Fastac 50 g/L EC (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-730-019 France (S) - 31 - Muret (S/FR/E89/161)	maize (Sabrina)	1. 07.05.1989 2. n. a. 3. 15.09. (silage) and 06.10.1989 (harvest)	spray	0.0060	500	0.0300	2 17.08.1989	71 ("beginning drying")	silage grain	0.19 <0.01	29 50	analytical method used: SAM 383-1 LOQ 0.01 mg/kg
AL-730-019 France (S) - 47 - Bouglon (S/FR/E89/161)	maize (Sabrina)	1. 02.05.1989 2. n. a. 3. 29.08. (silage) and 27.09.1989 (harvest)	spray	0.0053	500	0.0300	2 16.08.1989	P0	silage grain	0.32 <0.01	13 42	analytical method used: SAM 383-1 LOQ 0.01 mg/kg
AL-730-027 France (S) - 82340 - St Michel (S/FR/E90/160)	sweet maize (Jubilé)	1. 06.06.1990 2. n. a. 3. 05.09.1990	spray	0.0080	500	0.0400	2 23.08.1990	82	grain	<0.01	13	analytical method used: SAM 351-1 LOQ 0.01 mg/kg
AL-730-027 France (S) - Lot et Garonne (S/FR/E90/424)	maize (Ibisco)	1. 15.05.1990 2. n. a. 3. 18.09.1990	spray	0.0070	520	0.0400	2 09.08.1990	82	grain	<0.01	13	analytical method used: SAM 351-1 LOQ 0.01 mg/kg
AL-730-027 France (S) - 33 - Lubbon (S/FR/E90/447)	maize (Jubilé)	1. 31.05.1990 2. n. a. 3. 04.09.1990	spray	0.0080	500	0.0400	2 13.08.1990	61-69 ("flowering")	grain	<0.01	22	analytical method used: SAM 351-1 LOQ 0.01 mg/kg

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2009/1125196 Spain - 50180 Utebo (L090229)	GC 0645 (Sancia)	1. 04.05.2009 2. n. a. 3. 16.09.-19.10.2009	boom spray	0.015	215	0.0323	1 16.09.09	83-85	whole plant cobs w/o husks rest of plant with husks (w/o roots) cobs w/o husks rest of plant with husks (w/o roots) grain rest of plant w/o root grain rest of plant w/o root	0.177 nd 0.318 nd 0.266 nd 0.168 nd 0.118	0 7 7 13 13 21 21 33 33	analytical method used: 567/0 LOQ 0.01 mg/kg

n. a. not available
n d not detectable

Non-EU

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Maize / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	South Africa, Brazil	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	WL85871 (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
AL-730-014 South Africa - A.J. Friedman Settlers (S/SA/E81/443)	maize (Pioneer 473)	1. N/A 2. N/A 3. 05. - 19.05.1982	spray	no information	no information	0.003	1 05.05.1982	no information	kernels	0.01 <0.01 <0.01 <0.01	0 2 7 14	analytical method used: SAMS 351-1 LOQ 0.01 mg/kg
AL-730-015 Brazil - SP- Ribeirao Preto (I/RE/MI-01/82)	maize (Agroceres)	1. 20.11.1982 2. N/A 3. 06.04.1983	spray	0.003	470	0.012	2 15.03.1983	79-89 ("consistent")	grain	<0.01	22	analytical method used: SAMS 351-1 LOQ 0.01 mg/kg
	maize (Agroceres)	1. 20.11.1982 2. N/A 3. 06.04.1983	spray	0.005	470	0.024	2 15.03.1983	79-89 ("consistent")	grain	<0.01	22	

n. a. not available

- ***Rice***

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Rice / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1026848 Spain - Seville, Coria del Rio, Nueva No. 31 (AF/8823/BA/1)	rice (Puntal)	1. 20.05.2005 2. n. a. 3. 12.09. - 10.10.2005	foliar spray	0.006	200	0.0125	1 12.09.2005	77	whole plant panicles rest of plant grain straw grain straw	0.181 0.097 0.091 0.059 0.077 0.05 0.052	0 14 14 21 21 28 28	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Puntal)	1. 20.05.2005 2. n. a. 3. 12.09. - 10.10.2005	foliar spray	0.006	200	0.0125	2 12.09.2005	77	whole plant panicles rest of plant grain straw grain straw	0.335 0.140 0.091 0.148 0.131 0.011 0.082	0 14 14 21 21 28 28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Rice / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1026848 France (S) - 11800 Domaine Saint Gabriel (AF/8823/BA/2)	rice (Ligalon)	1. 04.05.2005 2. n. a. 3. 25.08. - 21.09.2005	foliar spray	0.006	200	0.0125	1 25.08.2005	73-75	whole plant panicles rest of plant grain straw	0.265 0.160 0.088 0.015 0.072	0 13 13 21 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Ligalon)	1. 04.05.2005 2. n. a. 3. 25.08. - 21.09.2005	foliar spray	0.006	200	0.0125	2 25.08.2005	73-75	whole plant panicles rest of plant grain straw	0.452 0.200 0.123 0.011 0.194	0 13 13 21 21	
2006/1026848 Italy - Bologna 40058, Malalbergo, Via Ponticelli 2 (AF/8823/BA/3)	rice (Lido)	1. 04.05.2005 2. n. a. 3. 29.08. - 29.06.2005	foliar spray	0.006	200	0.0125	1 29.08.2005	75-77	whole plant panicles rest of plant grain straw grain straw	0.511 0.144 0.134 0.176 0.349 0.206 0.330	0 14 14 21 21 28 28	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Lido)	1. 04.05.2005 2. n. a. 3. 29.08. - 29.06.2005	foliar spray	0.006	200	0.0125	2 29.08.2005	75-77	whole plant panicles rest of plant grain straw grain straw	0.463 0.357 0.194 0.205 0.354 0.151 0.351	0 14 14 21 21 28 28	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Rice / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1026848 Greece - 573000 Central Macedonia, Thessaloniki, Nea Malgara (AF/8823/BA/4)	rice (Thai Bonnet)	1. 10.05.2005 2. n. a. 3. 26.08. - 23.09.2005	foliar spray	0.006	200	0.0125	1 26.08.2005	75	whole plant panicles rest of plant grain straw grain straw	0.416 0.102 0.093 0.039 0.044 0.020 0.056	0 14 14 21 21 28 28	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Thai Bonnet)	1. 10.05.2005 2. n. a. 3. 26.08. - 23.09.2005	foliar spray	0.006	200	0.0125	2 26.08.2005	75	whole plant panicles rest of plant grain straw grain straw	0.291 0.207 0.086 <0.01 0.141 0.039 0.077	0 14 14 21 21 28 28	
2007/1007946 France (S) - 11800 Marseilett, Domaine Saint Gabriel (AF/10496/BA/1)	rice (Cigalon)	1. 10.05.2006 2. n. a. 3. 17.08. - 13.09.2006	foliar spray	0.006	200.00	0.0125	1 17.08.2006	65-69	whole plant panicles rest of plant grain straw grain straw	0.270 0.063 0.120 0.049 0.060 0.027 0.053	0 14 14 21 21 27 27	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Cigalon)	1. 10.05.2006 2. n. a. 3. 17.08. - 13.09.2006	foliar spray	0.006	200.00	0.0125	2 17.08.2006	65-69	whole plant panicles rest of plant grain straw grain straw	0.380 0.092 0.140 0.066 0.110 0.036 0.061	0 14 14 21 21 27 27	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Rice / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2007/1007946 Spain - Isla Menor-Los, Cerro de Parlade, Cita 9024 Poligno Industrial (AF/10496/BA/2)	rice (Puntal)	1. 15.05.2006 2. n. a. 3.10.10. - 07.11.2006	foliar spray	0.006	200.00	0.0125	1 10.10.2006	83	whole plant panicles rest of plant grain straw grain straw	0.250 0.036 0.068 0.064 0.049 0.046 0.052	0 13 13 20 20 28 28	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Puntal)	1. 15.05.2006 2. n. a. 3.10.10. - 07.11.2006	foliar spray	0.006	200.00	0.0125	2 10.10.2006	83	whole plant panicles rest of plant grain straw grain straw	0.350 0.120 0.120 0.130 0.091 0.076 0.075	0 13 13 20 20 28 28	
2007/1007946 Italy - Bologna 40058, Malalbergo, Via Ponticelli 2 (AF/10496/BA/3)	rice (Cadet)	1. 12.04.2006 2. n. a. 3. 20.09. - 19.10.2006	foliar spray	0.006	200.00	0.0125	1 20.09.2006	87	whole plant panicles rest of plant grain straw grain straw	0.063 0.028 0.015 0.028 0.064 0.020 0.048	0 15 15 22 22 29 29	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Cadet)	1. 12.04.2006 2. n. a. 3. 20.09. - 19.10.2006	foliar spray	0.006	200.00	0.0125	2 20.09.2006	87	whole plant panicles rest of plant grain straw grain straw	0.320 0.180 0.086 0.110 0.190 0.084 0.190	0 15 15 22 22 29 29	

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Rice / cereal grains	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2007/1007946 Greece - 573000 Central Macedonia, Thessaloniki, Nea Malgara (AF/10496/BA/4)	rice (Claudio)	1. 07.05.2006 2. n. a. 3. 09.08. - 05.09.2006	foliar spray	0.006	200.00	0.0125	1 09.08.2006	72-74	whole plant panicles rest of plant grain straw grain straw	0.260 0.085 0.084 0.040 0.049 0.023 0.021	0 14 14 21 21 27 27	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
	rice (Claudio)	1. 07.05.2006 2. n. a. 3. 09.08. - 05.09.2006	foliar spray	0.006	200.00	0.0125	2 09.08.2006	72-74	whole plant panicles rest of plant grain straw grain straw	0.590 0.140 0.190 0.073 0.065 0.054 0.130	0 14 14 21 21 27 27	

n. a. not available

- **Sugar beet**

Northern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	Mageos MD
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	150 g/kg	**(common name and content)**	none
Formulation (e.g. WP)	Mageos MD (WG)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of : 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment			No of Treatm. and Last Date	Growth Stage (BBCH) at last treatment	Portion Analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
2003/1021716 France (North) - 45300 Manchecourt, Dossainville (AF/6792/BA/1)	sugar beet (Angelina)	1. 29.03.2002 2. n. a. 3. 27.09.2002	foliar spray	0.00373 0.00376	201 205	0.0075 0.0077	2 27.06.2002	39	roots tops	<0.01 <0.01	92	Method SAMS 351-2
2003/1021716 France (North) - 45300 Manchecourt, Dossainville (AF/6792/BA/2)	sugar beet (Crocodile)	1. 30.03.2002 2. n. a. 3. 27.09.2002	foliar spray	0.00376 0.00376	202 206	0.0076 0.0077	2 27.06.2002	39	roots tops	<0.01 <0.01	92	Method SAMS 351-2
2003/1021716 France (North) - 45300 Yevre-la-Ville (AF/6792/BA/3)	sugar beet (Angelina)	1. 27.03.2002 2. n. a. 3. 23.09.2002	foliar spray	0.00377 0.00374	199 198	0.0075 0.0074	2 27.06.2002	39	roots tops	<0.01 <0.01	88	Method SAMS 351-2

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	Mageos MD
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	150 g/kg	**(common name and content)**	none
Formulation (e.g. WP)	Mageos MD (WG)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	Commodity/ Variety	Date of : 1. Sowing/Planting 2. Flowering 3. Harvest	Method of Treatment	Application Rate per Treatment			No of Treatm. and Last Date	Growth Stage (BBCH) at last treatment	Portion Analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
2003/1021716 France (North) - 45300 Thignonville (AF/6792/BA/4)	sugar beet (Rafole)	1. 26.03.2002 2. n. a. 3. 19.09.2002	foliar spray	0.00373 0.00374	209 195	0.0078 0.0073	2 27.06.2002	39	roots tops	<0.01 <0.01	84	Method SAMS 351-2

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 03 I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2004/1006468 Germany . 16833 Brunne, Brandenburg (ACK/02/03)	sugar beet (Milan)	1. 24.04.2003 2. n. a. 3. 03. - 04.11.2003	foliar spray	0.00500	200	0.010	1 30.05.2003	18	plants leaves & tops roots leaves & tops roots	0.446 <0.05 <0.05 <0.05 <0.05	0 25 25 108 108	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg
2004/1006468 Germany - 47574 Goch-Nierswalde, Nordrhein-Westfalen (AGR/02/03)	sugar beet (Dorena)	1. 16.04.2003 2. n. a. 3. 12. - 18.09.2003	foliar spray	0.00500	200	0.010	1 06.06.2003	18	plants leaves & tops roots leaves & tops roots	0.273 <0.05 <0.05 <0.05 <0.05	0 14 14 98 98	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg
2004/1006468 Germany - 74193 Stetten a.H., Baden-Württemberg (DU2/02/03)	sugar beet (Tatjana)	1. 20.03.2003 2. n. a. 3. 01.10.2003	foliar spray	0.00500	200	0.010	1 23.05.2003	18	plants leaves & tops roots leaves & tops roots	0.138 0.292 0.228 <0.05 <0.05	0 21 21 131 131	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg
2004/1006468 Germany - 67229 Gerolsheim, Rheinland-Pfalz (DU4/02/03)	sugar beet (Tatjana)	1. 15.05.2003 2. n. a. 3. 02.10.2003	foliar spray	0.00500	200	0.010	1 14.05.2003	18	plants leaves & tops roots leaves & tops roots	0.208 <0.05 <0.05 <0.05 <0.05	0 30 30 140 140	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg

n. a. not available

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	Germany	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 QC I (SC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2004/1006468 Germany . 16833 Brunne, Brandenburg (ACK/02/03)	sugar beet (Milan)	1. 24.04.2003 2. n. a. 3. 03. - 04.11.2003	foliar spray	0.00500	200	0.010	1 30.05.2003	18	plants leaves & tops roots leaves & tops roots	0.312 <0.05 <0.05 <0.05 <0.05	0 25 25 108 108	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg
2004/1006468 Germany - 47574 Goch-Nierswalde, Nordrhein-Westfalen (AGR/02/03)	sugar beet (Dorena)	1. 16.04.2003 2. n. a. 3. 12. - 18.09.2003	foliar spray	0.00500	200	0.010	1 06.06.2003	18	plants leaves & tops roots leaves & tops roots	0.334 0.07 <0.05 <0.05 <0.05	0 14 14 98 98	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg
2004/1006468 Germany - 74193 Stetten a.H., Baden-Württemberg (DU2/02/03)	sugar beet (Tatjana)	1. 20.03.2003 2. n. a. 3. 01.10.2003	foliar spray	0.00500	200	0.010	1 23.05.2003	18	plants leaves & tops roots leaves & tops roots	0.098 <0.05 <0.05 <0.05 <0.05	0 21 21 131 131	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg
2004/1006468 Germany - 67229 Gerolsheim, Rheinland-Pfalz (DU4/02/03)	sugar beet (Tatjana)	1. 15.05.2003 2. n. a. 3. 02.10.2003	foliar spray	0.00500	200	0.010	1 14.05.2003	18	plants leaves & tops roots leaves & tops roots	0.335 <0.05 <0.05 <0.05 <0.05	0 30 30 140 140	analytical method used: BASF method 564-0 LOQ 0.05 mg/kg

n. a. not available

Southern Europe

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1026851 Italy - Budrio, 40054 Bologna (AF/8832/BA/1)	sugar beet (Dorotea)	1. 15.02.2005 2. n. a. 3. 26.07. - 16.08.2005	foliar spray	0.00600	400	0.025	1 26.07.2005	39-49	leaves & tops leaves & tops leaves & tops leaves & tops roots roots roots roots	0.724 0.227 0.159 0.045 <0.01 <0.01 <0.01 <0.01	0 7 14 21 0 7 14 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
2006/1026851 Spain - Coris del Rio, 41100 Seville (AF/8832/BA/2)	sugar beet (Dina)	1. 12.11.2005 2. n. a. 3. 23.06. - 14.07.2005	foliar spray	0.00600	400	0.025	1 23.06.2005	49	leaves & tops leaves & tops leaves & tops leaves & tops roots roots roots roots	0.482 0.079 0.059 0.02 <0.01 <0.01 <0.01 <0.01	0 7 14 21 0 7 14 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
2006/1026851 Spain - Las Marismas de Lebrija, 41740 (AF/8832/BA/3)	sugar beet (Sidonia)	1. 25.11.2004 2. n. a. 3. 28.06. - 19.07.2005	foliar spray	0.00600	400	0.025	1 28.06.2005	49	leaves & tops leaves & tops leaves & tops leaves & tops roots roots roots roots	0.462 0.035 0.048 <0.01 <0.01 <0.01 <0.01 <0.01	0 7 14 21 0 7 14 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2006/1026851 Greece, Thessaloniki, Central-Macedonia, GR-57300 (AF/8832/BA/4)	sugar beet (Riose)	1. 10.03.2005 2. n. a. 3. 18.08. - 08.09.2005	foliar spray	0.00600	400	0.025	1 18.08.2005	47	leaves & tops leaves & tops leaves & tops leaves & tops roots roots roots roots	0.22 0.152 0.073 0.052 <0.01 <0.01 <0.01 <0.01	0 7 14 21 0 7 14 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
2007/1007941 France - La Creuze, Mizérieux, Feurs, 42110 Loire (AF/10501/BA/1)	sugar beet (Laetitia)	1. 16.03.2006 2. n. a. 3. 12.09. - 03.10.2006	foliar spray	0.00600	400	0.025	1 12.09.2006	39	tops roots tops roots tops roots tops roots	0.77 0.01 0.1 <0.01 0.06 <0.01 0.05 <0.01	0 0 7 7 14 14 21 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
2007/1007941 Italy - Via Legnana 1b, Castel San Pietro, 40002 Bologna (AF/10501/BA/2)	sugar beet (Rizor)	1. 15.02.2006 2. n. a. 3. 07. - 28.08.2006	foliar spray	0.00300	400	0.013	1 07.08.2006	38	tops roots tops roots tops roots tops roots	0.78 <0.01 0.17 <0.01 0.07 <0.01 0.05 <0.01	0 0 7 7 14 14 21 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg

RESIDUE DATA SUMMARY FROM SUPERVISED TRIALS (SUMMARY)

Active substance (common name)	Alpha-cypermethrin	**Commercial Product (name)**	-
Crop/crop group:	Sugar beet / root and tuber vegetables	**Producer of commercial product**	67056 Ludwigshafen, Germany
Responsible body for reporting (name, address)	BASF SE, 67056 Ludwigshafen	**Indoor/Glasshouse/Outdoor**	Outdoor
Country	France, Greece, Italy, Spain	**Other active substance in the formulation**	
Content of active substance (g/kg or g/L)	100 g/L	**(common name and content)**	none
Formulation (e.g. WP)	BAS 310 40 I (EC)	**Residues calculated as:**	Alpha-Cypermethrin (BAS 310 I)

1	2	3	4	5			6	7	8	9	10	11
Report-No. Location (Trial No.)	**Commodity/ Variety**	**Date of : 1. Sowing/Planting 2. Flowering 3. Harvest**	**Method of Treatment**	**Application Rate per Treatment**			**No of Treatm. and Last Date**	**Growth Stage (BBCH) at last treatment**	**Portion Analysed**	**Residues (mg/kg)**	**PHI (days)**	**Remarks**
				kg a.s./hL	**Water (L/ha)**	**kg a.s./ha**						
2007/1007941 Italy - Via Mori, Budrio Prunaro, 40054 Bologna (AF/10501/BA/3)	sugar beet (Omelia)	1. 09.03.2006 2. n. a. 3. 04. - 24.08.2006	foliar spray	0.00300	400	0.013	1 04.08.2006	39	tops roots tops roots tops roots tops roots	0.49 <0.01 0.24 <0.01 0.09 <0.01 0.06 <0.01	0 0 6 6 13 13 20 20	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg
2007/1007941 Greece, Galatades, Pella, Central Macedonia, GR-58300 (AF/10501/BA/4)	sugar beet (Doria)	1. 01.03.2006 2. n. a. 3. 03. - 24.08.2006	foliar spray	0.00600	400	0.025	1 03.08.2006	43-44	tops roots tops roots tops roots tops roots	0.38 <0.01 0.08 <0.01 0.03 <0.01 0.02 <0.01	0 0 6 6 14 14 21 21	analytical method used: BASF method 567/0 LOQ 0.01 mg/kg

n. a. not available

BASF
The Chemical Company

Alpha-Cypermethrin

Document M-CA, Section 7

FATE AND BEHAVIOUR IN THE ENVIRONMENT

Compiled by:

[redacted]

[redacted]

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
March 20, 2015	Summary tables of kinetic evaluation of laboratory degradation and metabolism studies have been updated with the results of the visual assessment and the type I error rate: Table 7.1.1.1-8 and Table 7.1.1.1-9 Table 7.1.2.1.1-11 and Table 7.1.2.1.1-12 Table 7.1.2.1.1-28 to 7.1.2.1.1-31	
January 20, 2016	Report amendments containg graphs obtained from KinGUI that were missing in the report were included under CA 7.1.1.1/1 and CA 7.1.1.3/1 The information provided under CA 7.1.2.1.2/ was replaced by reports CA 7.1.2.1.2/1 and CA 7.1.2.1.2/1	
January 20, 2016	CA 7.1.2.2.1/1 Replacement of interim report (storage stability until day 360) by final report on storage stability; document 2014/1152598 replaced by document 2015/1249074 (storage stability unitl day 727)	
July 10, 2017	CA 7.1.2.2.1/1 Typo in BASF DocID was corrected. Wording in executive summary was adapted for the sake of clarification.	BASF DocID 2017/1134416
July 10, 2017	CA 7.1.2.2.1/5 Report amendment containing corrections of typing and calculation errors was included under CA 7.1.2.2.1/5. Respective data in study summary were replaced.	BASF DocID 2017/1134416
July 10, 2017	Results of requested additional kinetic evaluations for several studies were included under CA 7.1.1.1/3, CA 7.1.1.2/2, CA 7.1.1.3/3, CA 7.2.1.1/3, and CA 7.2.2.2/2.	BASF DocID 2017/1134416

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 7 FATE AND BEHAVIOUR IN THE ENVIRONMENT

A concordance list of structures and designations of reference compounds used during environmental fate studies is given below. During the registration history of alpha-cypermethrin several different code systems were used for the designation of cypermethrin isomers and metabolites. Some of the structures and codes in Table 7-1 were only used in studies that are no longer considered valid for the evaluation of alpha-cypermethrin.
The already peer-reviewed studies as well as the new studies were performed using cyclopropyl-ring and benzyl-ring ^{14}C-labelled alpha-cypermethrin.

Table 7-1: Substances and metabolites; structures, codes, synonyms

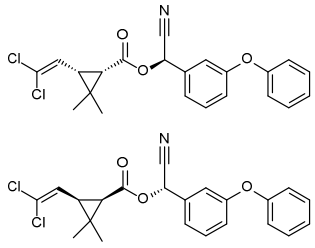
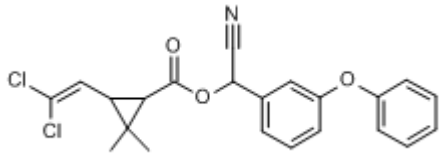
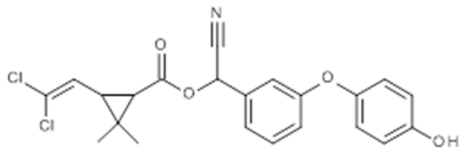
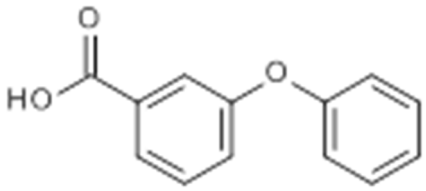
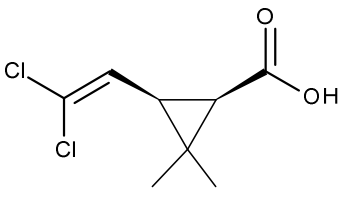
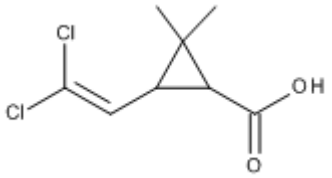
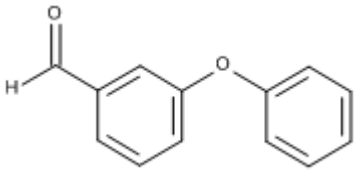
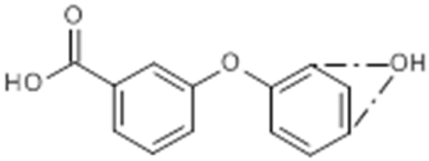
Code Number [a] (Synonyms)	Description	Compound found in:	Structure
BAS 310 I Reg. No. 4078193 CAS-No. 67375-30-8 WL 85871 CL 900049 Cis-2-Cypermethrin	Alpha-cypermethrin Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R) 3 (2,2 dichlorovinyl)-2,2 dimethylcyclopropanecarboxylate and (R)-cyano-3 phenoxybenzyl (1S,3S) 3 (2,2 dichlorovinyl)-2,2 dimethylcyclopropanecarboxylate	Soil Water Sediment	
BAS 311 I Reg. No.127266 CAS-No. 52315-07-8 WL43467	Cypermethrin Cyano-3 phenoxybenzyl 3 (2,2 dichlorovinyl)-2,2 dimethylcyclopropanecarboxylate		
M310I017 Reg. No. 6002320 WL 48394 (cis) WL 48393 (trans) CL 194198	4'hydroxy-alpha-cypermethrin	Soil	
M310I011 Reg. No. 130213 CAS number: 3739-38-6 PBA m-PB acid 3-PB acid WL 44607 CL 206128	3-phenoxybenzoic acid	Soil Water Sediment	
M310I001 Reg. No. 4080830 CAS number: 59042-49-8 DCVA DCVC acid WL 43480 CL 912554	cis-3-(2,2-dichlorovinyl)2,2-dimethylcyclopropanecaroxylic acid	Soil Water Sediment	

Table 7-1: Substances and metabolites; structures, codes, synonyms

Code Number [a] (Synonyms)	Description	Compound found in:	Structure
CL 194198 (cis)			
Reg. No. 180011 DCVA CL 196336 *(trans)*	3-(2,2-dichlorovinyl)-2,2-dimethyl-cyclopropanecarboxylic acid	Soil, Water Sediment	
M310I018 Reg. No. 4080665 WL 42049	3-phenoxy-benzaldehyd	Water (photolysis)	
M310I013 WL 46114 CL 213336	(Most likely) 3-(4-hydroxyphenoxy)benzoic acid	Water Sediment	

[a] For RP codes, different formats are in use:
"RP xxxxx" or or "RPxxxxx" or "RP0xxxxx"
Independent from these format differences, the last 5 digit "xxxxx" are unique for every compound

CA 7.1 Fate and behaviour in soil

Alpha-cypermethrin is the cis-2 isomer of cypermethrin. In the past many studies performed with cypermethrin were submitted to support the registration of alpha-cypermethrin. In the current approach it was tried to rely on studies performed with alpha-cypermethrin wherever possible.

CA 7.1.1 Route of degradation in soil

Many studies were performed with cypermethrin or alpha-cypermethrin during its registration history. An overview is given in Table 7-1. Most of them were no longer considered as relevant, due to high overdosing, insufficient duration or no longer state of the art identification techniques.

The degradation of alpha-cypermethrin was addressed by the following studies, which were no longer considered valid:

1. The degradation of the insecticide WL 43467 in soil under laboratory conditions (Standen 1976, AL-620-008, published by Roberts 1977, AL-905-062)
In this study, the degradation of cypermethrin and its isomers WL 43481 (both cis isomers) and WL 42641 (both trans isomers) were studied in three soils, two from Spain and one UK soil. The study was conducted with both labels and under aerobic and anaerobic conditions. The main route of degradation was the cleavage of the ester linkage forming DCVA (SD 44776) and 3-PBA (36750) as major metabolites. In one soil the degradation was very slow, because soil moisture was too low. A separate balance study was conducted for 182 days.
2. Further studies on the degradation of the insecticide WL 43467 (cypermethrin) in soil under laboratory conditions (Standen 1978, CY-620-003).
The same soils were used as in Standen (1976) but the study was prolonged to 1 year. No intermediate samples were taken. In a second experiment an additional UK soil was used to investigate the degradation of cis-DCVA (WL 43480) and trans-DCVA (WL 42640). It was incubated for 56 days and a dicarboxylic acid was identified as degradation product.

The studies of Standen (1976) and Standen (1978) were also submitted in the application for zeta-cypermethrin. These were evaluated by EFSA (EFSA Scientific Report (2008) 196, 1-119) and it was concluded that these studies should not be relied on for regulation. In the study no material balance is available, as no volatiles were trapped and samples were only taken at 4 intervals.

3. Degradation and leaching behaviour of the pyrethroid insecticide cypermethrin in soil (Sakata et al. 1986, published literature, CY-620-012)
The study was conducted with benzyl- or cyclopropyl- ^{14}C labelled cis and trans isomers of cypermethrin. Two experiments were described; a degradation in two acidic Japanese soils and a column leaching experiment in four Japanese soils. The comparison of cis and trans isomers shows that trans isomers degrade faster that cis isomers.
The study of Sakata (1985) was considered as supportive information. No information on the history of the soils was given. As the study was not conducted under GLP, no raw data are available and not all results were included in the publication.

The following studies were already not regarded in the existing Annex 1 listing due to their poor quality or because they did not follow current guidelines (Alpha-cypermethrin - Annex B - pages 387-389):

4. Degradation of pyrethroid optical isomers in soils (Sakata et al., 1992, published literature, AL-905-078)
5. The metabolism of WL 43467 (Cypermethrin) in lettuce and soil under outdoor conditions (Wright, 1977; AL-790-002)

6. The degradation of the pyrethroid insecticides WL 85871 (FASTAC) and WL 43481 in soil (McMinn, 1983; AL-620-005)

7. The bioavailability and further degradation of bound residues arising from WL 43867 (Standen 1978,)

8. Soil percolation studies with WL 43467 (Noble, 1976; AL-620-009)

9. Persistence of five pyrethroid insecticides in sterile and natural mineral and organic soil (Chapman et al. 1981, published literature)

10. Metabolism of alphamethrin (Alphaguard 25% EC M/M) in soil (Raghunahan, 1989, Gharda)

11. Studies on the persistence of alphamethrin (Alphaguard 10% EC M/M) in water (Raghunahan, 1991, Gharda)

Table 7.1.1-1: List of degradation studies in soil performed with (alpha-) cypermethrin

DocID	Parent compound	Soil	Application rate [mg kg^{-1}]	Incubation temperature	Incubation period [days]	Remark
CY-620-012	Cypermethrin	Clay loam Sandy clay loam	1.0	25±2°C	168	Sakata 1986
AL-620-009	Mixture of all cis isomers of cypermethrin; mixture of all trans isomers of cypermethrin	Garden soil	20	Room temperature	150	Noble 1976
CY-620-001	Cypermethrin	Sandy clay loam	18.9	n/a	28	Standen 1977
AL-790-002	Cypermethrin	Field soil	2 x 0.4	outside temperature	40	Wright 1977
CY-620-003	Mixture of all cis isomers of cypermethrin; mixture of all trans isomers of cypermethrin	Sandy clay Clay Sandy loam Sandy clay loam	2.5 13.5 2.7 9.6 2.0 10.0	25±2°C	65/ 364	Standen 1978
CY-620-004	Cypermethrin; mixture of all cis isomers of cypermethrin; mixture of all trans isomers of cypermethrin	Sandy clay loam	1.0	outside temperature 25±2°C	168	Standen 1978
AL-620-008	Cypermethrin; mixture of all cis isomers of cypermethrin; mixture of all trans isomers of cypermethrin	Sandy clay Sandy loam Clay	2.5	25±2°C (aerobic and anaerobic)	182	Standen 1976
AL-620-005	Alpha-cypermethrin; mixture of all cis isomers of cypermethrin	Sandy clay loam Clay loam	10	25°C	364	McMinn 1983
AL-905-078	Cypermethrin	Light clay Sandy clay loam Silty loam Clay loam	1.0	25±2°C	112	Sakata 1992

n/a = not available

CA 7.1.1.1 Aerobic degradation

Report:	CA 7.1.1.1/1 Michel, A., Hassink, J., 2014 a Metabolism of BAS 310 I in Soil under Aerobic Conditions 2014/1000641
Guidelines:	OECD 307, EPA 835.4100, SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 7.1.1.1/2 Hassink J., 2015 a Report amendment no. 1 to final report: Metabolism of BAS 310 I in soil under aerobic conditions 2015/1107644
Guidelines:	EPA 835.4100, OECD 307, BBA IV 4-1, SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

The amendment to the report contains KinGUI graphs of the parameter estimation section that were missing in the final report.

Executive Summary

The objective of this study was to investigate the rate of degradation of cyclopropane-1-^{14}C-labeled-alpha-cypermethrin (cyclopropane label) and benzyl-^{14}C-labeled-alpha-cypermethrin (benzyl label) in soil, to propose a degradation pathway under aerobic conditions and to define potentially occurring metabolites.

Each test item was incubated aerobically in a loamy sand (sandy loam, according to USDA) soil at 20ºC and 50% maximum water holding capacity (pF 2.5) in the dark for 120 days. The nominal application rate was 0.3 mg kg^{-1} dry soil. Soil aliquots were weighed into test vessels and placed into an incubation cabinet. A closed incubation system with continuous aeration was used with an attached trapping system for the determination of volatile compounds. Absorption solutions were removed at each sampling event.

At about 60 DAT and at the end of the incubation period after 120 days the microbial biomass was determined by the substrate induced respiration method.

Soil samples were taken at 0, 1, 3, 7, 10, 14, 31, 59, 92 and 120 days after treatment (DAT) for the benzyl label and at 0, 1, 3, 7, 10, 14, 31, 63, 91 and 122 DAT for cyclopropane label. At sampling times 0 and 60 DAT, soil samples were worked up in duplicate. The soil samples were extracted four times with acetonitrile/water (7/3, v/v). The extracts were analyzed by LSC and HPLC. The remaining soil after extraction was combusted, in order to determine the amount of non-extractable soil bound residues. A full material balance was provided for each sampling interval.

The amount of extractable radioactivity continuously decreased from 97.8% TAR at day 0 to 9.7% TAR after 122 days of incubation for the cyclopropane label and from 98.1% TAR at day 0 to 9.1% TAR after 120 days of incubation for the benzyl-^{14}C-label. The non-extractable radioactive residues (NER) increased from 2.2% TAR on day 0 to a maximum of 48.7% TAR after 122 days of incubation for the cyclopropane label and from 1.9% TAR on day 0 to a maximum of 47.4% TAR after 120 days of incubation for the benzyl-^{14}C-label. Mineralization to $^{14}CO_2$ reached a total of 33.9% TAR after 122 days of incubation for the cyclopropane label and 34.0% TAR for the benzyl-^{14}C-label. Other volatile compounds could not be detected in significant amounts at any time. The material balance ranged from 78.9 to 100.0% TAR for the cyclopropane label and from 88.2 to 103.3% TAR for the benzyl-^{14}C-label.

The NER fraction was further characterized for one replicate from each sampling. Upon extraction with NaOH and water, about 8.6% TAR of the NER remained unextractable from the soil treated with the cyclopropane label and 13.4% TAR remained unextractable from the soil treated with the benzyl-^{14}C-label. This portion of unextractable radioactivity was assigned to the humin fraction. The alkali-soluble radioactivity was further fractionated by precipitation with concentrated HCl to distinguish between acid-insoluble humic acids and acid-soluble fulvic acids. For the cyclopropane label, 19.9% of TAR could be assigned to the fulvic acid fraction at the end of the incubation. For the benzyl-^{14}C-label, a maximum of 18.2% of TAR could be assigned to the fulvic acid fraction after 60 days. At 90 DAT, 7.0% TAR and 9.4% TAR were assigned to the humic acids fraction for the cyclopropane label and the benzyl label, respectively. The fulvic acid fraction was partitioned with ethyl acetate for three exemplary samples (10, 63 and 122 DAT (cyclopropane label), 10, 59 and 120 DAT (benzyl label)). A maximum of about 9.0% and 5.6% of the fulvic acid fraction were soluble in the ethyl acetate phase at 60 DAT for the cyclopropane label and the benzyl label, respectively.

Besides the parent compound, two metabolites DCVA (3-(2,2-dichlorovinyl)-2,2-dimethyl cyclopropanecarboxylic acid) and M310I017 were observed. Parent compound and the metabolites were positively identified by time-of-flight mass spectrometry with electrospray ionization (ESI-Q-TOF). For the metabolite DCVA, the identification was confirmed by comparison of retention times with reference compounds.

The parent compound decreased continuously from 96.7% TAR at 0 DAT to 4.2% TAR after 122 days of incubation (for the cyclopropane label) and from 96.2% TAR at 0 DAT to 2.7% TAR after 120 days of incubation (for the benzyl label).

The metabolite DCVA was only observed in the extracts of the soil spiked with the cyclopropane label. It was first observed on day 3 after application, increased to about 13.6% TAR at 7 DAT and decreased to 4.9% TAR at 14 DAT. The metabolite M310I017 was observed with both labels. For the soil spiked with cyclopropane label, it was first detected at day 1 after application, increased to max. 8.4% TAR at 7 DAT and declined to 0.3% TAR at the end of the study. For the soil spiked with benzyl label, it was first detected at day 3 after application, increased to max. 7.5 % TAR at 7 DAT and declined to 2.0% TAR at the end of the study.

Kinetic evaluation was performed following the recommendations of the FOCUS Kinetics workgroup in order to derive degradation parameters as triggers for additional work (trigger endpoints) as well as modeling endpoints. The $DegT_{50}$/$DegT_{90}$ values for alpha-cypermethrin and its metabolites, DCVA, and M310I017 were determined as summarized in the table below (Table 7.1.1.1-1).

Table 7.1.1.1-1: Trigger and Modeling Endpoints for alpha-cypermethrin, DCVA, and M310I017 (non-GLP)

Compound	Label	Trigger endpoints			Modeling endpoints	
		Best-fit model	$DegT_{50}$ [d]	$DegT_{90}$ [d]	Best-fit model	$DegT_{50}$ [d]
alpha-cypermethrin	Cyclopropane-^{14}C	DFOP	3.1	42.2	FOMC	14.5[a]
	Benzyl-^{14}C	FOMC	3.9	43.5	FOMC	13.1[a]
DCVA	Cyclopropane-^{14}C	SFO[b]	5.2	17.1	SFO[b]	5.2
M310I017	Cyclopropane-^{14}C	SFO[b]	8.5	28.4	SFO[b]	8.5
	Benzyl-^{14}C	SFO[b]	19.9	66.1	SFO[b]	19.9

[a] Calculated as $DegT_{50} = DegT_{90} / 3.32$
[b] Evaluation of metabolite decline

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

BAS-Code: BAS 310 I (alpha-cypermethrin)
Chemical name: Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molecular formula: $C_{22}H_{19}Cl_2NO_3$
Molar mass: 416.3 g mol^{-1} (unlabeled)

Label 1 (cyclopropane label)
Label: cyclopropane-1-^{14}C
Batch No.: 986-1046
Specific radioactivity of a.s.: 4.9 MBq mg^{-1}
Radiochemical purity: 98.2%
Chemical purity: 93.6%

Label 2 (benzyl label)

Label:	benzyl-^{14}C
Batch No.:	775-0401
Specific radioactivity of a.s.:	4.94 MBq mg^{-1}
Radiochemical purity:	96.1%
Chemical purity:	93.7%

2. Soil

The German soil LUFA 5M from LUFA (Landwirtschaftliche Untersuchungs- und Forschungsanstalt, Speyer, Germany) was used in this study. It was sampled from 0-20 cm depth. The soil was passed through a 2 mm sieve and stored for no longer than three months at 4°C before use. The soil characteristics are summarized in Table 7.1.1.1-2.

Table 7.1.1.1-2: Characteristics of soil LUFA 5M

Soil designation	**LUFA 5 M** **12/1651/03** (Germany, Origin: LUFA Speyer)
DIN Particle size distribution [%]	
Sand 0.063 – 2 mm	54.9
Silt 0.002 – 0.063 mm	32.9
Clay < 0.002 mm	12.2
Textural class	loamy sand (Sl4)
USDA Particle size distribution [%]	
Sand 0.050 – 2 mm	58.8
Silt 0.002 – 0.050 mm	28.9
Clay < 0.002 mm	12.2
Textural class	sandy loam
Organic C [%]	1.99
Organic matter [%] *	3.43
pH [H_2O]	7.8
pH [$CaCl_2$]	7.2
Cation exchange capacity [cmol$^+$ kg^{-1}]	10.1
Max. water holding capacity [g 100g^{-1} dry weight]	28.9
Water holding capacity at pF 2.0 [g g^{-1} dry weight]	0.220
Water holding capacity at pF 2.5 [g g^{-1} dry weight]	0.154
Microbial biomass (start of study) [mg C 100g^{-1} dry soil] -	32.0 (certificate)
Untreated soil	50.5**
Soil treated with solvent	63.4**
Soil treated with unlabeled test item and solvent	66.1**
Microbial biomass (after 60 days) [mg C 100g^{-1} dry soil]	
Untreated soil	30.5**
Soil treated with solvent	28.3**
Soil treated with unlabeled test item and solvent	14.9**

Table 7.1.1.1-2: Characteristics of soil LUFA 5M

Soil designation	LUFA 5 M 12/1651/03 (Germany, Origin: LUFA Speyer)
Microbial biomass (end of study [120 days]) [mg C $100g^{-1}$ dry soil]	
Untreated soil	24.3**
Soil treated with solvent	22.2**
Soil treated with unlabeled test item and solvent	15.4**

* Organic matter = organic carbon x 1.724
** Determined at BASF test facility Limburgerhof

B. STUDY DESIGN

1. Experimental conditions

The test substance was applied at a nominal concentration of 0.08 mg alpha-cypermethrin per kg dry soil which corresponds to a field application rate of 30 g a.s. ha^{-1} (calculated on the basis of an equal distribution in the top 2.5 cm soil layer and a soil density of 1.5 g cm^{-3}). Because of the low specific radioactivity of the test items and to ensure a better detection of possible degradation products, the amount of test item applied was increased to 0.3 mg test item kg^{-1} dry soil.

Before application the soil was readjusted to 50% of the respective maximum water holding capacity (MWHC). After application to bulk soil, portions of 100 g soil (dry weight basis) were filled into test vessels. The actual application rate was 0.31 mg kg^{-1} for the cyclopropane-1-^{14}C-labeled alpha-cypermethrin (cyclopropane label) and 0.29 mg kg^{-1} for the benzyl-^{14}C-labeled alpha-cypermethrin (benzyl label). The mean value of the sum of extractable (ERR) and non-extractable radioactive residues (NER) at day 0 in 100 g dry soil was set to 100% TAR (total applied radioactivity).

For incubation, all test vessels were connected in line with aeration tubes in the soil metabolism apparatus. During the study, samples were continuously aerated with a slight stream of moistened synthetic air. For removing carbon dioxide, the air was passed through a bottle with NaOH before passing the test vessels. For trapping of volatiles possibly evolving from soil during the incubation, test vessels were connected to three gas washing flasks containing 50 mL ethylene glycol, 50 mL 0.5 M H_2SO_4, and 50 mL 0.5 M NaOH. The treated soils were incubated at 20 ± 2°C in the dark.

To determine the microbial biomass at 0, 60 and 120 days after treatment, three additional soil portions (1 kg dry soil equivalents, 50% MWHC) were incubated under the same conditions as the ^{14}C-treated samples. The first series was incubated without additional treatment. A second series was treated with 300 µL acetonitrile (ACN; without test item). A third series was treated with 280 µL of the specific application solution containing the unlabeled test item.

2. Sampling

Sampling dates were 0, 1, 3, 7, 10, 14, 31, 63, 91 and 122 days after treatment (DAT) for the cyclopropane labeled test item and 0, 1, 3, 7, 10, 14, 31, 59, 92 and 120 DAT for the benzyl labeled test item. At each sampling date two replicate samples were collected. One soil sample was completely worked up, the second replicate was stored in a freezer. On day 0 and day 60 two soil samples were worked up and two were stored.

Besides day 0, the volatile trapping solutions were sampled and replaced by new flasks with fresh solutions. The sampled trapping solutions were measured for radioactivity by LSC.

3. Description of analytical procedures

For the determination of ERR, the 100 g (dry weight basis) soil samples were consecutively extracted four times with 120 mL acetonitrile (ACN)/water (7/3, v/v) by mechanical shaking for 1 h. After centrifugation, the four ACN/water-extracts were pooled and analyzed by liquid scintillation counting (LSC).

Radio-HPLC was used to show the purity of the test items and to investigate the metabolism of the cyclopropane- and the benzyl-labeled test item. HPLC system 1 was used to quantify the test item and the metabolites, HPLC system 3 was applied for the separation of cis and trans isomers, and HPLC system 5 was used for the separation of enantiomers. The HPLC system 4 was used for the analysis of the water phase separated from the ethyl acetate for the characterization of the fulvic acids fraction.

Depending on the subsequent used HPLC-method, different sample workups were performed.

To determine the metabolite pattern, aliquots of the soil extracts were concentrated (rotary evaporator T ≈ 35°C). Afterwards, the residues were diluted with ACN and analyzed by radio-HPLC (HPLC system 1). Separation of cis and trans isomers, as well as enantiomers of alpha-cypermethrin was performed on HPLC system 3 and 5, respectively. For this, aliquots of the soil extracts were evaporated to dryness (rotary evaporator T ≈ 35°C), redissolved in ACN/water (7/3, v/v) and analyzed by LSC and radio-HPLC.

The soil residues remaining after extraction were air-dried, homogenized by means of a small mill, and three aliquots were combusted in an oxidizer. The evolved $^{14}CO_2$ from each combusted aliquot was trapped in Oxysolve C-400 scintillator and measured by LSC to determine the amount of the non-extractable residues (NER).

The fraction of NER was further characterized by NaOH extractions (0.5 M NaOH, three times). NaOH extracts were processed to determine the distribution of radioactivity to the humin, humic, and fulvic acid fractions. The humic acid fraction and the fulvic acid fraction were analyzed by LSC. The fulvic acid fraction was further partitioned with ethyl acetate. Concentrated ethyl acetate phases were analyzed by HPLC. The remaining radioactivity in the soil residues was determined by combustion analyses (humin fraction). Further details are given in the report.

The fulvic acid fraction was partitioned three times with ethyl acetate. After pooling, the ethyl acetate phase was analyzed by LSC.

The microbial biomass in the soil samples treated with and without solvent was determined at 0, 60 and 120 DAT. The method was based on the determination of oxygen consumption upon addition of glucose. The microbial biomass declined over the incubation phase. However, the results demonstrate that the soil was still viable and microbially active at days 60 and 120 (end) of the study.

4. Calculation of the degradation rate

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints) as well as modeling endpoints. Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006) "Guidance Document on Estimating Persistence and Degradation Kinetics from Environmental Fate Studies on Pesticides in EU Registration" Report of the FOCUS Work Group on Degradation Kinetics, EC Document Reference Sanco/10058/2005 version 2.0, 434 pp.*].

The software package KinGUII (version 2.2014.224.1704) was used for parameter fitting [*SCHÄFER, D., MIKOLASCH, M., RAINBIRD, P., HARVEY, B. (2007) KinGUI: A new kinetic software tool for evaluations according to FOCUS Degradation Kinetics. In: Del Re, A.A.M. et al. (Eds.): Proceedings of the XIII Symposium on Pesticide Chemistry, Piacenza, 2007, p. 916-923*., *WITT, J., GAO, Z., MEYER, H. (2014) KinGUII, Version 2.2014.224.1704 Bayer CropScience AG*]. The error tolerance and the number of iterations of the optimization tool were set to 0.00001 and 100, respectively.

For each data set, the kinetic models proposed by the FOCUS Kinetics guidance document were tested in order to identify the best-fit model. The recommended kinetic models, i.e. the single first order kinetics (SFO), the Gustafson-Holden model (FOMC), and bi-exponential (DFOP) kinetics are already implemented in KinGUI. The Goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test) [for details see Chapter 6.3 in FOCUS (2006)].

Where available, replicate measurements were considered for the parameter estimation. The initial concentration of the applied test item was set to the material balance recovered at day 0. The χ^2 value for the kinetic model was calculated as recommended by FOCUS, considering the average of replicate measurements. For the individual parameters, the t-test statistics were based on number of individual measurements.

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The detailed results on the extractability of radioactive residues and the overall mass balances are presented in Table 7.1.1.1-3. The mass balance ranged from 78.9% TAR to 100.0% TAR and from 88.2% TAR to 103.3% TAR for the cyclopropane label and the benzyl label, respectively.

B. EXTRACTABLE AND BOUND RESIDUES

The amount of extractable radioactive residues continuously decreased from 97.8% TAR at 0 DAT to 9.7% TAR at 122 DAT for the cyclopropane label and from 98.1% TAR at 0 DAT to 9.1% TAR at 120 DAT for the benzyl label.

The amount of non-extractable residues increased from 2.2% TAR immediately after treatment to 48.7% TAR after 122 days of incubation for the cyclopropane label and from 1.9% TAR immediately after treatment to 47.4% TAR after 120 days of incubation for the benzyl label. The NER fraction was further characterized by fractionation into humins, humic and fulvic acids.

Table 7.1.1.1-3: Mass balance and distribution of radioactivity after application of ^{14}C-labeled-alpha-cypermethrin to soil [% TAR]

Cyclopropane-1-^{14}C-label					Benzyl-^{14}C-label				
DAT	**ERR**	**NER**	**Volatiles***	**Material balance**	**DAT**	**ERR**	**NER**	**Volatiles***	**Material balance**
0 I	98.8	2.2	n.a.	101.0	0 I	102.1	1.7	n.a.	103.8
0 II	96.8	2.3	n.a.	99.0	0 II	94.1	2.1	n.a.	96.2
0 mean	97.8	2.2	n.a.	100.0	**0 mean**	98.1	1.9	n.a.	100.0
1	82.6	7.2	1.6	91.4	1	86.9	10.4	2.0	99.3
3	72.8	11.7	4.3	88.7	3	82.0	15.7	5.6	103.3
7	60.9	20.9	9.9	91.6	7	58.4	24.2	12.6	95.2
10	54.5	25.5	13.3	93.3	10	49.9	28.0	16.4	94.3
14	43.2	30.4	13.3	86.9	14	39.9	37.7	20.4	97.9
31	27.1	37.4	24.0	88.5	31	22.8	40.4	28.6	91.9
63 I	16.4	39.4		84.3	59 I	15.0	45.4		92.1
63 II	12.0	38.3	28.4	78.7	59 II	17.4	45.9	31.7	95.0
63 mean	14.2	38.9		81.5	**59 mean**	16.2	45.6		93.5
91	10.0	36.9	32.0	78.9	92	10.5	44.7	33.0	88.2
122	9.7	48.7	33.9	92.3	120	9.1	47.4	34.0	90.5

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
n.a. = Not analyzed
* No other volatiles than CO_2 were found

C. VOLATILIZATION

Carbon dioxide was the only trapped volatile degradation product found in the sodium hydroxide traps reaching 33.9% TAR and 34.0% TAR after 122 days of incubation for the cyclopropane label and the benzyl label, respectively. In sulfuric acid and ethylene glycol no significant radioactivity could be measured at any time.

D. TRANSFORMATION OF PARENT COMPOUND

Aliquots of all sample extracts were analyzed by radio-HPLC (see Table 7.1.1.1-4).

Peak assignment of alpha-cypermethrin was performed by comparison to the retention time of the application solution and by mass spectrometry (10 DAT sample). One metabolite (DCVA) was identified by mass spectrometry (10 DAT sample) and comparison of retention times to a reference item. The identification of the metabolite M310I017 was achieved by mass spectrometry. The identity of M310I017 was additionally confirmed by chromatographic comparison to a reference sample from the aerobic soil degradation study [*CA 7.1.2.1.1/4, BASF DocID 2014/1159491*].

The amount of alpha-cypermethrin decreased throughout the incubation from 96.7 % TAR at 0 DAT to 4.2% TAR at the end of the study for the cyclopropane label. For the soils spiked with benzyl label, the amount of alpha-cypermethrin decreased from 96.2% TAR at 0 DAT to 2.7% TAR at the end of the study.

The metabolite DCVA was only observed in the extracts of the soil treated with the cyclopropane label. It was detected with max. 13.6% TAR at 7 DAT and decreased to 4.9% TAR at 14 DAT.

The metabolite M310I017 was observed with both labels. For the soil samples spiked with the cyclopropane label, the amount of metabolite M310I017 increased to max. 8.4% TAR at 7 DAT and decreased to 0.3% TAR at the end of the study. For the soil samples spiked with the benzyl label, the amount of metabolite M310I017 increased to max. 7.5% TAR at 7 DAT and decreased to 2.0% TAR at 59 DAT.

Table 7.1.1.1-4: Radio-HPLC analysis of extracts of soil LUFA 5M treated with ^{14}C-labeled-alpha-cypermethrin[% TAR]

Cyclopropane-1-^{14}C-label						Benzyl-^{14}C-label				
DAT	**Total**	**DCVA**	**M310I017**	**BAS310I**	**Sum others***	**DAT**	**Total**	**M310I017**	**BAS310I**	**Sum others***
0 I	98.8	n.d.	n.d.	98.8	0.0	0 I	102.1	n.d.	98.2	3.9
0 II	96.8	n.d.	n.d.	94.6	2.1	0 II	94.1	n.d.	94.1	0.0
0 mean	**97.8**	**-**	**-**	**96.7**	**1.1**	**0 mean**	**98.1**	**-**	**96.2**	**1.9**
1	82.6	n.d.	4.5	76.2	2.0	1	86.9	n.d.	79.7	7.2
3	72.8	3.1	4.4	48.1	17.1	3	82.0	4.0	46.7	31.3
7	60.9	13.6	8.4	38.9	0.0	7	58.4	7.5	40.1	10.8
10	54.5	10.4	6.4	35.6	2.0	10	49.9	4.1	40.2	5.6
14	43.2	4.9	5.6	24.0	8.7	14	39.9	5.1	22.5	12.3
31	27.1	n.d.	0.5	14.6	12.0	31	22.8	1.4	8.3	13.1
63 I	16.4	n.d.	n.d.	6.3	10.1	59 I	15.0	1.7	4.7	8.5
63 II	12.0	n.d.	n.d.	3.3	8.7	59 II	17.4	2.3	8.1	6.9
63 mean	**14.2**	**-**	**-**	**4.8**	**10.0**	**59 mean**	**16.2**	**2.0**	**6.4**	**7.7**
91	10.0	n.d.	0.7	3.5	5.8	92	10.5	n.d.	4.1	6.4
122	9.7	n.d.	0.3	4.2	5.2	120	9.1	n.d.	2.7	6.4

TAR = Total applied radioactivity
DAT = Days after treatment
n.d. = Not detected
* Individual peaks < 5.0% TAR

Further HPLC analysis revealed that only the cis isomer of alpha-cypermethrin was present in the sample extracts throughout the study and that no trans isomer was formed.

No change of the enantiomeric ratio could be observed during the study irrespective of the radiolabel.

E. CHARACTERIZATION OF NON-EXTRACTABLE RESIDUES (NER)

Results of the non-extractable residues characterization performed by humic substance fractionation are given in Table 7.1.1.1-5 to Table 7.1.1.1-7.

Upon extraction with NaOH and water, about 8.6% and 13.4% of the NER remained unextractable for the cyclopropane label and the benzyl label, respectively. This portion was assigned to the humin fraction. The alkali-soluble radioactivity was further fractionated. For the cyclopropane label, 19.9% TAR could be assigned to the fulvic acids fraction (after 122 days) and maximum 7.0% TAR to the humic acids fraction at 91 DAT. For the benzyl label, 18.2% TAR could be assigned to the fulvic acids fraction (at 59 DAT) and maximum 9.4% TAR to the humic acids fraction at 92 DAT. The humic acid fraction was not further analyzed.

The fulvic acid fraction was partitioned with ethyl acetate for three exemplary samples (10, 59/63 and 120/122 days). About 9.0% and 5.6% of the fulvic acid fraction were soluble in the ethyl acetate phase at 63/59 DAT for the cyclopropane label and the benzyl label, respectively. HPLC analyses of the water phase of the fulvic acid fraction revealed the presence of one main peak and two small peaks. The main peak was found to occur with a maximum of 5.3 % TAR at 63 DAT for the cyclopropane label and 6.3% TAR at 120 DAT for the benzyl label. No further attempts were made to identify the compounds observed in the water phase of the fulvic acid.

During the extraction of NERs by NaOH, strong alkaline conditions were reached. Alpha-cypermethrin is known to be quickly hydrolyzed in alkaline conditions [*CA 7.2.1.1/1, BASF DocID 2005/1016375*]. It is therefore likely that the compounds observed by HPLC in the water phase of the fulvic acid fraction were created during the extraction by NaOH, i.e. alkaline hydrolysis.

Table 7.1.1.1-5: Characterization of bound residues [% TAR] of ^{14}C-labeled-alpha-cypermethrin

Cyclopropane-1-^{14}C-label							Benzyl-^{14}C-label						
DAT	NER*	ERR			NER II (Humins)	Sum ERR + NER II*	DAT	NER*	ERR			NER II (Humins)	Sum ERR + NER II*
		NaOH	H_2O	Total					NaOH	H_2O	Total		
1 I	7.2	5.9	0.2	6.1	1.2	7.3	1 I	10.4	6.5	0.4	6.8	2.5	9.4
3 I	11.7	9.0	0.3	9.3	2.1	11.4	3 I	15.7	11.5	0.4	11.9	4.7	16.6
7 I	20.9	14.1	0.4	14.5	3.4	17.9	7 I	24.2	17.5	1.0	18.5	7.0	25.6
10 I	25.5	17.7	0.7	18.4	4.5	22.9	10 I	28.0	19.1	1.0	20.1	8.3	28.4
14 I	30.4	22.8	0.7	23.5	5.8	29.3	14 I	37.7	23.0	1.3	24.3	9.7	34.0
31 I	37.4	26.4	1.0	27.4	7.8	35.2	31 I	40.4	27.1	1.5	28.6	12.3	40.9
63 I	39.4	26.8	0.9	27.8	8.5	36.2	59 I	45.4	28.4	1.5	29.9	13.8	43.8
63 II	38.3	24.4	0.9	25.4	7.8	33.2	59 II	45.9	28.0	1.9	29.9	13.2	43.1
63 mean	**38.9**	**25.6**	**0.9**	**26.6**	**8.2**	**34.7**	**59 mean**	**45.7**	**28.2**	**1.7**	**29.9**	**13.5**	**43.5**
91 I	36.9	24.7	0.9	25.6	7.9	33.4	92 I	44.7	26.1	1.7	27.8	12.5	40.3
122 I	48.7	24.9	0.8	25.7	8.6	34.3	120 I	47.4	26.1	1.3	27.4	13.4	40.8

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
* Deviations from initial NER values have to be attributed to differing LSC results

Table 7.1.1.1-6: Fractionation of alkali-soluble residues [% TAR] of ^{14}C-labeled-alpha-cypermethrin

Cyclopropane-1-^{14}C-label					Benzyl-^{14}C-label				
DAT	**Sum of NaOH and water extracts***	**Humic acids**	**Fulvic acids**	**Sum humic and fulvic acids***	**DAT**	**Sum of NaOH and water extracts***	**Humic acids**	**Fulvic acids**	**Sum humic and fulvic acids***
1 I	6.1	1.0	4.6	5.6	1 I	6.8	2.0	4.2	6.2
3 I	9.3	1.4	7.6	9.0	3 I	11.9	3.4	8.0	11.4
7 I	14.5	2.8	10.2	13.1	7 I	18.5	5.7	10.9	16.6
10 I	18.3	3.1	14.4	17.6	10 I	20.1	5.5	13.7	19.2
14 I	23.5	5.4	16.6	21.9	14 I	24.3	7.8	14.2	22.0
31 I	27.4	7.2	18.1	25.3	31 I	28.6	9.3	16.9	26.2
63 I	27.8	5.3	20.3	25.6	59 I	29.9	8.8	19.3	28.1
63 II	25.4	7.1	16.6	23.8	59 II	29.9	9.9	17.1	27.0
63 mean	**26.6**	**6.2**	**18.5**	**24.7**	**59 mean**	**29.9**	**9.4**	**18.2**	**27.6**
91 I	25.6	7.0	16.9	23.9	92 I	27.8	9.4	16.0	25.4
122 I	25.7	4.9	19.9	24.8	120 I	27.4	7.9	17.9	25.7

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
* Deviations from initial NER values have to be attributed to differing LSC results

Table 7.1.1.1-7: Fractionation of residues from the fulvic acids [% TAR] of ^{14}C-labeled-alpha-cypermethrin

Cyclopropane-1-^{14}C-label					Benzyl-^{14}C-label				
DAT	**Fulvic acids***	**Ethyl acetate**	**Aqueous phase**	**Sum ethyl acetate and aqueous phase***	**DAT**	**Fulvic acids***	**Ethyl acetate**	**Aqueous phase**	**Sum ethyl acetate and aqueous phase***
10 I	14.4	7.0	6.6	13.6	10 I	13.7	4.2	8.3	12.4
63 I	16.6	9.0	10.8	19.8	59 I	14.2	5.6	12.3	17.9
122 I	19.9	7.8	11.0	18.8	120 I	17.9	5.1	12.2	17.3

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
* Deviations from initial NER values have to be attributed to differing LSC results

F. KINETIC MODELING RESULTS

The degradation of alpha-cypermethrin could be best described by bi-phasic kinetic fit (FOMC or DFOP). For the metabolites DCVA and M310I017 the first-order kinetic model was selected. A summary of the $DegT_{50}$ and $DegT_{90}$ values of alpha-cypermethrin and its metabolites DCVA and M310I017 derived as trigger and modeling endpoints are given in Table 7.1.1.1-8 and Table 7.1.1.1-9. The kinetic evaluation for the metabolite was conducted by fitting the decline curve of the metabolite starting from the peak (maximum measured amount) as proposed by FOCUS. Therefore, no formation fraction could be delineated.

Table 7.1.1.1-8: Trigger endpoints for alpha-cypermethrin, DCVA, and M310I017 (non-GLP)

Compound	Label	Best-fit model	χ^2 error	type I error rate (Prob. > t)	Visual fit	$DegT_{50}$ [d]	$DegT_{90}$ [d]
alpha-cypermethrin	Cyclopropane-^{14}C	DFOP	6.0	k1: <0.01 k2: <0.01 g: <0.01	Good	3.1	42.2
	Benzyl-^{14}C	FOMC	12.1	β: <0.05	Good	3.9	43.5
DCVA	Cyclopropane-^{14}C	SFO[a]	7.6	k: <0.05	Good	5.2	17.1
M310I017	Cyclopropane-^{14}C	SFO[a]	12.8	k: <0.01	Acceptable	8.5	28.4
	Benzyl-^{14}C	SFO[a]	24.9	k: <0.05	Acceptable	19.9	66.1

[a] Evaluation of metabolite decline

Table 7.1.1.1-9: Modeling endpoints for alpha-cypermethrin, DCVA, and M310I017 (non-GLP)

Compound	Label	Best-fit model	χ^2 error	type I error rate (Prob. > t)	Visual fit	$DegT_{50}$ [d]
alpha-cypermethrin	Cyclopropane-^{14}C	FOMC	8.1	β: <0.01	Good	14.5[a]
	Benzyl-^{14}C	FOMC	12.0	β: <0.05	Good	13.1[a]
DCVA	Cyclopropane-^{14}C	SFO[b]	7.6	k: <0.05	Good	5.2
M310I017	Cyclopropane-^{14}C	SFO[b]	12.8	k: <0.01	Acceptable	8.5
	Benzyl-^{14}C	SFO[b]	24.9	k: <0.05	Acceptable	19.9

[a] Calculated as $DT_{50} = DT_{90}/3.32$
[b] Evaluation of metabolite decline

III. CONCLUSION

Alpha-cypermethrin was rapidly degraded in a loamy sand soil. The fraction of non-extractable residues increased to about 48% TAR at the end of the study after 120 days. $^{14}CO_2$ was formed in amounts up to 33.9 - 34.0% TAR. No other volatile compounds were detected in significant amounts.

Two metabolites DCVA and M310I017 were confirmed with maximum concentrations of 13.6% TAR and 8.4 - 7.5% TAR, respectively. The metabolite DCVA was only observed in the extracts of soil spiked with the cyclopropane-^{14}C-label. No benzyl-specific metabolites could be identified.

According to the best fit kinetics, $DegT_{50}$ values of 3.1 days and 3.9 days were calculated as trigger endpoints for cyclopropane-1-^{14}C-labeled-alpha-cypermethrin and benzyl-^{14}C-labeled-alpha-cypermethrin, respectively. The corresponding calculated modeling endpoints were 14.5 and 13.1 days for the cyclopropane-labeled and the benzyl labeled test item, respectively.

For DCVA, a $DegT_{50}$ value of 5.2 days was calculated as trigger and modeling endpoint, according to the best fit kinetics.

For M310I017, $DegT_{50}$ values of 8.5 and 19.9 days were calculated as trigger and modeling endpoints for the experiment using the cyclopropane-labeled and benzyl-labeled test item, respectively.

The RMS BE has asked for additional kinetic evaluations on 27. Oct. 2016. The results are presented and discussed below.

Report:	CA 7.1.1.1/3 Anonymous, 2016 a Response to RMS BE - Request concerning kinetic re-evaluations – Alpha-cypermethrin 2016/1324158
Guidelines:	none
GLP:	no

RMS: RMS notes that the Notifier performed a separate kinetic assessment for each label. In peer review, it is common practice to handle residuals from the same soil with different radiolabelled positions as replicate values. Please Notifier consider that residue data from two tested labels from the same soil have to be processed as replicates, for the determination of the kinetic parameters of alpha-cypermethrin and its metabolites.

BASF response: A kinetic evaluation was performed by processing the residue data of the two tested labels as replicates. The results are summarized in Table 7.1.1.1-10 and the corresponding Kingui fits are presented in Appendix 1. in the original response document. The trigger and modeling endpoints (DT_{50} = 3.8 d, DT_{90} = 45.7 d, modeling DT_{50} = 13.8 d, FOMC model) are comparable with those derived by performing a separate kinetic assessment for each label (geomean DT_{50} = 3.5 d, geomean DT_{90} = 42.8 d, geomean modeling DT_{50} = 13.8 d). Therefore, the endpoints presented in the dossier under CA 7.1.1.1/1 are considered adequate.

Table 7.1.1.1-10: Statistical and visual assessment of kinetic models for degradation of alpha-cypermethrin in soil under aerobic conditions

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	Trigger endpoints		Modeling endpoints
				$DegT_{50}$ [d]	$DegT_{90}$ [d]	$DegT_{50}$ [d]
SFO	21.4	k: <0.01	poor	6.0	19.9	6.0
FOMC	11.2	β: <0.01	good	3.8	45.7	13.8 [a]
DFOP	9.6	k1: <0.01 k2: <0.01 g: <0.01	acceptable	3.2	39.1	17.1 [b]

[a] Calculated as $DegT_{50} = DegT_{90} / 3.32$
[b] Calculated as $DegT_{50} = \ln(2)/k_{slow}$

CA 7.1.1.2 Anaerobic degradation

Report:	CA 7.1.1.2/1 Staudenmaier H., Kuhnke G., 2014 a Anaerobic soil metabolism of alpha-cypermethrin (BAS 310 I) 2013/1386602
Guidelines:	OECD 307 (2002), EPA OPPTS 835.4200
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

The anaerobic soil metabolism of alpha-cypermethrin (BAS 310 I) was investigated in a German loamy sand soil (according to the German soil classification scheme DIN) in the laboratory. The nominal application rate was 0.3 mg ^{14}C-labeled alpha-cypermethrin per kg dry soil. Two different labeling positions, benzyl-U-^{14}C and cyclopropane-1-^{14}C, were tested in separate experiments. A mixture of ^{14}C- and ^{13}C-labeled test items (1/1) was applied at an exaggerated rate to obtain samples allowing for the identification of unknown metabolites. After application, the treated soil was incubated in the dark under aerobic conditions at 20°C at a moisture content close to pF 2.5. A closed incubation system with continuous aeration was used.

After 9 days (approximately one half-life of the test item) the soil was covered by a water layer of about 1-2 cm depth and the incubation system was flushed with nitrogen in order to establish anaerobic conditions. The establishment of anaerobic conditions was monitored by measuring the oxygen content, the redox potential, and the pH of the test system at each sampling time from 9 days after treatment (DAT) onwards. The degradation of the test item was followed until 120 DAT. Samples were taken at 0, 1, 3, 6, 8, 10, 14, 27, 45, 62, 90 and 120 DAT. Volatiles were collected in traps attached to the incubation system.

The soil samples were extracted four times with acetonitrile/water and twice with acetonitrile. During the anaerobic phase for the first extraction, acetonitrile was added to the soil-water-slurry to obtain an acetonitrile/water ratio of 1/1 (v/v). This extraction was followed by three extractions with acetonitrile/water (7/3, v/v) and two extractions with acetonitrile. The individual extracts were analyzed by means of liquid scintillation counting (LSC) and the combined extracts by means of High Performance Liquid Chromatography (HPLC). The remaining soil after extraction was combusted in order to determine the amount of non-extractable residues. The latter were further characterized by subsequent NaOH treatment of samples from 3 DAT onwards.

The amount of parent compound decreased continuously from 95.1% of the total applied radioactivity (TAR) and 99.5% TAR at the beginning to 13.0% TAR and 11.1% TAR at 120 DAT with the benzyl and cyclopropane label, respectively. With each label, one prominent label specific metabolite was detected: M310I011 (3-PBA) with the benzyl label and M310I001 (DCVA) with the cyclopropane label. M310I011 increased from 1.5% TAR at day 0 to 30.0% TAR at 120 DAT (end of incubation). M310I001 increased from 0.9% TAR at day 0 to 55.6% TAR at 120 DAT. The label specific metabolite M310I013 was detected with the benzyl label up to 2.2% TAR. Two further metabolites were detected with both labels: M310I022 and M310I017. M310I022 was found from 62 DAT onwards with maximal amounts of 2.9% TAR for the benzyl label and 4.3% TAR for the cyclopropane label at 120 DAT. M310I017 was found from 1 DAT onwards. It reached its maximum at 8 DAT with 7.0% TAR for both the benzyl and the cyclopropane label and was found in amounts of 0.9% TAR (benzyl label) and 1.1% TAR (cyclopropane label) at the end of incubation.

Formation of CO_2 (mineralization) was observed throughout the study reaching 8.7% TAR for the benzyl label and 7.4% TAR for the cyclopropane label after 120 days. No other volatile compounds were detected in significant amounts throughout the incubation period of 120 days.

Non-extractable radioactive residues (NER) were formed with maximum amounts of 31.9% TAR (benzyl label) and 15.1% TAR (cyclopropane label) after 120 days of incubation. The non-extractable radioactive residues were further characterized by fractionation into fulvic acids, humic acids, and humins. Upon extraction with NaOH roughly one fifth (cyclopropane label) to one third (benzyl label) of the NER were assigned to the humin fraction. The NaOH released radioactivity was attributed in a ratio of about 3:2 (benzyl label) and 2:3 (cyclopropane label) to the humic acid and the fulvic acid fraction, respectively.

Chiral analysis of selected extracts showed that both enantiomers of alpha-cypermethrin were degraded at a similar rate. The ratio between the two enantiomers of alpha-cypermethrin was only very slightly shifted towards the R-enantiomer over time.

Kinetic analyses and calculations of DT_{50} and DT_{90} values were performed following the recommendations of the FOCUS Kinetics workgroup. For alpha-cypermethrin, $DegT_{50}$ values accounted for 48.7 and 48.1 days, the $DegT_{90}$ values for 280.0 and 159.7 days for the benzyl (FOMC) and for the cyclopropane label (SFO), respectively. For the metabolite M310I017 the DT_{50} was calculated to 36.1 and 26.5 days, the DT_{90} to 120.0 and 169.3 days for the benzyl (SFO) and the cyclopropane label (FOMC), respectively. No decline was observed for the metabolites M310I001 and M310I011, and consequently no DT_{50} values could be derived from this study.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

BAS code:	BAS 310 I (alpha-cypermethrin)
Reg. No.:	4078193
Chemical name (IUPAC):	racemate of (S)-cyano-3-phenoxybenzyl (1R,3R) 3 (2,2 dichlorovinyl)-2,2-dimethyl cyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S) 3 (2,2 dichlorovinyl) 2,2 dimethyl cyclopropanecarboxylate
Molar mass:	416.3033 g mol^{-1} (unlabeled)

Benzyl-label

Position of radiolabel:	benzyl-U-^{14}C
Specific radioactivity:	4.64 MBq mg^{-1}
Chemical purity:	93.4%
Radiochemical purity:	97.5% (radio-HPLC)
Batch No.:	775-0501

Cyclopropane-^{14}C-label

Position of radiolabel:	cyclopropane-1-^{14}C
Specific radioactivity:	4.7 MBq mg^{-1}
Chemical purity:	95.8%
Radiochemical purity:	98.9% (radio-HPLC)
Batch No.:	986-2015

Phenoxy-^{13}C-label

Position of radiolabel:	Phenoxy-1,2,3,4,5,6-^{13}C
Chemical purity:	95.1%
Batch No.:	1025-1034

Cyclopropane-^{13}C-label

Position of radiolabel:	cyclopropane-1-^{13}C
Chemical purity:	97.7%
Batch No.:	990-1024

2. Soil

The soil used (LUFA 5M) was a loamy sand soil (according to the German soil classification scheme DIN). After sampling from the field, the soil was allowed to dry at room temperature for one day and then sieved though a 2 mm sieve before use. After moistening to about 10 g per 100 g moist soil, the soil was stored at about 4°C. Prior to the application, the soil was acclimatized at ambient temperature, adjusted to a soil moisture of approximately 40% of the maximum water holding capacity (MWHC) and stored in the incubation chamber (20°C, dark) until application. The soil characteristics are summarized in Table 7.1.1.2-1.

Table 7.1.1.2-1: Characteristics of soil LUFA 5M

Designation	**LUFA 5M**
Origin	Mechtersheim, Rhineland-Palatinate, Germany
DIN particle size distribution [%]	
sand 0.063 – 2 mm	52.4
silt 0.002 – 0.063 mm	34.6
clay < 0.002 mm	13.0
textural class	loamy sand
USDA particle size distribution [%]	
sand 0.050 – 2 mm	59.7
silt 0.002 – 0.050 mm	27.4
clay < 0.002 mm	13.0
textural class	sandy loam
Total organic carbon [%]	2.03
CEC [$cmol^+ kg^{-1}$]	11.4
pH ($CaCl_2$) [-]	7.3
pH (H_2O) [-]	7.9
MWHC [g 100 g^{-1} dry soil]	27.0
Water retention at pF 2.0 [$g_{soil\ moisture}\ g^{-1}$ dry soil]	0.158
Water retention at pF 2.5 [$g_{soil\ moisture}\ g^{-1}$ dry soil]	0.150
Bulk density [kg L^{-1}]	1348
Microbial biomass [mg C 100 g^{-1} dry soil]	24.0

MWHC = Maximum water holding capacity

B. STUDY DESIGN

1. Experimental conditions

Generally, four applications were performed. Two applications with ^{14}C-benzyl-labeled and ^{14}C-cyclopropane-labeled test item were both performed at a nominal concentration of 0.3 mg a.s. per kg dry soil. To obtain samples allowing for the identification of unknown metabolites, soil was also treated at an exaggerated rate of about 3 mg test item per kg dry soil in additional experimental setups. The application solutions contained 1:1 mixtures of ^{14}C- and ^{13}C-labeled test item. Selected samples were worked up and the extracts subjected to LC-MS/MS for mass spectrometric identification of metabolites.

Prior to treatment with test item, the soil was checked for moisture content and re-adjusted to approximately 40% MWHC. Portions of 100 g soil (dry weight basis) were then filled into test vessels. The samples were incubated in the dark at a temperature of 20 ± 2°C and were connected to a continuous flow-through (CO_2-free synthetic air) system with traps for the collection of volatile organics (ethylene glycol and 0.5 M H_2SO_4) and CO_2 (0.5 M NaOH) during the aerobic phase of the incubation.

Test vessels containing the treated soil samples were switched to anaerobic conditions after 9 days of aerobic incubation by flooding each test vessel with 50 mL deionized water (forming a 1-2 cm water layer over the soils) and changing the flow-through gas to nitrogen instead of synthetic air. All incubation vessels were further kept in the dark at 20 ± 2ºC. During anaerobic incubation, the redox potentials (soil and water phases), the pH, and the dissolved oxygen in the water layer were determined at each sampling interval. Throughout the study, the water content of the soil was monitored periodically by weighing, and water was added when needed.

2. Sampling

Sampling times were 0 (no sampling of volatiles), 1, 3, 6, 8, 10, 14, 27, 45, 62, 90 and 120 days after treatment (DAT) for the soils treated at 0.3 mg a.s. kg^{-1} application rate.

At each sampling time two replicate vessels with soil were sampled (three replicate samples at 0, 62 and 120 DAT). At 10 DAT (benzyl label) and 1 DAT (cyclopropane label), reserve samples for were worked up like the regular samples. Moreover, at each sampling time except day 0, the flasks of the corresponding trapping system for volatiles were sampled as well and replaced with flasks containing fresh trapping solutions.

For the soils treated at the higher application rate of 3 mg a.s. kg^{-1} one replicate of each trial was sampled at 9, 35, 84 and 127 DAT. The trapping flasks were replaced as described above.

3. Description of analytical procedures

Aerobic incubation phase
Prior to work-up, the moisture of the samples was re-adjusted to the original content (close to pF 2.5). Soil samples were consecutively extracted with 4 x 100 mL acetonitrile/water (7/3, v/v) and 2 x 100 mL acetonitrile on a laboratory shaker (30 min). After each extraction step, the sample was centrifuged (15 min). Aliquots of each supernatant were measured by LSC.

Anaerobic incubation phase
For extraction during the anaerobic phase, the soil water slurry was extracted by adding acetonitrile to obtain an acetonitrile/water ratio of 1/1 (v/v). The slurry was extracted on a laboratory shaker for 30 min, the sample was centrifuged and the supernatant transferred into a 100 mL volumetric flask. Further extractions with 3 x 100 mL of acetonitrile/water (7/3, v/v) and 2 x 100 mL of acetonitrile followed. Aliquots of each extract were measured by LSC.

For HPLC, the acetonitrile/water and acetonitrile extracts were combined, concentrated to dryness by means of a rotary evaporator (40°C) and subjected to radio-HPLC.

To determine the amount of non-extractable residues, the extracted soil was dried in a drying oven at 60°C and aliquots thereof were combusted. The evolved $^{14}CO_2$ was measured by LSC.

The fraction of NER was further characterized by NaOH extractions (0.5 M NaOH, three times) of selected samples (from 3 DAT to 120 DAT). NaOH extracts were analyzed by LSC and processed to determine the distribution of radioactivity to the humin, humic, and fulvic acid fraction. The humic acid fraction and the fulvic acid fraction were analyzed by LSC. The fulvic acid fraction was further partitioned with ethyl acetate. Each ethyl acetate and water phase was measured by LSC. The ethyl acetate phases were combined, concentrated, and analyzed by LSC and HPLC. The remaining radioactivity in the soil residues was determined by combustion analyses (humin fraction). Further details are given in the report.

Aliquots of the ethylene glycol, 0.5 M H_2SO_4 or 0.5 M NaOH trapping solutions were subjected to LSC measurement.

Soil samples treated with an exaggerated rate of test item were worked up as described above for samples with a lower application rate. Combined soil extracts from 9, 35 and 84 DAT (benzyl-^{14}C- and phenoxy-^{13}C-labeled test item) and from 9 and 35 DAT (cyclopropane-^{14}C- and cyclopropane-^{13}C-labeled test item) were subjected to LC-MS/MS analysis.

4. Calculation of the degradation rate

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints). Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values were performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006) "Guidance Document on Estimating Persistence and Degradation Kinetics from Environmental Fate Studies on Pesticides in EU Registration" Report of the FOCUS Work Group on Degradation Kinetics, EC Document Reference Sanco/10058/2005 version 1.0, November 2011, 436 pp*].

The software package KinGUII (version 2.2014.224.1704) was used for parameter fitting [*SCHÄFER, D., MIKOLASCH, M., RAINBIRD, P., HARVEY, B. (2007)*; *WITT, J., GAO, Z., MEYER, H. (2014)*]. The error tolerance and the number of iterations of the optimization tool were set to 10^{-6} and 100, respectively.

For each data set, the kinetic models proposed by the FOCUS Kinetics guidance document were tested in order to identify the best-fit model. The recommended kinetic models, i.e. the single first order kinetics (SFO), the Gustafson-Holden model (FOMC), and bi-exponential (DFOP) kinetics as already implemented in KinGUI. The Goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test).

II. RESULTS AND DISCUSSION

A. TEST CONDITIONS

From 9 DAT to the end of the experiment the redox potential in the soil layer and the water phase of both experiments (exaggerated and lower rate) decreased continuously ranging from 161.6 to 270.9 mV at 9 DAT to a range of -178.7 to -350.8 mV at 120 DAT. The O_2 content in the water phase diminished quickly from 6.41/5.77 mg L^{-1} at 9 DAT to 0.34/0.50 mg L^{-1} at 10 DAT. The pH of the water phase increased during the experiment from 8.13/8.10 to 9.18 in both experimental setups. The development of these parameters demonstrated that anaerobic conditions were established during the experiment.

B. MASS BALANCE

Overall recovery of radiolabeled material averaged 94.1% TAR for the benzyl label and 98.7% TAR for the cyclopropane label. The distribution of radioactivity in extractable residues (ERR), non-extractable residues (NER), CO_2, and other volatiles is shown in Table 7.1.1.2-2 and Table 7.1.1.2-3.

C. EXTRACTABLE AND BOUND RESIDUES

The amount of extractable radioactive residues decreased from its initial percentage of > 99% TAR at 0 DAT to 51.4% TAR (benzyl label) and to 73.9% TAR (cyclopropane label) at 120 DAT. The decrease of the ERR can be mainly attributed to the aerobic phase of the incubation.

The different extractability in the two radiolabels was mirrored in the formation of non-extractable residues. Significant amounts of NER were formed in the soil treated with the benzyl label which increased continuously to 31.9% TAR after 120 days of incubation. In the samples treated with the cyclopropane label, lower amounts of NER were observed which increased to a maximum of 15.1% TAR at 120 DAT.

Table 7.1.1.2-2: Distribution of radioactivity and material balance after application of benzyl-U-^{14}C-labeled alpha-cypermethrin to soil LUFA 5M [% TAR]

DAT	Water/acetonitrile (3/7)	Acetonitrile	ERR	CO_2	Other volatiles	NER	Mass balance
0	99.7	0.5	100.2	n.a.	n.a.	1.2	101.4
0	99.2	0.5	99.7	n.a.	n.a.	1.2	100.9
0 mean	**99.5**	**0.5**	**100.0**	**-**	**-**	**1.2**	**101.2**
1	93.3	0.6	93.9	0.4	0.0	4.7	99.0
3	86.1	0.6	86.7	1.1	0.0	10.2	98.0
6	77.3	0.5	77.8	1.4	0.0	17.2	96.4
8	66.8	0.5	67.3	2.6	0.0	21.5	91.4
10*	61.2	0.5	61.7	3.6	0.0	24.7	90.0
14	61.4	0.7	62.1	3.9	0.0	25.1	91.0
27	60.8	0.8	61.6	4.9	0.0	26.8	93.3
45	55.9	0.8	56.7	5.7	0.0	27.8	90.3
62	56.8	0.8	57.6	6.6	0.0	29.1	93.4
62	56.1	0.9	57.0	6.6	0.0	29.4	93.1
62 mean	**56.5**	**0.9**	**57.3**	**6.6**	**0.0**	**29.3**	**93.3**
90	55.4	1.2	56.6	7.8	0.0	29.6	94.0
120	51.1	1.0	52.0	8.7	0.0	32.0	92.8
120	49.7	1.0	50.7	8.7	0.0	31.8	91.1
120 mean	**50.4**	**1.0**	**51.4**	**8.7**	**0.0**	**31.9**	**91.9**

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
n.a. = Not analyzed
* Work-up of reserve sample

Table 7.1.1.2-3: Distribution of radioactivity and material balance after application of cyclopropane-1-^{14}C-labeled alpha-cypermethrin to soil LUFA 5M [% TAR]

DAT	Water/acetonitrile (3/7)	Acetonitrile	ERR	CO_2	Other volatiles	NER	Mass balance
0	102.5	0.5	103.1	n.a.	n.a.	1.3	104.4
0	102.0	0.5	102.5	n.a.	n.a.	1.3	103.8
0 mean	**102.3**	**0.5**	**102.8**	**-**	**-**	**1.3**	**104.1**
1*	98.3	0.6	98.9	0.3	0.0	4.3	103.5
3	93.7	0.7	94.4	1.0	0.0	7.1	102.6
6	85.1	0.8	85.8	2.6	0.0	11.2	99.7
8	80.1	0.7	80.8	3.2	0.0	14.1	98.1
10	76.9	0.8	77.7	3.5	0.0	15.7	96.9
14	74.0	0.8	74.8	3.6	0.0	15.6	94.1
27	78.1	0.9	79.0	4.3	0.0	15.8	99.1
45	74.9	0.8	75.7	5.1	0.0	15.4	96.2
62	74.7	0.8	75.5	5.7	0.0	14.8	96.1
62	77.2	0.8	78.0	5.7	0.0	15.2	99.0
62 mean	**75.9**	**0.8**	**76.7**	**5.7**	**0.0**	**15.0**	**97.5**
90	74.4	0.8	75.2	6.6	0.0	14.0	95.9
120	71.7	0.9	72.6	7.4	0.0	15.0	95.0
120	74.4	0.9	75.2	7.4	0.0	15.2	97.9
120 mean	**73.0**	**0.9**	**73.9**	**7.4**	**0.0**	**15.1**	**96.4**

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
n.a. = Not analyzed
* Work-up of reserve sample

D. VOLATILISATION

Formation of CO_2 (mineralization) was observed throughout the study reaching 8.7% TAR for the benzyl label and 7.4% TAR for the cyclopropane label after 120 days. Although mineralization was observed in both, the aerobic and the anaerobic phases of incubation, the formation of CO_2 markedly slowed down during the anaerobic phase. No other volatile compounds were detected in significant amounts throughout the incubation period of 120 days.

E. TRANSFORMATION OF PARENT COMPOUND

The summarized results of radio-HPLC analyses are presented in Table 7.1.1.2-4 and Table 7.1.1.2-5.

The extracted amount of parent compound decreased from 95.1% TAR and 99.5% at the beginning to 13.0% TAR and 11.1% TAR at 120 DAT with the benzyl and cyclopropane labels, respectively. The degradation was fast during the aerobic phase of the experiment, reaching 48.9% TAR for the benzyl label and 52.7% TAR for the cyclopropane label at 10 DAT. However, it slowed down during the anaerobic phase.

With each label, one prominent label specific metabolite was detected: M310I011 (3-PBA) with the benzyl label and M310I001 (DCVA) with the cyclopropane label. Both metabolites occurred already at 0 DAT, and their concentrations increased further until 120 DAT, seeming to reach a plateau. The concentration of M310I011 increased from 1.5% TAR at day 0 to 30.0% TAR at 120 DAT (end of incubation). The concentration of M310I001 increased from 0.9% TAR at day 0 to 55.6% TAR at 120 DAT. The label specific metabolite M310I013 was first detected at 27 DAT (benzyl label) and found in concentrations up to 2.2% TAR.

Two further metabolites were detected with both labels: M310I022 and M310I017. M310I022 was found from 62 DAT onwards with maximum amounts of 2.9% TAR (benzyl label) and 4.3% TAR (cyclopropane label) at 120 DAT. M310I017 was found from 1 DAT onwards. It reached its maximum at 8 DAT with 7.0% TAR for both the benzyl and cyclopropane label and was found in amounts of 0.9% TAR (benzyl label) and 1.1% TAR (cyclopropane label) at the end of incubation.

Table 7.1.1.2-4: Radio HPLC analysis of combined acetonitrile/water + acetonitrile extracts of soil LUFA 5M treated with benzyl-U-^{14}C-labeled alpha-cypermethrin [% TAR]

DAT	Extract	M310I013	M310I011	M310I022	M310I017	Alpha-cypermethrin	Uk	Uk	Sum others*
		t_R~42.0‘	t_R~64.8‘	t_R~73.9‘	t_R~74.2‘	t_R~76.8‘	t_R~78.8‘	t_R~79.2‘	
0	100.2	n.d.	1.6	n.d.	n.d.	95.6	2.1	0.9	n.d.
0	99.7	n.d.	1.5	n.d.	n.d.	94.7	2.1	1.5	n.d.
0 mean	**100.0**	**-**	**1.5**	**-**	**-**	**95.1**	**2.1**	**1.2**	**-**
1	93.9	n.d.	1.5	n.d.	2.1	85.8	2.7	1.8	n.d.
3	86.7	n.d.	2.3	n.d.	4.7	75.5	2.1	1.3	0.8
6	77.8	n.d.	2.6	n.d.	6.6	63.7	2.3	1.5	1.2
8	67.3	n.d.	2.5	n.d.	7.0	51.6	2.1	2.5	1.5
10**	61.7	n.d.	3.2	n.d.	5.7	48.9	1.6	0.9	1.3
14	62.1	n.d.	6.1	n.d.	5.7	46.3	2.2	1.2	0.7
27	61.6	1.0	14.6	n.d.	4.0	37.8	2.5	1.5	0.3
45	56.7	1.0	21.8	n.d.	2.6	28.4	1.4	1.3	0.2
62	57.6	1.6	26.7	0.6	2.1	23.8	1.6	1.3	0.0
62	57.0	1.5	26.1	0.8	2.2	23.1	1.8	1.4	0.0
62 mean	**57.3**	**1.6**	**26.4**	**0.7**	**2.2**	**23.5**	**1.7**	**1.3**	**0.0**
90	56.6	1.8	29.3	1.7	1.4	20.2	1.6	0.6	0.0
120	52.0	2.1	30.9	2.1	0.9	13.8	1.5	0.8	0.0
120	50.7	2.4	29.2	3.6	0.8	12.2	1.4	1.0	0.0
120 mean	**51.4**	**2.2**	**30.0**	**2.9**	**0.9**	**13.0**	**1.4**	**0.9**	**0.0**

TAR = Total applied radioactivity
DAT = Days after treatment
Uk = Unknown
t_R = retention time [min]
n.d. = Not detected
* Each peak below or equal to 1.0% TAR
** Work-up of reserve sample

Table 7.1.1.2-5: Radio HPLC analysis of combined acetonitrile/water + acetonitrile extracts of soil LUFA 5M treated with cyclopropane-1-^{14}C-labeled alpha-cypermethrin [% TAR]

DAT	Extract	M310I001	M310I022	M310I017	Alpha-cypermethrin	Uk	Uk	Sum others*
		t_R~65.7‘	t_R~73.9‘	t_R~74.2‘	t_R~76.8‘	t_R~78.8‘	t_R~79.2‘	
0	103.1	0.8	n.d.	n.d.	100.5	1.1	0.7	n.d.
0	102.5	1.0	n.d.	n.d.	98.6	1.3	1.6	n.d.
0 mean	**102.8**	**0.9**	**-**	**-**	**99.5**	**1.2**	**1.2**	**-**
1**	98.9	1.7	n.d.	2.0	93.0	1.1	1.0	n.d.
3	94.4	5.1	n.d.	4.6	82.6	0.9	0.4	0.8
6	85.8	10.4	n.d.	6.9	65.6	0.5	0.9	1.6
8	80.8	11.5	n.d.	7.0	57.8	1.2	1.4	1.9
10	77.7	15.9	n.d.	5.9	52.7	0.9	1.2	1.2
14	74.8	20.5	n.d.	5.5	46.0	0.8	0.6	1.3
27	79.0	33.0	n.d.	3.6	39.6	1.0	1.3	0.5
45	75.7	40.9	n.d.	2.8	29.7	0.8	0.6	0.9
62	75.5	48.2	0.7	1.9	22.9	0.7	0.4	0.6
62	78.0	49.0	0.7	1.8	24.0	0.9	0.7	0.7
62 mean	**76.7**	**48.6**	**0.7**	**1.9**	**23.5**	**0.8**	**0.6**	**0.7**
90	75.2	53.6	3.0	1.3	16.5			0.8
120	72.6	54.6	3.6	0.8	11.7	0.6	0.4	0.9
120	75.2	56.6	4.9	1.3	10.5	0.5	0.5	1.0
120 mean	**73.9**	**55.6**	**4.3**	**1.1**	**11.1**	**0.5**	**0.4**	**0.9**

TAR = Total applied radioactivity
DAT = Days after treatment
Uk = Unknown
n.d. = Not detected
* Each peak below or equal to 1.0% TAR
** Work-up of reserve sample

Exclusion of trans-isomers of cypermethrin

Selected extracts (benzyl and cyclopropane label; 0, 8, 62 and 120 DAT) were investigated using a second reverse-phase HPLC system for the presence of trans isomers of cypermethrin. No noticeable amounts of trans isomers of cypermethrin were detected in the soil extracts, i.e. no conversion of the cis isomers to trans isomers occurred in the soil. The detailed results are presented in the report.

Investigation of the isomerization of cis isomers

Alpha-cypermethrin is a mixture of two enantiomers. In order to investigate the ratio of enantiomers over time a chiral HPLC method was used to analyze soil extracts from 0, 8, 27, 62 and 120 DAT. The data showed that both enantiomers of alpha-cypermethrin were degraded at a similar rate. The ratio between the two enantiomers of alpha-cypermethrin was only very slightly shifted towards the R-enantiomer over time. The detailed results are presented in the report.

F. CHARACTERIZATION OF NON-EXTRACTABLE RESIDUES (NER)

From 3 DAT to 120 DAT, NER were further characterized by NaOH treatment and subsequent fractionation into fulvic acids, humic acids, and humins. Results are shown in Table 7.1.1.2-6 and Table 7.1.2.1.1-7.

Table 7.1.1.2-6: Characterization of non-extractable residues in soil treated with benzyl-U-^{14}C-alpha-cypermethrin [% TAR]

Days after treatment	Non-extractable residues	NaOH solutions	Fulvic acids			Humic acids	Humins
			Total	Ethyl acetate soluble	Acidic water soluble		
3	10.2	7.4	2.9	1.4	1.4	4.3	2.9
6	17.2	12.3	4.7	2.0	2.5	7.8	5.1
8	21.5	15.5	5.5	2.4	3.1	9.7	6.4
10	24.7	17.7	6.1	2.5	3.3	10.9	7.9
14	25.1	17.6	5.5	2.6	3.0	11.2	8.3
27	26.8	18.7	6.4	3.2	3.1	11.6	8.5
45	27.8	19.1	6.5	3.5	2.9	12.3	9.1
62*	29.1	19.6	6.9	3.8	2.7	12.5	9.3
90	29.6	21.1	7.9	5.0	2.8	12.2	9.9
120*	32.0	21.1	8.8	5.8	2.8	11.5	10.4

TAR = Total applied radioactivity
* Replicate 1

Table 7.1.2.1.1-7: Characterization of non-extractable residues in soil treated with cyclopropane-1-^{14}C-alpha-cypermethrin [% TAR]

Days after treatment	Non-extractable residues	NaOH solutions	Fulvic acids			Humic acids	Humins
			Total	Ethyl acetate soluble	Acidic water soluble		
3	7.1	6.0	3.9	2.5	1.3	1.9	1.3
6	11.2	9.6	5.7	3.7	1.9	3.5	2.1
8	14.1	11.7	7.0	4.7	2.2	4.4	2.7
10	15.7	13.0	7.2	5.2	2.3	5.2	3.1
14	15.6	12.7	7.1	5.1	2.2	5.2	3.1
27	15.8	12.4	6.9	4.8	2.1	5.0	3.4
45	15.4	11.7	6.5	4.7	1.9	4.5	3.3
62*	15.2	11.3	6.5	4.7	1.8	4.4	3.4
90	14.0	11.2	6.9	5.0	1.9	4.1	3.5
120**	15.0	11.2	6.5	4.7	1.6	4.2	3.6

TAR = Total applied radioactivity
* Replicate 2
** Replicate 1

G. KINETIC MODELING RESULTS

The kinetic evaluation was performed according to FOCUS in order to derive trigger endpoints for alpha-cypermethrin and its degradation products. As a decline of residues was observed only for M310I017, it was the only metabolite which could be included in the kinetic evaluation. The kinetic analysis was based on the decline of the measured residues under anaerobic incubation conditions. Trigger endpoints for the metabolites M310I001 and M310I011 could not be estimated since no decline of the measured residues was observed.

The resulting $DegT_{50}$ and $DegT_{90}$ values for alpha-cypermethrin and the DT_{50} and DT_{90} values for M310I017 are summarized in Table 7.1.1.2-8 and Table 7.1.1.2-9.

Table 7.1.1.2-8: Trigger endpoints for alpha-cypermethrin

Compound	Label	Kinetic model	χ^2 error	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Alpha-cypermethrin	Benzyl	FOMC	2.8	48.7	280.0
	Cyclopropane	SFO	2.9	48.1	159.7

Table 7.1.1.2-9: Trigger endpoints for metabolites of alpha-cypermethrin

Compound	Label	Kinetic model	χ^2 error	DT_{50} [d]	DT_{90} [d]
M310I017	Benzyl	SFO[a]	5.4	36.1	120.0
	Cyclopropane	FOMC[a]	3.8	26.5	169.3
M310I001	-	-	-	Derivation of endpoints not possible	
M310I011		-	-	Derivation of endpoints not possible	

[a] Evaluation of metabolite decline

III. CONCLUSION

Alpha-cypermethrin degraded fast under aerobic conditions followed by a slightly slower degradation under anaerobic conditions. Significant amounts of non-extractable radioactive residues were formed in the soil treated with the benzyl label which increased continuously to 31.9% TAR after 120 days of incubation. In the samples treated with the cyclopropane label, lower amounts of non-extractable radioactive residues were observed which increased to a maximum of 15.1% TAR at 120 DAT. $^{14}CO_2$ was formed in moderate amounts up to 8.7% TAR. No other volatile compounds were detected in significant amounts.

Several metabolites were formed. The most prominent label specific metabolites were M310I011 (benzyl label) and M310I001 (cyclopropane label) reaching 30.0% TAR and 55.6% TAR, respectively at the end of the study. The metabolites M310I017 and M310I022 were found with both labels at up to 7.0% TAR and 4.3% TAR, respectively.

Using a chiral HPLC method it was shown that both enantiomers of alpha-cypermethrin were degraded at a similar rate.

$DegT_{50}$ values for the benzyl and the cyclopropane labeled alpha-cypermethrin were calculated to be 48.7 and 48.1 days and $DegT_{90}$ values were estimated to be 280.0 and 159.7 days according to the best fit kinetics. For the metabolite M310I017, best fit kinetic calculations resulted in DT_{50} values of 36.1 and 26.5 days and DT_{90} values of 120.0 and 169.3 days. No decline was observed for the metabolites M310I001 and M310I011 and therefore no DT_{50} values could be derived from this study.

The RMS BE has asked for additional kinetic evaluations on 27. Oct. 2016. The results are presented and discussed below.

Report:	CA 7.1.1.2/1 Anonymous, 2016 a Response to RMS BE - Request concerning kinetic re-evaluations – Alpha-cypermethrin 2016/1324158
Guidelines:	none
GLP:	no

RMS: RMS notes that the Notifier performed a separate kinetic assessment for each label. In peer review, it is common practice to handle residuals from the same soil with different radiolabelled positions as replicate values. Please Notifier consider that residue data from two tested labels from the same soil need to be processed as replicates for the active substance and its metabolites.

BASF response: A kinetic evaluation was performed by processing the residue data of the two tested labels as replicates. The results are summarized in Table 7.1.1.2-10 and the corresponding Kingui fits are presented in Appendix 2 in the original response document. The endpoints (DT_{50} = 46.8 d, DT_{90} = 222 d, FOMC model) are comparable with those derived by performing a separate kinetic assessment for each label (geomean DT_{50} = 48.4 d, geomean DT_{90} = 211.5 d). Therefore, the endpoints presented in the dossier are considered adequate.

Table 7.1.1.2-10: Statistical and visual assessment of kinetic models for degradation of alpha-cypermethrin in soil under anaerobic condition

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	3.0	k: <0.01	good	51.4	170.7
FOMC	1.9	β: <0.1	good	46.8	222.0
DFOP	2.0	k1: <0.01 k2: <0.01 g: <0.01	good	46.6	191.6

CA 7.1.1.3 Soil photolysis

Studies already submitted and peer reviewed:

1. Takahashi N. (1985), Photodegradation of the pyrethroid insecticide cypermethrin in water and on soil surfaces
Photodegradation of cypermethrin was investigated on three Japanese soils using cypermethrin labelled in the cyclopropyl- the cyano- and the benzyl position. Treated soils were exposed to natural sunlight for 7 days. No balance could be obtained and the soils were drying during the experiment, so that an influence on metabolite degradation could not be excluded. The study was not conducted according to OECD guideline and examples of raw data are missing. Though a lot of effort was undertaken, the study should not be included in the evaluation.

2. Hall, J.S. et al. (1981) Cypermethrin: Photodegradation on a soil surface
The test was carried out with mixtures of cis and trans cypermethrin isomers. Treated soil plates were exposed to natural sunlight during fine weather periods and protected with a polythene shelter during rainfall. The shelter allowed the transmission of UV radiation from sunlight. The study was rejected because of its poor quality in the evaluation.

3. Van Dijk A. et al. (1993) ^{14}C- Alpha-cypermethrin: Study of its photodegradation on soil
The study was performed with alpha-cypermethrin labelled in the benzyl ring only. The treated soil was exposed to artificial sunlight (wavelength above 290 nm) for 30 days during a 12 hour day and night cycle. Dark control samples were also analysed. The irradiated samples showed a DT_{50} of 31 days compared to 193 days of the dark control. The main metabolite detected was 3-PBA with traces of 3-PBAldehyde.

Table 7.1.1.3-1: List of soil photolysis studies in soil performed with (alpha-) cypermethrin

DocID	Parent compound	Soil	Application rate [mg kg^{-1}]	Incubation temperature	Irradiation period [hours]	Remark
CY-905-038	Cypermethrin	Light clay Sandy loam Sandy clay loam	1.1 µg/cm³	outside temperature	1 month (natural sunlight)	Takahashi et al. 1985 Literature
CY-620-007	Cypermethrin	Sandy loam	210 g/ha	outside temperature	32 (artificial sunlight)	Hall, J..S. 1981
AL-620-010	Alpha-cypermethrin	Sandy silty loam	4.0	n/a	12 (natural sunlight)	Van Dijk, Burri, 1991

n/a = not available

A new photolysis study of alpha-cypermethrin on soil was performed (Michel, A. Hassink, J. 2014), but the study of Van Dijk and Burri can be considered as additional information.

Report:	CA 7.1.1.3/1 Michel, A., Hassink, J., 2014 a Soil Photolysis of BAS 310 I 2014/1000642
Guidelines:	EPA 161-3, EPA 835.2410, EEC 91/414 Annex II, OECD Draft Guideline Phototransformation of Chemicals on Soil Surfaces (January 2002), SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 7.1.1.3/2 Hassink J., 2015 b Report Amendment No. 1 - Soil photolysis of BAS 310 I 2015/1107645
Guidelines:	EPA 161-3, EPA 835.2410, Draft OECD Guideline: Phototransformation of Chemicals in Soil Surfaces (Jan. 2002), SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

The amendment to the report contains KinGUI graphs of the parameter estimation section that were missing in the final report.

Executive Summary

A soil photolysis was conducted with cyclopropane-1-^{14}C-labeled-alpha-cypermethrin (cyclopropane label) and benzyl-^{14}C-labeled-alpha-cypermethrin (benzyl label) to investigate their behavior in soil under the influence of light.

The overall mean values for the material balance in the photolysis and the dark control were in the range of 95.1 - 118.8% of the total applied radioactivity (TAR). Carbon dioxide was the only volatile degradation product trapped. It reached 5.4% TAR after 15 days in the photolysis test and 7.6% TAR in the dark control for the cyclopropane label. For the benzyl label, it reached 1.9% TAR after 15 days in the photolysis test and 6.7% TAR in the dark control.

The sample extractability for the photolysis test differed from the dark control. At the end of the study, 15.0% TAR and 10.8% TAR of the cyclopropane and benzyl label, respectively, were not extractable from the illuminated soil samples.. At the end of the incubation of the dark control samples, about 12.4% TAR and 17.3% TAR of the cyclopropane and benzyl label, respectively, were non-extracted. The alkali-soluble radioactivity amounted to 11% TAR 15 days after treatment (DAT) in the photolysis and was further fractionated to distinguish between acid-insoluble humic acids and acid-soluble fulvic acids. The major part of the radioactivity could be assigned to the fulvic acid fraction (max. 8.4% TAR at 15 DAT for the cyclopropane label and 4.6% TAR at 15 DAT for the benzyl label). In the dark control samples, the amount of non-extractable residues reached 12.4% TAR and 17.3% TAR for the cyclopropane label and the benzyl label, respectively, after 15 days. It was confirmed that the alkali-soluble fraction consisted of radioactive material mainly assigned to the fulvic fraction (6.1% TAR at 15 DAT for the cyclopropane label and 5.9% TAR for the benzyl label).

The concentration of the cyclopropane label declined to 65.5% TAR in the course of the photolysis experiment under continuous irradiation and to 74.5% TAR in the dark control samples. The concentration of the benzyl label declined in the photolysis experiment to 64.1% TAR within 15 days and to 55.7% TAR in the dark control samples.

Aliquots of all samples were analyzed by radio-HPLC. One metabolite was observed in the photolysis study with the cyclopropane label as well as in the corresponding dark control samples. It accounted for 5.4% TAR at 15 DAT in the irradiated samples and reached a maximum of 10% TAR at 10 DAT in the dark control samples. This metabolite was also observed in the aerobic soil metabolism study [*CA 7.1.1.2, BASF DocID 2014/1000641*], and was identified by MS measurement as DCVA. Structure elucidation is fully described in the final report of the aerobic soil metabolism study. No other degradation products with more or equal 5% TAR occurred in the photolysis and the dark controls. No modification of the enantiomeric ratio and no formation of the trans isomer of alpha-cypermethrin were observed during the study.

Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values (non-GLP) for alpha-cypermethrin in soil was performed following the recommendations of the FOCUS Kinetics workgroup. The analysis was conducted by non-linear regression methods employing the software tool KinGUII. The following $DegT_{50}$ and $DegT_{90}$ values used as trigger endpoints were calculated (Table 7.1.1.3-2).

Table 7.1.1.3-2: **Trigger endpoints for alpha-cypermethrin**

Test system	Label	Best-fit model	χ^2 error	Trigger endpoints	
				$DegT_{50}$ [d]	$DegT_{90}$ [d]
Irradiated	Cyclopropane-1-^{14}C	SFO	3.2	29.7	98.6
	Benzyl-^{14}C-label	DFOP	3.7	27.6	119.8
Dark control	Cyclopropane-1-^{14}C	SFO	4.1	30.9	102.6
	Benzyl-^{14}C	DFOP	5.0	No reliable endpoints derived	

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

BAS-Code:	BAS 310 I (alpha-cypermethrin)
Registry No.:	4078193
Chemical name:	Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molecular formula:	$C_{22}H_{19}CI_2NO_3$
Molar mass:	416.3 g mol^{-1} (unlabeled)

Label 1 (cyclopropane label)

Label:	cyclopropane-1-^{14}C
Batch No.:	986-1046
Specific activity of a.s.:	4.9 MBq mg^{-1}
Radiochemical purity:	98.2%
Chemical purity:	93.6%

Label 2 (benzyl label)

Label:	benzylring-U-^{14}C
Batch No.:	775-0401
Specific activity of a.s.:	4.94 MBq mg^{-1}
Radiochemical purity:	96.1%
Chemical purity:	93.7%

2. Soil

The study was conducted with two soil samples classified as sandy loam (USDA) and originated from the Landwirtschaftliche Untersuchungs- und Forschungsanstalt (LUFA) Speyer, South Western Germany. The sampling depth was 0-20 cm. The soil was passed through a 2 mm sieve, remoistened to approximately 8-12% soil moisture and then stored at about 4°C in the dark no longer than three months before use. An overview of the soil parameters is listed in Table 7.1.1.3-3.

Table 7.1.1.3-3: Characteristics of soil LUFA 5M used for soil photolysis study with ^{14}C-alpha-cypermethrin

Soil designation	**LUFA 5M 12/1651/04** Germany (Origin LUFA Speyer)	**LUFA 5M 12/1651/05** Germany (Origin LUFA Speyer)
DIN 4220 Particle size distribution [%]		
sand 0.063 – 2 mm	52.4	54.1
silt 0.002 – 0.063 mm	34.6	31.7
clay < 0.002 mm	13.0	14.2
textural class	loamy sand	loamy sand
USDA Particle size distribution [%]		
sand 0.050 – 2 mm	59.7	58.4
silt 0.002 – 0.050 mm	27.4	27.4
clay < 0.002 mm	13.0	14.2
textural class	sandy loam	sandy loam
Organic C [%]	2.03	2.18
pH [H_2O]	7.9	8.0
pH [$CaCl_2$]	7.3	7.4
cation exchange capacity [$cmol^+ kg^{-1}$]	11.4	8.8
Max. water holding capacity [g $100g^{-1}$ dry weight]	27.0	26.2
microbial biomass (start of study) [mg C $100g^{-1}$ dry soil]	24.0	31.1

B. STUDY DESIGN

1. Experimental conditions

Twelve and ten small steel dishes (approximately 8.8 cm x 4.3 cm x 1.2 cm) were filled with soil for photolysis test and for dark control, respectively. The dishes, which were later treated with the test item, were arranged in a rectangular bowl with a connected thermostat. The temperature of the dishes used for photolysis was adjusted and controlled by an external unit (22°C ± 1°C) while the dishes for dark control were put into an incubator at 22°C ± 1°C. The light intensity for the photolysis test group was set to 3 mW cm^{-2} (UVA range).The bowl was closed airtight with a quartz glass covering and the whole incubation device was continuously aerated with CO_2-depleted (0.5 M NaOH) and remoistened air via an air inlet and outlet.

In order to trap potentially evolving volatiles (including $^{14}CO_2$) the emergent air was bubbled through three different trapping solutions located between dish and pump: 1. NaOH (0.5 M); 2. ethylene glycol; 3. H_2SO_4 (0.5 M).

The incubation bowl for photolysis was placed under a SUNTEST CPS plus (Atlas) equipped with a Xenon lamp emitting light with a sunlight similar spectrum at a light intensity of about 3 mW cm^{-2} (UVA range). This corresponds to a clear summer day in Southern Germany (about 49°N). Wavelengths < 290 nm were filtered off to simulate natural sunlight.

To maintain the temperature especially on the quartz glass surface in order to avoid a rapid drying of the soil surface, the air space between lamp and quartz glass within the SUNTEST device was cooled by an external apparatus (Yeti, Seveso). To maintain the initial water content as constant as possible, dishes were weighed at each incubation day and the evaporated water was replaced.

The amount of test item to be applied on the soil surface was calculated based on the field application rate of 30 g ha^{-1}. If a soil layer of 1 cm and bulk density of 1.5 kg L^{-1} is assumed, the application rate corresponds to about 0.2 mg test item kg^{-1} dry soil. Because of the low specific radioactivity of the test items and to ensure a better detection of possible degradation products, the amount of test item applied was increased to 0.3 mg test item per kg dry soil, which corresponds to 9 µg of test item per dish.

2. Sampling

The sampling dates were 0, 1, 3, 7, 10 and 15 days after treatment (DAT). Two vessels were taken at each sampling time from each photolysis test system and the dark control (with exception of DAT 0, where no dark control samples were taken). At each sampling time, the respective volatile trapping solutions were removed.

3. Description of analytical procedures

Each soil sample was consecutively extracted four times with 40 mL of acetonitrile/water (7:3, v/v) by mechanical shaking for 1 h. After each extraction step, solid and extract were separated by centrifugation and filtered. The four corresponding extracts were combined and measured by liquid scintillation counting (LSC).

After the last acetonitrile/water extraction, the soil residues were air-dried and homogenized by milling. Aliquots of each sample were combusted in a sample oxidizer. Trapped $^{14}CO_2$ was analyzed by LSC.

All extracts were analyzed by radio-HPLC to determine the metabolite pattern. Prior to injection, the solvent of the extracts was completely evaporated by a rotary evaporator at 35°C. Then the residues were redissolved in a defined volume of the respective extraction solvent and subjected to HPLC analysis. Separation of cis and trans isomers, as well as enantiomers of alpha-cypermethrin was performed on two separate HPLC systems.

Non-extractable residues (NER) were found in amounts of 5-10% TAR at sampling days 3, 7, 10, and 15 in the photolysis experiment. The NER were further characterized by NaOH extraction. Samples were extracted three times with 0.5 M NaOH on a rotary shaker (270 rpm) and twice washed with water. Aliquots of the NaOH and the water extracts were analyzed by LSC. NaOH extracts and water extracts were pooled, representing together the fulvic and humic acid fraction.

Acidic precipitation (pH 1) of the humic acids from the pooled extracts was performed. After centrifugation, supernatants (fulvic acids) were analyzed by LSC. The precipitates were redissolved and measured by LSC.

The remaining soil samples after NaOH and water extraction were air-dried at room temperature, aliquots were combusted, and the formed $^{14}CO_2$ was trapped and analyzed by LSC to determine the ^{14}C residues in the humin fraction.

4. Calculation of the degradation rate

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints). Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006)*]. The software package KinGUII (version 2.2014.224.1704) was used for parameter fitting [*Witt et al. (2014)*]. The error tolerance and the number of iterations of the optimization tool were set to 0.00001 and 100, respectively.

For each data set, the kinetic models proposed by the FOCUS Kinetics guidance document were tested to identify the best-fit model. The recommended kinetic models, i.e. the single first order kinetics (SFO) and the Gustafson-Holden model (FOMC), are already implemented in KinGUI. The Goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test) [for details see *Chapter 6.3 in FOCUS (2006)*].

Replicate measurements were considered for the parameter estimation. The initial concentration of the applied test item was set to the material balance recovered at day 0. The χ^2 value was calculated for the kinetic model as recommended by FOCUS, considering the average of replicate measurements. For the individual parameters, the t-test statistics were based on number of individual measurements.

II. RESULTS AND DISCUSSION

A. MASS BALANCE

Total recoveries of radioactivity extracted from soil are summarized in Table 7.1.1.3-4 and Table 7.1.1.3-5. The overall values for the mean mass balance in the photolysis and in the dark control were for both labels in the range of 95.1 - 118.8% TAR.

Table 7.1.1.3-4: Material balance and distribution of radioactivity for the photolysis and the dark control test with cyclopropane-1-^{14}C-labeled alpha-cypermethrin [% TAR]

DAT	Photolysis test				Dark control test			
	ERR	NER	Volatiles*	Material balance	ERR	NER	Volatiles*	Material balance
0 I**	96.0	1.4	n.a.	97.4	96.0	1.4	n.a.	97.4
0 II**	100.7	1.9	n.a.	102.6	100.7	1.9	n.a.	102.6
0 mean**	**98.3**	**1.7**	n.a.	**100.0**	**98.3**	**1.7**	n.a.	**100.0**
1 I	105.8	4.5	0.7	111.0	109.1	5.0	0.8	115.0
1 II	103.2	4.5	0.7	108.4	108.0	4.8	0.8	113.6
1 mean	**104.5**	**4.5**	**0.7**	**109.7**	**108.6**	**4.9**	**0.8**	**114.3**
3 I	91.3	6.2	1.5	98.9	96.5	7.6	2.0	106.1
3 II	130.4	7.0	1.5	138.8	99.3	7.7	2.0	109.0
3 mean	**110.8**	**6.6**	**1.5**	**118.8**	**97.9**	**7.7**	**2.0**	**107.5**
7 I	93.9	9.0	2.7	105.6	95.0	9.4	3.9	108.3
7 II	94.4	8.2	2.7	105.2	95.3	9.8	3.9	109.0
7 mean	**94.2**	**8.6**	**2.7**	**105.4**	**95.2**	**9.6**	**3.9**	**108.6**
10 I	95.8	10.6	3.7	110.2	100.9	12.5	5.3	118.7
10 II	97.0	12.6	3.7	113.3	101.3	11.8	5.3	118.4
10 mean	**96.4**	**11.6**	3.7	**111.7**	**101.1**	**12.2**	**5.3**	**118.5**
15 I	93.8	15.7	5.4	114.9	91.3	12.9	7.6	111.8
15 II	91.4	14.2	5.4	111.0	89.9	11.9	7.6	109.5
15 mean	**92.6**	**15.0**	**5.4**	**113.0**	**90.6**	**12.4**	**7.6**	**110.6**

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
n.a. = Not analyzed
* No other volatiles than CO_2 were found
** Day 0 samples of the dark control are the day 0 samples of the photolysis test

Table 7.1.1.3-5: Material balance and distribution of radioactivity for the photolysis and the dark control test with benzyl-^{14}C-labeled alpha-cypermethrin [% TAR]

DAT	Photolysis test				Dark control test			
	ERR	NER	Volatiles*	Material balance	ERR	NER	Volatiles*	Material balance
0 I**	97.2	1.8	n.a.	99.1	97.2	1.8	n.a.	99.1
0 II**	98.9	2.0	n.a.	100.9	98.9	2.0	n.a.	100.9
0 mean**	**98.1**	**1.9**	**n.a.**	**100.0**	**98.1**	**1.9**	**n.a.**	**100.0**
1 I	97.7	4.1	0.2	101.9	95.7	4.7	0.4	100.7
1 II	96.8	4.3	0.2	101.3	88.8	5.0	0.4	94.2
1 mean	**97.2**	**4.2**	**0.2**	**101.6**	**92.2**	**4.9**	**0.4**	**97.5**
3 I	96.7	6.2	0.4	103.3	89.6	9.1	1.4	100.1
3 II	94.3	5.6	0.4	100.3	89.4	9.4	1.4	100.2
3 mean	**95.5**	**5.9**	**0.4**	**101.8**	**89.5**	**9.3**	**1.4**	**100.2**
7 I	91.1	8.1	0.8	100.1	83.0	14.0	3.3	100.3
7 II	91.2	7.7	0.8	99.8	81.1	11.8	3.3	96.2
7 mean	**91.2**	**7.9**	**0.8**	**99.9**	**82.0**	**12.9**	**3.3**	**98.3**
10 I	89.4	8.8	1.3	99.5	76.2	14.3	4.7	95.3
10 II	89.4	9.2	1.3	99.8	80.0	13.5	4.7	98.3
10 mean	**89.4**	**9.0**	**1.3**	**99.6**	**78.1**	**13.9**	**4.7**	**96.8**
15 I	87.5	11.1	1.9	100.5	70.6	16.8	6.7	94.0
15 II	87.3	10.6	1.9	99.9	71.8	17.7	6.7	96.2
15 mean	**87.4**	**10.8**	**1.9**	**100.2**	**71.2**	**17.3**	**6.7**	**95.1**

TAR = Total applied radioactivity
DAT = Days after treatment
ERR = Extractable radioactive residues
NER = Non-extractable radioactive residues
n.a. = Not analyzed
* No other volatiles than CO_2 were found
** Day 0 samples of the dark control are the day 0 samples of the photolysis test

B. EXTRACTABLE AND BOUND RESIDUES

For the cyclopropane label, the amount of extractable radioactive residues (ERR) decreased to 92.6 and 90.6% TAR after 15 days of incubation (photolysis and dark control, respectively). For the benzyl label, the amount of ERR decreased to 87.4 and 71.2% TAR after 15 days of incubation (photolysis and dark control, respectively).

For the cyclopropane label, the amount of non-extractable residues (NER) increased from 1.7% TAR on day 0 to 15.0% TAR in the photolysis compared to 12.4% TAR in the dark control after 15 days. For the benzyl label, the amount of NER increased from 1.9% TAR on day 0 to 10.8% TAR in the photolysis compared to 17.3% TAR in the dark control after 15 days.

C. VOLATILIZATION

Carbon dioxide was the only trapped volatile degradation product. After 15 days of treatment, 5.4% TAR and 1.9% TAR of the cyclopropane- and benzyl-labeled test item, respectively, were detected in the photolysis experiments and 7.6% TAR and 6.7% TAR were detected in the dark controls. In sulfuric acid and ethylene glycol no significant radioactivity could be measured at any time.

D. TRANSFORMATION OF PARENT COMPOUND

Aliquots of all sample extracts were analyzed by radio-HPLC (see Table 7.1.1.3-6 and Table 7.1.1.3-7).

After 15 days, the amount of cyclopropane label decreased to 65.5% TAR in the photolysis and to 74.5% TAR in the dark control experiment. The benzyl label decreased to 64.1%TAR in the photolysis and to 55.7% TAR in the dark control experiment.

Several degradation products were detected in the extracts, but only 3-(2,2-dichlorovinyl)-2,2-dimethyl cyclopropanecarboxylic acid (DCVA) appeared in amounts higher than 5% TAR in extracts of soil samples treated with the cyclopropane label. It was found up to 5.4% TAR in irradiated conditions and up to 10% TAR in the dark control. All other degradation products never exceeded 5% TAR.

The identity of DCVA was confirmed by chromatographic comparison with the retention time of the reference item Reg. No. 180011 in the aerobic soil metabolism study [*CA 7.1.1.2, BASF DocID 2014/1000641*], in which DCVA was also detected in extracts of soil samples also treated with the cyclopropane labeled test item. Verification of the identification of DCVA was obtained by mass spectrometry in the mentioned study.

Table 7.1.1.3-6: HPLC analysis of ERR extracts: photolysis and dark control of cyclopropane-1-^{14}C-labeled-alpha-cypermethrin [% TAR]

DAT	Photolysis test				Dark control test			
	Total	DCVA	BAS 310 I	Sum others*	Total	DCVA	BAS 310 I	Sum others*
0 I**	96.0	n.d.	96.0	0.0	96.0	n.d.	96.0	0.0
0 II**	100.7	n.d.	92.6	8.1	100.7	n.d.	92.6	8.1
0 mean**	**98.3**	**-**	**94.3**	**4.0**	**98.3**	**-**	**94.3**	**4.0**
1 I	105.8	n.d.	97.6	8.2	109.1	n.d.	104.5	4.6
1 II	103.2	n.d.	88.7	14.6	108.0	n.d.	108.0	0.0
1 mean	**104.5**	**-**	**93.1**	**11.4**	**108.6**	**-**	**106.2**	**2.7**
3 I	91.3	n.d.	73.8	17.6	96.5	1.9	90.6	4.0
3 II	130.3	n.d.	116.8	13.6	99.3	0.0	99.3	0.0
3 mean	**110.8**	**-**	**95.3**	**15.6**	**97.9**	**0.9**	**94.9**	**2.0**
7 I	93.9	n.d.	88.6	5.3	95.0	0.0	75.5	19.6
7 II	94.4	n.d.	84.6	9.8	95.3	3.0	82.7	9.7
7 mean	**94.2**	**-**	**86.6**	**7.6**	**95.2**	**1.5**	**79.1**	**14.6**
10 I	95.8	n.d.	83.0	12.8	100.9	5.7	88.7	6.4
10 II	97.0	n.d.	84.8	12.2	101.3	14.2	82.3	4.8
10 mean	**96.4**	**-**	**83.9**	**12.5**	**101.1**	**10.0**	**85.5**	**5.5**
15 I	93.8	5.8	68.7	19.2	91.3	9.0	79.9	2.5
15 II	91.3	5.1	62.2	24.1	89.9	4.1	69.2	16.6
15 mean	**92.6**	**5.4**	**65.5**	**22.1**	**90.6**	**6.6**	**74.5**	**9.6**

ERR = Extractable radioactive residues
TAR = Total applied radioactivity
DAT = Days after treatment
n.d. = Not detected
* Individual peaks < 5.0%TAR
** Day 0 samples of the dark control are the day 0 samples of the photolysis test

Table 7.1.1.3-7: HPLC analysis of ERR extracts: photolysis and dark control of benzyl-^{14}C-labeled-alpha-cypermethrin [% TAR]

DAT	Photolysis test			Dark control test		
	Total	BAS 310 I	Sum others*	Total	BAS 310 I	Sum others*
0 I**	97.2	86.0	11.2	97.2	86.0	11.2
0 II**	98.9	86.0	12.9	98.9	86.0	12.9
0 mean**	**98.1**	**86.0**	**12.1**	**98.1**	**86.0**	**12.1**
1 I	97.7	85.7	11.9	95.7	78.6	17.1
1 II	96.8	79.8	17.1	88.8	68.9	20.0
1 mean	**97.2**	**82.7**	**14.5**	**92.2**	**73.7**	**18.5**
3 I	96.7	81.4	15.4	89.6	75.9	13.7
3 II	94.3	79.9	14.4	89.4	65.6	23.9
3 mean	**95.5**	**80.6**	**14.9**	**89.5**	**70.8**	**18.8**
7 I	91.1	62.3	28.8	83.0	52.9	30.1
7 II	91.2	69.7	21.6	81.1	60.4	20.7
7 mean	**91.2**	**66.0**	**25.2**	**82.0**	**56.6**	**25.4**
10 I	89.4	68.9	20.5	76.2	61.5	14.7
10 II	89.4	67.3	22.0	80.0	67.8	12.2
10 mean	**89.4**	**68.1**	**21.3**	**78.1**	**64.6**	**13.5**
15 I	87.5	60.9	26.5	70.6	49.0	21.6
15 II	87.3	67.3	20.0	71.8	62.5	9.2
15 mean	**87.4**	**64.1**	**23.3**	**71.2**	**55.7**	**15.4**

ERR = Extractable radioactive residues
TAR = Total applied radioactivity
DAT = Days after treatment
* Individual peaks < 5.0%TAR
** Day 0 samples of the dark control are the day 0 samples of the photolysis test

Throughout the study, only one peak was observed in the chromatograms obtained by the isomer selective HPLC system. This confirms that no trans isomers were formed during the photolysis study.

Additionally, no change of the enantiomeric ratio could be observed during the study, irrespective of the radiolabel.

F. KINETIC MODELING RESULTS

The calculated $DegT_{50}$ and $DegT_{90}$ values of alpha-cypermethrin used as trigger endpoints are given in Table 7.1.1.3-8. For the metabolite DCVA no reliable endpoints could be derived.

Table 7.1.1.3-8: Trigger endpoints for alpha-cypermethrin

Test system	Label	Best-fit model	χ^2 error	Trigger endpoints	
				$DegT_{50}$ [d]	$DegT_{90}$ [d]
Irradiated	Cyclopropane-^{14}C	SFO	3.2	29.7	98.6
	Benzyl-^{14}C	DFOP	3.7	27.6	119.8
Dark control	Cyclopropane-^{14}C	SFO	4.1	30.9	102.6
	Benzyl-^{14}C	DFOP	5.0	No reliable endpoints derived	

III. CONCLUSION

Irradiation in the soil photolysis experiment with cyclopropane-1-^{14}C-labeled-alpha-cypermethrin and benzyl-^{14}C-labeled alpha-cypermethrin did not show an influence of sunlight on the degradation behavior and metabolite formation in soil. The known metabolite DCVA was observed in the soil samples treated with the cyclopropane-^{14}C-labeled alpha-cypermethrin, both, in the irradiated samples and in the dark controls.

The RMS BE has asked for additional kinetic evaluations on 27. Oct. 2016. The results are presented and discussed below.

Report: CA 7.1.1.3/3
Anonymous, 2016 a
Response to RMS BE - Request concerning kinetic re-evaluations – Alpha-cypermethrin
2016/1324158
Guidelines: none
GLP: no

^{14}C alpha-cypermethrin: study of its photodegradation on soil (Van Dijk *et al.*, 1993) BASF DocID: AL-620-010

RMS: Please Notifier perform a kinetic assessment according to FOCUS kinetics guidance document (2006) for the data from Van Dijk.

BASF response: A kinetic evaluation was performed according to FOCUS kinetics guidance document (2006) for the data from Van Dijk et al. (1993). The results are summarized in Table 7.1.1.3-9 and Table 7.1.1.3-10 and the corresponding Kingui fits are presented in Appendix 3 in the original response document. The endpoints (DT_{50}: 34.1 and 177.9 d for irradiated and dark control test system, SFO model) are comparable with those derived in the original study (DT_{50}: 31 and 193 d for irradiated and dark control test system). For this reason and taking into account that kinetic evaluation according to FOCUS is not formally required for soil photolysis studies, the endpoints of the original study are considered still valid.

Table 7.1.1.3-9: Statistical and visual assessment of kinetic models for soil photolysis of alpha-cypermethrin, irradiated test system

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	5.5	k: < 0.01	acceptable	34.1	113.3
FOMC	6.0	β: < 0.01	acceptable	34.1	113.3
DFOP	6.9	k1: < 0.01 k2: < 0.05 g: < 0.01	acceptable	34.1	113.3

Table 7.1.1.3-10: Statistical and visual assessment of kinetic models for soil photolysis of alpha-cypermethrin, dark control test system

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	3.1	k: < 0.05	acceptable	177.9	591.1
FOMC	2.6	β: not sig.	acceptable	>1000	>1000
DFOP	2.0	k1: not sig. [a] k2: not sig. g: < not sig.	acceptable	217.4	800.9

[a] Hessian not invertible – NA was calculated for standard deviation, confidence interval and t-test.

CA 7.1.1.3/1 Michel, A., Hassink, J. (2014b). "Soil Photolysis of BAS 310 I". BASF DocID: 2014/1000642

RMS: RMS notes that the Notifier performed a separate kinetic assessment for each label. In peer review, it is common practice to handle residuals from the same soil with different radiolabelled positions as replicate values. Please Notifier consider that residue data from two tested labels from the same soil need to be processed as replicates, and please Notifier perform a new kinetic assessment.

BASF response: A kinetic evaluation was performed by processing the residue data of the two tested labels as replicates. The results are summarized in Table 7.1.1.3-11 and Table 7.1.1.3-12 and the corresponding Kingui fits are presented in Appendix 4**Fehler! Verweisquelle konnte nicht gefunden werden.** in the original response document. The endpoints (DT_{50}: 26.9 and 25.1 d for irradiated and dark control test system, SFO) are comparable with those derived by performing a separate kinetic assessment for each label (geomean DT_{50}: 28.6 d for irradiated test system; DT_{50}: 30.9 d for dark control test system). Therefore, the endpoints presented in the dossier are considered adequate.

Table 7.1.1.3-11: Statistical and visual assessment of kinetic models for soil photolysis of alpha-cypermethrin in the irradiated test system

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	3.1	k: <0.01	good	26.9	89.2
FOMC	3.1	β: not sig.	good	54.9	>1000
DFOP	2.2	k1:< 0.01. k2: < 0.05 g: not sig.	good	27.6	101.2

Table 7.1.1.3-12: Statistical and visual assessment of kinetic models for soil photolysis of alpha-cypermethrin in the dark control test system

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	5.2	k: <0.01	good	25.1	83.4
FOMC	3.4	β: not sig.	good	106.4	>1000
DFOP	3.9	k1: not sig. k2: not sig g: not sig.	good	46.1	219.7

CA 7.1.2 Rate of degradation in soil

CA 7.1.2.1 Laboratory studies

The studies listed in Table 7.1.2.1-1 were evaluated and peer reviewed in the registration process.

1. Andre (1989): Versuchsbericht zum Verbleib von Pflanzenschutzmitteln in Boden gemäß BBA-Richtlinien Teil IV, 4-1

Andre (1990a) Determination of the stability of Cyperkill 40 in a sand soil and

Andre (1990b) Determination of the stability of Cyperkill 40 in a loamy sand soil

The three trials reported by Andre 1989, 1990a and 1990b were submitted by Mitchell Cotts and are available for BASF. The studies were not conducted with alpha-cypermethrin but with a mixture of cis and trans isomers that were separately quantified. The application rates a highly exaggerated (6 kg a.s. ha^{-1} more than 100 fold overdosed) and not agricultural soils were used, but forest soils. Therefore these studies are not considered suitable for the determination of the DT_{50} of alpha-cypermethrin.

Table 7.1.2.1-1: List of laboratory degradation studies in soil performed with (alpha-) cypermethrin

DocID	Parent compound	Soil	Application rate [mg kg^{-1}]	Incubation temperature	Incubation period [days]	Remark
Andre, 1989, 1990a, 1990b	Cypermethrin	Sandy loam Sand Loamy sand	8.0	20-22°C		Submitted by Mitchell Cotts
AL-620-008	Cypermethrin; mixture of all cis isomers of cypermethrin; mixture of all trans isomers of cypermethrin	Sandy clay Sandy loam Clay	2.5	25±2°C (aerobic and anaerobic)	160	Standen 1976
AL-620-013	Alpha-cypermethrin	Sandy loam	0.3 1.5	10±2°C 20±2°C 20±2°C	120	Gedik, Keirs 2001

2. The degradation of the insecticide WL 43467 in soil under laboratory conditions (Standen 1976 AL-620-008, published by Roberts 1977, AL-905-062)
In this study, the degradation of cypermethrin and its isomers WL 43481 (both cis isomers) and WL 42641 (both trans isomers) were studied in three soils, two from Spain and one UK soil. The study of Standen (1976) was also submitted in the application for zeta-cypermethrin. These were evaluated by EFSA (EFSA Scientific Report (2008) 196, 1-119) and it was concluded that these studies should not be relied on for regulation. In the study no material balance is available, as no volatiles were trapped and samples were only taken at 4 intervals.
3. Gedik L. and Keirs D.C. (2001) [^{14}C]-Alpha-cypermethrin (BAS 310 I): Degradation in soil under aerobic conditions
This study was peer reviewed but a kinetic re-evaluation and an amendment describing the attempt for the identification of an unknown metabolite of the study were performed for this submission. Therefore the study will be resubmitted.

CA 7.1.2.1.1 Aerobic degradation of the active substance

Report:	CA 7.1.2.1.1/1 Gedik L.,Keirs D.C., 2001 a [14C]-Alphacypermethrin (BAS 310 I): Degradation in soil under aerobic conditions AL-620-013
Guidelines:	EEC 91/414 Annex II 7.1.1.1.1, SETAC Europe Part 1 Section 1.1 (March 1995), EEC 95/36 of 14 July 1995 amending 91/414/EEC, OECD Draft Guideline for aerobic and anaerobic transformation in soil (October 1999)
GLP:	yes (certified by United Kingdom Department of Health, UK)

Executive Summary

The objective of this study was to investigate the rate of breakdown of ^{14}C-alpha-cypermethrin in a sandy loam soil under aerobic conditions at two temperatures (20°C and 10°C). The rate of evolution of volatiles and the profile of degradation products was also investigated.

A freshly collected sample of a typical agricultural soil was obtained from within the UK for use on the study. Soil samples were treated with ^{14}C-alpha-cypermethrin at a rate equivalent to 0.307 mg kg^{-1} soil. The maximum anticipated field application rate for alpha-cypermethrin is 30 g a.s. ha^{-1} equivalent to 0.03 mg kg^{-1}. However, as the specific activity of alpha-cypermethrin was low, the application rate was increased to 0.3 mg kg^{-1} in order to facilitate the detection of alpha-cypermethrin and its degradation products. The samples were incubated in the dark at a soil moisture of 50% of the maximum water holding capacity and a temperature of 20 ± 2°C (Group A) or 10 ± 2°C (Group B) for up to 120 days under aerobic conditions. Ethanediol and sodium hydroxide were used to collect non-specific volatiles and $^{14}CO_2$, respectively. Traps were sampled and replenished at regular intervals throughout the study. Duplicate samples were collected for analysis of total radioactivity at zero time (immediately after application of test substance) and at 3, 7, 14, 28, 42, 70 and 120 days of incubation. Samples were extracted with acetonitrile : water (70:30, v/v) and analyzed by HPLC.

The mean overall recovery of applied radioactivity ranged from 93% to 103% other all samples. The total levels of solvent extractable radioactivity declined over the incubation period in both groups from a maximum of 98% of the total applied radioactivity (TAR) at zero time to 12% at day 120 at 20°C and from 100% to 36% TAR at 10°C. As the total levels of extractable radioactivity decreased with time, concomitant increases of $^{14}CO_2$ and non-extractable residues were observed over the course of the study. These accounted for 51% and 37% TAR, respectively at study termination at 20°C and 32% and 34% TAR at 10°C. Radioactivity recovered in the ethanediol traps and apparatus washings were low (less than 1% TAR).

Following extraction, the organic matter from day 120 samples incubated at both temperatures was fractionated into humin, fulvic acid, and humic acid. The radioactivity associated with the humin, fulvic acid, and humic acid accounted for means of 19, 5 and 6% TAR in soil samples incubated at 20°C, respectively. The corresponding values in soil samples incubated at 10°C were 17, 6 and 6% TAR.

The test item was rapidly degraded at each temperature. Alpha-cypermethrin quantitatively accounted for the TAR at zero time and these levels subsequently decreased to a mean of 7.5% at 20°C and 26.7% TAR at 10°C at day 120.
Low levels of CL 206128 (3-PBA), three unknown components, and polar material were also extracted from the soil. Two other minor unknown components (<1% TAR) were associated with the fulvic acid fraction of the organic matter extracted from Group B samples.

In addition to parent compound, low levels of CL 206128 (3-PBA) were detected in samples at day 7 in both groups accounting for less than 9% TAR. A minor unknown component, designated A, was detected in all samples after day 3. At 20°C, levels of this component accounted for a maximum of 5.1% TAR in a single day 7 replicate (mean value of 4.9% TAR) and subsequently decreased to a mean value of 1.7% TAR at study termination. Unknown A accounted for a maximum of 8.1% TAR in a single 10°C replicate at day 14 and decreased to a mean value of 5.3% TAR at day 120. Two other minor components, designated B and C, and polar material were detected at intervals throughout the incubation period in both groups each accounting for less than 5% TAR.

HPLC analysis of the fulvic acid fractions following fractionation of the organic matter indicated the presence of a polar component in samples from both groups. Two minor components, designated D and E, were also detected in the samples from Group B accounting for less than 1% TAR. Alpha-cypermethrin was not detected in any of the fulvic acid fractions.

At the time of study conduction, the rate of degradation of alpha-cypermethrin was calculated using ModelMaker v 4.0 assuming simple first-order or biphasic first-order kinetics. The DT_{50} and DT_{90} values for the degradation of alpha-cypermethrin in soil incubated at 20°C were estimated as 19 - 20 and 68 - 104 days, respectively. The corresponding values for soil incubated at 10°C were 50 - 55 and 183 - 233 days, respectively.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

BAS code:	BAS 310 I (alpha-cypermethrin)
Reg.No.:	4078193
Chemical name (IUPAC):	A racemate comprising (S)-α-cyano-3-phenoxybenzyl(1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-α-cyano-3-phenoxybenzyl(1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molecular weight:	416.32 g mol^{-1} (unlabeled)

Radio-labeled alpha-cypermethrin

Position of radiolabel:	Benzyl-ring-U-^{14}C
Specific radioactivity:	84.5 µCi mg^{-1}
Radiochemical purity:	99.5% (according to certificate)

97.7% (determined by radio-HPLC)
99.1% (determined by radio-TLC)

Carbon-13 labeled alpha-cypermethrin

Position of radiolabel:	Benzyl-ring-U-^{14}C / benzyl-7-^{13}C
Specific radioactivity:	27.04 µCi mg^{-1}
Radiochemical purity:	98.08% (according to certificate)

2. Soils

Top-soil samples (0-20 cm) of typical agricultural soil were used for treatment and incubation. The soils were sieved through a 2 mm sieve before use, remoistened to approximately 50% of the maximum water holding capacity, and stored in the dark at about 4°C. The soil characteristics are summarized in Table 7.1.2.1.1-1.

Table 7.1.2.1.1-1: Soil properties of soil used to investigate degradation rate of ^{14}C-alpha-cypermethrin

Soil designation	Ipswich (Levington Soil Code PT190; Inveresk Soil Code S332)
Origin	Ipswich, UK
UK Particle size distribution [%]	
Sand 0.060 – 2 mm	65.8
Silt 0.002 – 0.060 mm	26.9
Clay < 0.002 mm	7.3
Textural class	Sandy loam
USDA Particle size distribution [%]	
Sand 0.050 – 2 mm	66.6
Silt 0.002 – 0.050 mm	26.7
Clay < 0.002 mm	6.7
Textural class	Sandy loam
Total organic carbon [%]	0.9
pH (KCl)	6.5
Cation exchange capacity [meq/100g dry soil]	9.7
Maximum water holding capacity [%]	42.2
Microbial biomass (initial) [mg C/100g dry soil]	25.8
Microbial biomass at 20°C[mg C/100g dry soil]	20.3
Microbial biomass at 10°C [mg C/100g dry soil]	27.4

B. STUDY DESIGN

1. Experimental conditions

The soils were treated with ^{14}C-labeled alpha-cypermethrin at a nominal concentration of 0.307 mg kg^{-1} dry soil. After treatment, the soil in the test vessels was carefully shaken by hand in order to distribute the test item as homogeneously as possible within the soil. 100 g soil portions (dry weight equivalents) were prepared including duplicate samples at each sampling point.

The test vessels were incubated in the dark for up to 120 days at a temperature of either 20 ± 2°C (Group A) or 10 ± 2°C (Group B). Throughout the incubation period, the test vessels were continuously aerated with a slight stream of moistened, CO_2-free air. The outgoing air was led through a series of three gas-washing bottles. The first trap was a safety trap to prevent back flow, the remaining two contained trapping solutions for potential volatiles (ethanediol and 0.5M NaOH). Soil moisture was maintained at about 50% of the maximum water holding capacity for the duration of the study.

For the determination of the microbial biomass, extra test vessels with untreated soil treated with 100 µL acetonitrile and incubated as described above.

2. Sampling

Duplicate samples were taken at 0, 3, 7, 14, 28, 42, 70 and 120 days after treatment (DAT). Traps were sampled weekly up to 49 days and at each sampling occasion. Traps were also sampled at 98 days.

3. Description of the analytical procedures

The soil samples were extracted consecutively four times with about 100 mL acetonitrile : water (70:30, v/v) by shaking for about one hour.

After each extraction step, the suspension was centrifuged, the supernatant was filtered, and aliquots of each solution were radio-assayed. After removal of samples from flasks, flasks were soaked in acetone to remove any residual radioactivity. Aliquots of each apparatus wash were submitted for liquid scintillation counting (LSC).

Aliquots (about 25% by volume) of each extract were combined, concentrated, and analyzed by LSC as well as HPLC for the characterization of ^{14}C alpha-cypermethrin and its degradation products. TLC analysis of selected concentrated extracts was performed for confirmatory purposes. Procedural recoveries for sample concentration were in the range of 89 to 102% of the total applied radioactivity (TAR).

The extracted soil was dried at room temperature and aliquots were combusted to determine the amount of non-extractable radioactive residues (NER). Combustion products were trapped in Carbosorb mixed with Permaflour E+ scintillator and measured by LSC.

Trap solution aliquots and apparatus wash aliquots were analyzed by LSC as well. To confirm the presence of $^{14}CO_2$ in the trap samples, the radioactivity associated with the sodium hydroxide traps from a single soil sample from day 120 was precipitated as barium ^{14}C-carbonate. The radioactive content of the supernatant was then determined by LSC.

The organic matter from the 120 day extracted residues was exhaustively extracted, twice with 0.5 M NaOH by shaking and sonication. After centrifugation, radioactivity of the residue (humin fraction) was determined by combustion analysis. The humic acid fraction in the NaOH extracts was precipitated by acidification (pH 1) using concentrated HCl. After centrifugation, the supernatant (fulvic acid fraction) was removed. Both, the humic and the fulvic acid fractions were analyzed by LSC. The fulvic acid fractions were further analyzed by HPLC. All samples were stored at -20°C in a freezer.

4. Calculation of the degradation rate of alpha-cypermethrin

At the time of study conduction the DT_{50} and DT_{90} values for alpha-cypermethrin in the aerobic soil systems at 20°C and 10°C were calculated based on first-order kinetics. Two mathematical compartment models were developed using ModelMaker v 4.0 to describe the observed data with simple first-order kinetics (SFO) and biphasic first-order kinetics (DFOP). An actual kinetic evaluation of the study data following FOCUS recommendations [*FOCUS (2006)*] can be found in section CA 7.1.2.1.1/3 [*BASF DocID 2014/1159505*].

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The detailed results on the extractability of radioactive residues in the soils are presented in Table 7.1.2.1.1-2 and Table 7.1.2.1.1-3. The average material balance ranged from 93% to 101% TAR in soil samples incubated at 20°C and from 95% to 103% TAR in soil samples incubated at 10°C.

B. EXTRACTABLE AND BOUND RESIDUES

The distribution of radioactivity in extractable residues (ERR), non-extractable residues (NER), CO_2, and other volatiles is shown in Table 7.1.2.1.1-2 and Table 7.1.2.1.1-3, respectively. The total levels of extractable radioactivity (ERR) declined over the incubation period from a mean of 97.6% TAR at zero time to 12.2% TAR at 120 DAT (20°C) and from 99.8% to 36.1% TAR (10°C), respectively.

The level of NER increased during the study from about 0.7% TAR at zero time to about 36.8% TAR at 120 DAT in samples incubated at 20°C and from about 0.4% to about 33.6% TAR at 120 DAT in samples incubated at 10°C. After fractionation of the organic matter, the NER fraction was split up into humin, fulvic acid, and humic acid fraction accounting for 18.8, 5.2, and 6.0% TAR at 20°C, respectively, and 16.6, 5.6 and 5.7% TAR at 10°C, respectively. Results of the fractionation are presented in Table 7.1.2.1.1-4.

Table 7.1.2.1.1-2: Distribution of radioactivity and material balance after application of ^{14}C-alpha-cypermethrin to soil at 20°C (Group A) [% TAR]

Sampling Time	Sample No.	Volatiles		Extractable Radioactivity	Non-Extractable Residues	Apparatus wash	Total Recovery
		Ethanediol	NaOH	Acetonitrile : water (70:30, v/v)			
Zero Time	1	n.s.	n.s.	96.82	0.73	n.s.	97.55
	2	n.s.	n.s.	98.37	0.70	n.s.	99.07
	Mean	-	-	97.60	0.71	-	98.31
3 DAT	3	n.d.	1.59	94.02	4.17	0.03*	99.82
	4	0.01	3.28	89.92	6.34	0.04*	99.58
	Mean	n.d.	2.44	91.97	5.26	0.04	99.70
7 DAT	5	0.02	8.07	79.00	12.71	0.07*	99.87
	6	0.01	1.62	84.04	8.27	0.06*	94.01
	Mean	0.02	4.84	81.52	10.49	0.07	96.94
14 DAT	7	0.02	14.66	62.98	19.74	0.07*	97.47
	8	0.02	13.08	66.62	17.32	0.06*	97.10
	Mean	0.02	13.87	64.80	18.53	0.07	97.28
28 DAT	9	0.06	25.27	45.01	23.25	0.03*	93.62
	10	0.05	26.00	40.51	29.44	0.07*	96.07
	Mean	0.05	25.64	42.76	26.35	0.05	94.85
42 DAT	11	0.08	34.66	27.40	33.29	0.21	95.64
	12	0.06	34.80	28.98	27.25	0.07*	91.15
	Mean	0.07	34.73	28.19	30.27	0.14	93.40
70 DAT	13	0.08	41.36	20.38	34.21	0.03*	96.05
	14	0.10	40.94	22.07	39.14	0.06*	102.32
	Mean	0.09	41.15	21.22	36.68	0.04	99.18
120 DAT	15	0.19	51.68	11.33	39.48	0.02*	102.69
	16	0.11	51.12	13.16	34.06	0.01*	98.45
	Mean	0.15	51.40	12.24	36.77	0.01	100.57

DAT = Days after treatment
n.s.= No sample
n.d. = Not detected
* Results calculated from data less than 30 dpm above background

Table 7.1.2.1.1-3: Distribution of radioactivity and material balance after application of ^{14}C-alpha-cypermethrin to soil at 10°C (Group B) [% TAR]

Sampling Time	Sample No.	Volatiles		Extractable Radioactivity	Non-Extractable Residues	Apparatus wash	Total Recovery
		Ethanediol	NaOH	Acetonitrile : water (70:30, v/v)			
Zero Time	101	n.s.	n.s.	100.63	0.36	n.s.	100.98
	102	n.s.	n.s.	98.92	0.46	n.s.	99.38
	Mean	-	-	99.77	0.41	-	100.18
3 DAT	103	n.d.	1.13	99.24	2.53	0.04*	102.95
	104	n.d.	1.13	98.84	3.21	0.06*	103.24
	Mean	-	1.13	99.04	2.87	0.05	103.09
7 DAT	105	0.01	2.21	95.36	2.78	0.12*	100.49
	106	0.01	2.51	94.49	4.93	0.06*	102.01
	Mean	0.01	2.36	94.93	3.86	0.09	101.25
14 DAT	107	0.02	6.56	84.36	9.44	0.12*	100.51
	108	0.02	6.34	84.62	11.29	0.07*	102.34
	Mean	0.02	6.45	84.49	10.37	0.10	101.42
28 DAT	109	0.05	11.79	72.62	8.85	0.03*	93.35
	110	0.05	12.87	69.73	14.27	0.16	97.07
	Mean	0.05	12.33	71.17	11.56	0.10	95.21
42 DAT	111	0.05	17.00	60.13	20.03	0.14*	97.35
	112	0.05	15.12	63.76	16.95	0.42	96.30
	Mean	0.05	16.06	61.94	18.49	0.28	96.82
70 DAT	113	0.06	22.93	48.57	29.08	n.d.	100.64
	114	0.07	21.86	49.55	27.22	0.04*	98.73
	Mean	0.07	22.39	49.06	28.15	0.02	99.69
120 DAT	115	0.13	33.96	31.03	34.77	0.03*	99.93
	116	0.11	29.58	41.10	32.46	0.05*	103.30
	Mean	0.12	31.77	36.07	33.62	0.04	101.61

DAT = Days after treatment
n.s.= No sample
n.d. = Not detected
* Results calculated from data less than 30 dpm above background

Table 7.1.2.1.1-4: Organic matter fractionation of extracted residues of ^{14}C-alpha-cypermethrin [% TAR]

Sampling Time	Temperature (Group)	Sample No.	Percent Applied Dose Characterized as:		
			Humin	Fulvic Acid	Humic Acid
120 DAT	20°C (Group A)	15	21.54	4.99	5.29
		16	16.02	5.33	6.69
		Mean	18.78	5.16	5.99
120 DAT	10°C (Group B)	115	17.68	5.94	6.04
		116	15.42	5.27	5.31
		Mean	16.55	5.61	5.68

DAT = Days after treatment

C. VOLATILIZATION

Besides the formation of NER, the formation of CO_2 was an important degradation pathway of alpha-cypermethrin. Hence, the fraction of $^{14}CO_2$ accounted for means of 51.4% and 31.8% TAR for soil samples incubated at 20°C and 10°C, respectively. Other radioactive residues recovered in the ethanediol traps were negligible (less than 1% TAR).

D. TRANSFORMATION OF PARENT COMPOUND

All combined soil extracts were analyzed by radio-HPLC. The results obtained with this system are summarized in Table 7.1.2.1.1-5 and Table 7.1.2.1.1-7.

HPLC analysis of the soil extracts indicated that alpha-cypermethrin was rapidly degraded at each temperature. Alpha-cypermethrin quantitatively accounted for the applied radioactivity at zero time and these levels subsequently decreased to a mean of 7.5% TAR at 20°C and 26.7% TAR at 10°C, respectively, after 120 days of incubation. In addition to parent material, low levels of CL 206128 (3-PBA) were detected in samples at 7 DAT in both groups accounting for less than 9% TAR. A minor unknown component, designated A, was detected in all samples after 3 DAT. At 20°C, levels of this component accounted for a maximum of 4.9% TAR in a single 7 DAT replicate and subsequently decreased to a mean value of 1.7% TAR at study termination. Unknown A accounted for a maximum of 8.1% TAR in a single 10°C replicate at 14 DAT and decreased to a mean value of 5.3% TAR at 120 DAT. Two further minor components, designated B and C, and polar material were detected at intervals throughout the incubation period in both groups each accounting for less than 5% TAR.

TLC analysis of soil extracts from 7 DAT and 120 DAT confirmed the results obtained by HPLC.

HPLC analysis of the fulvic acid fractions following fractionation of the organic matter indicated the presence of a polar component in samples from both groups. Two minor components, designated D and E, were also detected in the sample from Group B accounting for less than 1% TAR. Alpha-cypermethrin was not detected in any of the fulvic acid fractions.

Table 7.1.2.1.1-5: Summary of metabolite identity in soil for ^{14}C-alpha-cypermethrin at 20°C (Group A) [% TAR]

Compound	Sampling Time											
	0 DAT			3 DAT			7 DAT			14 DAT		
	1	2	Mean	3	4	Mean	5	6	Mean	7	8	Mean
Alpha-cypermethrin	96.82	98.37	97.60	92.59	85.51	89.05	72.91	76.66	74.79	57.51	62.16	59.84
CL 206128 (3-PBA)	n.d.	n.d.	-	n.d.	n.d.	-	1.51	2.26	1.89	n.d.	n.d.	-
Unknown A	n.d.	n.d.	-	1.43	4.41	2.92	4.58	5.12	4.85	4.06	3.18	3.62
Unknown B	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	1.42	1.28	1.35
Unknown C	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
Polar	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
CO_2	n.s.	n.s.	-	1.59	3.28	2.44	8.07	1.62	4.84	14.66	13.08	13.87
Volatiles	n.s.	n.s.	-	n.d.	0.01	n.d.	0.02	0.01	0.02	0.02	0.02	0.02
Bound Residues	0.73	0.70	0.71	4.17	6.34	5.26	12.71	8.27	10.49	19.74	17.32	18.53
Total % Recovery	97.55	99.07	98.31	99.78	99.55	99.67	99.80	93.94	96.88	97.41	97.04	97.23

DAT = Days after treatment
n.d. = Not detected
n.s. = No sample

Table 7.1.2.1.1-6: Summary of metabolite identity in soil for ^{14}C-alpha-cypermethrin at 20°C (Group A) [% TAR] (continued)

Compound	Sampling Time											
	28 DAT			42 DAT			70 DAT			120 DAT		
	9	10	Mean	11	12	Mean	13	14	Mean	15	16	Mean
Alpha-cypermethrin	36.99	34.96	35.98	20.71	20.78	20.75	14.92	16.05	15.49	7.44	7.53	7.49
CL 206128 (3-PBA)	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
Unknown A	4.54	4.12	4.33	3.66	3.74	3.70	2.44	2.48	2.46	1.17	2.25	1.71
Unknown B	1.38	1.43	1.41	0.98	0.64	0.81	0.83	0.73	0.78	0.71	0.82	0.77
Unknown C	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	0.39	0.20
Polar	2.10	n.d.	1.05	2.05	3.82	2.94	2.19	2.81	2.50	2.00	2.17	2.09
CO_2	25.27	26.00	25.64	34.66	34.80	34.73	41.36	40.94	41.15	51.68	51.12	51.40
Volatiles	0.06	0.05	0.05	0.08	0.06	0.07	0.08	0.10	0.09	0.19	0.11	0.15
Bound Residues	23.25	29.44	26.35	33.29	27.25	30.27	34.21	39.14	36.68	39.48	34.06	36.77
Total % Recovery	93.59	96.00	94.81	95.43	91.09	93.27	96.03	102.25	99.15	102.67	98.45	100.58

DAT = Days after treatment
n.d. = Not detected

Table 7.1.2.1.1-7: Summary of metabolite identity in soil for ^{14}C-alpha-cypermethrin at 10°C (Group B) [% TAR]

Compound	Sampling Time											
	0 DAT			3 DAT			7 DAT			14 DAT		
	101	102	Mean	103	104	Mean	105	106	Mean	107	108	Mean
Alpha-cypermethrin	100.63	98.92	99.78	97.01	93.77	95.39	86.24	79.63	82.94	76.23	76.48	76.36
CL 206128 (3-PBA)	n.d.	n.d.	-	n.d.	n.d.	-	2.75	8.13	5.44	n.d.	n.d.	-
Unknown A	n.d.	n.d.	-	2.23	5.07	3.65	6.37	6.74	6.56	8.13	8.14	8.14
Unknown B	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
Unknown C	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
Polar	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
CO_2	n.s.	n.s.	-	1.13	1.13	1.13	2.21	2.51	2.36	6.56	6.34	6.45
Volatiles	n.s.	n.s.	-	n.d.	n.d.	-	0.01	0.01	0.01	0.02	0.02	0.02
Bound Residues	0.36	0.46	0.41	2.53	3.21	2.87	2.78	4.93	3.86	9.44	11.29	10.37
Total % Recovery	100.99	99.38	100.19	102.90	103.18	103.04	100.36	101.95	101.17	100.38	102.27	101.34

DAT = Days after treatment
n.d. = Not detected
n.s. = No sample

Table 7.1.2.1.1-8: Summary of metabolite identity in soil for ^{14}C-alpha-cypermethrin at 10°C (Group B) [% TAR] (continued)

Compound	Sampling Time											
	28 DAT			42 DAT			70 DAT			120 DAT		
	109	110	Mean	111	112	Mean	113	114	Mean	115	116	Mean
Alpha-cypermethrin	65.35	60.16	62.76	50.40	55.23	52.82	38.84	42.68	40.76	21.40	31.93	26.67
CL 206128 (3-PBA)	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-	n.d.	n.d.	-
Unknown A	7.27	7.23	7.25	7.08	5.61	6.35	6.45	6.87	6.66	5.85	4.73	5.29
Unknown B	n.d.	2.34	1.17	1.20	1.91	1.56	n.d.	n.d.	-	0.99	1.55	1.27
Unknown C	n.d.	n.d.	-	1.44	1.01	1.23	1.22	n.d.	0.61	0.86	0.64	0.75
Polar	n.d.	n.d.	-	n.d.	n.d.	-	2.06	n.d.	1.03	1.94	2.26	2.10
CO_2	11.79	12.87	12.33	17.00	15.12	16.06	22.93	21.86	22.39	33.96	29.58	31.77
Volatiles	0.05	0.05	0.05	0.05	0.05	0.05	0.06	0.07	0.07	0.13	0.11	0.12
Bound Residues	8.85	14.27	11.56	20.03	16.95	18.49	29.08	27.22	28.15	34.77	32.46	33.62
Total % Recovery	93.31	96.92	95.12	97.20	95.88	96.56	100.64	98.70	99.67	99.90	103.26	101.59

DAT = Days after treatment
n.d. = Not detected

Calculation of the degradation rates

Using simple first-order kinetics, the predicted concentrations after 40 days are below the measured values, even though the overall r^2 values were relatively high. Using biphasic first-order kinetics significantly improved the fit, as all time points could be well described. The r^2 values with biphasic kinetics were higher than with simple first-order kinetics, and the residuals were evenly distributed with time. Therefore, first-order DT_{50} values of 19.3 d at 20°C and 49.9 d at 10°C, and DT_{90} values of 104.0 d at 20°C and 233.0 d at 10°C, obtained from the biphasic fits were preferred (Table 7.1.2.1.1-9**:**).

Table 7.1.2.1.1-9: Estimated first-order DT_{50} and DT_{90} values of alpha-cypermethrin optimized with ModelMaker 4.0*

Temperature	Kinetic model	DT_{50} [d]	DT_{90} [d]	r^2
20°C	Simple first-order	20.6	68.3	0.987
	Biphasic first-order	19.3	104.0	0.997
10°C	Simple first-order	54.9	182.5	0.962
	Biphasic first-order	49.9	233.0	0.982

* calculated at the time of study conduction; an actual kinetic evaluation of the study data following FOCUS recommendations [FOCUS (2006) "Guidance Document on Estimating Persistence and Degradation Kinetics from Environmental Fate Studies on Pesticides in EU Registration" Report of the FOCUS Work Group on Degradation Kinetics, EC Document Reference Sanco/10058/2005 version 1.0 (November 2011), 436 pp.] can be found in section CA 7.1.2.1.1/3.

III. CONCLUSION

Under the conditions of the study, alpha-cypermethrin degraded quickly in a sandy loam soil at two incubation temperatures. $^{14}CO_2$ was the principal degradation product detected and accounted for ca 51% and ca 32% TAR in soils incubated at 20°C and 10°C, respectively. Low levels of CL 206128 (3-PBA), three unknown components, and polar material were also extracted from the soil.

The DT_{50} and DT_{90} values for the degradation of alpha-cypermethrin in soil incubated at 20°C were estimated as 19 - 20 and 68 - 104 days, respectively. The corresponding values for soil incubated at 10°C were 50 - 55 and 183 - 233 days.

In conclusion, based on the results of this study and the estimated DT_{50} data, it is unlikely that alpha-cypermethrin will persist in soil.

Report:	CA 7.1.2.1.1/2 Singh M., 2003 a Isolation and identification of an unknow metabolite A of Alphacypermethrin (BAS 310 I) found in the Inveresk study 399307 (Degradation in soil under aerobic conditions) 2003/5000474
Guidelines:	EEC 91/414 Annex II 7.1.1.1.1
GLP:	yes (certified by United States Environmental Protection Agency)

Executive Summary

An aerobic degradation study was conducted by INVERESK RESEARCH using ^{14}C-alpha-cypermethrin for about 120 days [*CA 7.1.2.1.1/1, BASF DocID 2001/7001614 (AL-620-013)*]. The test soil was sandy loam and was supplied and characterized by Levington Agriculture (lpswich, UK). One of the metabolites, designated as unknown metabolite A, was a minor degradation product in both experiments (10°C and 20°C) and declined over time. The maximum amount of unknown A found in the 20°C experiment was 4.85% TAR at 7 DAT. The maximum amount of unknown A found in the 10°C experiment was 8.14% TAR, but was in excess of 5% TAR at several sampling intervals. Therefore, an attempt was made to isolate and identify the unknown metabolite A. Based on the chromatographic characteristics and the MS/MS data, the unknown metabolite A has been assigned the molecular formula $C_{22}H_{19}0_4NCl_2$ (Nominal Molecular Weight 432). The proposed chemical structure is shown in the discussion section.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Substance

No test substance was applied to the test soils in this study. Soil samples extracted and analyzed in this study for the identification of the unknown metabolite A were generated in the aerobic degradation study of alpha-cypermethrin [*CA 7.1.2.1.1/1, BASF DocID 2001/7001614 (AL-620-013)*].

2. Soil

The soil samples extracted in this study were generated in the aerobic degradation study of ^{14}C-alpha-cypermethrin [*CA 7.1.2.1.1/1; BASF DocID 2001/7001614 (AL-620-013)*]. Additionally, some of the unused extracts from this study were also utilized for the isolation of unknown metabolite A.

B. STUDY DESIGN

1. Description of analytical procedures

Extraction

The soil samples available for extraction, were split almost equally and transferred into four centrifuge tubes. The samples were then extracted with a mixture of acetonitrile and water (70/30, v/v; ~150 mL). Each extraction was performed by shaking the soil and the solvent mixture for about one hour at 300 rpm followed by centrifugation for 15 minutes. Three additional extractions were performed following this procedure. The supernatants from all four extractions were combined, measured and aliquots of the combined supernatant were assayed by liquid scintillation counting (LSC) to determine the amount total radioactivity available for the isolation of the unknown A and to track the material balance during further processing of the combined extract.

Isolation

The acetonitrile/water extract was concentrated on a rotary evaporator to remove most of the acetonitrile. Then, the aqueous solution was transferred to a separatory funnel and extracted with ethyl acetate (3x). The ethyl acetate extracts were combined and measured. Aliquots (3x) of the combined ethyl acetate extracts and aqueous layers were assayed by LSC to estimate the amount of radioactivity present in the ethyl acetate fraction and the aqueous fraction. Next, the ethyl acetate extract was concentrated on a rotary evaporator to almost dryness. The residual material was dissolved in acetonitrile. The acetonitrile solution was loaded on a pre-conditioned silica gel column and successively eluted with n-hexane, acetonitrile, and ethyl acetate. All eluants (n-hexane, acetonitrile, and ethyl acetate) were measured and assayed by LSC to determine the amount of the radioactivity in each eluant. Most of the radioactivity was found in the acetonitrile eluant.
The unprocessed acetonitrile/water extracts, received from INVERESK RESEARCH, were also processed in the same manner as described above.

The acetonitrile eluates obtained as mentioned above were washed with n-hexane to remove the majority of parent material (alpha-cypermethrin) present in the acetonitrile eluate. Next, the acetonitrile fraction was concentrated to approximately 5.0 mL.

ldentification of the isolated unknown metabolite A by mass spectrometry

An aliquot of the isolated ^{14}C-unknown A was analyzed by mass spectrometry (APCI, LC-MS/MS positive ion mode).

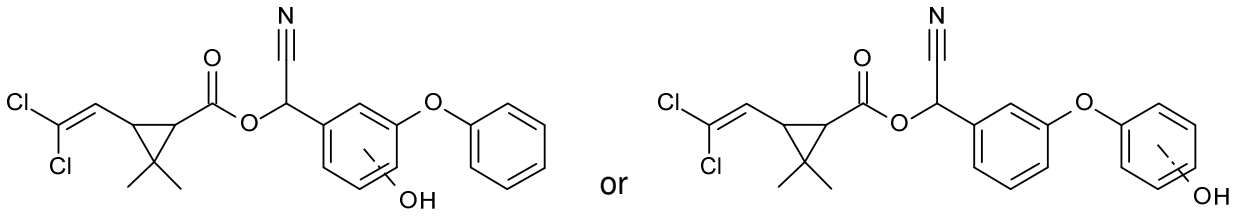
II. RESULTS AND DISCUSSION

A. STORAGE STABILITY

For the identification work, samples (both extracts and unextracted soil samples) generated in the aerobic degradation study [*CA 7.1.2.1.1/1; BASF DocID 2001/7001614 (AL-620-013)*] were used in this study. The soil extract was analyzed by HPLC after and before storage of about 2 years. The HPLC results show no change in the profiles. Therefore, the unknown metabolite A was stable during the storage period.

B. IDENTIFICATION OF THE UNKNOWN METABOLITE A

The identity of the degradation product unknown A found in the study was established by a combination of HPLC data and mass spectrometric analysis. The chromatographic data suggest that the unknown metabolite A has all the structural features of alpha-cypermethrin and no cleavage of either the ester bond or ether bond in alpha-cypermethrin has taken place. These conclusions are also supported by mass spectroscopy data.
The MS/MS data for the isolated unknown metabolite A is consistent for the following structures:



III. CONCLUSION

The metabolite unknown A, found in the aerobic degradation of alpha-cypermethrin, is a mono hydroxylation product of alpha-cypermethrin. Hydroxylation has taken place at one of the carbon atoms in the diphenoxy ether moiety of alpha-cypermethrin. The position could not be confirmed by MS. As in the recent metabolism studies, a metabolite with the hydroxylation in the para position of the phenyl ring was identified (M310I017), it seems likely that metabolite A has the same structure and is identical to M310I017.

Report:	CA 7.1.2.1.1/3 Sachers S., 2014 a Kinetic evaluation of aerobic soil degradation of BAS 310 I - alpha-cypermethrin: Determination of trigger and modeling endpoints according to FOCUS 2014/1159505
Guidelines:	FOCUS Kinetics (2006) SANCO/10058/2005 version 1.0 of Nov. 2011
GLP:	no

Executive Summary

The degradation behavior of the insecticide alpha-cypermethrin (BAS 310 I) in soil has been investigated in a laboratory study under aerobic degradation conditions with one soil at two different temperatures (10°C and 20°C). The purpose of this evaluation was to analyze the degradation kinetics of alpha-cypermethrin observed in the study according to current guidance of the FOCUS workgroup on degradation kinetics.

The appropriate kinetic models to derive trigger and modeling endpoints were identified considering the procedures and kinetic models proposed by the FOCUS kinetics guidance.

The best-fit model to derive trigger endpoints was selected based on a visual and statistical assessment. Normalization of $DegT_{50}$ values suitable for modeling to reference moisture was not necessary as the soil moisture at study conditions was higher than the reference moisture.

The kinetic evaluation showed that the DFOP model provided the best fit to the measured data for alpha-cypermethrin incubated at 10°C while the SFO model was appropriate for derivation of modeling endpoints. For the experiment conducted at 20°C the FOMC model provided the best fit and was also appropriate for derivation of modeling endpoints.

Best-fit $DegT_{50}$ as well as $DegT_{90}$ values, derived for the use as trigger endpoints, were calculated as 48.1 and 228.7 days at 10°C and 18.3 and 84.8 days at 20°C, respectively. Modeling $DegT_{50}$ values were calculated as 54.7 days at 10°C and 25.5 days at 20°C.

I. MATERIAL AND METHODS

The degradation of alpha-cypermethrin in one soil at two different temperatures [see *CA 7.1.2.1.1/1, BASF DocID 2001/7001614 (AL-620-013)*] was analyzed taking into account the current guidance of the FOCUS workgroup on degradation kinetics [*FOCUS (2006)*].

Kinetic modeling strategy

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints) as well as modeling endpoints. The best-fit model was selected based on visual and statistical assessment and the corresponding $DegT_{50}$ and $DegT_{90}$ values are reported as trigger endpoints. Appropriate $DegT_{50}$ values for use in environmental fate models were derived depending on the kinetic model.

Kinetic models included in the evaluations

For each data set, the kinetic models proposed by FOCUS Kinetics [*FOCUS (2006)*] were tested in order to identify the best-fit model, i.e. single first order (SFO) kinetics, the Gustafson-Holden model (FOMC), and the bi-exponential (DFOP) kinetics. The respective model descriptions and corresponding equations for calculating endpoints ($DegT_{50}$, $DegT_{90}$) are shown in the FOCUS Kinetics guidance [*FOCUS (2006)*].

A kinetic model is considered appropriate if the residuals are randomly distributed around zero, the χ^2 error indicates a sufficient quality of the fit (e.g. value is ideally < 15% but may be higher if the visual fit represents the degradation behavior well), and the estimated degradation parameters differ significantly from zero.

The goodness-of-fit of the kinetic models was assessed by visual inspection and statistical measures, as recommended by the FOCUS Kinetics guidance [*FOCUS (2006)*].

Data handling and software for kinetic evaluation

The experimental data were derived from the study report [*CA 7.1.2.1.1/1, BASF DocID 2001/7001614 (AL-620-013)*] and the initial concentration of alpha-cypermethrin was set to the material balance. For all samplings, duplicate measurements were considered for the parameter estimation.

The software package KinGUI version 2.2014.224.1704 was used for parameter fitting. The error tolerance and the number of iterations of the optimization tool (IRLS) were set to $1x10^{-6}$ and 100, respectively.

Normalization to reference conditions

According to FOCUS, $DegT_{50}$ values suitable for modeling should be normalized to reference moisture at field capacity (pF 2) if the experiment was conducted at a lower soil moisture.

In the evaluated study the actual soil moisture of 21.1 g per 100 g dry soil (i.e. 50% of the maximum water holding capacity (MWHC) of 42.2 g per 100 g dry soil) is higher than the reference soil moisture of 19 g per 100 g dry soil (moisture at pF 2 of a sandy loam of according to FOCUS. Hence, further normalization of the calculated $DegT_{50}$ suitable for modeling was not necessary.

Experimental data

The kinetic evaluation was conducted for one aerobic laboratory soil degradation study [see *CA 7.1.2.1.1/1, BASF DocID 2001/7001614 (AL-620-013)*]. Detailed soil characteristics in each trial are reported in the cited study.

The test soil treated with benzyl ring-U-^{14}C labeled alpha-cypermethrin at a nominal application rate of 0.3 mg kg^{-1} dry soil, corresponding to 300 g a.s. ha^{-1} (i.e. tenfold of the maximum recommended field application rate), was incubated under dark aerobic conditions at two temperatures (20ºC and 10°C, respectively) and 50% MWHC for 120 days. Soil samples were taken at 0, 3, 7, 14, 28, 42, 70 and 120 days after treatment (DAT).

The measured data as well as resulting datasets submitted to kinetic analysis are given in Table 7.1.2.1.1-10**:** .

Table 7.1.2.1.1-10: Experimental data for Ipswich sandy loam at 10°C and 20°C

DAT	10°C		20°C	
	Experimental data [% TAR]	Input data according to FOCUS [% TAR]	Experimental data [% TAR]	Input data according to FOCUS [% TAR]
0	100.6	101.0 [a]	96.8	97.6 [a]
0	98.9	99.4 [a]	98.4	99.1 [a]
3	97.0	97.0	92.6	92.6
3	93.8	93.8	85.5	85.5
7	86.2	86.2	72.9	72.9
7	79.6	79.6	76.7	76.7
14	76.2	76.2	57.5	57.5
14	76.5	76.5	62.2	62.2
28	65.4	65.4	37.0	37.0
28	60.2	60.2	35.0	35.0
42	50.4	50.4	20.7	20.7
42	55.2	55.2	20.8	20.8
70	38.8	38.8	14.9	14.9
70	42.7	42.7	16.1	16.1
120	21.4	21.4	7.4	7.4
120	31.9	31.9	7.5	7.5

DAT = Days after treatment
TAR = Total Applied Radioactivity
[a] Set to material balance

II. RESULTS AND DISCUSSION

The kinetic evaluation showed that the DFOP model provided the best fit to the measured data for alpha-cypermethrin incubated at 10°C while the SFO model was appropriate for derivation of modeling endpoints. For the experiment conducted at 20°C, the FOMC model provided the best fit and was also appropriate for derivation of modeling endpoints.

A summary of the calculated trigger and modeling endpoints for alpha-cypermethrin is given in Table 7.1.2.1.1-11 and Table 7.1.2.1.1-12.

Table 7.1.2.1.1-11: Trigger endpoints for alpha-cypermethrin

Study	Incubation temperature [°C]	Best-fit model	$\chi 2$ error	p (t-test)	Visual fit	$DegT_{50}$ [d]	$DegT_{90}$ [d]
2001/7001614	10	DFOP	1.8	k1: <0.05 k2: <0.05 g: <0.05	Good	48.1	228.7
	20	FOMC	3.5	β: <0.05	Good	18.3	84.8

Table 7.1.2.1.1-12: Modeling endpoints for alpha-cypermethrin

Study	Incubation temperature [°C]	Kinetic model	$\chi 2$ error	p (t-test)	Visual fit	$DegT_{50}$ [d]
2001/7001614	10	SFO	4.8	k: <0.05	Good	54.7
	20	FOMC	3.5	β: <0.05	Good	25.5 [a]

[a] Calculated as DegT50 = DegT90/3.32

III. CONCLUSION

Trigger and modeling endpoints were derived for alpha-cypermethrin in a laboratory degradation study at two different temperatures.

The kinetic evaluation revealed that the DFOP model provided the best fit to the measured data for alpha-cypermethrin incubated at 10°C while the SFO model was appropriate for derivation of modeling endpoints. For the experiment conducted at 20°C, the FOMC model provided the best fit and was also appropriate for derivation of modeling endpoints.

Best-fit $DegT_{50}$ as well as $DegT_{90}$ values, derived for the use as trigger endpoints, were calculated as 48.1 and 228.7 days at 10°C and 18.3 and 84.8 days at 20°C, respectively. Modeling $DegT_{50}$ values were calculated as 54.7 days at 10°C and 25.5 days at 20°C.

Report:	CA 7.1.2.1.1/4 Michel, A., Hassink, J., 2014 b Rate of degradation of alpha-cypermethrin (BAS 310 I) in soil under aerobic conditions 2014/1159491
Guidelines:	OECD 307, EPA 835.4100, SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

The degradation rate of alpha-cypermethrin (BAS 310 I) was investigated under aerobic conditions in three German soils (Li10, LUFA 2.2, and LUFA 2.3).

The soils were treated with cyclopropane-1-^{14}C-labeled-alpha-cypermethrin and benzyl-^{14}C-labeled-alpha-cypermethrin at a nominal rate of 0.3 mg kg^{-1} dry soil. Soil aliquots were incubated in the laboratory in the dark under aerobic conditions with soil moistures between 40% and 50% of the maximum water holding capacity (MWHC) and a temperature of 20°C ± 2°C. An incubation system with continuous aeration was used with an attached trapping system for the determination of volatile compounds. The microbial biomass was determined by the substrate induced respiration method before the start of the study, after 71 and 136 days of incubation.

Samples were taken 0, 1, 3, 7, 10, 14, 60, 87 and 120 days after treatment (DAT). All soil samples were worked up in duplicate. The soil samples were extracted four times with ACN/H_2O/HCOOH (700/300/1, v/v/v) and the individual extracts analyzed by liquid scintillation counting (LSC). The combined extracts were then concentrated and analyzed by LSC and high performance liquid chromatography (HPLC). The remaining soil after extraction was combusted in order to determine the amount of non-extractable soil bound residues. Additionally, the amount of formed volatiles was determined for each sampling point except on day 0. A full mass balance was provided for each sampling interval.

The amount of extractable radioactivity continuously decreased in the aerated soils to 27.9%, 39.9%, and 16.6% TAR after 120 days of incubation for the soils Li10, LUFA 2.2, and LUFA 2.3, respectively. The amount of non-extractable radioactive residues (NER) increased during the study from 0.6%, 1.5%, and 1.3% TAR at day 0 to 32.2%, 23.5%, and 34.8% TAR after 120 days of incubation for the soils Li10, LUFA 2.2, and LUFA 2.3, respectively. Mineralization reached a total of up to 48.8% TAR after 120 days. The mass balance ranged from 88.3% to 111.0% TAR.

The NER fraction was further characterized exemplarily for one replicate at 60 DAT and 120 DAT. After 120 days, 7.0%, 3.8%, and 7.8%, TAR remained unextractable for the soil Li10, LUFA 2.2, and LUFA 2.3 respectively. This portion of unextractable radioactivity was assigned to the humin fraction. The alkali-soluble radioactivity was further fractionated by precipitation with concentrated HCl. After 120 days, 10.8% TAR (Li10), 10.5%TAR (LUFA 2.2), and 14.9% TAR (LUFA 2.3) could be assigned to the fulvic acids fraction and 14.1% TAR (Li10), 7.9%TAR (LUFA 2.2), and 6.8% TAR (LUFA 2.3) to the humic acids fraction.

The humic acid fraction was not further analyzed. The fulvic acid fraction was partitioned with ethyl acetate for two exemplary samples (60 and 120 days). At 120 DAT, about 6.9% and 3.3% TAR (Li10), 5.3% and 4.3% TAR (LUFA 2.2), and 8.1% and 6.7% TAR (LUFA 2.3) of the fulvic acid fraction were soluble in the water phase and the ethyl acetate phase, respectively.

The parent compound decreased continuously from 99.4%, 98.5%, and 98.7% TAR at day 0, to 20.8%, 30.1%, and 10.5% TAR at the end of the incubation for the soils Li10, LUFA 2.2, and LUFA 2.3 respectively.

One metabolite was observed in the soil extracts, at maximum amounts of 5.8, 5.9, and 5.3% TAR in soils Li10, LUFA 2.2 and LUFA 2.3, respectively. The maxima were reached between 10 and 30 days after application, with a decrease thereafter. This metabolite was identified by mass spectrometry (MS) and nuclear magnetic resonance (NMR) measurements as M310I017.

Kinetic evaluation was performed following the recommendations of the FOCUS Kinetics workgroup in order to derive trigger and modeling endpoints. The following $DegT_{50}$ and $DegT_{90}$ values were calculated (Table 7.1.2.1.1-13 to Table 7.1.2.1.1-16):

Table 7.1.2.1.1-13: Trigger endpoints for alpha-cypermethrin

Soil	Compound	Best-fit model	χ^2 error	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Li 10	alpha-cypermethrin	DFOP	1.3	28.3	196.3
LUFA 2.2		DFOP	1.7	35.0	329.4
LUFA 2.3		FOMC	1.2	24.3	134.6

Table 7.1.2.1.1-14: Trigger endpoints for the metabolite M310I017

Soil	Compound	Best-fit model	χ^2 error	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Li 10	M310I017	SFO[a]	13.5	22.6	75.1
LUFA 2.2		SFO[a]	13.2	42.3	140.4
LUFA 2.3		SFO[b]	16.1	4.9	16.2

[a] DFOP model selected for parent compound
[b] FOMC model selected for parent compound

Table 7.1.2.1.1-15: Modeling endpoints for alpha-cypermethrin

Soil	Compound	Kinetic model	χ^2 error	$DegT_{50}$ [d]
Li 10	Alpha-cypermethrin	DFOP	1.3	75.3[a]
LUFA 2.2		DFOP	1.7	133.3[a]
LUFA 2.3		SFO	4.9	29.4

[a] Calculated as $DT_{50} = \ln 2/k2$

Table 7.1.2.1.1-16: Modeling endpoints for the metabolite M310I017

Soil	Compound	Kinetic model	χ^2 error	$DegT_{50}$ [d]	Formation fraction [-]
Li 10	M310I017	SFO[a]	13.5	22.6	0.215
LUFA 2.2		SFO[a]	13.2	42.3	0.188
LUFA 2.3		SFO[b]	15.5	3.1	0.629

[a] DFOP model selected for parent compound
[b] SFO model selected for parent compound

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

BAS-Code: BAS 310 I (alpha-cypermethrin)
Chemical name: Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molecular formula: $C_{22}H_{19}Cl_2NO_3$
Molar mass: 416.3 g mol^{-1} (unlabeled)

Label 1 (cyclopropane label)
Label: cyclopropane-1-^{14}C
Batch No.: 986-2101
Specific activity of a.s.: 4.71 MBq mg^{-1}
Radiochemical purity: 96.9%
Chemical purity: 91.8%

Label 2 (benzyl label)
Label: benzyl-1-^{14}C
Batch No.: 775-0601
Specific activity of a.s.: 4.74 MBq mg^{-1}
Radiochemical purity: 99.5%
Chemical purity: 96.1%

2. Soils

Three German agricultural soils (Li10, LUFA 2.2, and LUFA 2.3) were used for treatment and incubation. The soils were sieved through a 2 mm sieve before use and stored at about 4°C in the dark no longer than 3 months before use. The soil characteristics are summarized in Table 7.1.2.1.1-17.

Table 7.1.2.1.1-17: Properties of soils used for investigation of alpha-cypermethrin degradation rates under aerobic conditions

Soil designation	**Li10 Limburgerhof 13/1680/04** (Germany)	**LUFA 2.2 Mechtersheim 13/736/04** (Germany)	**LUFA 2.3 Hanhofen 13/570/04** (Germany)
DIN Particle size distribution [%]			
sand 0.063 – 2 mm	81.8	79.7	60.9
silt 0.002 – 0.063 mm	13.3	14.7	32.1
clay < 0.002 mm	5.0	5.6	6.9
textural class	loamy sand (SI2)	loamy sand (Sl2)	silty sand (Su3)
USDA Particle size distribution [%]			
sand 0.050 – 2 mm	84.0	82.7	64.5
silt 0.002 – 0.050 mm	11.0	11.7	28.6
clay < 0.002 mm	5.0	5.6	6.9
textural class	loamy sand	loamy sand	sandy loam
Total organic C [%]	0.93	1.40	0.71
pH [H_2O]	6.6	6.0	6.5
pH [$CaCl_2$]	6.1	5.4	5.9
Cation exchange capacity [$cmol^+ kg^{-1}$]	3.7	4.0	3.0
Max. water holding capacity [g $100g^{-1}$ dry weight]	26.9	32.4	24.1
Water holding capacity at pF 2.0 [g g^{-1} dry weight]	0.105	0.175	0.136
Water holding capacity at pF 2.5 [g g^{-1} dry weight]	0.093	0.140	0.115
Microbial biomass (day 0) [mg C $100g^{-1}$ dry soil]	28.4	49.6	32.0
Microbial biomass (day 71) [mg C $100g^{-1}$ dry soil]	11.3	16.4	16.9
Microbial biomass (day 136) [mg C $100g^{-1}$ dry soil]	7.1	13.1	12.4

B. STUDY DESIGN

1. Experimental conditions

The soils were adjusted to about 40% (Li10), 45% (LUFA 2.2), and 50% (LUFA 2.3) of the maximum water holding capacity (MWHC) and treated at a nominal application rate of 0.08 mg alpha-cypermethrin per kg dry soil which corresponds to a field application rate of 30 g a.s. ha^{-1} (calculated on the basis of an equal distribution in the top 2.5 cm soil layer and a soil density of 1.5 g cm^{-3}). Because of the low specific radioactivity of the test items and to ensure a better detection of possible degradation products, the amount of test item applied was increased to 0.3 mg test item kg^{-1} dry soil.

After application to bulk soil, portions of 100 g soil (dry weight basis) treated with either benzyl-^{14}C-labeled test item (Li10) or cyclopropane-^{14}C-labeled test item (LUFA 2.2 and LUFA 2.3) were filled into test vessels. The number of vessels was sufficient to allow duplicate sampling at each sampling time and to keep a limited number of reserve vessels.

The vessels were incubated in the dark for 120 days at a temperature of 20°C ± 2°C. Throughout the incubation period, the test vessels were continuously aerated with a slight stream of moistened synthetic air. The outgoing air was led through a series of gas-washing bottles containing trapping solutions for potential volatiles (ethylene glycol, 0.5 M H_2SO_4, 0.5 M NaOH). The water content of the soils was monitored throughout the incubation period by weighing the sampled vessels and evaporated water was replaced.

2. Sampling

Sampling times for the three soil types Li10, LUFA 2.2 and LUFA 2.3 were 0, 1, 3, 7, 10, 14, 30, and 60, 87, and 120 days after treatment (DAT).

At each sampling time, two replicate vessels with soil were removed from the incubation cabinet. Besides day 0, the volatile trapping solutions were sampled and replaced by new flasks with fresh solutions. The sampled trapping solutions were measured for radioactivity by liquid scintillation counting (LSC).

3. Description of the analytical procedures

The soils were consecutively extracted four times with 120 mL acetonitrile/water/formic acid (700/300/1, v/v/v) by shaking for about one hour. After each extraction, the suspension was centrifuged, filtered, and aliquots of each solution were radioassayed.

The four acetonitrile/water/formic acid extracts were combined and concentrated to dryness, re-dissolved in the extraction solvent and analyzed by LSC. Aliquots of the combined extracts were analyzed by LSC. After an additional concentration step, HPLC was performed to obtain the metabolite pattern. Separation of cis and trans isomers, as well as enantiomers of alpha-cypermethrin was performed on two separate HPLC systems.

In order to identify the peak of the unknown metabolite at about 74 min, fraction collection was performed with replicate 2 of the 10 DAT sample. Additionally, also the parent peak was isolated. After two steps of concentration to dryness and redissolving, both fractions were measured by LSC, radio-HPLC, and HPLC-MS.

Furthermore, samples of rat feces from a rat metabolism study [*CA 5.1.1/2, BASF DocID 2013/1086630*] were extracted twice with 150 mL acetonitrile by shaking for 30 min. After centrifugation and combination of the two extracts, aliquots were analyzed by LSC. Aliquots were concentrated to dryness, redissolved in ACN/H_2O/HCOOH (7/3/1, v/v/v) and analyzed by LSC. Fraction collections were performed as described in detail in the study report. The fractions were combined, concentrated, and analyzed by LSC, HPLC, MS and NMR.

The extracted soil was dried under the fume hood at room temperature and afterwards grounded in an analytical mill. Aliquots were combusted in order to determine the amount of non-extractable radioactive residues. The evolved $^{14}CO_2$ from each combusted aliquot was trapped in an Oxysolve C-400 scintillator and measured by LSC.

The fraction of NER was further characterized by NaOH extractions (0.5 M NaOH, three times). NaOH extracts were processed to determine the distribution of radioactivity to the humin, humic, and fulvic acid fractions. The humic acid fraction and the fulvic acid fraction were analyzed by LSC. The fulvic acid fraction was further partitioned with ethyl acetate. Concentrated ethyl acetate phases were analyzed by HPLC. The remaining radioactivity in the soil residues was determined by combustion analyses (humin fraction). Further details are given in the report.

The microbial biomass in the soil samples treated with and without solvent was determined before the start of the study, at 61 and 137 DAT. The method was based on the determination of oxygen consumption upon addition of glucose. In all three soils, the microbial biomass declined over the incubation phase. However, the results demonstrate that the soils were still viable and microbially active after 71 and 136 days.

4. Calculation of the degradation rate

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints). Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006) "Guidance Document on Estimating Persistence and Degradation Kinetics from Environmental Fate Studies on Pesticides in EU Registration" Report of the FOCUS Work Group on Degradation Kinetics, EC Document Reference Sanco/10058/2005 version 2.0, 434 pp.*].

The software package KinGUII (version 2.2014.224.1704) was used for parameter fitting [*SCHÄFER, D., MIKOLASCH, M., RAINBIRD, P., HARVEY, B. (2007*, *WITT, J., GAO, Z., MEYER, H. (2014)*]. The error tolerance and the number of iterations of the optimization tool were set to 0.00001 and 100, respectively.

For each data set, the kinetic models proposed by the FOCUS Kinetics guidance document were tested in order to identify the best-fit model. The recommended kinetic models, i.e. the single first order kinetics (SFO), the Gustafson-Holden model (FOMC), and bi-exponential (DFOP) kinetics are already implemented in KinGUI. The Goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test) [for details see Chapter 6.3 in *FOCUS (2006)*].

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The detailed results on the radioactive residues in the soils are presented in Table 7.1.2.1.1-18 to Table 7.1.2.1.1-20. The material balance throughout the incubation period ranged from 98.8 to 111.0% TAR for soil Li10, from 100.0 to 104.8% TAR for soil LUFA 2.2 and from 88.3 to 103.9% TAR for soil LUFA 2.3.

B. EXTRACTABLE AND BOUND RESIDUES

The distribution of radioactivity in extractable residues (ERR), non-extractable residues, CO_2 and other volatiles for each soil is shown in Table 7.1.2.1.1-18 to Table 7.1.2.1.1-20.

The amount of extractable radioactivity decreased steadily in the aerated soils to 27.9%, 39.9%, and 16.6% TAR at the end of the incubation for the soils Li10, LUFA 2.2, and LUFA 2.3, respectively.

The non-extractable radioactive residues (NER) increased during the study from 0.6%, 1.5%, and 1.3% TAR on day 0 to 32.2%, 23.5%, and 34.8% TAR after 120 days of incubation for the soils Li10, LUFA 2.2, and LUFA 2.3, respectively.

Table 7.1.2.1.1-18: Material balance and distribution of radioactivity after application of benzyl-^{14}C-labeled alpha-cypermethrin to Soil Li10 [% TAR]

DAT	Extractable residues					NER	Volatiles*				Mass Balance
	ACN/ H_2O 1	ACN/ H_2O 2	ACN/ H_2O 3	ACN/ H_2O 4	Total		NaOH	Ethylene Glycol	H_2SO_4	Total	
0 (Rep 1)	80.5	16.7	3.5	0.7	101.3	0.5	n.a.	n.a.	n.a.	n.a.	101.9
0 (Rep 2)	76.7	16.8	3.3	0.8	97.5	0.6	n.a.	n.a.	n.a.	n.a.	98.1
0 (mean)	**78.6**	**16.7**	**3.4**	**0.8**	**99.4**	**0.6**	**n.a.**	**n.a.**	**n.a.**	**n.a.**	**100.0**
1 (Rep 1)	73.9	21.0		0.8	95.8	2.8	0.8	0.0	0.0	0.8	99.3
1 (Rep 2)	73.9	20.0		0.8	94.6	2.8	0.9	0.0	0.0	0.9	98.3
1 (mean)	**73.9**	**20.5**		**0.8**	**95.2**	**2.8**	**0.8**	**0.0**	**0.0**	**0.8**	**98.8**
3 (Rep 1)	71.1	17.9	3.6	0.8	93.3	5.9	2.1	0.0	0.0	2.1	101.4
3 (Rep 2)	70.1	18.0	3.6	0.8	92.5	5.5	2.1	0.0	0.0	2.1	100.0
3 (mean)	**70.6**	**17.9**	**3.6**	**0.8**	**92.9**	**5.7**	**2.1**	**0.0**	**0.0**	**2.1**	**100.7**
7 (Rep 1)	66.6	16.0	3.5	0.8	86.9	10.8	4.8	0.0	0.0	4.8	102.6
7 (Rep 2)	68.1	15.2	3.5	0.8	87.6	11.0	4.8	0.0	0.0	4.8	103.4
7 (mean)	**67.3**	**15.6**	**3.5**	**0.8**	**87.3**	**10.9**	**4.8**	**0.0**	**0.0**	**4.8**	**103.0**
10 (Rep 1)	60.7	16.0	3.3	0.8	80.8	14.5	7.0	0.0	0.0	7.0	102.3
10 (Rep 2)	60.3	10.5	3.6	0.8	75.2	16.3	6.9	0.0	0.0	6.9	98.4
10 (mean)	**60.5**	**13.3**	**3.4**	**0.8**	**78.0**	**15.4**	**7.0**	**0.0**	**0.0**	**7.0**	**100.4**
14 (Rep 1)	55.2	14.3	3.1	0.7	73.3	16.8	9.8	0.0	0.0	9.8	99.9
14 (Rep 2)	55.0	14.1	3.0	0.7	72.8	18.9	9.7	0.0	0.0	9.7	101.4
14 (mean)	**55.1**	**14.2**	**3.0**	**0.7**	**73.1**	**17.8**	**9.7**	**0.0**	**0.0**	**9.8**	**100.6**
30 (Rep 1)	44.5	11.4	2.5	0.7	59.2	25.1	21.1	0.0	0.0	21.1	105.4
30 (Rep 2)	44.7	11.6	2.7	0.7	59.8	24.8	21.1	0.0	0.0	21.1	105.6
30 (mean)	**44.6**	**11.5**	**2.6**	**0.7**	**59.5**	**24.9**	**21.1**	**0.0**	**0.0**	**21.1**	**105.5**
60 (Rep 1)	32.0	8.3	2.1	0.7	43.0	31.1	36.1	0.0	0.0	36.1	110.2
60 (Rep 2)	31.8	8.5	2.1	0.6	43.1	29.9	35.9	0.0	0.0	35.9	108.9
60 (mean)	**31.9**	**8.4**	**2.1**	**0.6**	**43.0**	**30.5**	**36.0**	**0.0**	**0.0**	**36.0**	**109.6**
87 (Rep 1)	25.4	7.0	1.7	0.5	34.6	31.7	44.0	0.0	0.0	44.0	110.4
87 (Rep 2)	26.4	7.0	1.6	0.6	35.6	32.5	43.5	0.0	0.0	43.5	111.7
87 (mean)	**25.9**	**7.0**	**1.7**	**0.6**	**35.1**	**32.1**	**43.8**	**0.0**	**0.0**	**43.8**	**111.0**
120 (Rep 1)	22.0	4.5	1.0	0.2	27.8	32.3	48.7	0.0	0.0	48.7	108.8
120 (Rep 2)	22.3	4.4	1.0	0.2	28.0	32.1	48.8	0.0	0.0	48.8	108.9
120 (mean)	**22.2**	**4.5**	**1.0**	**0.2**	**27.9**	**32.2**	**48.8**	**0.0**	**0.0**	**48.8**	**108.9**

TAR = Total applied radioactivity
DAT = Days after treatment
ACN = Acetonitrile
n.a. = Not analyzed
* No other volatiles than CO_2 were found

Table 7.1.2.1.1-19: Material balance and distribution of radioactivity after application of cyclopropane-1-^{14}C-labeled alpha-cypermethrin to Soil LUFA 2.2 [% TAR]

DAT	Extractable residues					NER	Volatiles*				Mass balance
	ACN/ H_2O 1	ACN/ H_2O 2	ACN/ H_2O 3	ACN/ H_2O 4	Total		NaOH	Ethylene glycol	H_2SO_4	Total	
0 (Rep 1)	71.6	20.0	4.9	1.3	97.8	1.5	n.a.	n.a.	n.a.	n.a.	99.3
0 (Rep 2)	72.6	20.5	4.9	1.2	99.2	1.5	n.a.	n.a.	n.a.	n.a.	100.7
0 (mean)	**72.1**	**20.3**	**4.9**	**1.2**	**98.5**	**1.5**	**n.a.**	**n.a.**	**n.a.**	**n.a.**	**100.0**
1 (Rep 1)	71.7	20.4	5.1	1.1	98.3	3.0	0.4	0.0	0.0	0.4	101.6
1 (Rep 2)	72.4	18.0	4.6	1.1	96.1	2.8	0.1	0.0	0.0	0.1	99.0
1 (mean)	**72.1**	**19.2**	**4.8**	**1.1**	**97.2**	**2.9**	**0.3**	**0.0**	**0.0**	**0.3**	**100.3**
3 (Rep 1)	69.4	19.4	4.3	1.1	94.1	5.2	1.4	0.0	0.0	1.4	100.7
3 (Rep 2)	69.7	18.4	4.3	1.1	93.5	4.7	1.9	0.0	0.0	1.9	100.1
3 (mean)	**69.5**	**18.9**	**4.3**	**1.1**	**93.8**	**4.9**	**1.7**	**0.0**	**0.0**	**1.7**	**100.4**
7 (Rep 1)	65.1	18.2	4.5	1.1	88.9	9.5	5.2	0.0	0.0	5.2	103.6
7 (Rep 2)	66.3	18.3	4.6	1.1	90.4	9.2	5.2	0.0	0.0	5.2	104.8
7 (mean)	**65.7**	**18.3**	**4.5**	**1.1**	**89.6**	**9.4**	**5.2**	**0.0**	**0.0**	**5.2**	**104.2**
10 (Rep 1)	60.6	16.5	4.2	1.1	82.3	12.6	7.6	0.0	0.0	7.6	102.5
10 (Rep 2)	61.1	16.9	4.3	1.2	83.5	11.5	7.6	0.0	0.0	7.6	102.6
10 (mean)	**60.8**	**16.7**	**4.3**	**1.2**	**82.9**	**12.0**	**7.6**	**0.0**	**0.0**	**7.6**	**102.6**
14 (Rep 1)	54.5	15.3	3.8	1.1	74.6	17.0	10.4	0.0	0.0	10.4	102.0
14 (Rep 2)	56.3	15.5	3.7	1.0	76.5	14.1	12.7	0.0	0.0	12.7	103.3
14 (mean)	**55.4**	**15.4**	**3.7**	**1.0**	**75.6**	**15.5**	**11.6**	**0.0**	**0.0**	**11.6**	**102.7**
30 (Rep 1)	45.6	12.7	3.4	1.0	62.7	19.8	18.3	0.0	0.0	18.3	100.8
30 (Rep 2)	46.7	13.0	3.4	1.0	64.2	18.7	20.9	0.0	0.0	20.9	103.8
30 (mean)	**46.2**	**12.8**	**3.4**	**1.0**	**63.4**	**19.3**	**19.6**	**0.0**	**0.0**	**19.6**	**102.3**
60 (Rep 1)	35.4	9.7	2.6	0.8	48.4	22.6	29.0	0.0	0.0	29.0	99.9
60 (Rep 2)	38.3	11.0	2.9	0.9	53.0	21.9	31.3	0.0	0.0	31.3	106.2
60 (mean)	**36.8**	**10.3**	**2.7**	**0.8**	**50.7**	**22.2**	**30.1**	**0.0**	**0.0**	**30.1**	**103.1**
87 (Rep 1)	33.1	9.8	2.4	0.8	46.1	22.9	35.4	0.0	0.0	35.4	104.4
87 (Rep 2)	34.0	10.0	2.7	0.9	47.6	12.8	37.6	0.0	0.0	37.6	98.1
87 (mean)	**33.5**	**9.9**	**2.5**	**0.9**	**46.9**	**17.9**	**36.5**	**0.0**	**0.0**	**36.5**	**101.3**
120 (Rep 1)	30.5	7.0	1.7	0.3	39.5	23.0	40.2	0.0	0.0	40.2	102.7
120 (Rep 2)	31.0	7.1	1.8	0.5	40.4	23.9	42.6	0.0	0.0	42.6	106.9
120 (mean)	**30.7**	**7.0**	**1.7**	**0.4**	**39.9**	**23.5**	**41.4**	**0.0**	**0.0**	**41.4**	**104.8**

TAR = Total applied radioactivity
DAT = Days after treatment
ACN = Acetonitrile
n.a. = Not analyzed
* No other volatiles than CO_2 were found

Table 7.1.2.1.1-20: Material balance and distribution of radioactivity after application of cyclopropane-1-^{14}C-labeled alpha-cypermethrin to Soil LUFA 2.3 [% TAR]

DAT	Extractable residues					NER	Volatiles*				Mass balance
	ACN/ H_2O 1	ACN/ H_2O 2	ACN/ H_2O 3	ACN/ H_2O 4	Total		NaOH	Ethylene glycol	H_2SO_4	Total	
0 (Rep 1)	79.5	16.8	3.4	0.6	100.3	1.3	n.a.	n.a.	n.a.	n.a.	101.6
0 (Rep 2)	76.2	16.8	3.4	0.7	97.1	1.3	n.a.	n.a.	n.a.	n.a.	98.4
0 (mean)	**77.8**	**16.8**	**3.4**	**0.7**	**98.7**	**1.3**	**n.a.**	**n.a.**	**n.a.**	**n.a.**	**100.0**
1 (Rep 1)	77.0	18.5		0.8	96.3	4.4	0.7	0.0	0.0	0.7	101.4
1 (Rep 2)	76.7	19.7		0.8	97.2	4.1	0.5	0.0	0.0	0.5	101.8
1 (mean)	**76.9**	**19.1**		**0.8**	**96.7**	**4.2**	**0.6**	**0.0**	**0.0**	**0.6**	**101.6**
3 (Rep 1)	70.8	16.9	3.3	0.7	91.6	7.5	1.5	0.0	0.0	1.5	100.6
3 (Rep 2)	71.7	17.3	3.3	0.7	93.0	6.8	1.4	0.0	0.0	1.4	101.2
3 (mean)	**71.2**	**17.1**	**3.3**	**0.7**	**92.3**	**7.1**	**1.5**	**0.0**	**0.0**	**1.5**	**100.9**
7 (Rep 1)	67.0	15.6	3.5	0.8	86.9	12.0	4.9	0.0	0.0	4.9	103.8
7 (Rep 2)	67.3	15.2	3.6	0.9	86.9	12.4	4.6	0.0	0.0	4.6	103.9
7 (mean)	**67.1**	**15.4**	**3.5**	**0.8**	**86.9**	**12.2**	**4.7**	**0.0**	**0.0**	**4.7**	**103.9**
10 (Rep 1)	62.1	15.1	3.5	0.8	81.5	15.1	7.5	0.0	0.0	7.5	104.1
10 (Rep 2)	61.3	15.2	3.7	0.9	81.0	15.4	7.1	0.0	0.0	7.1	103.5
10 (mean)	**61.7**	**15.1**	**3.6**	**0.9**	**81.3**	**15.2**	**7.3**	**0.0**	**0.0**	**7.3**	**103.8**
14 (Rep 1)	55.5	14.3	3.0	0.8	73.5	17.8	10.8	0.0	0.0	10.8	102.1
14 (Rep 2)	57.1	14.4	3.1	0.8	75.4	18.4	10.4	0.0	0.0	10.4	104.2
14 (mean)	**56.3**	**14.3**	**3.0**	**0.8**	**74.5**	**18.1**	**10.6**	**0.0**	**0.0**	**10.6**	**103.1**
30 (Rep 1)	40.3	9.6	2.4	0.7	52.9	26.7	19.4	0.0	0.0	19.4	99.0
30 (Rep 2)	41.3	10.9	2.6	0.8	55.6	27.1	18.6	0.0	0.0	18.6	101.4
30 (mean)	**40.8**	**10.3**	**2.5**	**0.8**	**54.3**	**26.9**	**19.0**	**0.0**	**0.0**	**19.0**	**100.2**
60 (Rep 1)	23.0	5.8	1.6	0.5	30.9	34.7	25.2	0.0	0.0	25.2	90.8
60 (Rep 2)	22.7	5.9	1.7	0.6	30.9	34.2	26.8	0.0	0.0	26.8	92.0
60 (mean)	**22.8**	**5.8**	**1.6**	**0.6**	**30.9**	**34.5**	**26.0**	**0.0**	**0.0**	**26.0**	**91.4**
87 (Rep 1)	16.9	4.5	1.2	0.5	23.1	36.6	31.1	0.0	0.0	31.1	90.7
87 (Rep 2)	17.2	4.5	1.3	0.5	23.5	35.4	32.5	0.0	0.0	32.5	91.4
87 (mean)	**17.1**	**4.5**	**1.2**	**0.5**	**23.3**	**36.0**	**31.8**	**0.0**	**0.0**	**31.8**	**91.1**
120 (Rep 1)	13.1	2.9	0.5	0.2	16.7	33.3	36.1	0.0	0.0	36.1	86.2
120 (Rep 2)	13.0	2.7	0.6	0.2	16.6	36.2	37.7	0.0	0.0	37.7	90.5
120 (mean)	**13.0**	**2.8**	**0.6**	**0.2**	**16.6**	**34.8**	**36.9**	**0.0**	**0.0**	**36.9**	**88.3**

TAR = Total applied radioactivity
DAT = Days after treatment
ACN = Acetonitrile
n.a. = Not analyzed
* No other volatiles than CO_2 were found

C. VOLATILIZATION

Mineralization reached a total of up to 48.8% TAR after 120 days of incubation (in soil Li10). No other volatiles than $^{14}CO_2$ occurred.

D. TRANSFORMATION OF PARENT COMPOUND

All combined soil extracts were analyzed by radio-HPLC. The results are summarized in Table 7.1.2.1.1-21 to Table 7.1.2.1.1-23.

Considering all three soils, the concentration of alpha-cypermethrin decreased throughout the incubation period from 99.4%, 98.5%, and 98.7% TAR at day 0, to 20.8%, 30.1%, and 10.5% TAR at the end of the incubation for the soils Li10, LUFA 2.2, and LUFA 2.3, respectively.

Several degradation products were detected in the extracts but only one peak appeared in amounts higher than 5% TAR. The unknown metabolite eluting after 74 min was present in the soil extracts in amounts of max. 5.8%, 5.9%, and 5.3% TAR in soils Li10, LUFA 2.2, and LUFA 2.3, respectively. The maxima were reached between 10 and 30 days after application with a decrease thereafter.

The unknown peak at around 74 min was isolated via HPLC fraction collection and MS analysis was performed. MS analysis identified the unknown substance as the product of the hydroxylation of alpha-cypermethrin on the benzyl ring. However, it did not provide information on the exact position of the hydroxylation (ortho, meta, or para position).

Since a hydroxylated metabolite was identified in the rat metabolism study [*CA 5.1.1/2, BASF DocID 2013/1086630*], the corresponding rat sample was newly extracted and the hydroxylated metabolite was isolated by fraction collection. The collection was analyzed by HPLC-MS and NMR, and it could be confirmed that the hydroxylation is located on the para position of the benzyl ring (M310I017). The identification of M310I017 in the soil extracts was confirmed for each soil by co-chromatography with an exemplary soil extract (10 DAT, replicate 1).

Table 7.1.2.1.1-21: Radio-HPLC Analysis of Extracts of Soil Li10 after application of benzyl-^{14}C-labeled alpha-cypermethrin [% TAR]

Days after treatment (DAT)	Total	M310I017	alpha-cypermethrin	Sum others*
0 [rep 1]	101.3	-	101.3	0.0
0 [rep 2]	97.5	-	97.5	0.0
0 [mean]	**99.4**	**-**	**99.4**	**0.0**
1 [rep 1]	95.8	-	95.8	0.0
1 [rep 2]	94.6	-	94.6	0.0
1 [mean]	**95.2**	**-**	**95.2**	**0.0**
3 [rep 1]	93.3	2.7	90.6	0.0
3 [rep 2]	92.5	2.9	89.6	0.0
3 [mean]	**92.9**	**2.8**	**90.1**	**0.0**
7 [rep 1]	86.9	4.9	80.1	2.0
7 [rep 2]	87.6	5.0	80.3	2.2
7 [mean]	**87.3**	**5.0**	**80.2**	**2.1**
10 [rep 1]	80.8	6.3	72.1	2.4
10 [rep 2]	75.2	5.2	68.4	1.6
10 [mean]	**78.0**	**5.8**	**70.2**	**2.0**
14 [rep 1]	73.3	5.7	65.4	2.2
14 [rep 2]	72.8	5.2	64.1	3.5
14 [mean]	**73.1**	**5.5**	**64.7**	**2.9**
30 [rep 1]	59.2	5.0	48.5	5.7
30 [rep 2]	59.8	6.1	48.9	4.8
30 [mean]	**59.5**	**5.6**	**48.7**	**5.2**
60 [rep 1]	43.0	3.9	34.4	4.7
60 [rep 2]	43.1	3.6	35.4	4.0
60 [mean]	**43.0**	**3.8**	**34.9**	**4.4**
87 [rep 1]	34.6	3.7	26.4	4.5
87 [rep 2]	35.6	3.6	27.8	4.2
87 [mean]	**35.1**	**3.7**	**27.1**	**4.4**
120 [rep 1]	27.8	2.8	21.4	3.7
120 [rep 2]	28.0	2.8	20.2	4.9
120 [mean]	**27.9**	**2.8**	**20.8**	**4.3**

TAR = Total applied radioactivity
* individual peaks < 3% TAR

Table 7.1.2.1.1-22: Radio-HPLC analysis of extracts of soil LUFA 2.2 after application of cyclopropane-1-^{14}C-labeled alpha-cypermethrin [% TAR]

Days after treatment (DAT)	Total	M310I017	alpha-cypermethrin	Sum others*
0 [rep 1]	97.8	-	97.8	-
0 [rep 2]	99.2	-	99.2	-
0 [mean]	**98.5**	**-**	**98.5**	**-**
1 [rep 1]	98.3	-	98.3	-
1 [rep 2]	96.1	-	96.1	-
1 [mean]	**97.2**	**-**	**97.2**	**-**
3 [rep 1]	94.1	1.6	92.5	-
3 [rep 2]	93.5	1.2	92.3	-
3 [mean]	**93.8**	**1.4**	**92.4**	**-**
7 [rep 1]	88.9	3.8	84.2	0.9
7 [rep 2]	90.4	4.0	85.4	1.0
7 [mean]	**89.6**	**3.9**	**84.8**	**0.9**
10 [rep 1]	82.3	6.1	74.7	1.5
10 [rep 2]	83.5	4.7	77.3	1.6
10 [mean]	**82.9**	**5.4**	**76.0**	**1.6**
14 [rep 1]	74.6	5.5	66.9	2.2
14 [rep 2]	76.5	5.8	68.7	2.0
14 [mean]	**75.6**	**5.6**	**67.8**	**2.1**
30 [rep 1]	62.7	5.6	52.4	4.6
30 [rep 2]	64.2	6.2	53.0	5.1
30 [mean]	**63.4**	**5.9**	**52.7**	**4.8**
60 [rep 1]	48.4	4.5	37.1	6.7
60 [rep 2]	53.0	4.9	44.2	3.9
60 [mean]	**50.7**	**4.7**	**40.7**	**5.3**
87 [rep 1]	46.1	4.7	36.5	4.9
87 [rep 2]	47.6	5.1	37.2	5.3
87 [mean]	**46.9**	**4.9**	**36.9**	**5.1**
120 [rep 1]	39.5	4.6	29.3	5.5
120 [rep 2]	40.4	3.9	30.9	5.6
120 [mean]	**39.9**	**4.2**	**30.1**	**5.6**

TAR = Total applied radioactivity
* individual peaks < 3% TAR

Table 7.1.2.1.1-23: Radio-HPLC analysis of extracts of soil LUFA 2.3 after application of cyclopropane-1-^{14}C-labeled alpha-cypermethrin [% TAR]

Days after treatment (DAT)	Total	M310I017	alpha-cypermethrin	Sum others**
0 [rep 1]	100.3	-	100.3	-
0 [rep 2]	97.1	-	97.1	-
0 [mean]	**98.7**	**-**	**98.7**	**-**
1 [rep 1]	96.3	-	96.3	-
1 [rep 2]	97.2	-	97.2	-
1 [mean]	**96.7**	**-**	**96.7**	**-**
3 [rep 1]	91.6	3.1	88.6	-
3 [rep 2]	93.0	3.0	90.0	-
3 [mean]	**92.3**	**3.0**	**89.3**	**-**
7 [rep 1]	86.9	4.5	81.2	1.3
7 [rep 2]	86.9	5.4	80.4	1.1
7 [mean]	**86.9**	**5.0**	**80.8**	**1.2**
10 [rep 1]	81.5	5.3	70.9	5.2
10 [rep 2]	81.0	5.2	73.4	2.4
10 [mean]	**81.3**	**5.3**	**72.2**	**3.8**
14 [rep 1]	73.5	4.7	63.0	5.9
14 [rep 2]	75.4	4.3	65.4	5.7
14 [mean]	**74.5**	**4.5**	**64.2**	**5.8**
30 [rep 1]	52.9	2.5	42.6	7.8
30 [rep 2]	55.6	3.3	45.4	6.9
30 [mean]	**54.3**	**2.9**	**44.0**	**7.4**
60 [rep 1]	30.9	2.2	24.9	3.7
60 [rep 2]	30.9	1.7	23.8	5.5
60 [mean]	**30.9**	**2.0**	**24.3**	**4.6**
87 [rep 1]	23.1	1.1	18.4	3.6
87 [rep 2]	23.5	1.1	18.6	3.7
87 [mean]	**23.3**	**1.1**	**18.5**	**3.7**
120 [rep 1]	16.7	0.7	10.8	5.2
120 [rep 2]	16.6	1.1	10.2	5.3
120 [mean]	**16.6**	**0.9**	**10.5**	**5.3**

TAR = Total applied radioactivity
* individual peaks < 3% TAR

Throughout the study, only one peak was observed in the chromatograms containing only cis isomers. This confirmed that no trans isomers were formed during the study.

Separation of enantiomers was performed by chiral HPLC. A change of the enantiomeric ratio from 1.0 to 1.5, 2.3, and 2.0 could be observed in the soils Li10, LUFA 2.2, and LUFA 2.3, respectively. The results of the determination of the R/S ratio of the CN-bearing carbon atom of alpha-cypermethrin are presented in Table 7.1.2.1-24, as well as in the study report.

Table 7.1.2.1-24: Chiral radio-HPLC analysis of extracts of soil Li10, LUFA 2.2, and LUFA 2.3 after application of 14C-labeled alpha-cypermethrin [% TAR]

Days after treatment (DAT)	Li10		LUFA 2.2		LUFA 2.3	
	Reg. No. 5835576 (R enant.)	Reg. No. 5835577 (S enant.)	Reg. No. 5835576 (R enant.)	Reg. No. 5835577 (S enant.)	Reg. No. 5835576 (R enant.)	Reg. No. 5835577 (S enant.)
0 [rep 1]	46.0	47.7	45.1	44.1	48.0	47.4
0 [rep 2]	46.6	45.7	45.3	45.9	45.7	45.4
0 [mean]	**46.3**	**46.7**	**45.2**	**45.0**	**46.9**	**46.4**
1 [rep 1]	44.9	45.2	45.9	45.2	46.0	46.6
1 [rep 2]	43.4	44.4	45.6	45.8	45.9	45.5
1 [mean]	**44.1**	**44.8**	**45.7**	**45.5**	**46.0**	**46.0**
3 [rep 1]	42.3	42.6	41.8	43.1	39.9	42.7
3 [rep 2]	41.7	42.9	40.8	43.1	38.6	41.3
3 [mean]	**42.0**	**42.8**	**41.3**	**43.1**	**39.3**	**42.0**
7 [rep 1]	34.8	39.3	35.4	40.5	30.6	39.0
7 [rep 2]	35.2	39.5	36.0	39.3	31.2	37.3
7 [mean]	**35.0**	**39.4**	**35.7**	**39.9**	**30.9**	**38.2**
10 [rep 1]	31.7	35.6	31.4	37.1	25.7	33.4
10 [rep 2]	29.5	32.7	32.6	38.5	26.3	33.9
10 [mean]	**30.6**	**34.2**	**32.0**	**37.8**	**26.0**	**33.7**
14 [rep 1]	27.0	30.5	25.4	32.0	20.8	29.6
14 [rep 2]	26.2	30.4	26.9	33.6	21.2	30.8
14 [mean]	**26.6**	**30.5**	**26.2**	**32.8**	**21.0**	**30.2**
30 [rep 1]	19.0	25.9	17.6	26.9	11.8	20.0
30 [rep 2]	18.9	24.2	18.0	27.3	12.5	22.4
30 [mean]	**19.0**	**25.1**	**17.8**	**27.1**	**12.1**	**21.2**
60 [rep 1]	11.1	16.8	10.9	20.4	5.8	10.3
60 [rep 2]	11.4	16.3	13.1	24.6	6.3	11.2
60 [mean]	**11.3**	**16.6**	**12.0**	**22.5**	**6.0**	**10.8**
87 [rep 1]	8.1	11.5	9.8	20.8	4.4	9.1
87 [rep 2]	7.8	12.0	10.8	21.7	5.5	10.8
87 [mean]	**7.9**	**11.8**	**10.3**	**21.3**	**5.0**	**10.0**
120 [rep 1]	7.7	11.1	7.8	18.3	3.3	6.0
120 [rep 2]	6.8	9.7	8.1	17.9	3.6	7.4
120 [mean]	**7.3**	**10.4**	**8.0**	**18.1**	**3.5**	**6.7**

TAR = Total applied radioactivity
Enant. = Enantiomer

E. CHARACTERIZATION OF NON-EXTRACTABLE ("BOUND") RESIDUES

Summaries of bound residue fractionating following extraction by NaOH and H_2O are given in Table 7.1.2.1.1-25 to Table 7.1.2.1.1-27.

After 120 days of incubation, 10.8% TAR could be assigned to the fulvic acids fraction and 14.1% TAR to the humic acids fraction in soil Li10. On soil LUFA 2.2, 10.5% TAR could be assigned to the fulvic acids fraction and 7.9% TAR to the humic acids fraction. On soil LUFA 2.3, 14.9% TAR could be assigned to the fulvic acids fraction and 6.8% TAR to the humic acids fraction at 120 DAT.

About 5.5%, 3.2%, and 7.8% TAR remained unextractable for the soils Li10, LUFA 2.2, and LUFA 2.3 after 60 days. After 120 days, 7.0%, 3.8%, and 7.8% TAR remained unextractable for the soil Li10, LUFA 2.2, and LUFA 2.3, respectively. This portion of unextractable radioactivity was assigned to the humin fraction.

The ethyl acetate phase was further analyzed by HPLC. The main peak was found to occur with a maximum of 5.5% TAR at 60 DAT for the cyclopropane-1-^{14}C-labeled-alpha-cypermethrin on soil LUFA 2.3. No further attends were made to identify the compounds observed in the water phase.

Table 7.1.2.1.1-25: Characterization of bound residues [% TAR]

Soil	DAT	NER*	ERR II			NER II (Humins)	Sum ERR II + NER II*
			NaOH	H_2O	Total		
Li10	60 [rep 2]	29.9	21.0	0.8	21.8	5.5	27.3
Li10	120 [rep 2]	32.1	24.3	1.0	25.3	7.0	32.3
LUFA 2.3	60 [rep 2]	34.2	23.0	0.9	23.9	7.8	31.7
LUFA 2.3	120 [rep 2]	36.2	22.2	1.1	23.2	7.8	31.0
LUFA 2.2	60 [rep 2]	21.9	14.9	0.0	14.9	3.2	18.1
LUFA 2.2	120 [rep 2]	23.9	14.8	0.1	15.0	3.8	18.8

TAR = Total applied radioactivity
DAT = Days after treatment
NER = Non-extractable residues
ERR = Extractable radioactive residues
* Deviations from initial NER values have to be attributed to differing LSC results

Table 7.1.2.1.1-26: Fractionation of alkali-soluble residues [% TAR]

Soil	DAT	Sum of NaOH and water extracts*	Humic acids	Fulvic acids	Sum humic and fulvic acids*
Li10	60 [rep 2]	21.8	12.1	9.5	21.5
Li10	120 [rep 2]	25.3	14.1	10.8	24.9
LUFA 2.3	60 [rep 2]	23.9	6.8	15.8	22.6
LUFA 2.3	120 [rep 2]	23.2	6.8	14.9	21.7
LUFA 2.2	60 [rep 2]	14.9	6.9	9.9	16.8
LUFA 2.2	120 [rep 2]	15.0	7.9	10.5	18.4

TAR = Total applied radioactivity
DAT = Days after treatment
* Deviations have to be attributed to differing LSC results

Table 7.1.2.1.1-27: Fractionation of residues from the fulvic acids [% TAR]

Soil	DAT	Fulvic acids*	Ethyl acetate	Aqueous phase	Sum ethyl acetate and aqueous phase*
Li10	60 [rep 2]	9.5	3.2	5.9	9.1
Li10	120 [rep 2]	10.8	3.3	6.9	10.3
LUFA 2.3	60 [rep 2]	15.8	7.8	7.8	15.6
LUFA 2.3	120 [rep 2]	14.9	6.7	8.1	14.8
LUFA 2.2	60 [rep 2]	9.9	4.4	4.8	9.2
LUFA 2.2	120 [rep 2]	10.5	4.3	5.3	9.6

TAR = Total applied radioactivity
DAT = Days after treatment
* Deviations from initial NER values have to be attributed to differing LSC results

F. KINETIC MODELING RESULTS

A summary of the $DegT_{50}$ and $DegT_{90}$ values of alpha-cypermethrin and its metabolite M310I017 estimated as trigger endpoints are given in Table 7.1.2.1.1-28 and Table 7.1.2.1.1-29.

Table 7.1.2.1.1-28: Trigger endpoints for alpha-cypermethrin

Soil	Compound	Best-fit model	χ^2 error	p (t-test)	Visual fit	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Li10	Alpha-cypermethrin	DFOP	1.3	k1: <0.01 k2: <0.01 g: <0.01	Good	28.3	196.3
LUFA 2.2		DFOP	1.7	k1: <0.01 k2: <0.01 g: <0.01	Good	35.0	329.4
LUFA 2.3		FOMC	1.2	β: <0.01	Good	24.3	134.6

Table 7.1.2.1.1-29: Trigger endpoints for the metabolite M310I017

Soil	Compound	Best-fit model	χ^2 error	p (t-test)	Visual fit	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Li10	M310I017	SFO[a]	13.5	k: <0.01	Acceptable	22.6	75.1
LUFA 2.2		SFO[a]	13.2	k: <0.01	Acceptable	42.3	140.4
LUFA 2.3		SFO[b]	16.1	k: <0.01	Acceptable	4.9	16.2

[a] DFOP model selected for parent compound
[b] FOMC model selected for parent compound

A summary of the $DegT_{50}$ and $DegT_{90}$ values of alpha-cypermethrin and its metabolite M310I017 estimated as modeling endpoints are given in Table 7.1.2.1.1-30 and Table 7.1.2.1.1-31.

Table 7.1.2.1.1-30: Modeling endpoints for alpha-cypermethrin

Soil	Compound	Kinetic model	$\chi 2$ error	p (t-test)	Visual fit	$DegT_{50}$ [d]
Li 10	Alpha-cypermethrin	DFOP	1.3	k1: <0.01 k2: <0.01 g: <0.01	Good	75.3[a]
LUFA 2.2		DFOP	1.7	k1: <0.01 k2: <0.01 g: <0.01	Good	133.3[a]
LUFA 2.3		SFO	4.9	k: <0.01	Acceptable	29.4

[a] Calculated as $DT_{50} = \ln 2/k2$

Table 7.1.2.1.1-31: Modeling endpoints for the metabolite M310I017

Soil	Compound	Kinetic model	$\chi 2$ error	p (t-test)	Visual fit	$DegT_{50}$ [d]	Formation fraction [-]
Li 10	M310I017	SFO[a]	13.5	k: <0.01	Acceptable	22.6	0.215
LUFA 2.2		SFO[a]	13.2	k: <0.01	Acceptable	42.3	0.188
LUFA 2.3		SFO[b]	15.5	k: <0.01	Acceptable	3.1	0.629

[a] DFOP model selected for parent compound
[b] SFO model selected for parent compound

III. CONCLUSION

Alpha-cypermethrin degraded in aerobic soils with DT_{50} values of 28.3, 35.0 and 24.3 days in the soils Li10, LUFA 2.2, and LUFA 2.3, respectively. At the same time the fraction of non-extractable residues increased reaching between 23.5 and 34.8% TAR at the end of the study after 120 days. $^{14}CO_2$ was formed in significant amounts (between 36.9% and 48.8% TAR).

The metabolite M310I017 was observed with maximum concentrations in the range 5.3% - 5.9% TAR. The metabolite M310I017 was observed in the extracts of all three soils. Some enantioselectivity was observed in the degradation with fater degradation of the R enantiomer.

Normalization of laboratory degradation endpoints to reference conditions

Since for environmental fate modeling soil $DegT_{50}$ values at reference conditions (temperature of 20°C and soil moisture at field capacity, i.e. pF2) are required, the modeling endpoints ($DegT_{50}$) reported in CA 7.1.1.1/1 [*BASF Doc ID 2014/1000641*], CA 7.1.2.1.1/4 [*BASF Doc ID 2014/1159491*], and CA 7.1.2.1.2 [*CLASS T. and DORN U. (2003)*] were normalized following the recommendations of FOCUS [*FOCUS (2012): Generic Guidance for Tier 1 FOCUS Ground Water Assessments, version 2.1 (Dec. 2012), 64 pp.*]. Since the laboratory studies were performed at 20°C, a temperature correction was not necessary. The soil moisture normalization was performed using the moisture dependency equations by Walker as described in Equation 7.1.2.1.1-1.

Equation 7.1.2.1.1-1: Calculation of the moisture correction factor according to Walker

$$f_{moist} = \begin{cases} \left(\frac{\theta_{act}}{\theta_{ref}}\right)^{0.7} & if \quad \theta_{act} < \theta_{ref} \\ 1 & if \quad \theta_{act} \geq \theta_{ref} \end{cases}$$

with:	f_{moist}	moisture correction factor	[-]
	θ_{ref}	reference soil moisture at field capacity (pF2, 10 kPa)	[g / 100 g dry soil]
	θ_{act}	actual soil moisture during incubation	[g / 100 g dry soil]

For [*BASF Doc ID 2014/1000641*] and [*BASF Doc ID 2014/1159491*], the actual soil moisture and the reference moisture were taken from the study reports. For Class and Dorn [*CLASS T. and DORN U. (2003)*], the actual soil moisture was taken from the study report and the reference soil moisture was derived from FOCUS [*FOCUS (2012)*]. The normalized $DegT_{50}$ values were calculated by multiplying the $DegT_{50}$ values at study conditions by the correction factor f_{moist} as described in Equation 7.1.2.1.1-2.

Equation 7.1.2.1.1-2: Calculation of the $DegT_{50}$ at reference conditions (20°C, pF2)

$$DegT_{50,ref} = DegT_{50,act} \times f_{moist}$$

with:	$DegT_{50,ref}$	normalized $DegT_{50}$	[d]
	$DegT_{50,act}$	$DegT_{50}$ at study conditions	[d]
	f_{moist}	moisture correction factor	[-]

Parameters included in the normalization procedure and the resulting normalized $DegT_{50}$ values for modeling are summarized in Table 7.1.2.1.1-32.

Table 7.1.2.1.1-32: Normalization of alpha-cypermethrin $DegT_{50}$ values to reference conditions

Study	Soil	Kinetic model	θ_{act}	θ_{ref}	f_{moist}	$DegT_{50,act}$ [d]	$DegT_{50,ref}$ [d]
2014/1000641	LUFA 5M (c)	FOMC	14.5	22	0.75	14.5[a]	10.8
	LUFA 5M (b)	FOMC				13.1[a]	9.8
2014/1159491	Li 10	DFOP	10.8	10.5	1.00	75.3[b]	75.3
	LUFA 2.2	DFOP	14.6	17.5	0.88	133.3[b]	117.3
	LUFA 2.3	SFO	12.1	13.6	0.92	29.4	27.0

θ_{act} = Actual soil moisture [g 100 g^{-1} dry soil]
θ_{ref} = Reference soil moisture at field capacity (pF 2) according to [*FOCUS (2012)*] [g 100 g^{-1} dry soil]
f_{moist} = Moisture correction factor [-]
$DegT_{50,act}$ = DT_{50} at study conditions [d]
$DegT_{50,ref}$ = DT_{50} at reference conditions [d]
(c) = Cyclopropyl label
(b) = Benzyl label
[a] Calculated as $DegT_{50}$ = DT_{90} / 3.32
[b] Calculated as $DegT_{50}$ = ln(2)/k2

Summary of degradation endpoints for alpha-cypermethrin in different soils under aerobic conditions

In the following tables an overview on the results of the laboratory soil degradation studies with alpha-cypermethrin is provided.

Table 7.1.2.1.1-33: Summary table on best-fit degradation endpoints of alpha-cypermethrin obtained in laboratory soil studies

BASF DocID	Soil / Soil type	pH	Org. C [%]	Temp. [°C]	Moisture [% MWHC]	Best-fit $DegT_{50}$ / $DegT_{90}$ [d]	Method of calculation
AL-620-013 2014/1159505	Ipswich / Sandy loam (b)	6.5 (KCl)	0.9	10	50	48.1 / 228.7	DFOP
	Ipswich / Sandy loam (b)	6.5 (KCl)	0.9	20	50	18.3 / 84.8	FOMC
2014/1000641	LUFA 5M / Sandy loam (c)	7.2 ($CaCl_2$)	2.0	20	50	3.1 / 42.2	DFOP
	LUFA 5M / Sandy loam (b)	7.2 ($CaCl_2$)	2.0	20	50	3.9 / 43.5	FOMC
2014/1159491	Li10 / Loamy sand (b)	6.1 ($CaCl_2$)	0.9	20	40	28.3 / 196.3	DFOP
	LUFA 2.2 / Loamy sand (c)	5.4 ($CaCl_2$)	1.4	20	45	35.0 / 329.4	DFOP
	LUFA 2.3 / Sandy loam (c)	5.9 ($CaCl_2$)	0.7	20	50	24.3 / 134.6	FOMC

(b), (c) = Benzyl or cyclopropane-labeled test item used
MWHC = Maximum water holding capacity

Table 7.1.2.1.1-34: Summary table on degradation endpoints for modeling of alpha-cypermethrin obtained in laboratory soil studies (20°C, pF2)

BASF DocID	Soil / Soil type	pH	Org. C [%]	Temp. [°C]	Moisture [% MWHC]	$DegT_{50}$ at study conditions [d]	Method of calculation	$DegT_{50}$ normalized to 20°C, pF2 [d]
AL-620-013 2014/1159505	Ipswich / Sandy loam (b)	6.5 (KCl)	0.9	10	50	54.7	SFO	-
	Ipswich / Sandy loam (b)	6.5 (KCl)	0.9	20	50	25.5[a]	FOMC	25.5
2014/1000641	LUFA 5M / Sandy loam (c)	7.2 ($CaCl_2$)	2.0	20	50	14.5[a]	FOMC	10.8
	LUFA 5M / Sandy loam (b)	7.2 ($CaCl_2$)	2.0	20	50	13.1[a]	FOMC	9.8
2014/1159491	Li10 / Loamy sand (b)	6.1 ($CaCl_2$)	0.9	20	40	75.3[b]	DFOP	75.3
	LUFA 2.2 / Loamy sand (c)	5.4 ($CaCl_2$)	1.4	20	45	133.3[b]	DFOP	117.3
	LUFA 2.3 / Sandy loam (c)	5.9 ($CaCl_2$)	0.7	20	50	29.4	SFO	27.0

(b), (c) = Benzyl or cyclopropane-labeled test item used
MWHC = Maximum water holding capacity
[a] Calculated as $DegT_{50} = DegT_{90}/3.32$
[b] Calculated as $DT_{50} = \ln 2/k2$

CA 7.1.2.1.2 Aerobic degradation of metabolites, breakdown and reaction products

Report: CA 7.1.2.1.2/1
Sacchi R.R., 2015 a
Rate of Degradation of 3-Phenoxybenzoic Acid (Reg. No. 130213) (metabolite of BAS 310 I) on European Soils at 20 ± 2ºC under aerobic conditions
2015/3003981

Guidelines: INMETRO NIT DICLA- 035 Rev. 02 (Sep. 2011), OECD 307 (2002), POP-PA.1006, EPA 835.4100

GLP: yes
(certified by Instituto Nacional de Metrologia, Normalizacao e Qualidade Industrial - INMETRO, Rio de Janeiro, Brazil)

Executive Summary

The objective of this study was to examine the aerobic degradation of 3-PBA (3-phenoxybenzoic acid, metabolite of alpha-cypermethrin, BAS 310 I) in three soils.

According to the USDA scheme, the soils were characterized as sandy loam (LUFA 5M), and loamy fine sand (LUFA 2.2, Li 10). Prior to application, the soil samples were acclimatized for 15 days and adjusted to 40% of their maximum water holding capacities.

The application rate (based on dry soil weight) of 3-PBA was 0.16 mg kg^{-1}. Assuming a soil depth of 2.5 cm and a soil density of 1.5 g cm^{-3} this corresponds to a theoretical field application rate of about 60 g ha^{-1}. The soil was filled into incubation flasks which were then placed in thermostated cabinet set to 20°C in the dark. Loss of soil water was controlled by weighing and re-adjusted with distilled water if necessary. The dosed soil samples were incubated for various intervals up to 14 days prior to extraction.

The analytical method was validated and revealed a limit of quantification (LOQ) of 0.001 mg kg^{-1} (0.6% of the initially applied residue (AR)) and a limit of detection (LOD) of 0.0003 mg kg^{-1} (0.1% AR) for 3-PBA.

Procedural recoveries were analysed together with treated samples.The mean recovery values were found to be within the acceptable ranged from 70 % to 120 % for all soils tested.

The residues observed for the analyte (expressed as mg kg^{-1}) in the incubated soil samples were fitted by using the software package KinGUI. The kinetic models employed for this evaluation were described by FOCUS Kinetics workgroup. The trigger endpoints for 3-PBA in the three soils examined were in the range of 0.3 to 1.7 days ($DegT_{50}$) and 2.6 to 5.5 days ($DegT_{90}$). The modeling endpoints for ETU were in the range of 0.8 to 1.7 days ($DegT_{50}$).

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test compound: 3-PBA (Reg No.130213)
Chemical name (IUPAC): 3-Phenoxybenzoicacid
Batch No.: AC12251-34
Molar mass: 214.2 g mol^{-1}
Molecular formula: $C_{13}H_{10}O_3$
Purity: 100%

2. Soil

Three field-fresh soils were used in the study. The soils were kept under aerobic conditions and the evaporated water was added if necessary. The soil characteristics are summarized in Table 7.1.2.1.2-1.

Table 7.1.2.1.2-1: Characteristics of three soils

Soil designation	LUFA 2.2 (15/736/01)	Li10 (15/1680/01)	LUFA 5M (15/1651/01)
DIN Particle size distribution [%]			
Sand 0.063 – 2 mm	82.9	82.5	55.7
Silt 0.002 – 0.063 mm	12.0	12.6	32.3
Clay < 0.002 mm	5.1	4.8	12.1
Textural class	loamy sand (S12)	silty sand (Su2)	loamy sand (Sl4)
USDA Particle size distribution [%]			
Sand 0.050 – 2 mm	83.8	83.8	60.2
Silt 0.002 – 0.050 mm	11.1	11.4	27.8
Clay < 0.002 mm	5.1	4.8	12.1
Textural class	loamy fine sand	loamy fine sand	sandy loam
Total carbon [%]	1.73	0.86	2.11
Total organic carbon [%]	1.73	0.86	1.18
pH [H_2O]	5.6	6.7	7.8
pH [$CaCl_2$]	5.3	6.2	7.4
Cation exchange capacity [$cmol^+ kg^{-1}$]	7.7	6.3	10.2
Max. water holding capacity [g per 100g dry weight]	34.7	26.8	31.1
pF 2.0 [$g_{soil\ moisture}\ g_{dry\ soil}^{-1}$]	0.165	0.106	0.208
pF 2.5 [$g_{soil\ moisture}\ g_{dry\ soil}^{-1}$]	0.149	0.092	0.152
Microbial biomass [mg C per 100g dry soil]	39.1	22.3	32.4

B. STUDY DESIGN

1. Experimental conditions

Soil samples were adjusted to 40% of their maximum water holding capacities and incubated at room temperature in the dark for 15 days prior to application.

The application rate (based on dry soil weight) of the analyte dosed to soil was 0.16 mg kg^{-1}. Assuming a soil depth of 2.5 cm and a density of 1.5 g cm^{-3} this rate correspond to a theoretical field application rate of about 60 g ha^{-1}. This rate was chosen under consideration of a reasonable good detectability in the present study.
One kg of each soil (dry soil) was treated with 1.0 mL of the treatment solution containing 0.160 mg 3-PBA). Samples were homogenized, and 50 g (dry weight based) were filled into test vessels. These were sealed with special caps with air inlet and outlet tubes. The samples were incubated in the dark at a temperature of (20 ± 2)°C and were aerated with a slight stream of moistened and CO2 free air. Loss of soil water was controlled by weighing and was re-adjusted with distilled water if necessary.

The treated soil samples were incubated for various intervals up to 14 days prior to extraction. Additionally, untreated moist bulks of soil were kept in the thermostated cabinet.

2. Sampling

Soil samples were removed from the thermostated cabinet for subsequent extraction after 0, 1, 3, 7, 9, and 14 days of incubation. Six samples were taken at time zero to prove the homogeneous distribution of the test substance in the soils. Duplicate samples were taken at all other sampling times.

3. Description of analytical procedures

BASF Analytical Method R0034/01 (BASF DocID 2015/7000103) was used for analysis. 5 g aliquots of the samples were extracted consecutively twice with 25 mL acetonitrile/water (70:30, v/v) on a laboratory shaker at 400 rpm for about 30 minutes. After each extraction step, the sample was centrifuged for 5 minutes at 4000 rpm and the supernatant was decanted into a 50 mL disposable centrifuge tube. An aliquot (10%) from the combined extract was evaporated to dryness using nitrogen at about 50ºC and then dissolved in methanol/water (20:80, v/v) with 0.1% formic acid. The concentrated extracts were sonicated for about 1-2 minutes and vortexed for about 15 seconds to ensure that all residues was dissolved. Prior to LC-MS/MS analysis, concentrated extracts were transferred into a HPLC vial using a syringe filter

The analytical method achieves a limit of quantification (LOQ) of 0.001 mg kg^{-1} and a limit of detection (LOD) of 0.0003 mg kg^{-1} for 3-PBA.

4. Calculation of the degradation rate

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints) as well as modeling endpoints. Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006)*].

The software package KinGUI (version 2.2012.320.1629) was used for parameter fitting [*SCHÄFER et al. (2007)*].

Modeling strategy

The kinetic models employed for this evaluation were described by FOCUS [*FOCUS (2006)*].

Trigger endpoints were derived from the kinetic model that provides the best fit to the measured data, generally indicated by the lowest χ^2 - error. Modeling endpoints were derived preferably from the SFO model. If the SFO model is not appropriate, pragmatic procedures were used to derive conservative pseudo-SFO degradation rates from the FOMC model by dividing $DegT_{90}$ by 3.32.

The appropriateness of the distinct kinetic model to describe soil degradation was tested by visual assessment. Furthermore, a kinetic model was considered appropriate for deriving trigger or modeling endpoints of χ^2 – error value was low (ideally below 15%).

Experimental data used for kinetic analysis

The experimental data were derived from the study reports and adjusted according to FOCUS [*FOUCS (2006)*].

The datasets submitted to kinetic analyses are given in Table 7.1.2.1.2-2.

Table 7.1.2.1.2-2: Input data of 3-PBA expressed in mg kg^{-1} included in the kinetic analysis

Time [days]	Li10 [mg kg^{-1}]	LUFA 2.2 [mg kg^{-1}]	LUFA 5M [mg kg^{-1}]
0	0.138	0.105	0.121
	0.158	0.103	0.119
	0.162	0.106	0.121
	0.156	0.106	0.121
	0.161	0.106	0.119
	0.147	0.099	0.122
1	0.067	0.024	0.078
	0.066	0.024	0.08
3	0.024	0.009	0.038
	0.025	0.009	0.034
7	0.007	0.005	0.005
	0.008	0.005	0.004
9	0.006	0.003	0.003
	0.006	0.004	0.003
14	0.0005*	0.0003*	0.0002*
	0.0005*	0.0002*	0.0002*

* Value between LOQ and LOD

II. RESULTS AND DISCUSSION

The three soils were incubated at (20 ± 2)°C and soil moisture of approximately 40 % of the maximum water holding capacity. A nominal application rate of 0.160 mg a.i./kg dry soil was used. The extractable residues at day 0 and ranged from 0.104 to 0.154 mg/kg dry soil. This corresponds to mean recovery values at day 0 of 94.2 %, 64.0 % and 73.8 % for Li10, LUFA 2.2 and LUFA 5M, respectively. The low recovery obtained for two soils is deemed to be caused by the very fast degradation of the compound, which already started during the sample preparation and application process. The degradation in each soil was followed for 14 days.
Procedural recoveries were determined for each soil and sampling date from day 1 onwards in triplicate. Overall mean recoveries were 93% (Li10), 97% (LUFA 2.2) and 99% (LUFA 5M). A potential matrix effect was investigated and was found to be below 20% in all cases.

Residues of the test item extracted from soil after given incubation periods are summarized in Table 7.1.2.1.2-3.

Table 7.1.2.1.2-3: Extractable 3-PBA residues from soils LUFA 2.2, Li10 and LUFA 5M

Days [a]	LUFA 2.2		Li10		LUFA 5M	
	[mg kg^{-1}]	[%] [b]	[mg kg^{-1}]	[%] [b]	[mg kg^{-1}]	[%] [b]
0[c]	0.105	100.0	0.138	100.0	0.121	100.0
0[c]	0.103	100.0	0.158	100.0	0.119	100.0
0[c]	0.106	100.0	0.162	100.0	0.121	100.0
0[c]	0.106	100.0	0.156	100.0	0.121	100.0
0[c]	0.106	100.0	0.161	100.0	0.119	100.0
0[c]	0.099	100.0	0.147	100.0	0.122	100.0
1	0.024	23.4	0.067	43.5	0.078	64.7
1	0.024	22.6	0.066	43.1	0.080	66.5
3	0.009	8.4	0.024	15.9	0.038	31.6
3	0.009	8.8	0.025	16.4	0.034	28.4
7	0.005	4.7	0.007	4.8	0.005	3.8
7	0.005	4.6	0.008	4.9	0.004	3.4
9	0.003	2.9	0.006	4.1	0.003	2.5
9	0.004	3.4	0.006	3.8	0.003	2.3
14	0.0003*	0.3	0.0005*	0.3	0.0002*	0.2
14	0.0002*	0.2	0.0005*	0.3	0.0002*	0.2

[a] Days after application
[b] Percent of the normalized applied residue
[c] Six replicates to demonstrate homogeneous distribution of test substance
* Value between LOQ and LOD

The calculated degradation parameters and kinetic endpoints of 3-PBA for use as triggers for additional work and for modeling are summarized in the following tables (Table 7.1.2.1.2-4 and Table 7.1.2.1.2-5).

For the evaluated soil degradation experiments, the visual assessment and the goodness-of-fit statistics show plausible fits. Therefore, the resulting $DegT_{50}$ / DT_{50} values can be considered reliable.

Table 7.1.2.1.2-4: 3-PBA trigger endpoints

Soil	Kinetic model	χ^2 error [%]	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Li 10	FOMC	1.9	0.8	4.3
Lufa 2.2	FOMC	3.0	0.3	2.6
Lufa 5M	SFO	2.04	1.7	5.5

Table 7.1.2.1.2-5: 3-PBA modeling endpoints

Soil	Kinetic model	χ^2 error [%]	$DegT_{50}$ [d]
Li 10	FOMC	1.9	1.3[a]
Lufa 2.2	FOMC	3.0	0.78[a]
Lufa 5M	SFO	2.04	1.7

[a] Calculated as $DT_{50} = DT_{90}/3.32$

III. CONCLUSION

The objective of the study was to examine aerobic degradation of 3-PBA (metabolite of alpha-cypermethrin) in three soils. The trigger endpoints for 3-PBA in the six soils examined were in the range of 0.3 to 1.7 days ($DegT_{50}$) and 2.6 to 5.5 days ($DegT_{90}$). The modeling endpoints for 3-PBA were in the range of 0.8 to 1.7 days ($DegT_{50}$).
It could be demonstrated that 3-PBA is rapidly degraded in soil.

Report: CA 7.1.2.1.2/2
Sacchi R. R., 2015 a
Rate of degradation of cis-DCVA (Reg. No. 4080830) (metabolite of BAS 310 I) on european soils at 20 ± 2ºC under aerobic conditions
2015/3003982
Guidelines: INMETRO NIT DICLA- 035 Rev. 02 (Sep. 2011), OECD 307 (2002)
GLP: yes
(certified by Instituto Nacional de Metrologia, Normalizacao e Qualidade Industrial - INMETRO, Rio de Janeiro, Brazil)

Executive Summary

The objective of this study was to examine the aerobic degradation of cis-DCVA (cis-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylic acid, metabolite of alpha-cypermethrin, BAS 310 I) in three soils.

According to the USDA scheme, the soils were characterized as sandy loam (LUFA 5M), and loamy fine sand (LUFA 2.2, Li 10). Prior to application, the soil samples were acclimatized for 15 days and adjusted to 40% of their maximum water holding capacities.

The nominal application rate (based on dry soil weight) of cis-DCVA was 0.160 mg kg^{-1}. Assuming a soil depth of 2.5 cm and a soil density of 1.5 g cm^{-3} this corresponds to a theoretical field application rate of about 60 g ha^{-1}. The soil was filled into incubation flasks, which were does individually and were then placed in thermostated cabinet set to 20°C in the dark. Loss of soil water was controlled by weighing and re-adjusted with distilled water if necessary. The dosed soil samples were incubated for various intervals up to 31 or 41 days prior to extraction.

The analytical method was validated and revealed a limit of quantification (LOQ) of 0.001 mg kg^{-1} (0.6% of the initially applied residue (AR)) and a limit of detection (LOD) of 0.0003 mg kg^{-1} (0.1% AR) for cis-DCVA.

Procedural recoveries were analysed together with treated samples.The mean recovery values were found to be within the acceptable ranged from 70 % to 110 % for all soils tested.

The residues observed for the analyte (expressed as mg kg^{-1}) in the incubated soil samples were fitted by using the software package KinGUI. The kinetic models employed for this evaluation were described by FOCUS Kinetics workgroup. The trigger and modeling endpoints for cis-DCVA in the three soils examined were in the range of 4.7 to 13.5 days ($DegT_{50}$) and 15.7 to 45.0 days ($DegT_{90}$).

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test compound:	cis-DCVA (Reg No. 4080830)
Chemical name (IUPAC):	cis-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-carboxylic acid
Batch No.:	AC12717-65
Molar mass:	209.01 g mol^{-1}
Molecular formula:	$C_8H_{10}Cl_2O_2$
Purity:	99.5%

2. Soil

Three field-fresh soils were used in the study. The soils were kept under aerobic conditions and the evaporated water was added if necessary. The soil characteristics are summarized in Table 7.1.2.1.2-6.

Table 7.1.2.1.2-6: Characteristics of three soils

Soil designation	LUFA 2.2 (15/736/03)	Li10 (15/1680/03)	LUFA 5M (14/1651/03)
DIN Particle size distribution [%]			
Sand 0.063 – 2 mm	84.3	83.5	59.3
Silt 0.002 – 0.063 mm	10.3	11.4	28.3
Clay < 0.002 mm	5.3	5.1	12.4
Textural class	loamy sand (S12)	loamy sand (S12)	loamy sand (Sl4)
USDA Particle size distribution [%]			
Sand 0.050 – 2 mm	85.5	86.2	53.4
Silt 0.002 – 0.050 mm	9.2	8.6	34.3
Clay < 0.002 mm	5.1	5.1	12.4
Textural class	loamy fine sand	loamy fine sand	sandy loam
Total carbon [%]	1.45	1.03	2.03
Total organic carbon [%]	1.45	1.03	1.05
pH [H_2O]	6.4	6.9	7.4
pH [$CaCl_2$]	6.4	6.0	6.9
Cation exchange capacity [$cmol^+ kg^{-1}$]	10.5	7.3	12.5
Max. water holding capacity [g per 100g dry weight]	32.0	26.5	29.4
pF 2.0 [$g_{soil\ moisture}\ g_{dry\ soil}^{-1}$]	0.170	0.107	0.200
pF 2.5 [$g_{soil\ moisture}\ g_{dry\ soil}^{-1}$]	0.141	0.091	0.158
Microbial biomass [mg C per 100g dry soil]	37.4	41.2	29.9

B. STUDY DESIGN

1. Experimental conditions

Soil samples were adjusted to 40% of their maximum water holding capacities and incubated at room temperature in the dark prior to application.

The application rate (based on dry soil weight) of the analyte dosed to soil was 0.16 mg kg^{-1}. Assuming a soil depth of 2.5 cm and a density of 1.5 g cm^{-3} this rate correspond to a theoretical field application rate of about 60 g ha^{-1}. This rate was chosen under consideration of a reasonable good detectability in the present study.
50 g (dry weight based) of soil were filled into test vessels. The soil in each vessel was treated with 50 µL of a solution containing 0.160 µg/mL cis-DCVA. The soil in the vessels was mixed to achieve a homogenous distribution of the test compound in the soil. Then these vessels were sealed with special caps with air inlet and outlet tubes. The samples were incubated in the dark at a temperature of (20 ± 2)°C and were aerated with a slight stream of moistened and CO_2 free air. Loss of soil water was controlled by weighing and was re-adjusted with distilled water if necessary.

The treated soil samples were incubated for various intervals up to 41 days prior to extraction. Additionally, untreated moist bulks of soil were kept in the thermostated cabinet.

2. Sampling

Soil samples were removed from the thermostated cabinet for subsequent extraction after 0, 1, 3, 7, 10, 21, 31 and 41 days (LUFA 2.2 only 31 days) of incubation. Three samples were taken at time zero to prove the homogeneous distribution of the test substance in the soils. Duplicate samples were taken at all other sampling times.

3. Description of analytical procedures

BASF Analytical Method R0034/01 (BASF DocID 2015/7000103) was used for analysis. 5 g aliquots of the samples were extracted consecutively twice with 25 mL acetonitrile/water (70:30, v/v) on a laboratory shaker at 400 rpm for about 30 minutes. After each extraction step, the sample was centrifuged for 5 minutes at 4000 rpm and the supernatant was decanted into a 50 mL disposable centrifuge tube. An aliquot (10%) from the combined extract was evaporated to dryness using nitrogen at about 50ºC (in water bath) and then re-dissolved in methanol/water (20:80, v/v) with 0.1% formic acid. The concentrated extracts were sonicated for about 1-2 minutes and vortexed for about 15 seconds to ensure that all residues was dissolved. Prior to LC-MS/MS analysis, concentrated extracts were transferred into a HPLC vial using a syringe filter (0.45 µm PTFE). Some of the samples were diluted with methanol/water (20:80, v/v) and 0.1% formic acid to reach lower concentrations falling into the range of the calibration curve.

The analytical method achieves a limit of quantification (LOQ) of 0.001 mg kg^{-1} and a limit of detection (LOD) of 0.0003 mg kg^{-1} for cis-DCVA.

4. Calculation of the degradation rate

Kinetic evaluation was performed in order to derive degradation parameters as triggers for additional work (trigger endpoints) as well as modeling endpoints. Kinetic analysis and calculation of $DegT_{50}$ and $DegT_{90}$ values was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006)*].

The software package KinGUI (version 2.2012.320.1629) was used for parameter fitting [*SCHÄFER et al. (2007)*].

Modeling strategy

The kinetic models employed for this evaluation were described by FOCUS [*FOCUS (2006)*].

Trigger endpoints were derived from the kinetic model that provides the best fit to the measured data, generally indicated by the lowest χ^2 - error. Modeling endpoints were derived from the SFO model.

The appropriateness of the distinct kinetic model to describe soil degradation was tested by visual assessment. Furthermore, a kinetic model was considered appropriate for deriving trigger or modeling endpoints of χ^2 – error value was low (ideally below 15%).

Experimental data used for kinetic analysis

The experimental data were derived from the study reports and adjusted according to FOCUS [*FOUCS (2006)*].

The datasets submitted to kinetic analyses are given in Table 7.1.2.1.2-7.

Table 7.1.2.1.2-7: Input data of cis-DCVA expressed in mg kg^{-1} included in the kinetic analysis

Time [days]	Li10 [mg kg^{-1}]	LUFA 2.2 [mg kg^{-1}]	LUFA 5M [mg kg^{-1}]
0	0.170*	0.176*	0.175*
	0.173*	0.171*	0.174*
1	0.152	0.158	0.152
	0.147	0.158	0.156
3	0.137	0.115	0.149
	0.141	0.113	0.151
7	0.137	0.068	0.138
	0.134	0.070	0.139
10	0.126	0.037	0.100
	0.128	0.038	0.101
14	0.096	0.018	0.092
	0.099	0.019	0.094
21	0.054	0.007	0.028
	0.050	0.007	0.031
31	0.016	0.005	0.006
	0.014	0.006	0.006
41	0.003	n.a.	0.003
	0.003	n.a.	0.003

* Mean of three replicates taken to demonstrate homogeneous distribution

II. RESULTS AND DISCUSSION

The three soils were incubated at (20 ± 2)°C and soil moisture of approximately 40 % of the maximum water holding capacity. A nominal application rate of 0.160 mg a.i./kg dry soil was used. The extractable residues at day 0 were found to be higher than the nominal concentration and ranged from 0.170 to 0.176 mg/kg dry soil. This corresponds to mean recovery values at day 0 of 107.2 %, 108.4 % and 109.1 % for Li10, LUFA 2.2 and LUFA 5M, respectively. The degradation in each soil was followed for 41 days, with the exception of LUFA 2.2 soil, which was only followed for 31 days.
Procedural recoveries were determined for each soil and sampling date from day 1 onwards in triplicate. Overall mean recoveries were 93% (Li10), 97% (LUFA 2.2) and 99% (LUFA 5M). A potential matrix effect was investigated for all soils and was found to be below 10% in all cases.

Residues of the test item extracted from soil after given incubation periods are summarized in Table 7.1.2.1.2-8.

Table 7.1.2.1.2-8: Extractable cis-DCVA residues from soils LUFA 2.2, Li10, and LUFA 5M

Days [a]	LUFA 2.2		Li10		LUFA 5M	
	[mg kg^{-1}]	[%] [b]	[mg kg^{-1}]	[%] [b]	[mg kg^{-1}]	[%] [b]
0	0.176	100.0	0.170	100.0	0.175	100.0
0	0.171	100.0	0.173	100.0	0.174	100.0
1	0.158	91.0	0.152	88.3	0.152	87.2
1	0.158	91.1	0.147	85.7	0.156	89.3
3	0.114	65.9	0.137	79.9	0.148	85.0
3	0.113	64.8	0.141	82.2	0.150	86.0
7	0.068	39.3	0.137	80.0	0.138	79.2
7	0.070	40.4	0.134	77.9	0.139	79.7
10	0.037	21.4	0.126	73.5	0.100	57.3
10	0.038	21.7	0.128	74.5	0.101	57.7
14	0.018	10.5	0.096	55.8	0.092	52.7
14	0.019	10.8	0.099	57.6	0.094	53.8
21	0.007	4.2	0.054	31.3	0.028	16.1
21	0.007	4.1	0.050	29.3	0.031	17.7
31	0.005	3.1	0.016	9.1	0.006	3.7
31	0.006	3.4	0.014	8.3	0.006	3.3
41	n.a.	-	0.003	2.0	0.003	1.5
41	n.a.	-	0.003	2.0	0.003	1.6

[a] Days after application
[b] Percent of the measured day zero residue
* Value between LOQ and LOD

The calculated degradation parameters and kinetic endpoints of cis-DCVA for use as triggers for additional work and for modeling are summarized in the following table (Table 7.1.2.1.2-9)

For the evaluated soil degradation experiments, the visual assessment and the goodness-of-fit statistics show plausible fits. Therefore, the resulting $DegT_{50}$ / DT_{50} values can be considered reliable.

Table 7.1.2.1.2-9: cis-DCVA trigger and modeling endpoints

Soil	Kinetic model	χ^2 error [%]	$DegT_{50}$ [d]	$DegT_{90}$ [d]
Li 10	SFO	12.16	13.5	44.96
Lufa 2.2	SFO	4.1	4.7	15.7
Lufa 5M	SFO	12.53	11.1	36.8

Normalization of laboratory degradation endpoints to reference conditions

Since for environmental fate modeling soil $DegT_{50}$ values at reference conditions (temperature of 20°C and soil moisture at field capacity, i.e. pF2) are required, the modeling endpoints ($DegT_{50}$) reported in CA 7.1.1.1/1 [*BASF Doc ID 2014/1000641*], CA 7.1.2.1.1/4 [*BASF Doc ID 2014/1159491*], CA 7.1.2.1.2/1 [BASF Doc ID 2015/3003981] and CA 7.1.2.1.2/2 [BASF Doc ID 2015/3003982] were normalized as described in CA 7.1.2.1.1.

Parameters included in the normalization procedure and the resulting normalized $DegT_{50}$ values for modeling are summarized in Table 7.1.2.1.2-10.

Table 7.1.2.1.2-10: Normalization of alpha-cypermethrin metabolites M310I017, DCVA, and 3-PBA $DegT_{50}$ values to reference conditions

Study	Soil	Kinetic model	θ_{act}	θ_{ref}	f_{moist}	$DegT_{50,act}$ [d]	$DegT_{50,ref}$ [d]
M310I017							
2014/1000641	LUFA 5M (c)	SFO	14.5	22	0.75	8.5	6.3
	LUFA 5M (b)	SFO				19.9	14.8
2014/1159491	Li10	SFO	10.8	10.5	1.00	22.6	22.6
	LUFA 2.2	SFO	14.6	17.5	0.88	42.3	37.2
	LUFA 2.3	SFO	12.1	13.6	0.92	3.1	2.8
DCVA							
2014/1000641	LUFA 5M (c)	SFO	14.5	22	0.75	5.2	3.9
2015/3003982	LUFA 2.2	SFO	12.8	17	0.82	4.7	3.9
	Li10	SFO	10.6	10.7	0.99	13.5	13.4
	LUFA 5M	SFO	11.8	20	0.69	11.1	7.7
3-PBA							
2015/3003981	LUFA 2.2	FOMC	10.7	10.6	1.00	1.3	1.3
	Li10	FOMC	13.9	18.5	0.82	0.8	0.6
	LUFA 5M	SFO	12.4	20.8	0.70	1.7	1.2

θ_{act} = Actual soil moisture [g 100 g^{-1} dry soil]
θ_{ref} = Reference soil moisture at field capacity (pF 2) according to [*FOCUS (2012)*] [g 100 g^{-1} dry soil]
f_{moist} = Moisture correction factor [-]
$DegT_{50,act}$ = DT_{50} at study conditions [d]
$DegT_{50,ref}$ = DT_{50} at reference conditions [d]
(c) = Cyclopropyl label
(b) = Benzyl label

Summary on metabolite occurrence and degradation rates in aerobic soil

Table 7.1.2.1.2-11: Maximum occurrence of alpha-cypermethrin metabolites in laboratory aerobic soil studies (20°C, 40-50% MWHC)

Metabolite	BASF DocID	Parent label	Soil	Maximum [% TAR]
M310I017	2014/1000641	cyclopropane benzyl	LUFA 5M	8.4 7.5
	2014/1159491	benzyl cyclopropane cyclopropane	Li10 LUFA 2.2 LUFA 2.3	5.8 5.9 5.3
DCVA	2014/1000641	cyclopropane	LUFA 5M	13.6

MWHC = Maximum water holding capacity
TAR = Total applied radioactivity

Table 7.1.2.1.2-12: Trigger endpoints of alpha-cypermethrin metabolites in aerobic soil studies (laboratory, 20°C, 40-50% MWHC)

Metabolite	Data source BASF DocID	Test item (label)	Soil	Trigger DT_{50}/DT_{90} [d]	Method of calculation
M310I017	2014/1000641	parent (c) parent (b)	LUFA 5M	8.5 / 28.4 19.9 / 66.1	SFO [a] SFO [a]
	2014/1159491	parent (b) parent (c) parent (c)	Li10 LUFA 2.2 LUFA 2.3	22.6 / 75.1 42.3 / 140.4 4.9 / 16.2	SFO [b] SFO [b] SFO [c]
DCVA	2014/1000641	parent (c)	LUFA 5M	5.2 / 17.1	SFO [a]
	2015/3003982	cis-DCVA (non-labeled)	LUFA 2.2 Li10 LUFA 5M	4.7 / 15.7 13.5 / 45.0 11.1 / 36.8	SFO SFO SFO
3-PBA	2015/3003981	3-PBA (non-labeled)	LUFA 2.2 Li10 LUFA 5M	0.3 / 2.6 0.8 / 4.3 1.7 / 5.5	FOMC FOMC SFO

(b), (c) = Benzyl or cyclopropane-labeled test item used
MWHC = Maximum water holding capacity
[a] Decline fit for the metabolite
[b] DFOP kinetics for parent
[c] FOMC kinetics for parent

Table 7.1.2.1.2-13: Modeling endpoints for alpha-cypermethrin metabolites in aerobic soil studies (laboratory, 20°C, 40-50% MWHC)

Metabolite	Data source BASF DocID	Test item (label)	Soil	DT_{50} at study conditions [d]	Method of calculation	DT_{50} normalized to 20°C / pF2 [d]	Formation fraction
M310I017	2014/1000641	parent (c)	LUFA 5M	8.5	SFO [a]	6.3	n.c.
		parent (b)		19.9	SFO [a]	14.8	n.c.
	2014/1159491	parent (b)	Li10	22.6	SFO [b]	22.6	0.215
		parent (c)	LUFA 2.2	42.3	SFO [b]	37.2	0.188
		parent (c)	LUFA 2.3	3.1	SFO [c]	2.8	0.629
DCVA	2014/1000641	parent (c)	LUFA 5M	5.2	SFO [a]	3.9	n.r.
	2015/3003982	cis-DCVA (non-labeled)	LUFA 2.2	4.7	SFO	3.9	n.c.
			Li10	13.5	SFO	13.4	n.c.
			LUFA 5M	11.1	SFO	7.7	n.c.
3-PBA	2015/3003981	3-PBA (non-labeled)	LUFA 2.2	0.8	SFO	0.6	n.c.
			Li10	1.3	FOMC	1.3	n.c.
			LUFA 5M	1.7	FOMC	1.2	n.c.

(b), (c) = Benzyl or cyclopropane-labeled test item used
MWHC = Maximum water holding capacity
n.c. = Not calculated
n.r. = Not relevant
[a] Decline fit for the metabolite
[b] DFOP kinetics for parent
[c] SFO kinetics for parent

CA 7.1.2.1.3 Anaerobic degradation of the active substance

Information on the rate of degradation under anaerobic conditions is provided in CA 7.1.1.2: Staudenmaier H. Kuhnke G., 2014: Anaerobic soil metabolism of alpha-cypermethrin (BAS 310 I) 2013/1386602.

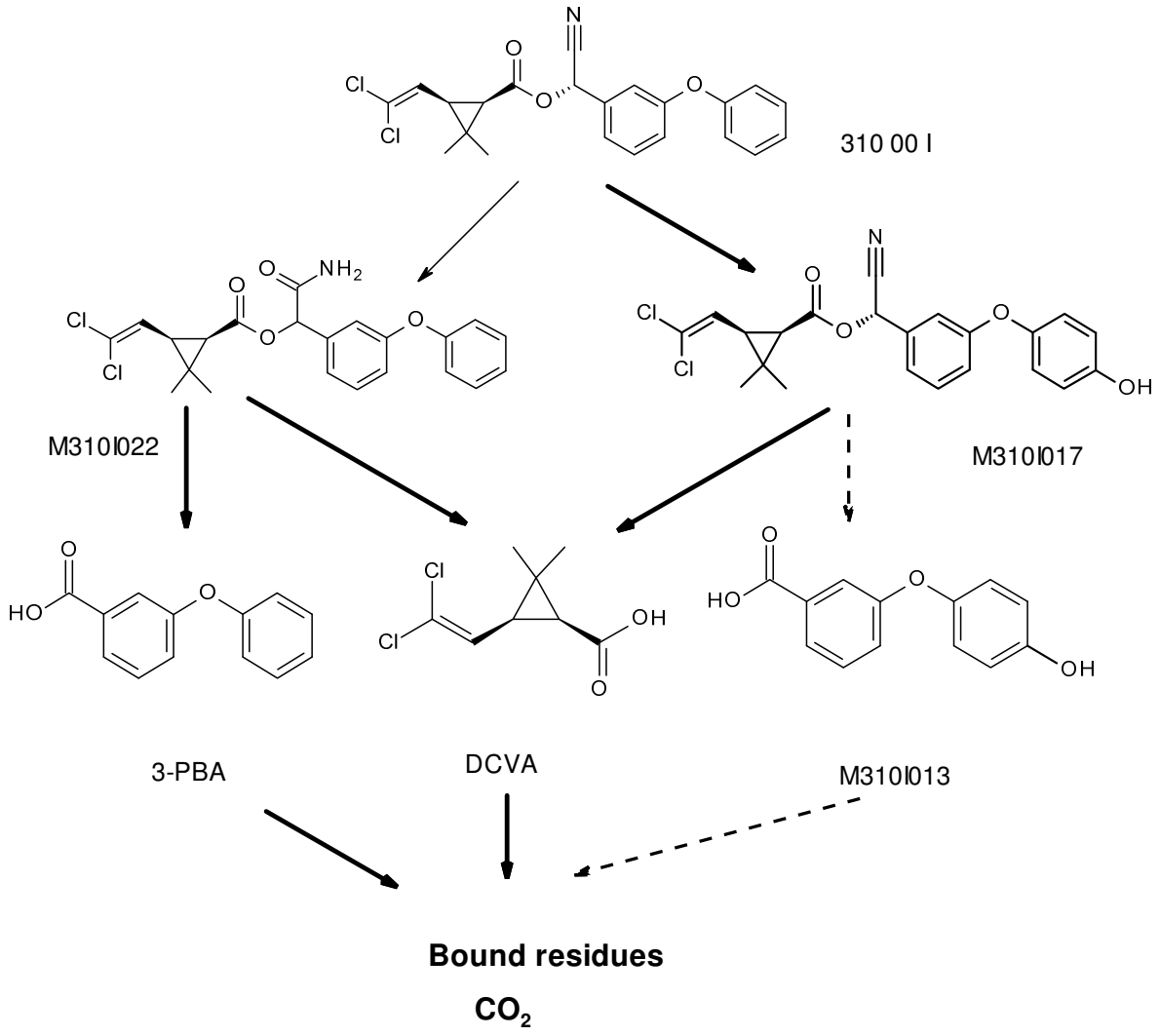
CA 7.1.2.1.4 Anaerobic degradation of metabolites, breakdown and reaction products

No study was performed for metabolites. Information on the rate of degradation of metabolite M310I017 under anaerobic conditions is provided in CA 7.1.1.2: Staudenmaier H. Kuhnke G., 2014: Anaerobic soil metabolism of alpha-cypermethrin (BAS 310 I) 2013/1386602. DCVA and 3-PBA are also aerobic metabolites and information on their aerobic egradation is available. No metabolites specific to anaerobic conditions were detected.

Metabolic pathway of alpha-cypermethrin in soil

Based on the aerobic and anawerobic metabolism studies, as well as the soil photolysis the following route of degradation is proposed for alpha-cypermethrin:

Figure 7.1.2.1.4-1: Route of degradation of alpha-cypermethrin in soil



CA 7.1.2.2 Field studies

Numerous field dissipation studies were conducted either with cypermethrin or alpha-cypermethrin. An overview is presented in Table Table 7.1.2.2-1. These studies were no longer considered adequate and new field trials were performed. The study on six sites in the EU was conducted according to new guidance using a sand cover to exclude photolytic effects or volatilization. This study was used to derive modelling endpoints. To obtain a more realistic picture of the behaviour of alpha.cypermethrin in soil for persistence endpoints, a study conducted in the US was submitted.

Table 7.1.2.2-1: List of terrestrial field dissipation studies in soil performed with (alpha-) cypermethrin

DocID	Parent compound	Sites	Application rate [kg ha^{-1}]	Crop	Incubation period [days]	Remark
CY-790-011	Cypermethrin	France (1st year)	0.5	Sugar beets	224	Bosio 1977
CY-790-010	Cypermethrin	France	1 3	Sugar beets	217	Bosio 1977
CY-790-001	Cypermethrin	Spain (1st year)	0.5	Bare soil	365	Gilham 1977
CY-790-003	Cypermethrin	Spain (2nd year)	0.5	Bare soil		Petry 1977
CY-790-004	Cypermethrin	Spain (3rd year)	0.5	Bare soil	375	Knight 1979
CY-790-002	Cypermethrin	UK (1st year)	0.5	Bare soil	365	Knight, 1978
CY-790-005	Cypermethrin	UK (2nd year)	0.5	Bare soil		Cole, 1980
CY-790-007	Cypermethrin	UK (3rd year)	0.5	Bare soil		Knight, 1981
CY-790-015	Cypermethrin	Germany	0.5	Bare soil	100	Bosio 1988
AL-790-006	Alpha-cypermethrin	Reculver, UK (1st year)	0.5	Bare soil	363	Forbes 1983
AL-790-007	Alpha-cypermethrin	Reculver, UK (2nd year)	0.5	Bare soil	345	Forbes 1983
AL-790-008	Alpha-cypermethrin	Reculver, UK (3rd year)	0.5	Bare soil	364	Forbes 1985
AL-790-009	Alpha-cypermethrin	Hoath, UK (1st year)	0.5	Bare soil	282	Bosio 1983
AL-790-010	Alpha-cypermethrin	Hoath, UK (2nd year)	0.5	Bare soil	345	Forbes 1983
AL-790-011	Alpha-cypermethrin	Hoath, UK (3rd year)	0.5	Bare soil	363	Forbes 1985
AL-790-012	Alpha-cypermethrin	Coates, UK (1st year)	0.5	Bare soil	386	Forbes 1983
AL-790-013	Alpha-cypermethrin	Coates, UK (2nd year)	0.5	Bare soil	335	Forbes 1985
AL-790-014	Alpha-cypermethrin	Coates, UK (3rd year)	0.5	Bare soil	351	Coveny 1986
Fischer et al., 1993	Cypermethrin					Mitchell Cotts
Schulz, 1993a	Cypermethrin	Schwichteler, Germany	0.5	Bare soil	365	Mitchell Cotts
Schulz, 1993b	Cypermethrin	Obernburg, Germany	0.5	Bare soil	365	Mitchell Cotts
Schulz, 1993c	Cypermethrin	Kleve-Reichswalde, Germany	0.5	Bare soil (prev. Forest)	365	Mitchell Cotts
Schulz, 1993d	Cypermethrin	Eschau-Wildenstein, Germany	0.5	Bare soil (prev. Forest)	365	Mitchell Cotts

Trials performed with cypermethrin
(All trials non GLP.)
1. Bosio P. G. (1977) Residues of WL 43467 in soil from France -Long term trials - first year 1976
Short description of highly overdosed (0.5 kg ha^{-1}) dissipation trial in France. Product was not incorporated into soil. Rapid dissipation in soil observed. Poor quality report and data not suitable to derive endpoints.

2.Bosio P. G. (1977) Residues of WL 43467 in soil from France - 1976 trials
Short description of highly overdosed (1 and 3 kg ha^{-1}) dissipation trial in France. Product was incorporated into soil. Rapid dissipation in soil observed. Poor quality report and data not suitable to derive endpoints.

3. Gilham J. A. (1977) Residues of WL 43467 in soil from Spain - Soil persistence trial -First year
Short description of highly overdosed (0.5 kg ha^{-1}) dissipation trial in Spain. Side by side comparison of incorporated and non-incorporated product. Inconsistent results were observed from incorporated trial, probably due to homogeneity problems. In both cases dissipation below LOQ within 34 weeks. Poor quality report and data not suitable to derive endpoints.

4. Petry J. E. et al. (1977) Residues of WL 43467 in soil from Spain - Soil persistence trial - Second year
Short description of highly overdosed (0.5 kg ha^{-1}) dissipation trial in Spain. Application on the same plot as in Gilham (1977). Side by side comparison of incorporated and non-incorporated product. Poor quality report and data not suitable to derive endpoints.

5. Knight C. J. et al. (1979) Residues of WL 43467 in soil from Spain - Soil persistence trial - Third year
Short description of highly overdosed (0.5 kg ha^{-1}) dissipation trial in Spain. Application on the same plot as in Gilham (1977). Side by side comparison of incorporated and non-incorporated product. Poor quality report and data not suitable to derive endpoints.

6. Knight C. J. et al. (1978) Analysis of soil from UK for residues of WL 43467 - Soil persistence trial - First year
Short description of two highly overdosed (0.5 kg ha^{-1}) dissipation trials in the UK. Poor quality report and data not suitable to derive endpoints.

7. Cole E. R. et al. (1980) Analysis of soil from UK for residues of WL 43467 - Soil persistence trial - Second year
Short description of two highly overdosed (0.5 kg ha^{-1}) dissipation trials in the UK. Application in second year on the same plot as in Knight (1978). Poor quality report and data not suitable to derive endpoints

8. Knight C. J. et al. (1981) Analysis of soil from UK for residues of WL 43467 - Soil persistence trial - Third year
Short description of two highly overdosed (0.5 kg ha^{-1}) dissipation trials in the UK. Application in third year on the same plot as in Knight (1978). Poor quality report and data not suitable to derive endpoints.

9. Bosio P. G. (1988) Residues of cypermethrin in soil from Germany treated with Ripcord - 1988 trial -
Short description of two highly overdosed (0.5 kg ha^{-1}) dissipation trials in Germany. Poor quality report and data not suitable to derive endpoints.

Trials performed with alpha-cypermethrin
(All trials non GLP.)
1. Forbes S. (1983) Analysis of soil from UK for residues of WL 85871 - Soil persistence trial - First year
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a sandy clay soil in Reculver (UK). Application done in August. Poor quality report, method not suitable to determine 10% of initial, therefore data are not suitable to derive endpoints.

2. Forbes S. (1983) Analysis of soil from UK for residues of WL 85871 - Soil persistence trial - Second year
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a sandy clay soil in Reculver (UK). Second application at the same site. Application done in August. Poor quality report, only 5 data points, therefore data are not suitable to derive endpoints.

3. Forbes S. (1985) Analysis of soil from UK (Reculver) for residues of FASTAC (WL 85871) - Soil persistence trial - Third year
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a sandy clay soil in Reculver (UK). Third application at the same site. Application done in August. Samples were shipped at ambient temperature. Poor quality report. Therefore data are not suitable to derive endpoints.

4. Bosio P.G. (1983) Residues of WL 85871 and metabolites in soil from UK treated with FASTAC - 1981/82 trials -
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a loam in Hoath (UK). Application done in August. Poor quality report, only three values above LOQ, metabolites not detected. Therefore data are not suitable to derive endpoints.

5. Forbes S. (1983) Analysis of soil from UK (Hoath) for residues of FASTAC (WL 85871) - Soil persistence trial - Second year -
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a loam in Hoath (UK). Application done in August, second application to the same site. Poor quality report, only five values above LOQ. Therefore data are not suitable to derive endpoints.

6. Forbes S. (1983) Analysis of soil from UK (Hoath) for residues of FASTAC (WL 85871) - Soil persistence trial - third year -
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a loam in Hoath (UK). Application done in August, third application to the same site. Poor quality report, inconsistent resukts in the three trial years. Therefore data are not suitable to derive endpoints.

7. Forbes S. (1983) Analysis of soil from UK (Coates) for residues of WL 85871 - Soil persistence trial - Second year year
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a now described as silty clay soil in Coates (UK). Second application after one year on the same site. Poor quality of report. The soil is not well described, therefore data are not suitable to derive endpoints.

8. Coveney P. C. (1986) Analysis of soil from UK (Coates) for residues of WL 85871 - Soil persistence trial - Third year
Short description of a highly overdosed (0.5 kg ha^{-1}) dissipation trial in a peat soil in Coates (UK). Application done in June. Poor quality report. The soil is not well described, therefore data are not suitable to derive endpoints.

CA 7.1.2.2.1 Soil dissipation studies

Report:	CA 7.1.2.2.1/1 Geschke, S., 2015 Freezer Storage Stability of the Diastereomeric Forms of BAS 311 I and its Metabolites 3-Phenoxybenzoic acid and DCVA (Cis and Trans Isomers) in Soil ~~2015/1249075~~ 2015/1249074
Guidelines:	EC guideline 7032/VI/95 rev. 5 OECD 506 (2007) EPA guideline OPPTS 860.1380
GLP:	yes

Executive Summary

In the present study, the storage stability of cypermethrin isomers cis 1, cis 2, trans 3 and trans 4) and its metabolites 3-phenoxybenzoic acid and DCVA (cis and trans isomers) was investigated by fortifying untreated soil samples (six different soil types) with the test items and analysing the samples after different periods of storage (0, 30, 60, 120, 240, 360, 630, and 720 days) in the deep-freezer.

Description of the Analytical Method

Residues of cypermethrin isomers were extracted with 0.1 % formic acid in acetonitrile. An aliquot of the extract was evaporated to dryness and reconstituted in 0.1 % formic acid in acetonitrile/demineralized water (1:1, v/v) prior analysis by HPLC with MS/MS detection.
Residues of 3-phenoxybenzoic acid (3-PBA) and DCVA isomers cis and trans were extracted twice with acetonitrile/ demineralized water (70:30, v/v). An aliquot of the extracts was evaporated to dryness and reconstituted in 0.1 % formic acid in methanol/ demineralized water (20:80, v/v) prior to analysis by HPLC with MS/MS detection.

Untreated soil control samples were analyzed for residues of the analytes. Residues were below the limit of detection (< 30 % of LOQ).

Analysis of Storage Stability Samples for cypermethrin isomers (values reported as % of initial)

~~After~~ During 727 days of storage, 80 % to 100 % of the initial concentration of cypermethrin isomer cis 1 was found in spiked soil samples fortified with the mixture of the cis isomers. In case of the cis 2 isomer the range was 79% to 103%.

~~After~~ During 727 days of storage 75% to 114% of the initial concentration of the trans 3 isomer was found in samoles spiked with a mixture of both trans isomers. In case of trans 4 isomer the range was 80% to 107%.

~~After~~ During 727 days of storage, the residues of alpha-cypermethrin ~~were~~ remained in the range of 82% to 112 % of the initial concentration.

Analysis of Storage Stability Samples for PBA and DCVA (CIS and TRANS isomers)

~~After~~ During 721 days of storage, residues ~~were still~~ remained in the range of 87 % to 118 % of the initial concentration of cis-DCVA isomer in spiked soil samples fortified with DCVA isomers. The range for the trans-DCVA isomer was 74% to 110% of the initial concentration.

~~After~~ During 721 days of storage residues of 3-PBA ~~were still~~ remained in the range of 88% to 149 % of the initial concentration of 3-PBA.

According to EC guideline 7032/VI/95 and U.S. EPA guideline OPPTS 860.1380, residues can be regarded as stable if the mean recovery at a given storage period does not fall below 70 % of the initial value. The results indicate that alpha-cypermethrin, its isomers and metabolites were stable under deep-frozen (≤ -18°C) conditions in soil for at least 727 days of storage.

I. MATERIAL AND METHODS

A. MATERIALS

Test materials:	Cypermethrin cis isomers	Cypermethrin trans isomers	Alpha Cypermethrin	3-PBA	DCVA
Synonyms:					
Batch No.:	AC8949-76	AC8949-77	COD-000595	AC12251-34	AC9966-87
Purity:	99.8%	99.8%	99.2%	100%	99.0%
CAS #:	211504-93-7	211504-94-8	67375-30-8	3739-38-6	55701-05-8

B. STUDY DESIGN

1. Experimental Conditions

The storage stability of BAS 311 I (cypermethrin isomers cis-1, cis-2, trans-3, and trans-4) and its metabolites 3-phenoxybenzoic acid and DCVA (cis and trans isomers) was investigated by fortifying untreated soil samples (six different soil types) with the test items (cypermethrin cis isomers (cis-1/cis-2 = 45:55), cypermethrin trans isomers (trans-3 : trans-4 = 43.5 : 56.5), DCVA (mixture of cis/trans Isomers = 51.5:48.5), alpha-cypermethrin, and 3-PBA) and analysing the samples after different periods of storage (cis I, cis II, trans III, trans IV, and alpha-cypermethrin: 0, 31, 62, 122, 241, 361, 634, and 727 days; DCVA and 3-PBA: 0, 31, 60, 123, 241, 360, 633, and 721 days) at -18°C.

2. Description of analytical procedures

For determination of BAS 311 I (cypermethrin isomers cis-1, cis-2, trans-3, and trans-4) and its metabolites 3-PBA and DCVA (cis and trans isomers), soil samples were analysed by method R0034/01. The limit of quantification (LOQ) was 0.001 mg kg^{-1} for each analyte.

Determination of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4

Residues of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4 were extracted with 0.1% formic acid in acetonitrile. An aliquot of the extract was evaporated to dryness and reconstituted in 0.1% formic acid in acetonitrile/demineralized water (1:1, v/v) prior analysis by HPLC-MS/MS detection.

Determination of 3-phenoxybenzoic acid (3-PBA) and DCVA isomers cis and trans

Residues of 3-phenoxybenzoic acid (3-PBA) and DCVA isomers cis and trans were extracted twice with acetonitrile/ demineralized water (70:30, v/v). An aliquot of the extracts was evaporated to dryness and reconstituted in 0.1% formic acid in methanol/ demineralized water (20:80, v/v) prior to analysis by HPLC-MS/MS detection.

The analytical method used for determination of residues of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4, 3-PBA, and DCVA isomers cis and trans in soil samples was fully validated during a parallel GLP-compliant study (Geschke, 2014, S12-01863).

The method was concurrently validated with analyses of stored soil samples within the summarized study. Procedural recovery samples were obtained by fortification (10 x LOQ) of untreated soil samples with cypermethrin isomers cis-1, cis-2, trans-3, and trans-4, 3-PBA, and DCVA isomers cis and trans test items prior to extraction.

II. RESULTS AND DISCUSSION

Procedural recoveries for cypermethrin isomers cis-1, cis-2, trans-3, and trans-4

Overall mean recoveries ranged from 89% to 113% for cypermethrin isomer cis-1 for soil L120555 to L120560 at a fortification level of 0.009 mg kg^{-1} (n = 16); from 93% to 113% for cypermethrin isomer cis-2 for soil L120555 to L120560 at a fortification level of 0.011 mg kg^{-1} (n = 16); from 91% to 114% for cypermethrin isomer trans-3 for soil L120555 to L120560 at a fortification level of 0.0087 mg kg^{-1} (n = 16), and from 95% to 111% for cypermethrin isomer trans-4 for soil L120555 to L120560 at a fortification level of 0.0113 mg kg^{-1} (n = 16). The maximum relative standard deviation of 11.9% at the given fortification levels was observed for the four different isomers. The results are shown from Table 7.1.2.2.1-1 to Table 7.1.2.2.1-5.

Analysis of storage stability samples for cypermethrin isomers cis-1, cis-2, trans-3, and trans-4

After 727 days of storage, 83% (L120555), 83% (L120556), 89% (L120557), 82% (L120558), 84% (L120559) and 85% (L120560) of the initial concentration of cypermethrin isomer cis-1 was found in spiked soil samples fortified with the mixture of the cis isomers (cis-1/ cis-2 = 45:55).

After 727 days of storage, 89% (L120555), 90% (L120556), 90% (L120557), 88% (L120558), 89% (L120559) and 91% (L120560) of the initial concentration of cypermethrin isomer cis-2 was found in spiked soil samples fortified with mixture of the cis isomers (cis-1/ cis-2 = 45:55).

After 727 days of storage, 86% (L120555), 88% (L120556), 85% (L120557), 75% (L120558), 83% (L120559) and 84% (L120560) of the initial concentration of cypermethrin isomer trans-3 was found in spiked soil samples fortified with the mixture of the trans isomers (trans-3/ trans-4 = 43.5:56.5).

After 727 days of storage, 86% (L120555), 88% (L120556), 88% (L120557), 81% (L120558), 80% (L120559) and 86% (L120560) of the initial concentration of cypermethrin isomer trans-4 was found in spiked soil samples fortified with mixture of the trans isomers (trans-3/ trans-4 = 43.5:56.5).

After 727 days of storage, 89% (L120555), 90% (L120556), 86% (L120557), 88% (L120558), 88% (L120559) and 88% (L120560) of the initial concentration of cypermethrin isomer cis-2 was found in spiked soil samples fortified with the test item alpha-cypermethrin.

Recovery rates for storage samples is reported not corrected for procedural recoveries and recovery obtained at time zero is set to 100%. The results are shown from Table 7.1.2.2.1-1 to Table 7.1.2.2.1-5.

Procedural recoveries for 3-PBA and DCVA (cis and trans isomers)
Overall mean recoveries were 74% to 110% for 3-PBA for soil L120555 to L120560 at a fortification level of 0.01 mg kg^{-1} (n = 16), 75% to 100% for DCVA isomer cis for soil L120555 to L120560 at a fortification level of 0.0103 mg kg^{-1} (n = 16), and 79% to 105% for DCVA isomer trans for soil L120555 to L120560 at a fortification level of 0.0097 mg kg^{-1} (n = 16). The maximum relative standard deviation of 12.8% at the given fortification levels was observed for the three analytes. The results are shown from Table 7.1.2.2.1-6 to Table 7.1.2.2.1-8.

Analysis of Storage Stability Samples for PBA and DCVA (cis and trans isomers)
After 721 days of storage, 87% (L120555), 96% (L120556), 88% (L120557), 93% (L120558), 107% (L120559) and 93% (L120560) of the initial concentration of DCVA isomer cis was found in spiked soil samples fortified with the test item DCVA isomers (cis/ trans = 51.5:48.5).
After 721 days of storage, 75% (L120555), 82% (L120556), 91% (L120557), 88% (L120558), 87% (L120559) and 84% (L120560) of the initial concentration of DCVA isomer trans was found in spiked soil samples fortified with the test item DCVA isomers (cis/ trans = 51.5:48.5).
After 721 days of storage, 97% (L120555), 95% (L120556), 92% (L120557), 114% (L120558), 103% (L120559) and 101% (L120560) of the initial concentration of 3-PBA was found in spiked soil samples fortified with the test item 3-PBA.

Recovery rates for storage samples is reported not corrected for procedural recoveries and recovery obtained at time zero is set to 100%. The results are shown from Table 7.1.2.2.1-6 to Table 7.1.2.2.1-8.

Table 7.1.2.2.1-1: Storage stability of cypermethrin isomers cis-1, fortified with the test item cypermethrin cis isomers (cis-1/cis-2 = 45:55) from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	116	107	112	100	104±6.3
	31	103	109	106	95	
	62	111	108	110	98	
	122	106	98	102	91	
	241	105	105	105	94	
	361	95	98	97	87	
	634	108	106	107	96	
	727	93	92	93	83	
L120556	0	111	110	106	100	101±6.2
	31	100	103	102	96	
	62	103	104	104	98	
	122	98	102	100	95	
	241	96	97	97	91	
	361	106	104	105	100	
	634	100	103	102	96	
	727	89	87	88	83	
L120557	0	114	117	116	100	103±6.3
	31	102	107	105	90	
	62	100	100	100	87	
	122	104	106	105	91	
	241	100	96	98	85	
	361	99	100	100	86	
	634	91	94	93	80	
	727	103	102	103	89	
L120558	0	105	110	108	100	101±5.6
	31	104	106	105	98	
	62	102	101	102	94	
	122	103	104	104	96	
	241	99	103	101	94	
	361	101	101	101	94	
	634	102	106	104	97	
	727	88	88	89	82	
L120559	0	111	110	111	100	104±5.8
	31	105	106	106	95	
	62	110	111	111	100	
	122	103	104	104	94	
	241	111	107	109	99	
	361	100	98	99	90	
	634	101	99	100	90	
	727	93	92	93	84	
L120560	0	108	112	110	100	103±5.6
	31	107	108	108	98	
	62	110	111	111	100	
	122	104	104	104	95	
	241	103	101	102	93	
	361	98	101	100	90	
	634	99	99	99	90	
	727	92	94	93	85	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-2: Storage stability of cypermethrin isomers cis-2, fortified with the test item cypermethrin cis isomers (cis-1/cis-2 = 45:55) from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	106	98	102	100	100±6.0
	31	103	107	105	103	
	62	102	102	102	100	
	122	96	89	93	91	
	241	105	105	105	103	
	361	98	97	98	96	
	634	108	105	107	104	
	727	92	89	91	89	
L120556	0	101	91	96	100	94±5.0
	31	93	95	94	98	
	62	93	93	93	97	
	122	90	92	91	95	
	241	101	101	101	105	
	361	99	98	99	103	
	634	95	97	96	100	
	727	87	85	86	90	
L120557	0	108	112	110	100	97±6.8
	31	101	104	103	93	
	62	92	92	92	84	
	122	95	96	96	87	
	241	96	97	97	88	
	361	93	97	95	86	
	634	86	88	87	79	
	727	100	99	100	90	
L120558	0	95	99	97	100	94±5.1
	31	95	96	96	98	
	62	91	91	91	94	
	122	94	93	94	96	
	241	97	104	101	104	
	361	93	96	95	97	
	634	97	100	99	102	
	727	85	85	85	88	
L120559	0	101	100	101	100	98±5.4
	31	102	102	102	101	
	62	98	101	100	99	
	122	92	91	92	91	
	241	108	105	107	106	
	361	98	95	97	96	
	634	96	95	96	95	
	727	90	89	90	89	
L120560	0	98	101	104	100	97±4.3
	31	99	99	102	99	
	62	97	100	99	99	
	122	94	93	97	94	
	241	104	103	104	104	
	361	94	97	99	96	
	634	92	93	93	93	
	727	89	93	91	91	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-3: Storage stability of cypermethrin isomers trans-3, fortified with the test item cypermethrin trans isomers (trans-3 : trans-4 = 43.5 : 56.5) from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	108	109	109	100	109±8.1
	31	110	107	109	100	
	62	124	124	124	114	
	122	112	114	113	104	
	241	103	99	101	93	
	361	107	109	108	100	
	634	114	117	116	106	
	727	92	94	93	86	
L120556	0	97	98	98	100	98±6.5
	31	101	100	101	103	
	62	110	112	111	114	
	122	98	97	98	100	
	241	98	96	97	99	
	361	101	101	101	104	
	634	99	102	101	103	
	727	86	86	86	88	
L120557	0	117	114	116	100	101±9.2
	31	104	106	105	91	
	62	110	109	110	95	
	122	104	113	109	94	
	241	93	93	93	81	
	361	89	88	89	77	
	634	95	90	93	80	
	727	102	95	99	85	
L120558	0	105	105	105	100	97±11.9
	31	105	107	106	101	
	62	108	114	111	106	
	122	105	106	106	100	
	241	90	87	89	84	
	361	83	81	82	78	
	634	99	99	99	94	
	727	77	80	79	75	
L120559	0	106	106	106	100	100±8.1
	31	113	103	108	102	
	62	113	108	111	104	
	122	105	106	106	100	
	241	97	98	98	92	
	361	94	100	97	92	
	634	93	89	91	86	
	727	86	90	88	83	
L120560	0	107	111	109	100	100±8.9
	31	102	97	100	91	
	62	115	118	117	107	
	122	106	102	104	95	
	241	95	97	96	88	
	361	97	85	91	83	
	634	92	93	93	85	
	727	92	92	92	84	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-4: Storage stability of cypermethrin isomers trans-4, fortified with the test item cypermethrin trans isomers (trans-3 : trans-4 = 43.5 : 56.5) from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	104	110	107	100	104±6.6
	31	105	99	102	95	
	62	110	109	110	102	
	122	96	104	100	93	
	241	101	101	101	94	
	361	113	117	116	107	
	634	107	108	108	100	
	727	94	91	93	86	
L120556	0	98	102	100	100	97±4.6
	31	96	103	100	100	
	62	97	98	98	98	
	122	95	90	93	93	
	241	99	97	98	98	
	361	99	101	100	100	
	634	99	98	99	99	
	727	86	90	88	88	
L120557	0	112	112	112	100	100±5.9
	31	104	104	104	93	
	62	96	102	99	88	
	122	97	100	99	88	
	241	94	96	95	85	
	361	97	98	98	87	
	634	93	91	92	82	
	727	99	97	98	88	
L120558	0	110	108	109	100	100±6.9
	31	104	104	104	95	
	62	100	100	100	92	
	122	99	97	98	90	
	241	96	95	96	88	
	361	95	93	94	86	
	634	106	110	108	99	
	727	85	91	88	81	
L120559	0	110	112	111	100	102±6.5
	31	109	104	107	96	
	62	102	96	99	89	
	122	100	99	100	90	
	241	98	105	102	91	
	361	104	109	107	96	
	634	100	99	100	90	
	727	89	89	889	80	
L120560	0	109	112	111	100	101±5.1
	31	104	104	104	94	
	62	104	101	103	93	
	122	102	102	102	92	
	241	96	103	100	90	
	361	104	96	100	90	
	634	94	97	96	86	
	727	96	93	95	86	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-5: Storage stability of cypermethrin isomers cis-2, fortified with the test item alpha-cypermethrin from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	108	115	112	100	113±7.0
	31	112	110	111	100	
	62	131	119	126	112	
	122	107	107	107	96	
	241	118	118	116	106	
	361	115	111	113	101	
	634	119	120	120	107	
	727	99	99	99	89	
L120556	0	109	110	110	100	108±4.1
	31	104	108	106	97	
	62	106	107	107	97	
	122	108	107	108	98	
	241	113	114	114	104	
	361	111	113	112	102	
	634	109	106	108	98	
	727	98	99	99	90	
L120557	0	122	121	122	100	108±5.5
	31	108	107	108	88	
	62	105	106	106	87	
	122	107	110	109	89	
	241	108	112	110	91	
	361	107	112	110	90	
	634	99	101	100	92	
	727	106	104	105	86	
L120558	0	101	107	104	100	103±5.7
	31	105	102	104	100	
	62	97	98	98	94	
	122	104	103	104	100	
	241	110	114	112	108	
	361	103	107	105	101	
	634	106	106	106	102	
	727	95	89	92	88	
L120559	0	110	114	112	100	106±4.6
	31	109	110	110	98	
	62	102	102	102	91	
	122	103	103	103	92	
	241	111	113	112	100	
	361	106	103	105	93	
	634	104	105	105	93	
	727	99	97	98	88	
L120560	0	104	110	107	100	104±4.7
	31	107	109	108	101	
	62	99	104	102	95	
	122	104	99	102	95	
	241	106	110	108	101	
	361	105	107	106	99	
	634	108	104	106	99	
	727	97	92	95	88	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-6: Storage stability of DCVA isomer cis, fortified with the test item DCVA (mixture of cis/trans isomers = 51.5:48.5) from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	89	91	90	100	94±8.1
	31	92	101	97	107	
	60	96	94	95	106	
	123	100	103	102	113	
	241	100	105	103	114	
	360	95	100	98	108	
	633	88	96	92	102	
	721	80	77	79	87	
L120556	0	90	89	90	100	96±9.9
	31	108	101	105	117	
	60	108	104	106	118	
	123	103	97	100	112	
	241	96	104	100	112	
	360	96	100	98	109	
	633	70	85	78	87	
	721	92	90	86	96	
L120557	0	96	86	91	100	89±7.6
	31	91	84	88	96	
	60	98	92	95	104	
	123	103	86	95	104	
	241	90	91	91	99	
	360	90	93	92	101	
	633	81	83	82	90	
	721	86	74	80	88	
L120558	0	99	97	98	100	96±8.5
	31	97	108	103	105	
	60	109	102	106	108	
	123	105	90	98	99	
	241	75	89	82	84	
	360	98	92	95	97	
	633	91	95	88	90	
	721	89	93	91	93	
L120559	0	84	92	88	100	94±7.2
	31	105	88	97	110	
	60	95	104	100	113	
	123	100	105	103	116	
	241	97	98	98	111	
	360	90	91	91	103	
	633	84	86	85	97	
	721	93	95	94	107	
L120560	0	89	91	90	100	96±9.6
	31	79	100	90	99	
	60	87	96	92	102	
	123	102	110	106	118	
	241	103	104	104	115	
	360	99	109	104	116	
	633	100	92	96	107	
	721	81	86	84	93	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-7: Storage stability of DCVA isomer trans, fortified with the test item DCVA (mixture of cis/trans isomers = 51.5:48.5) from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	104	102	103	100	94±9.5
	31	92	101	97	94	
	60	103	97	100	97	
	123	94	98	96	93	
	241	97	101	99	96	
	360	92	89	91	88	
	633	90	89	90	87	
	721	66	88	77	75	
L120556	0	110	105	108	100	97±10.5
	31	106	103	105	97	
	60	107	106	107	99	
	123	103	94	99	92	
	241	102	103	103	95	
	360	88	85	87	80	
	633	76	83	80	74	
	721	88	89	89	82	
L120557	0	101	102	102	100	95±5.9
	31	97	96	97	95	
	60	103	100	102	100	
	123	95	93	94	93	
	241	98	95	97	95	
	360	94	90	92	91	
	633	82	86	84	83	
	721	95	90	93	91	
L120558	0	108	108	108	100	101±9.1
	31	106	106	106	98	
	60	116	122	119	110	
	123	99	95	97	90	
	241	99	99	99	92	
	360	89	91	90	83	
	633	92	92	92	85	
	721	98	93	96	88	
L120559	0	102	102	102	100	94±8.6
	31	108	92	100	98	
	60	103	103	103	101	
	123	99	96	98	96	
	241	96	91	94	92	
	360	84	83	84	82	
	633	82	84	83	81	
	721	90	88	89	87	
L120560	0	108	108	108	100	96±7.2
	31	95	103	99	92	
	60	101	103	102	94	
	123	91	95	93	86	
	241	103	99	101	94	
	360	88	88	88	81	
	633	90	89	90	83	
	721	90	91	91	84	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

Table 7.1.2.2.1-8: Storage stability of 3-PBA, fortified with the test item 3-PBA from deep-frozen soil

Soil	Storage period [days]	Recovery [%]		Mean Recovery [%]	% of Initial*	Overall mean Recovery ± RSD (%)
L120555	0	88	82	85	100	88±6.5
	31	89	84	87	102	
	60	94	101	98	115	
	123	84	86	85	100	
	241	95	94	95	111	
	360	89	91	90	106	
	633	86	80	83	98	
	721	86	79	83	97	
L120556	0	89	91	90	100	91±7.1
	31	89	91	90	100	
	60	101	103	102	113	
	123	99	93	96	107	
	241	96	95	96	106	
	360	90	88	89	99	
	633	81	79	80	89	
	721	86	85	86	95	
L120557	0	88	86	87	100	84±8.0
	31	81	76	79	90	
	60	98	101	100	114	
	123	80	82	81	93	
	241	84	90	87	100	
	360	82	83	83	95	
	633	80	79	80	91	
	721	77	83	80	92	
L120558	0	83	83	83	100	96±12.8
	31	99	98	99	119	
	60	119	129	124	149	
	123	89	83	86	104	
	241	96	96	96	116	
	360	96	97	97	116	
	633	85	86	86	103	
	721	94	96	95	114	
L120559	0	87	87	87	100	92±6.3
	31	87	85	86	99	
	60	98	101	100	114	
	123	94	95	95	109	
	241	99	99	99	114	
	360	93	93	93	107	
	633	86	82	84	97	
	721	86	94	90	103	
L120560	0	96	97	97	100	93±9.3
	31	96	95	96	99	
	60	107	104	106	109	
	123	98	99	99	102	
	241	81	90	86	89	
	360	86	84	85	88	
	633	78	78	78	81	
	721	94	100	97	101	

* = The mean recovery at 0 days was set to 100 % of initial; RSD: relative standard deviation; values not corrected for procedural recoveries

III. CONCLUSION

According to EC guideline 7032/VI/95 and U.S. EPA guideline OPPTS 860.1380, residues can be regarded as stable if the mean recovery at a given storage period does not fall below 70% of the initial value. The results show that cypermethrin isomers cis-1, cis-2, trans-3, and trans-4 are stable under deep-frozen conditions in (≤ -18°C) soil for at least 721 days of storage.

According to EC guideline 7032/VI/95 and U.S. EPA guideline OPPTS 860.1380, residues can be regarded as stable if the mean recovery at a given storage period does not fall below 70% of the initial value. The results indicate that 3-PBA and DCVA isomers cis and trans are stable under deep-frozen (≤ -18°C) conditions in soil for at least 727 days of storage.

Report:	CA 7.1.2.2.1/2 Jacobsen B., Warren R., Robaugh D.A., Schmitt J.L., Perez S.R., Saha M., 2013a Terrestrial Field Dissipation study of the Insecticide Alpha-Cypermethrin (BAS 310 I) Following Broadcast Application of BAS 310 65 I Using a Row Crop Use Pattern 2013/7002604
Guidelines:	EPA OPPTS 835.6100
GLP:	yes (certified by United States Environmental Protection Agency, USA)

Executive Summary

The purpose of this study was to determine the mobility and persistence potential of alpha-cypermethrin (BAS 310 I) when applied under field conditions as a capsule suspension (CS) formulation to a bare soil plot. Field sites in Louisiana, Oklahoma, New York, and California were selected for this study and are geographically appropriate for the proposed use pattern of BAS 310 65 I in the U.S.. The BAS 310 65 I formulation utilized in this study is a capsule suspension formulation containing 100 g a.s. per liter.

The experiment was carried out in accordance with the terrestrial field dissipation requirements as outlined in the US EPA Fate, Transport and Transformation Test Guideline, OPPTS 835.6100: Terrestrial Field Dissipation.

At each test site, the designated treated plot received three uniform broadcast applications of BAS 310 65 I, with a targeted 7-day interval between applications, at a target rate of 28 g a.s. ha^{-1} per application, the maximum proposed label use rate. At each site there were two test plots: one treated bare soil plot and one control bare soil plot. The treated plot at each test site was divided into three replicate subplots. The plots were kept in a bare soil condition throughout the study. Rainfall was supplemented with irrigation at all four of the test sites.

The application rate was verified at each application at each test location by placing 10 environmental extraction disks in each of the three replicate subplots just prior to each application. Considering all sites, the overall mean recovery of alpha-cypermethrin (cis-2 isomer) plus the cis-1 isomer (a transformation product) from the extraction disk application monitor samples was 81% of the target (considering applications 1, 2, and 3).

Five soil core samples were taken from the soil surface to a depth of 36 inches (about 91 cm) at various times following each application and the final (third) application from each replicate subplot. Soil samples were targeted for collection at T1 (immediately after first application), T1+1, T1+3, -T2, T2, -T3, T3 and then 1, 3, 6, 10, 20, 30, 60, 90, 120, 180 and 270 days after last application (DALA). Soil cores were sectioned into segments of 0-2, 2-4, and 4-6, 6-12, 12-18, 18-24, 24-30 and 30-36 inches (about 0-5, 5-10, 10-15, 15-30, 30-46, 46-61, 61-76 and 76-91 cm). Core segments were composited by depth for each replicate subplot.

Soil samples were extracted with acetonitrile and extracts were analyzed for alpha-cypermethrin (cis-2 isomer) as well as the other cypermethrin isomers cis-1, trans-3, and trans-4 in order to evaluate potential isomer interconversion. Sample extracts were also analyzed for the soil transformation products cis- and trans-DCVA, and 3-PBA. High Performance Liquid Chromatography coupled with tandem mass spectrometry (HPLC-MS/MS) analytical method was used with a limit of quantification of 0.001 ppm and limit of detection of 0.0002 ppm for each analyte.

Analytical dry weight residue concentrations, undisturbed soil bulk density values, and segment lengths were used to calculate the mass of each analyte in the core segments on a gram per hectare basis. The metabolite values (3-PBA, cis-DCVA, and trans-DCVA) were converted to alpha-cypermethrin equivalent mass (by multiplying for the appropriate molecular weight factor). Total mass values of each analyte were used to determine the rate of dissipation of the analytes over time.

First-order (SFO) kinetic fits were selected to represent the dissipation of the sum of isomers at the California, Louisiana, and Oklahoma sites, while the biphasic FOMC model was selected for the New York site. Dissipation of the sum of isomers was rapid at all four sites. The $DisT_{50}$ values ranged from 3.4 days to 6.3 days with a median of 5.0 days, an average of 4.9 days, and a standard deviation of 1.4 days. The $DisT_{90}$ values ranged from 13.4 to 27.9 days with a median of 20.3 days, an average of 20.5 days, and a standard deviation of 5.9 days.

Metabolites 3-PBA, cis-DCVA, and trans-DCVA were observed at all four sites. Metabolite dissipation was characterized by simple calculation of the observed $DisT_{90}$. The interval between the maximum average observed level and the time at which the observed average dissipated to ≤ 10% of the maximum was determined. The maximum average observed level of 3-PBA ranged from 2.5 to 10% with an average of 6.0%. The observed $DisT_{90}$ of 3-PBA ranged from 5 to 37 days with an average of 23 days. The maximum average observed level of cis-DCVA ranged from 1.5 to 2.9% with an average of 2.1%. The observed $DisT_{90}$ of cis-DCVA ranged from 9 to 60 days with an average of 24 days. The maximum average observed level of trans-DCVA ranged from 0.2 to 1.7% with an average of 0.9%. The observed $DisT_{90}$ of trans-DCVA ranged from 1 to 9 days with an average of 6 days. The maximum average observed level of the sum of DCVA isomers ranged from 1.7 to 3.8% with an average of 3.0%. The observed $DisT_{90}$ of the DCVA isomer sum ranged from 9 to 60 days with an average of 24 days. These data indicate that the metabolites of alpha-cypermethrin were not persistent with 90% dissipation from peak average observed levels occurring within approximately one month.

Alpha-cypermethrin dissipated by aerobic soil processes and formed expected transformation products 3-PBA, cis-DCVA, and trans-DCVA at each site. Limited conversion of alpha-cypermethrin (cis-2 isomer) to other cypermethrin isomers (cis-1, trans-3, trans-4) was observed. While the cis-1 isomers were most prevalent, maximum average levels reached only 2.8 to 5.0% of the amount applied. Neither alpha-cypermethrin nor degradation products appeared to be inherently susceptible to leaching. They were not detected in soil below 2-4 inch (about 10 cm) soil depth at any of the four locations, even if site hydrologic conditions were generally favorable for leaching at all four sites. Overall, the aerobic soil transformation was the primary dissipation process in combination with a very limited leaching mobility.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item (formulation):	BAS 310 65 I
Active ingredient:	alpha-cypermethrin (Reg. No. 4078193)
Chemical name (IUPAC):	Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molar mass:	416.3 g mol^{-1}
Batch No.:	144315 (containing 99.3 g alpha-cypermethrin L^{-1})
Type of formulation:	CS

2. Test sites

The dissipation of alpha-cypermethrin and its metabolites 3-PBA, cis-DCVA, and trans-DCVA under field conditions was investigated at four sites in the U.S. The site characteristics including the basic soil parameters of the corresponding soil horizons are presented in Table 7.1.2.2.1-9 and Table 7.1.2.2.1-10.

Table 7.1.2.2.1-9: Characteristics of the trial sites used in the field dissipation study (Louisiana and Oklahoma sites)

Trial	**R120066**					
Location	**Louisiana, USA**					
Soil properties	**0-6 inches (0-15 cm)**	**6-12 inches (15-30 cm)**	**12-18 inches (30-46 cm)**	**18-24 inches (46-61 cm)**	**24-30 inches (61-76 cm)**	**30-36 inches (76-91 cm)**
Soil class (USDA)	silt loam	silty clay loam	silty clay loam	clay loam	silty clay loam	silty clay loam
sand [%]	20	12	14	20	18	18
silt [%]	57	55	47	43	51	53
clay [%]	23	33	39	37	31	29
Total organic C [%]	0.92	0.84	0.64	0.54	0.33	0.28
Organic matter [%]	1.6	1.4	1.10	0.92	0.57	0.48
pH ($CaCl_2$)	6.2	6.4	5.4	5.4	5.7	5.9
CEC [meq $100g^{-1}$]	12.2	15.7	18.6	20.9	17.7	16.5
Moisture (gravimetric) at 1/3 bar [%]	20.1	26.5	30.6	30.9	27.8	24.2
Trial	**R120067**					
Location	**Oklahoma, USA**					
Soil properties	**0-6 inches (0-15 cm)**	**6-12 inches (15-30 cm)**	**12-18 inches (30-46 cm)**	**18-24 inches (46-61 cm)**	**24-30 inches (61-76 cm)**	**30-36 inches (76-91 cm)**
Soil class (USDA)	sandy loam	sandy loam	sandy loam	loam	loam	loam
sand [%]	69	67	57	51	45	41
silt [%]	19	19	27	29	33	35
clay [%]	12	14	16	20	22	24
Total organic C [%]	0.55	0.46	0.55	0.50	0.44	0.44
Organic matter [%]	0.94	0.79	0.94	0.87	0.75	0.75
pH ($CaCl_2$)	6.7	5.3	6.0	6.2	6.5	6.5
CEC [meq $100g^{-1}$]	8.3	8.2	9.7	10.3	11.7	13.9
Moisture (gravimetric) at 1/3 bar [%]	10.2	12.1	15.4	17.2	20.4	23.4

CEC = Cation exchange capacity
MWHC = Maximum water holding capacity

Table 7.1.2.2.1-10: Characteristics of the trial sites used in the field dissipation study (New York and California sites)

Trial	**R120068**					
Location	**New York, USA**					
Soil properties	**0-6 inches (0-15 cm)**	**6-12 inches (15-30 cm)**	**12-18 inches (30-46 cm)**	**18-24 inches (46-61 cm)**	**24-30 inches (61-76 cm)**	**30-36 inches (76-91 cm)**
Soil class (USDA)	sand	sand	sand	sand	sand	sand
sand [%]	91	92	93	95	95	95
silt [%]	7	6	5	3	3	3
clay [%]	2	2	2	2	2	2
Total organic C [%]	0.78	0.83	0.25	0.15	0.25	0.13
Organic matter [%]	1.3	1.4	0.43	0.26	0.43	0.22
pH ($CaCl_2$)	7.1	6.0	6.5	6.6	6.2	5.9
CEC [meq $100g^{-1}$]	5.6	5.7	3.5	2.5	3.3	2.2
Moisture (gravimetric) at 1/3 bar [%]	6.7	7.3	3.7	2.5	3.6	2.6
Trial	**R120069**					
Location	**California, USA**					
Soil properties	**0-6 inches (0-15 cm)**	**6-12 inches (15-30 cm)**	**12-18 inches (30-46 cm)**	**18-24 inches (46-61 cm)**	**24-30 inches (61-76 cm)**	**30-36 inches (76-91 cm)**
Soil class (USDA)	sandy loam	sandy loam	sandy loam	sandy loam	sandy loam	sandy loam
sand [%]	69	67	61	65	67	73
silt [%]	24	28	30	28	26	22
clay [%]	7	5	9	7	7	5
Total organic C [%]	0.68	0.33	0.23	0.11	0.16	0.09
Organic matter [%]	1.20	0.57	0.40	0.19	0.28	0.15
pH ($CaCl_2$)	6.8	7.5	7.9	8.1	8.1	8.1
CEC [meq $100g^{-1}$]	11.3	8.5	10.7	10.9	11.0	10.1
Moisture (gravimetric) at 1/3 bar [%]	15.9	14.5	19.3	17.3	16.9	14.6

CEC = Cation exchange capacity
MWHC = Maximum water holding capacity

The selected fields represented typical regions of agricultural practice and had been under cultivation for many years. The sampling sites were fallow prior to the study. No product containing alpha-cypermethrin had been used on the test plots in the last three years.

B. STUDY DESIGN

1. Experimental conditions

Each trial area was divided into two plots. One plot (about 47-151 m²) was used as control plot (untreated) and the second plot (about 348-557 m²) was treated with the test item. Treated plots consisted of three subplots (A, B, C) that were assigned for replicates. The size of each treated replicate (subplot) was 139.4 m^2 (R120066), 116.2 m² (R120067), 174.1 m² (R120068), and 186.1 m² (R120069), respectively. The control plot was not subdivided, but was separated from the treated plot by a buffer zone of at least 15 m width.

The product, formulated as a CS, was broadcast applied to bare soil in three applications, each at a nominal rate of 28 g a.s. ha^{-1}. Depending on the trial site, the applications were conducted from the end of Jun-2012 until the end of Jul-2012 using a calibrated boom sprayer. The actual application rates determined by quantifying the amount of spray discharged ranged from 27 to 29 g a.s. ha^{-1}. Details of the application are presented in Table 7.1.2.2.1-11.

Three methods were used to verify the application of alpha-cypermethrin at each trial site: calibrated sprayer pass time calculations, application verification device analysis, and determination of the total mass of alpha-cypermethrin present in application day soil samples. Details are given in the study report.

Table 7.1.2.2.1-11: Application parameters of field trial sites treated with alpha-cypermethrin

Trial Location	Test item/ Nominal content/ Formulation type	Application method	No. of applications	Application rate per treatment			No. of treated replicates	Application date
				Nominal [g a.s. ha^{-1}]	Actual [g a.s. ha^{-1}]	Dose verification*		
R120066 Louisiana	BAS 310 65 I 100 g a.s. L^{-1} CS	Broadcast spray to bare soil	3	28	28 27 29	101% of nominal rate	3 (139.4 m^2 each)	26-Jun-12 03-Jul-12 10-Jul-12
R120067 Oklahoma	BAS 310 65 I 100 g a.s. L^{-1} CS	Broadcast spray to bare soil	3	28	28 29 29	103% of nominal rate	3 (116.2 m^2 each)	27-Jun-12 03-Jul-12 10-Jul-12
R120068 New York	BAS 310 65 I 100 g a.s. L^{-1} CS	Broadcast spray to bare soil	3	28	29 29 29	103% of nominal rate	3 (174.1 m^2 each)	10-Jul-12 17-Jul-12 24-Jul-12
R120069 California	BAS 310 65 I 100 g a.s. L^{-1} CS	Broadcast spray to bare soil	3	28	29 28 28	101% of nominal rate	3 (186.1 m^2 each)	17-Jul-12 24-Jul-12 31-Jul-12

No tillage was performed during the course of the study from first to last sampling and no crops were grown throughout any of the trials. The plots were kept free of vegetation via the application of glyphosate, glufosinate, and paraquat.

Climatic conditions were based on records of appropriate weather stations located at a distance of 8-88 km from each site. Irrigation was applied to supplement normal precipitation so that the plots would receive 120% of historical average rainfall for the study period.

A summary of monthly weather data (temperature and precipitation, as well as volumes of the supplementary irrigation) is presented in Table 7.1.2.2.1-12.

Table 7.1.2.2.1-12: Summary of monthly air temperature, precipitation, and irrigation at each field trial site

Trial	R120066			R120067		
Location	Louisiana, USA			Oklahoma, USA		
Month/ Year	T_{mean} Air [°C]	Precipitation* [mm]	Irrigation* [mm]	T_{mean} Air [°C]	Precipitation* [mm]	Irrigation* [mm]
Jun 12	28.89	0.00	0.00	30.56	0.00	16.26
Jul 12	27.72	187.96	0.00	29.94	21.59	71.37
Aug 12	27.50	118.87	48.26	27.11	68.58	55.88
Sep 12	25.11	127.25	22.86	22.94	93.98	54.61
Oct 12	19.33	8.13	15.75	14.94	22.86	73.66
Nov 12	14.39	52.83	129.29	11.22	12.70	31.75
Dec 12	12.61	185.42	78.49	4.33	27.94	0.00
Jan 13	11.67	337.06	0.00	3.44	24.13	0.00
Feb-13	13.17	183.64	0.00	4.11	88.90	0.00
Mar-13	13.28	51.05	0.00	8.39	33.02	62.23
Apr-13	15.94	41.40	50.80	7.06	66.04	0.00
Trial	**R120068**			**R120069**		
Location	**New York, USA**			**California, USA**		
Month/ Year	**T_{mean} Air [°C]**	**Precipitation* [mm]**	**Irrigation* [mm]**	**T_{mean} Air [°C]**	**Precipitation* [mm]**	**Irrigation* [mm]**
Jun 12	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Jul 12	23.61	62.74	62.74	24.67	0.00	152.40
Aug 12	21.22	51.31	50.29	26.83	0.00	355.60
Sep 12	16.50	123.19	12.70	24.06	0.00	279.40
Oct 12	11.50	109.73	12.70	17.22	0.00	228.60
Nov 12	2.67	57.40	0.00	11.44	4.57	50.80
Dec 12	1.83	72.14	0.00	7.67	41.15	50.80
Jan 13	-2.06	42.67	0.00	5.78	19.05	76.20
Feb-13	-3.06	45.72	0.00	7.94	21.34	76.20
Mar-13	0.61	23.62	0.00	14.06	18.80	152.40
Apr-13	5.78	74.68	12.70	16.56	2.03	228.60

* Weather data refer to time period from start of trial (day of application) until end of trial (day of last sampling)
n.d. = Not determined

2. Sampling

Replicate soil samples were taken randomly at different sampling intervals (see Table 7.1.2.2.1-13) with a soil corer, starting one day prior to the first application until about 270 days after the last application. Fifteen cores were collected in the treated plot (5 cores for each subplot) and also 15 cores were taken from the control plot. A soil depth of up to 36 inches (~ 91 cm) was taken for each soil. Detailed sampling intervals are presented in Table 7.1.2.2.1-13.

Table 7.1.2.2.1-13: Summary of sampling intervals of the treated plots at each field trial site

Trial	Location	Sampling intervals [days]
R120066	Louisiana, USA	-T1, T1; T1+1; T1+3, -T2, T2, -T3, T3, [1, 3, 6, 10, 20, 30, 63, 91, 120, 202, 272]*
R120067	Oklahoma, USA	-T1, T1; T1+1; T1+3, -T2, T2, -T3, T3, [1, 3, 6, 10, 20, 30, 63, 92, 125, 181, 270]*
R120068	New York, USA	-T1, T1; T1+1; T1+3, -T2, T2, -T3, T3, [1, 3, 6, 10, 20, 30, 59, 90, 120, 177, 270]*
R120069	California, USA	-T1, T1; T1+1; T1+3, -T2, T2, -T3, T3, [1, 3, 6, 10, 20, 30, 59, 90, 120, 181, 269]*

T = Day of application (immediately after treatment)
-T = Day before the first application
* Sampling dates refer to the day of the last application (T3)

Samples from untreated soils were collected from the control plot on five occasions one day before the application (-T1) and 3, 10, 30 and 63 days (for trials R120066 and R120067) or 59 days (for trials R120068 and R120069) after the last application.
At each sampling interval in the treated plots, five 0-6 inches (0-15 cm) and five 6-36 inches (15-91 cm) soil cores were collected for each subplot (for a total of 15 0-6 inches and 15 6-36 inches cores), except on T1 when only 0-6 inches cores were collected, and on the day of each application (T1, T2, and T3), when a duplicate set of 0-2 inches cores was taken.

At –T1 in control plots, five 0-6 inches and five 6-36 inches cores were taken from three randomly selected sampling blocks for a total of 15 cores, while at subsequent sampling events five 0-6 inches and five 6-36 inches were selected from one randomly selected sampling block (for a total of 10 cores).
The specimens were pooled according to soil depth. Immediately after sampling and before freezing, all soil cores collected with the soil probe were segmented to 0-2, 2-4 and 4-6 inches, and 6-36 inches samples were cut into 6-inch segments and pooled by depth.

All soil specimens were placed into freezer storage at about -18°C and remained frozen until processing or analysis of the samples.

3. Description of analytical procedure

The analysis for alpha-cypermethrin, cypermethrin, trans-3, and trans-4-cypermethrin residues in soil samples and analysis for alpha-cypermethrin and cypermethrin residues in application verification and shipping verification samples was performed by Pyxant Labs Inc. (Colorado Springs, CO, USA). Analysis of the metabolites cis-DCVA, trans-DCVA, and 3-PBA was conducted by ADPEN Laboratories Inc. (Jacksonville, FL, USA).

Analysis of soil core samples for alpha-cypermethrin, cypermethrin, cis-DCVA, trans-DCVA, and 3-PBA was conducted using the BASF analytical method R0034/01. Details on the method are given in the study report.

Analysis of the application verification (AV) samples with environmental extraction disks (one of the three verification methods mentioned above) was conducted for residues of alpha-cypermethrin (cis-2 isomer) plus the cis-1 isomer, which was evident in the AV samples. The extraction disks were analyzed directly by extracting with acetonitrile on a solid phase extraction (SPE) vacuum manifold. The combined extracts were further diluted with 50:50 acetonitrile/water (v/v) with 0.1% formic acid and analyzed by HPLC-MS/MS.

For each site, three composite samples, one from each replicate treated plot, were analyzed at each sampling interval and depth. On application days (Tl, T2, and T3) three additional composite samples (0-2 inches (about 0-5 cm) depth only), one from each replicate treated plot, were also collected for analysis. Composite samples were generally analyzed once, however, selected samples were analyzed multiple times.

For alpha-cypermethrin and isomers, a 5 g soil sample was extracted by shaking once with 50 mL of 0.1% formic acid in acetonitrile. The extracts were concentrated to dryness and then diluted with 50:50 acetonitrile/water (v/v) with 0.1% formic acid. For the metabolites cis-DCVA, trans-DCVA, and 3-PBA, a 5 g soil sample was extracted by shaking twice with 25 mL acetonitrile/water (70:30, v/v). An aliquot (10%) from the combined extract was evaporated to dryness and then dissolved in methanol/water (20:80, v/v) with 0.1% formic acid.

All residues were determined using HPLC-MS/MS in the positive ion mode. The limit of quantitation was 0.001 mg kg^{-1} (ppm) and the limit of detection was 0.0002 mg kg^{-1} (ppm) for all analytes in soil. Soil results were reported on a dry weight basis for residue determination.

The shipping verification samples were analyzed for residues of alpha-cypermethrin (cis-2 isomers) plus the cis-1 isomer, according to BASF method R0034/01. The shipping verification samples (20 g each, weighed in the field) were extracted with 0.1% formic acid in acetonitrile. Following extraction, alpha-cypermethrin and cis-1 isomer residues were determined by HPLC-MS/MS.

4. Storage stability experiments

The stability of alpha-cypermethrin and its transformation products in frozen soil has been previously demonstrated in three studies [*ARCHER S. AND FORBES, S. (1985): Storage stability of FASTAC (WL85871) residues in crops and soil deep frozen at -18°C, BASF DocID 1985/7001362; HITCHINGS, E.J. (1989a): Storage stability of residues of the pyrethroid metabolite WL44607 added to soils deep frozen at -l8°C, BASF DocID 19817001056; HITCHINGS, E.J. (1989b): Storage stability of residues of the RIPCORD metabolite WL44776 added to soils deep frozen at -18°C, BASF DocID 1981/7001057*].

The freezer storage stability of alpha-cypermethrin, DCVA, and 3-PBA in soils collected from the four trial sites was evidenced in another study *[CA 7.1.2.2.1/1, BASF DocID 2014/1152598*] and will cover the interval needed for this study.

5. Kinetic evaluation

In order to generate a conservative kinetic assessment for alpha-cypermethrin (cis-2 isomer), the g ha^{-1} values for all isomers of the parent (cis-1, cis-2, trans-3, and trans-4, i.e. the cypermethrin isomers) were summed at each sampling time. The dissipation of the sum of isomers was assessed following the peak average level following the last application.

The guidance of the FOCUS work group on degradation kinetics [*FOCUS (2006*] was used as the general basis for conducting the kinetic analysis, statistical assessment, and selection of the best fit kinetic model. The software package KinGUII (version 2.2012.202.925) was used for parameter fitting. Decline of the isomer sum was evaluated using a two-parameter single first-order (SFO) model and the three-parameter first-order multi compartment (FOMC) biphasic model. Where FOMC provided a better fit, the four-parameter double first-order in parallel (DFOP) biphasic model was additionally assessed.

The goodness-of-fit of the kinetic models was assessed by visual inspection and statistical measures recommended by the FOCUS guidance. The goodness-of-fit used for the identification of the best-fit kinetic model was the χ^2 minimum error level. The kinetic endpoints ($DisT_{50}$ and $DisT_{90}$ values) are reported in Table 7.1.2.2.1-19 for the kinetic models which were selected based on statistical and visual assessment. The reliability of individual parameters was judged by means of a single-sided t-test.

II. RESULTS AND DISCUSSION

1. Application verification

The application verification samples (environmental extraction disks) were analyzed for alpha-cypermethrin (cis-2 isomer) plus the cis-1 isomers, which were evident in the samples. The application verification sample recoveries were corrected for procedural recoveries. The mean percent recovery from the application verification samples was 81% of the test item for all trial sites. Mean percent recoveries of the individual trial sites are given in Table 7.1.2.2.1-14.

Table 7.1.2.2.1-14: Summary of application verification sample recoveries (n = 3) [%]

Application No.	R120066 Louisiana	R120067 Oklahoma	R120068 New York	R120069 California
T1	109±16.0	67±2.5	77±5.5	66±10.0
T2	88±15.0	83±15	91±4.9	69±4.4
T3	85±6.9	78±3.1	77±4. 2	76±12.0

2. Residues in field soil samples

Mean procedural recoveries from control soil samples fortified with alpha-cypermethrin, cypermethrin isomers, cis-DCVA, trans-DCVA, and 3-PBA at 0.001 and 0.1 ppm were generally within the acceptable range of 70-120%. In those instances where low recoveries were obtained (typically for the trans-3 and trans-4 isomers), the sample was reanalyzed in duplicate to confirm results (the average of all three results were reported). The recovery results were not corrected as apparent residues were non-detectable in the associated control samples. A summary of the individual procedural recovery results is provided in the study report.

Field soil samples taken from different depths were analyzed to a maximum of about 272 days after the last of the three applications. The analytical results are summarized in Table 7.1.2.2.1-15 to Table 7.1.2.2.1-18. All residue values presented in these tables are related to the dry weight of the soil and were not corrected for procedural recoveries. Residue concentrations are reported according to soil depth (0-2, 2-4, 4-6, 6-12, 12-18, 18-24, 24-30 and 30-36 inches, corresponding to about 0-5, 5-10, 10-15, 15-30, 30-46, 46-61, 61-76 and 76-91 cm).

Although samples were collected to a depth of 36 inches, samples below 6-12 inches were generally not analyzed due to the absence of residues in overlying segments. Residue concentrations in samples taken prior to the application (-T1) at both sites were non-detectable (< 0.0002 ppm) and were omitted from the following tables for brevity.

Table 7.1.2.2.1-15: Residues of alpha-cypermethrin and metabolites [µg kg^{-1} dry weight] in treated soil samples (mean of three replicates) – Lousiana site (R120066)

Compound	Soil depth [in]	Targeted days after last application																
		T1	T1+1	T1+3	-T2	T2	-T3	T3	1	3	6	10	20	30	60 (63)	90 (91)	120	180 (202)	270 (272)
alpha-cypermethrin (cis-2 isomer)	0-2	50	32	24	20	48	26	78	40	64	41	9.3	5.7	3.1	*0.72*	*0.83*	*0.48*	*0.36*	*0.46*
	2-4	1.0	*0.96*	*0.65*	*0.36*	1.1	2.0	2.6	4.6	4.7	2.0	*0.71*	*0.57*	*0.29**	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	*0.86*	*0.52*	*0.33*	n.d.	*0.32**	*0.33*	*0.60*	*0.74*	1.5	*0.63*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	*0.33**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
cis-1 isomer	0-2	2.0	1.3	*0.99*	*0.82*	2.2	1.2	3.2	1.7	2.5	1.7	*0.56*	*0.34*	*0.23*	n.d.	n.d.	n.d.	-	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	*0.17**	*0.24**	*0.58**	*0.23*	*0.24**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
trans-3 isomer	0-2	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.21**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
trans-4 isomer	0-2	n.d.	*0.27*	*0.36**	*0.23**	*0.22*	*0.28*	*0.33**	*0.42*	*0.58*	*0.39*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.46**	n.d.	n.d.	-	-
cis-DCVA	0-2	*0.4*	*0.7*	*0.5*	*0.4*	*0.5*	*0.7*	*0.7*	*0.7*	*0.8*	*0.6*	*0.3**	*0.3**	*0.2*	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	*0.3**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.3**	n.d.
	4-6	*0.4**	*0.2**	*0.3**	*0.3**	*0.3**	*0.5**	*0.4*	*0.3*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Trans-DCVA	0-2	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.2*	n.d.	n.d.	n.d.	n.d.	*0.5**	*0.3*	*0.3**	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	*0.4**	n.d.	*0.6*	*0.3**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
3-PBA	0-2	n.d.	3.9	3.4	3.3	3.5	1.9	2.1	1.4	1.1	*0.9*	*0.4*	*0.4*	*0.3*	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	*0.3**	*0.3**	*0.3**	n.d.	*0.3**	*0.3**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.

T = Application dates
n.d. = Not detected
- = Not analyzed
* Denotes instance where the mean for all replicates is < 0.0002 ppm (< LOD), but one or two replicates contained detectable residues, the maximum of which is shown.
Values > LOD (> 0.0002 ppm), but < LOQ (< 0.001 ppm) are shown in italics.

Table 7.1.2.2.1-16: Residues of alpha-cypermethrin and metabolites [µg kg^{-1} dry weight] in treated soil samples (mean of three replicates) – Oklahoma site (R120067)

Compound	Soil depth [in]	Targeted days after last application																	
		T1	T1+1	T1+3	-T2	T2	-T3	T3	1	3	6	10	20	30	60 (63)	90 (92)	120 (125)	180 (181)	270
alpha-cypermethrin (cis-2 isomer)	0-2	48	26	19	14	54	26	69	54	41	30	21	6.8	1.5	*0.43*	n.d.	*0.47*	*0.45*	*0.47*
	2-4	*0.54*	n.d.	n.d.	n.d.	*0.20*	n.d.	*0.29**	n.d.	*0.30**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
cis-1 isomer	0-2	1.9	1.6	1.8	1.8	3.6	3.9	5.3	7.0	6.8	5.7	5.0	2.3	*0.60*	n.d.	n.d.	n.d.	*0.22**	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
trans-3 isomer	0-2	n.d.	n.d.	n.d.	*0.21**	n.d.	*0.29**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
trans-4 isomer	0-2	*0.22**	*0.35*	*0.42*	*0.33*	*0.34*	*0.47*	*0.53*	1.0	*0.82*	*0.49*	*0.31*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
cis-DCVA	0-2	*0.3**	*0.9*	*0.6*	*0.5*	1.0	1.0	1.1	1.7	2.0	1.7	1.4	*0.4**	n.d.	n.d.	n.d.	n.d.	-	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
Trans-DCVA	0-2	n.d.	12.7	n.d.	n.d.	n.d.	n.d.	n.d.	*0.6**	*0.5*	*0.3*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
3-PBA	0-2	*0.4*	1.2	*0.9*	1.3	1.9	1.6	*0.9*	1.6	3.4	1.8	1.5	*0.5*	*0.5**	n.d.	n.d.	n.d.	-	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	n.d.

T = Application dates
n.d. = Not detected
- = Not analyzed
* Denotes instance where the mean for all replicates is < 0.0002 ppm (< LOD), but one or two replicates contained detectable residues, the maximum of which is shown.
Values > LOD (> 0.0002 ppm), but < LOQ (< 0.001 ppm) are shown in italics.

Table 7.1.2.2.1-17: Residues of alpha-cypermethrin and metabolites [µg kg^{-1} dry weight] in treated soil samples (mean of three replicates) – New York site (R120068)

Compound	Soil depth [in]	Targeted days after last application																	
		T1	T1+1	T1+3	-T2	T2	-T3	T3	1	3	6	10	20	30	60 (59)	90	120	180 (177)	270
alpha-cypermethrin (cis-2 isomer)	0-2	30	24	22	13	36	19	46	42	20	14	12	4.7	3.7	3.1	2.2	1.8	2.9	-
	2-4	*0.26*	*0.35**	*0.68*	n.d.	1.5	n.d.	*0.26**	n.d.	n.d.	*0.36**	*0.46**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	4-6	n.d.	n.d.	*0.23*	n.d.	*0.45*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
cis-1 isomer	0-2	1.6	1.5	2.9	2.1	3.2	3.5	4.5	4.6	3.0	2.8	2.5	1.5	1.2	1.1	*0.63*	*0.37*	-	-
	2-4	n.d.	n.d.	n.d.	*0.27**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
trans-3 isomer	0-2	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.29**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
trans-4 isomer	0-2	n.d.	*0.44*	*0.51*	*0.37*	*0.31*	*0.43*	*0.44*	1.0	*0.54*	*0.39*	*0.32**	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-
cis-DCVA	0-2	n.d.	*0.4**	*0.6*	*0.7*	*0.7*	*0.9*	*0.9*	*0.4*	*0.5*	*0.4*	*0.3*	n.d.	*0.4*	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	*0.3**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Trans-DCVA	0-2	n.d.	n.d.	*0.4**	n.d.	*0.3**	*0.4**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
3-PBA	0-2	*0.3**	*0.4*	1.0	1.2	1.5	1.4	1.0	*0.9*	*0.4*	*0.6*	*0.5*	*0.3*	*0.3**	n.d.	n.d.	n.d.	n.d.	n.d.
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.

T = Application dates
n.d. = Not detected
- = Not analyzed
* Denotes instance where the mean for all replicates is < 0.0002 ppm (< LOD), but one or two replicates contained detectable residues, the maximum of which is shown.
Values > LOD (> 0.0002 ppm), but < LOQ (< 0.001 ppm) are shown in italics.

Table 7.1.2.2.1-18: Residues of alpha-cypermethrin and metabolites [µg kg^{-1} dry weight] in treated soil samples (mean of three replicates) – California site (R120069)

Compound	Soil depth [in]	Targeted days after last application																
		T1	T1+1	T1+3	-T2	T2	-T3	T3	1	3	6	10	20	30	60 (59)	90	120	180 (181)	270 (269)
alpha-cypermethrin (cis-2 isomer)	0-2	38	33	12	7.7	39	11	22	35	24	9.9	6.8	2.3	*0.90*	*0.64*	*0.75*	*0.37*	*0.30*	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	*0.31**	*0.24*	*0.25*	n.d.	*0.45*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	4-6	*0.26**	n.d.	n.d.	n.d.	n.d.	n.d.	*0.29**	n.d.	*0.37**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	*0.33**	n.d.	*0.39**	n.d.	*0.34**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
cis-1 isomer	0-2	1.8	4.0	3.1	2.4	3.7	3.9	2.5	5.7	5.9	4.0	2.8	1.1	*0.30*	*0.24**	*0.24**	n.d.	n.d.	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.60*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
trans-3 isomer	0-2	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	*0.26**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
trans-4 isomer	0-2	*0.24**	1.7	1.8	*0.97*	*0.93*	1.2	*0.57*	1.4	1.3	*0.66*	*0.35*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
cis-DCVA	0-2	*0.4**	*0.5*	*0.3*	*0.3*	*0.3*	*0.8*	*0.5*	1.3	1.0	*0.8*	*0.3**	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-	-	-	-
Trans-DCVA	0-2	n.d.	*0.6*	1.0	*0.7*	*0.5**	*0.6*	*0.4*	1.0	*0.7*	*0.3*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-	-	-	-
3-PBA	0-2	n.d.	1.5	1.3	1.8	*0.6*	1.6	1.3	6.2	1.7	*0.6*	*0.3*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-
	2-4	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-
	4-6	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-
	6-12	-	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	-	-	-	-	-	-	-	-	-	-

T = Application dates
n.d. = Not detected
- = Not analyzed
* Denotes instance where the mean for all replicates is < 0.0002 ppm (< LOD), but one or two replicates contained detectable residues, the maximum of which is shown.
Values > LOD (> 0.0002 ppm), but < LOQ (< 0.001 ppm) are shown in italics.

Alpha-cypermethrin was degraded significantly in all trials. The analytical data showed that about 90% of the applied amount was degraded 20 days after the last application. Quantifiable levels (> 0.001 mg kg^{-1}) of alpha-cypermethrin were only found to a depth of 2 inches (~ 5 cm). Alpha-cypermethrin was detected in soil depths > 4 inches (~ 10 cm) in trial Louisiana, New York, and California. However, measured concentrations of alpha-cypermethrin were all < LOQ.

Limited conversion of alpha-cypermethrin (cis-2) to other cypermethrin isomers (cis-1, trans-3, trans-4) was observed. While the cis-1 isomer was most prevalent, maximum average levels reached only 2.8 to 5.0% of the amount applied.

Metabolites cis- and trans-DCVA, as well as 3-PBA were detected in the upper 2 inches (~ 0-5 cm) and sporadically in soil depths of 2-4 inches (~ 5-10 cm) up to 30 days after the last application. At trial site Louisiana the trans-4 isomer of cypermethrin, cis-, and trans-DCVA were detected in samples > 60 days after application. At trial site Louisiana, metabolite cis-DCVA was detected also in soil depths from 4 to 6 inches (~ 10-15 cm) and up to 180 days after the last application in a soil depth of 2-4 inches (~ 5-10 cm). However, concentrations of cis-DCVA > 2 inches (~ 5 cm) depth were always < LOQ.

Overall, metabolites cis- and trans-DCVA were only present in small concentrations with detected concentrations always below the LOQ. Only at trial sites Oklahoma and California concentrations above the LOQ were detected in a soil depth of 0-2 inches (~ 0-5 cm).

Considerable concentrations (> LOQ) of the metabolite 3-PBA could be observed in the upper 2 inches (~ 0-5 cm) and up to 10 days after the applications. Detectable amounts of 3-PBA could be measured up to 30 days after the last application.

Leaching was assessed by analysis of soil core samples collected at multiple sampling events following each of three applications of the test substance to a depth of 36 inches (~ 91 cm). From the results it appears that leaching of alpha-cypermethrin and its metabolites can be disregarded as major dissipation route, since the active substance seems to have no potential to appear in groundwater.

The volatilization dissipation route was not evaluated since alpha-cypermethrin is classified as "nonvolatile from moist soil" using the calculation procedure specified in the OPPTS 835.6100 guideline. A vapor pressure of 7.7 x 10^{-8} Pa at 20°C, water solubility of 0.02 mg L^{-1} at 20°C, lowest soil adsorption K_d of 26492 [*Hill, A. (1993): [Benzyl-14C] WL85871 (FASTAC): Adsorption/desorption in three soils. BASF Reg. DocID 1993/7002014*], and a molecular weight of 416.3 g mol^{-1} were used in the calculation.

Surface water run off was not evaluated. Except for the Louisiana site (silt loam) surface and near surface soil textures were generally not conducive to infiltration excess and/or saturation excess runoff (sand and sandy loam) Further, field sites were nearly level (≤ 1% slope) and the protocol dictated that irrigation be managed to avoid ponding. Therefore, it is assumed that loss of water due to runoff was minor compared to infiltration and evaporation, and runoff was not considered to be a relevant dissipation route at these sites.

3. Kinetic evaluation

The calculated $DisT_{50}$ and $DisT_{90}$ values for the sum of alpha-cypermethrin (cis-2 isomer) plus other cypermethrin isomers (cis-1, trans-3, trans-4) in soil under terrestrial field conditions for each site are reported in Table 7.1.2.2.1-19. The values are based on the total mass in the sampled soil profile (0-36 inches, about 91 cm) over time.

Kinetic models were not fit to metabolite data due to the low and/or variable levels observed. Rather, dissipation of the metabolites 3-PBA, cis-DCVA, trans-DCVA, and the sum of DCVA isomers was characterized by calculation of the observed $DisT_{90}$ value. The day of the maximum average metabolite level was subtracted from the day the average level dissipated to ≤ 10% of the maximum. This interval represents the time in which 90% dissipation occurred and is used to characterize the persistence, or lack thereof, of the metabolites. The results for the metabolites are shown in Table 7.1.2.2.1-20.

Table 7.1.2.2.1-19: Summary of the kinetic evaluation of the sum of alpha-cypermethrin (cis-2) and isomers (cis-1, trans-3, trans-4)

Test site	Kinetic model	χ^2 error [%]	Best-fit endpoints	
			$DisT_{50}$ [d]	$DisT_{90}$ [d]
Louisiana	SFO	33.8	5.9	19.6
Oklahoma	SFO	6.3	6.3	20.9
New York	FOMC	13.5	3.4	27.9
California	SFO	8.4	4.0	13.4

Table 7.1.2.2.1-20: Interval from the maximum observed metabolite level to ≤ 10% of maximum ($DisT_{90}$; mean of three replicates)

Site	3-PBA				Cis-DCVA			
	Max [%]	Max DA1A	≤ 10% DA1A	$DisT_{90}$ interval [d]	Max [%]	Max DA1A	≤ 10% DA1A	$DisT_{90}$ interval [d]
Louisiana	6.8	1	24	23	2.0	14	24	10
Oklahoma	4.8	16	43	27	2.9	16	33	17
New York	2.5	7	44	37	1.5	13	73	60
California	10.0	15	20	5	2.1	15	24	9
Average	**6.0**	**10**	**33**	**23**	**2.1**	**15**	**39**	**24**
Site	**Trans-DCVA**				**DCVA Sum**			
	Max [%]*	**Max DA1A**	**≤ 10% DA1A**	**$DisT_{90}$ interval [d]**	**Max [%]**	**Max DA1A**	**≤ 10% DA1A**	**$DisT_{90}$ interval [d]**
Louisiana	1.0	15	20	5	2.7	15	24	9
Oklahoma	0.7	16	23	7	3.7	16	33	17
New York	0.2	13	14	1	1.7	13	73	60
California	1.7	15	24	9	3.8	15	24	9
Average	**0.9**	**15**	**20**	**6**	**3.0**	**15**	**39**	**24**

Max = Maximum average metabolite level of n = 3 replicates (g ha^{-1}) divided by the total target application rate of alpha-cypermethrin (84 g ha^{-1}) and multiplied by 100.

DA1A = Days after first application. Alpha-cypermethrin was applied at 0, 7, and 14 DA1A at the California, Louisiana, and New York sites and 0, 6, and 13 DA1A at the Oklahoma site. The target application rate for all applications was 28 g a.s. ha^{-1}.

For each analyte and site the day of the average maximum observed level of n = 3 replicates (Max DA1A) was subtracted from the day at which the average level dissipated to 10% or less of the maximum (≤10% DA1A). This provides the interval in which 90% of the maximum average observed level had dissipated, i.e. the observed DT_{90}. For example, the maximum average 3-PBA level at the California site was observed on 15 DA1A (8.36 g ha^{-1}). The average level dropped to ≤ 10% of the maximum by 20 DA1A (0.74 g ha^{-1}). Therefore, 90% dissipation occurred within 20 – 15 = 5 days.

The average values of n = 3 replicates was used in the calculations for each site.

III. CONCLUSION

The major route of dissipation of alpha-cypermethrin under terrestrial field conditions in the bare soil was transformation by aerobic soil metabolism processes. Expected transformation products 3-PBA, cis-DCVA, and trans-DCVA were observed at levels up to 10% of the nominal 84 g a.s. ha^{-1} total application rate.

First-order (SFO) kinetic fits were selected to represent the dissipation of the sum of isomers at the California, Louisiana, and Oklahoma sites, while the biphasic FOMC model was selected for the New York site. Dissipation of the sum of isomers was rapid at all four sites. The $DisT_{50}$ values ranged from 3.4 days to 6.3 days with a median of 5.0 days, an average of 4.9 days, and a standard deviation of 1.4 days. The $DisT_{90}$ values ranged from 13.4 to 27.9 days with a median of 20.3 days, an average of 20.5 days, and a standard deviation of 5.9 days. These results were similar to those previously reported for cypermethrin at a Louisiana and California sites where the $DisT_{50}$ values ranged from 5 to 13 days following application of much higher rates (672 g a.s. ha^{-1} total).

Metabolites 3-PBA, cis-DCVA, and trans-DCVA were observed at all four sites. The maximum average observed level of 3-PBA ranged from 2.5 to 10.0% with an average of 6.0%. The observed $DisT_{90}$ of 3-PBA ranged from 5 to 37 days with an average of 23 days. The maximum average observed level of cis-DCVA ranged from 1.5 to 2.9% with an average of 2.1%. The observed $DisT_{90}$ of cis-DCVA ranged from 9 to 60 days with an average of 24 days. The maximum average observed level of trans-DCVA ranged from 0.2 to 1.7% with an average of 0.9%. The observed $DisT_{90}$ of trans-DCVA ranged from 1 to 9 days with an average of 6 days. The maximum average observed level of the sum of DCVA isomers ranged from 1.7 to 3.8% with an average of 3.0%. The observed $DisT_{90}$ of the DCVA isomer sum ranged from 9 to 60 days with an average of 24 days. These data indicate that the metabolites of alpha-cypermethrin were not persistent with 90% dissipation from peak average observed levels occurring within approximately one month.

Site hydrologic conditions were generally favorable for leaching. However, under these favorable conditions, mean residues of alpha-cypermethrin and the metabolites were not detected in soil (LOD = 0.0002 mg kg^{-1}) below the 2-4 inch (~ 5-10 cm) soil depth at any of the four locations. Neither alpha-cypermethrin nor degradation products appeared to be inherently susceptible to leaching.

Calculation of maximum occurrences of DCVA and 3-PBA from the US field dissipation study

The metabolites DCVA and 3-PBA occurred in relevant amounts in the US field dissipation study with alpha-cypermethrin [CA 7.1.2.2.1/2, *BASF DocID 2013/*7002604]. In the following, the derivation of the maximum occurrences of the metabolites in the US field dissipation study is described.

For the US study, the residue data in g p.e. ha^{-1} (p.e. = parent equivalents) given in the kinetic evaluation report (Appendix 2 of the study) were used considering the sum of isomers of alpha-cypermethrin and DCVA.

Alpha-cypermethrin was applied at three consecutive applications with a target interval of 7 days. Maximum occurrences were calculated for all trials and for each individual application event, considering the maximum amount of metabolite residues (mean of three plots) after an individual application and the respective maximum amount of parent residues (mean of three plots). The results are summarized in Table 7.1.2.2.1-21.

Table 7.1.2.2.1-21: Maximum occurrence of alpha-cypermethrin metabolites in US field dissipation study

Metabolite	Trial	Application number	Maximum amount of metabolite [g p.e. ha^{-1}] [a]	Maximum amount of parent [g ha^{-1}] [a,b]	Maximum occurrence [%]
DCVA [b]	Louisiana	1	1.17	39.25	3.0
		2	1.49	37.93	3.9
		3	2.24	61.98	3.6
	Oklahoma	1	1.07	30.60	3.5
		2	1.26	35.24	3.6
		3	3.09	45.35	6.8
	New York	1	0.99	23.23	4.3
		2	1.47	29.73	4.9
		3	1.27	36.70	3.5
	California	1	1.77	27.84	6.4
		2	1.93	30.07	6.4
		3	3.19	29.20	10.9
3-PBA	Louisiana	1	5.70	39.25	14.5
		2	4.93	37.93	13.0
		3	3.08	61.98	5.0
	Oklahoma	1	1.48	30.60	4.8
		2	2.18	35.24	6.2
		3	4.00	45.35	8.8
	New York	1	1.74	23.23	7.5
		2	2.11	29.73	7.1
		3	1.43	36.70	3.9
	California	1	2.36	27.84	8.5
		2	2.17	30.07	7.2
		3	8.36	29.20	28.6

p.e. = Parent equivalents
[a] Mean of three plots
[b] Sum of cis and trans DCVA

Report: CA 7.1.2.2.1/3
Hoogeweg G., 2014a
Similarity of Four United States Alpha-Cypermethrin Terrestrial Field Dissipation Trial Sites to European Conditions: A Crosswalk Exercise Using ENASGIPS v2.3
2014/7003560
Guidelines: EPA OPPTS 835.6100 - supplemental
GLP: no

Executive Summary

The objective of the project was to conduct an ecoregion comparison and GIS Crosswalk for four sites of an US terrestrial field dissipation (TFD) study [*CA 7.1.2.2.1/2, BASF DocID 2013/7002604*] and to determine which locations in Europe, if any, have similar conditions. These four sites are located in California, Louisiana, New York, and Oklahoma. All work was conducted using the Organisation for Economic Co-operation and Development (OECD) Europe – North America Soil Geographic Information for Pesticide Studies (ENASGIPS) v2.3 application. Ecoregion similarity and GIS crosswalks were conducted in support of (re)registration of alpha-cypermethrin in Europe.

ENASGIPS is a component of an OECD project titled "Harmonized International Guidance for Pesticide Terrestrial Field Dissipation Studies and Crosswalk of North American and European Eco-regions". Its goal is to maximize the use of pesticide field dissipation studies by developing harmonized international guidance for conducting the studies and identifying comparable North American and European ecoregions. The underlying premise of ENASGIPS is that the field dissipation behavior of a pesticide in a region depends primarily on environmental factors, such as soils and climate. If these environmental factors are similar between regions, then field dissipation of a pesticide is also expected to be similar in those regions.

ENASGIPS compares ecoregions based on five parameters: mean annual temperature, mean annual precipitation, mean soil pH, mean soil organic carbon, and soil texture. Depending on the properties of the pesticide of interest, not all parameters may be important for determining field dissipation and a subset of these five parameters may be used to refine the ecoregion comparison. When all five parameters are used the comparison is termed "holistic". When a subset of the parameters is used the comparison is termed "weights of evidence". With either comparison approach, ecoregions are considered similar when their weighted similarity scores are 80% or higher.

Both holistic and weights of evidence ecoregion comparisons were made for the US alpha-cypermethrin terrestrial field dissipation sites using ENASGIPS. Since alpha-cypermethrin is a synthetic pyrethroid insecticide, and pyrethroids are well known to be highly sorbed to soil solids (i.e. limited leaching potential), precipitation was considered to be of low importance for explaining the field dissipation behavior of alpha-cypermethrin. Therefore, precipitation was not included in the ecoregion comparison for the weights of evidence assessment.

Results from the holistic similarity assessment show that matching ecoregions exceeding the 80% similarity level exist in Europe for the California, New York, and Oklahoma study sites. No matching ecoregions were found for the Louisiana study site using the 80% similarity criteria in the holistic assessment.

Results from the weights of evidence similarity assessment show that for each of the four US TFD study sites 4 to 13 similar ecoregions exceeding the 80% similarity level exist in Europe.

In addition to the holistic and weights of evidence ecoregion level comparisons, a GIS crosswalk exercise was also conducted using the ENASGIPS site selection tool. Ranges of weather and soil properties were derived from the US alpha-cypermethrin terrestrial field dissipation study and other data sources. Areas in Europe that fall within the defined ranges were then identified using both the holistic approach (i.e. all five variables included) and a weight of evidence approach (i.e. precipitation excluded).

The GIS crosswalk based on all five variables indicated that California and Louisiana did not have any matching sites. For New York over 48,902,000 ha and for Oklahoma over 41,381,000 ha of matching regions were found. When areas within matching ecoregions were calculated, the values were 43,575,847 ha and 17,828,812 ha for New York and Oklahoma, respectively.

The GIS crosswalk based on the weights of evidence (precipitation not included) determined total areas of over 3,008,860 ha for California, 7,047,082 ha for Louisiana, 69,076,085 ha for New York and 20,866,423 ha for Oklahoma within matching ecoregions.

Based on the weights of evidence approach it is shown that all four US alpha-cypermethrin terrestrial field dissipation trial sites have similar ecoregions and site areas in Europe. Therefore, the results of the US terrestrial field dissipation study of alpha-cypermethrin are applicable to Europe.

I. MATERIAL AND METHODS

Site specific data from four sites of a US terrestrial field dissipation study [*CA 7.1.2.2.1/2, BASF DocID 2013/7002604*] were used in the ENASGIPS and GIS Crosswalk. The four sites were located in California, Louisiana, New York, and Oklahoma. A study summary is presented in section CA 7.1.2.2.1 Field data collected for the soils were provided at 0.15 m (6 inch) intervals. ENASGIPS topsoil data are for the 0 – 0.3 m depth. Therefore, the depth weighted average of the 0 - 0.15 m and 0.15 - 0.30 m samples were calculated. A summary of the site topsoil physical properties is provided in (Table 7.1.2.2.1-22). Full details on the site characterization can be found in the original TFD study report.

Table 7.1.2.2.1-22: Summary of the environmental conditions at the four terrestrial dissipation sites

	California	Louisiana	New York	Oklahoma
Weather				
Used NOAA Weather Station [a]	Porterville	Grand Coteau	Sodus Center	Geary
Average Temperature [ºC] study period [b]	15.6	19.1	7.9	14.9
Total Precipitation [mm] study period	106.7	1292.9	662.9	459.7
Topsoil Properties (0-0.3 m, Depth weighted average)				
Soil Series	Nord	Dundee	Oakville	Pond Creek
Texture (USDA)	Sandy loam	Silt clay loam	Sand	Sandy loam
Slope [%]	0 - 2	0 - 1	0 - 6	1 - 3
Organic matter content [%]	0.9	1.5	1.4	0.9
Soil pH	7.2	6.3	6.6	6.0

[a] Nearby NOAA weather stations were used to determine the needed climate data, because the field data did not cover a 12-month period to calculate annual averages.
[b] Study periods were 17-Jul-2012 through 26-Apr-2013 (283 days) for California, 26-Jun-2012 through 08-Apr-2013 (286 days) for Louisiana, 10-Jul-2012 through 20-Apr-2013 (284) for New York, and 27-Jun-2012 through 06-Apr-2013 (283 days) for Oklahoma. These periods represent the time from the first application through the last field sampling analyzed for alpha-cypermethrin residues.

ENASGIPS Application

ENASGIPS is a component of an OECD project to maximize the use of pesticide field dissipation studies by developing harmonized international guidance for conducting the studies and identifying comparable North American and European ecoregions [*OECD (2012) Report of the OECD Workshop on the Development of Harmonized International Guidance for Pesticide Terrestrial Field Dissipation Studies and Crosswalk of North American & European Eco-regions. Series on Pesticides. No. 68. ENV/JM/MOM(2012)11. Available at http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?cote=env/jm/mono(2012)11&doclanguage=en. Last accessed Dec 12, 2014*]. The underlying premise of ENASGIPS is that the field dissipation behavior of a pesticide in a region depends on environmental factors, such as soils and climate. If these environmental factors are similar between regions, then field dissipation of a pesticide is also expected to be similar in those regions.

ENASGIPS consist of five individual components including:

- A tool for identifying ecoregions with similar soil and climate data
- A tool for site selection based on user defined soil, climate and crop criteria
- A tool for displaying crop density (percentage of land covered by crop) for Canada, United States and the European Union
- A tool for entering user site coordinates
- A geographic database of soil, climate and crop information

A main component of ENASGIPS is the Ecoregion Similarity Modelling (ESM). ESM's goal is to compare and identify similar ecoregions between North America and the European Union. The underlying assumption of ENASGIPS is that site specific characteristics of soil and climate play a major role in determining the behavior and fate of pesticides introduced into the environment. Principle soil factors that influence the pesticide behavior to a large extent are pH, organic carbon, and texture, although other properties such as cation exchange capacity (CEC) play a role. Climatic parameters include precipitation and temperature. A pesticide is expected to behave similarly in ecoregions that have similar soils and climate.

Depending on the properties of the pesticide of interest, not all parameters may be important for determining field dissipation and a subset of these five parameters may be used to refine the ecoregion comparison. When all five parameters are used the comparison is termed "holistic". When a subset of the parameters is used the comparison is termed "weights of evidence". With either comparison approach, ecoregions are considered similar when their weighted similarity scores are 80% or higher.

Holistic Assessment

The first step in the holistic ecoregions similarity assessment was to determine the ecoregions (henceforth referred to as root ecoregions) that overlap the four US TFD test sites. Next, for each root ecoregion the holistic ecoregions crosswalk tool was executed with a target similarity level of 80%. Eighty percent is the proposed default similarity score for use with the ENASGIPS tool as specified in the draft OECD harmonization project [*OECD (2012)*].

Weights of Evidence Assessment

The weights of evidence assessment was conducted, using the root ecoregions determined in the holistic approach. Based on the conceptual model of cypermethrins field dissipation, hypothesizing that cypermethrins remain sorbed in the topsoil compartment where biotic processes convert it to non-persistent products, the weights of evidence approach used soil texture, topsoil organic matter, pH, and temperature to determine which ecoregions in Europe matched the four root ecoregions, excluding precipitation. The target similarity level was 80%.

GIS Crosswalk

In addition to the holistic and weights of evidence ecoregion similarity assessments, ENASGIPS's Site Selection tool was used in a GIS crosswalk. Using the key parameters identified in the cypermethrins conceptual model sites in Europe were selected that adhere to specific value ranges based on the US field site characteristics. Parameters included in the weights of evidence site selection were soil texture, topsoil organic matter, pH, and temperature. A separate, holistic assessment also included precipitation (although not a major factor according to the conceptual model) to provide a complete overview of areas in Europe that are similar. Value ranges for these parameters were chosen based on values recorded at the field sites, the US Department of Agriculture Natural Resources Conservation Service (USDA NRCS) Soil Survey Geographic Database [*SSURGO: SOIL SURVEY STAFF (2013): Gridded Soil Survey Geographic (gSSURGO) Database for the Conterminous United States, United States Department of Agriculture, Natural Resources Conservation Service, available online at http://datagateway.nrcs.usda.gov/, November 2013*] and NOAA long-term annual climatic data. Table 7.1.2.2.1-23 lists the selection criteria used.

Table 7.1.2.2.1-23: Selection criteria used in the site assessment

	California		**Louisiana**		**New York**		**Oklahoma**	
Weather								
	Min	Max	Min	Max	Min	Max	Min	Max
2012-2013 Average Annual Temperature [ºC]	16.5		20.7		9.8		14.9 [a]	
Average Annual Temperature [ºC] [b]	16.1	16.9	15.0	26.4	8.8	10.9	12.5	17.3
Total Precipitation [mm] [b]	72.5	242.7	1358.1	2036.9	726.7	1084.6	201.1	916.1
Topsoil Properties								
Texture [FAO]	Coarse		Medium		Coarse		Coarse	
Organic matter content [%]	0.9	4.0	0.5	2.0	0.5	6.0	0.9	3.0
Organic carbon content [%]	0.5	2.3	0.3	1.2	0.3	3.5	0.5	1.7
Soil pH	6.6	9.0	3.5	8.4	4.5	7.3	5.1	7.4

[a] Based on the study period. NOAA does not have complete data coverage for 2012-2013 for Geary, OK

[b] Range determined by calculating the standard deviation of the annual average data for 2001-2013 and adding or subtracting one standard deviation from the 2012-2013 average.

II. RESULTS AND DISCUSSION

Ecoregion Similarity Assessment

The first step in the ecoregion similarity assessment was to determine the root ecoregions for each of the four US terrestrial field dissipation sites located in the states of California, Louisiana, New York, and Oklahoma.

As a result, field dissipation study sites were located in the "California Central Valley grasslands" ecoregion (California), in the "Mississippi Lowland forests" ecoregion (Louisiana), the "Southern Great Lakes forest" ecoregion (New York) and in the "Central and Southern mixed grasslands" ecoregion (Oklahoma). Detailed characterizations of the ecoregions are given in the study report.

Holistic Similarity

The holistic ecoregions crosswalk resulted in 14 ecoregions in Europe that have similar conditions to the root ecoregion containing the California field dissipation trial site. Similar ecoregions are located in Portugal, Spain, Southern France, Italy, Greece, Romania, and Hungary. The similarity score ranged from 81 to 100%.

The holistic ecoregions crosswalk for the Louisiana site resulted in no ecoregions in Europe that have similar conditions based on the 80% cutoff, but has 5 matching ecoregions with a 65% or greater similarity score. The similarity scores ranged from 66 to 68%.

The holistic ecoregions crosswalk for the New York site resulted in 10 ecoregions in Europe that have similar conditions. Similar ecoregions are located in France, Belgium, Netherlands, and Germany and are more representative of the coastal temperate climates. The similarity score ranged from 80 to 92%.

The holistic ecoregions crosswalk for the Oklahoma study site resulted in 7 ecoregions in Europe that have similar conditions. Similar ecoregions are located in Italy, Greece, and Romania. The similarity score ranged from 81 to 91%.

Results from the holistic similarity assessment show that matching ecoregions exist in Europe for the California, New York and Oklahoma study sites. The total area of matching ecoregions in Europe ranged from 40,710,000 ha to 158,117,000 ha. However, no matching ecoregions were found for the Louisiana study site using the 80% similarity criteria and including all five similarity parameters in the holistic assessment.

Weights of Evidence Similarity Assessment

In the weights of evidence similarity assessment soil texture, organic matter, pH, and annual average temperature were considered. These are the primary factors affecting cypermethrins field dissipation according to the conceptual model. Precipitation was excluded from the weights of evidence assessment as previously discussed.

The weights of evidence crosswalk for the California study site resulted in 10 ecoregions in Europe that are similar. Matching ecoregions are location in southern Portugal, Spain, Italy, and Greece. The similarity scores ranged from 81 to 100%.

The weights of evidence crosswalk for the Louisiana study site resulted in 4 ecoregions in Europe that are similar. Matching ecoregions are located in southern Portugal, Spain and Greece. The similarity scores ranged from 80 to 82%.

The weights of evidence crosswalk for the New York study site resulted in 13 ecoregions in Europe that are similar. Similar ecoregions are located in a wide band stretching from Northern Spain, France to Poland, Hungary and Bulgaria in the east. The similarity scores ranged from 82 to 96%.

The weights of evidence crosswalk for the Oklahoma study site resulted in 11 ecoregions in Europe that are similar. Similar ecoregion are located in the coastal Spain and France, most of Italy and Greece. The similarity scores ranged from 83 to 94%.

Results from the weights of evidence similarity assessment show that for each of the four US TFD study sites, 4 to 13 different similar ecoregions exist in Europe.

GIS Crosswalk

Using the site selection tool a GIS crosswalk was conducted for the four terrestrial field dissipation sites using the holistic approach (all variables) and the weights of evidence approach (i.e. all variables except precipitation).

Results indicate that the total area of matching land is very similar when the holistic and weights of evidence approaches are compared. The GIS crosswalk indicated that California and Louisiana did not have any matching sites using all variables. For New York over 48,902,000 ha and for Oklahoma over 41,381,000 ha of matching regions were found. When areas within matching ecoregions were calculated, the values were to 43,575,847 ha and 17,828,812 ha for New York and Oklahoma, respectively.

The GIS crosswalk based on the weights of evidence determined total areas of 3,008,860 ha for California, 7,047,082 ha for Louisiana, 69,076,085 ha for New York and 20,866,423 ha for Oklahoma within areas in matching ecoregions based on the weights of evidence approach.

Based on the holistic approach, the total area of matching areas within matching ecoregions was 89% for the New York site and 43% for the Oklahoma study sites. No matching areas were found for the California and Louisiana trial sites. Using the weights of evidence approach, 43% to 86% of the areas with matching soil and climate criteria fall within matching ecoregions for all four US terrestrial field dissipation sites.

III. CONCLUSIONS

The objective of the project was to conduct an ecoregion similarity assessment and GIS Crosswalk for four US field dissipation test sites and determine which locations in Europe, if any, have similar conditions. All work was conducted using the Organisation for Economic Co-operation and Development (OECD) Europe – North America Soil Geographic Information for Pesticide Studies (ENASGIPS) application. Ecoregion similarity and GIS crosswalks were conducted in support of (re)registration of alpha-cypermethrin in Europe.

Results from the holistic similarity assessment show the matching ecoregions exceeding the 80% similarity level exist in Europe for the California, New York, and Oklahoma study sites. No matching ecoregions were found for the Louisiana study site using the 80% similarity criteria. In the holistic similarity assessment topsoil texture, topsoil organic matter, topsoil pH, annual average temperature, and average annual total precipitation were considered.

Results from the weights of evidence similarity assessment show that for each of the four US study sites, 4 to 13 similar ecoregions exceeding the 80% similarity level exist in Europe. In the weights of evidence similarity assessment topsoil texture, topsoil organic matter, topsoil pH, and annual average temperature were considered. Precipitation was not included as it was considered unimportant for explaining field dissipation behavior of strongly sorbed cypermethrins such as alpha-cypermethrin, especially since field dissipation test sites are irrigated to supplement natural precipitation.

The GIS crosswalk indicated that California and Louisiana did not have any matching sites using all five variables in a holistic assessment. For New York 43,575,847 ha and for Oklahoma over 17,828,812 ha of matching areas were found within the matching ecoregions.

The GIS crosswalk based on the weights of evidence determined total areas of 3,008,860 ha for California, 7,047,082 ha for Louisiana, 69,076,085 ha for New York, and 20,866,423 ha for Oklahoma within matching ecoregions based on the weights of evidence approach.

Based on the weights of evidence approach it is shown that all four US alpha-cypermethrin terrestrial field dissipation trial sites have similar ecoregions and site areas in Europe. Therefore, results for the US terrestrial field dissipation study of alpha-cypermethrin are applicable to Europe.

Report:	CA 7.1.2.2.1/4 Kellner T., Kröger F., 2014a Field soil dissipation study of BAS 310 I (Alpha-Cypermethrin) in the formulation BAS 310 55 I (ME) on bare soil at six sites in Europe in 2012 - 2013 2014/1161559
Guidelines:	NAFTA Guidance Document for Conducting Terrestrial Field Dissipation Studies Regulatory Directive DIR2006-01 (March 2006), EPA 835.6100, SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995), EFSA Guidance to obtain DegT50 values in soil (2010), SANCO/825/00 rev. 8.1 (16 November 2010), SANCO/3029/99 rev. 4 (11 July 2000), EU (1999): 1607/VI/97 rev. 2
GLP:	yes (certified by Staatliches Gewerbeaufsichtsamt, Hildesheim, Germany)

Report:	CA 7.1.2.2.1/5 Kroeger F., 2016 a Report Amendment No. 1: Field soil dissipation study of BAS 310 I (Alpha-Cypermethrin) in the formulation BAS 310 55 I (ME) on bare soil at six sites in Europe in 2012-2013 2016/1136938
Guidelines:	NAFTA Guidance Document for Conducting Terrestrial Field Dissipation Studies Regulatory Directive DIR2006-01 (March 2006), EPA 835.6100, EEC 1607/VI/97 rev. 2 10.06.1999, SANCO/3029/99 rev. 4 (11 July 2000), SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995), EFSA Guidance to obtain DegT50 values in soil (2010)
GLP:	yes (certified by Staatliches Gewerbeaufsichtsamt, Hildesheim, Germany)

The amendment to the report contains corrections of typos and calculation errors.

Executive Summary

The dissipation of alpha-cypermethrin (BAS 310 I) under field conditions was investigated at six sites in Europe representative of Northern and Southern EU conditions. One trial each was performed in Northern Germany, Southern Germany, United Kingdom, Bulgaria, Spain, and in Southern France. All sites represent typical regions of agricultural practice representative for growing crops including wheat. The trial sites consisted of an untreated and a treated plot, the latter being subdivided into 3 subplots that were assigned for replicates.

The product BAS 310 55 I, formulated as a micro-emulsion (ME), was broadcast applied to bare soil in a single application at a nominal rate of 60 g a.s. ha^{-1} using a target water volume of 400 L ha^{-1}. Applications were conducted between early June and late July 2012 using a calibrated boom sprayer. The actual application rates for each trial determined by quantifying the amount of spray discharged ranged from 56 to 64 g a.s. ha^{-1}, with an average of 61 g a.s. ha^{-1}. Results from spray broth analysis for the individual trial sites revealed concentrations between 89 and 105% of the nominal value with an average of 98% across all sites. Dose verification conducted via application verification samples yielded recovery values for the individual sites ranging from 88 to 130% of the target rate and an average recovery of 109% across all sites.

Immediately after application of the test item, the plots were covered with a layer of sand of at least 3 – 5 mm depth to protect the applied product from surface processes like photolysis or volatilization, and to exclude any potential impact on the degradation of the test item caused by any of these processes. Thickness and homogeneity of the sand cover were controlled and maintained at least until the field received a total precipitation (rain and irrigation) of ≥ 10 mm. Since the sand cover remained intact until then, it was not necessary to renew it. Within the time period of up to 28 days after application, the individual plots received a total precipitation (rain and irrigation) of ≥ 10 mm (Northern Germany: 25.4 mm after 9 days, Southern Germany: 16.8 mm after 3 days, United Kingdom: 12.8 mm after 28 days, Bulgaria: 48.4 mm within 2 days, Spain: 12.7 mm within 14 days and France: 15.8 mm after 1 day).

No tillage or fertilization was performed during the course of the study and no crops were grown throughout any of the trials. The plots were kept generally free of vegetation (< 10% of weeds) via the application of glyphosate.

Rainfall was supplemented with irrigation at sites in Northern Germany (130.7 mm), Southern Germany (57.9 mm), United Kingdom (93.3 mm), Bulgaria (408.5 mm), Spain (195.1 mm) and Southern France (61.5 mm) the total water input was at least 107% of the historical average rainfall during the study period at the trial sites.

Soil residue specimens were taken at intervals up to 368 days after treatment (DAT) and down to a maximum soil depth of 50 cm. Soil cores below 10 cm depth (excepting usually the doubles) were cut into 10 cm segments. Soil segments of the same depth and subplot from a defined sampling, plot, event, and site were pooled and homogenized. A representative sub-sample of each depth was taken for residue analysis. All soil specimens were generally stored at about -18°C within a maximum of 6 hours after end of sampling, except the untreated soil specimen of one sampling from trial Germany North (L120555) and France South (L120560) which were placed into freezer storage at generally -18°C within 7 hours and 8.5 hours after end of sampling, and remained frozen until analysis.

In order to demonstrate stability of the residues in soil during storage and shipment, shipment verification specimens were prepared at selected sampling occasions by fortifying untreated soil from the field sites with a standard solution of alpha-cypermethrin. These specimens were stored and shipped under the same conditions as the actual residue specimens. Analysis of the shipping verification specimens on alpha-cypermethrin yielded average recovery values of 90-131% across all sites confirming residue stability during all storage and shipment procedures.

Soil specimens, as well as application monitors (Petri dishes) and shipping verification specimens were analyzed for cypermethrin isomers cis-1, cis-2, trans-3, and trans-4, and its metabolites 3-PBA and DCVA isomers cis and trans to the BASF analytical method R0034/01.

The analytical method of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4 involved extraction of the soil with 0.1% formic acid in acetonitrile, evaporation to dryness, and reconstitution in 0.1% formic acid in acetonitrile/water (1:1, v/v). The analytical method of 3-PBA and DCVA isomers cis and trans involved two extractions of the soil with acetonitrile/water (70:30, v/v), evaporation to dryness of an aliquot of the extracts and reconstitution in 0.1% formic acid in methanol/water (20:80, v/v).

The final determination of the analytes was performed by HPLC-MS/MS with a limit of quantification (LOQ) of 0.001 mg kg^{-1} for each analyte. Field soil specimens from the treated plot were analyzed down to a depth until at least two consecutive soil segments were free of quantifiable residues (< LOQ). Analysis was performed until a maximum of 368 DAT.

No residues above 30% of the LOQ of any analyte were detected in any of the untreated control samples proving that there were no interferences of the untreated soil material with the analytical procedure used. Procedural recovery experiments performed with untreated soils spiked with the analytes at concentrations of 0.001, 0.01, and 0.5 mg kg^{-1} yielded overall mean recovery rates of 92 and 98% for the individual analytes, confirming the validity of the analytical method used in this study.

Residue values of cypermethrin isomer cis-2 and of the sum of cypermethrin isomers cis-1 and cis-2 in mg kg^{-1} dry were summed up for all depths between 0 and 50 cm analyzed and converted to µg kg^{-1} for an improved readability.

Alpha-cypermethrin degraded under field conditions in soil at all six European field sites. The total amount of the sum of cypermethrin isomers cis-1 and cis-2 residues detected in the soil profiles decreased from an average of 39 µg kg^{-1} at 0 DAT to an average of 8 µg kg^{-1} (range: 0 – 20 µg kg^{-1}) at 60 DAT. Until the end of the study at about 360 DAT, residues decreased further to concentrations equal or below LOQ at four of the six trials, which equals to about 0 µg kg^{-1}. The two trials left with residues above LOQ showed mean rates of 5 µg kg^{-1} (range: 3 – 6 µg kg^{-1}) at the end of the study. DT_{50} values are subject of a separate study and modeling report

Altogether, it can be concluded that cypermethrin isomer did not show any significant tendency to move into deeper soil layers indicating low potential for cypermethrin isomer residues to leach to groundwater in this study.

Residues of the metabolites 3-PBA and DCVA (cis isomer only) above LOQ were only found in the top 0 – 10 cm three out of six field sites (Bulgaria, Spain, and France). Residues above LOQ were exclusively found at trial sites Bulgaria and Spain.

Metabolite DCVA isomer cis was detected in small amounts reaching a maximum of 3.3 µg kg^{-1} (Bulgaria; 14 DAT) and 2.7 µg kg^{-1} (Spain; 60 DAT) on average for the three subplots. Thereafter, residues declined and were no longer detected in amounts > LOQ after 132 days at the latest.

Metabolite 3-PBA was detected above LOQ in small amounts reaching maximum amounts of 2.5 µg kg^{-1} (Bulgaria; 30 DAT) and 3.0 µg kg^{-1} (Spain; 60 DAT) on average for the three subplots. Thereafter, residues declined and were no longer detected in amounts > LOQ after 85 days at the latest.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item (formulation):	BAS 310 55 I
Active substance (a.s.):	Alpha-cypermethrin (BAS 310 I, Reg. No. 4078193)
Chemical name (IUPAC):	Racemate of (S)-α-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-α-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molar mass:	416.3 g mol^{-1}
Batch No.:	101209 /(containing 49.2 g alpha-cypermethrin L^{-1})
Type of formulation:	ME

2. Test sites

The dissipation of alpha-cypermethrin under field conditions was investigated at six sites in Europe representative of Northern and Southern EU conditions. One trial each was performed in Northern Germany, Southern Germany, United Kingdom, Bulgaria, Spain, and in Southern France. The visual homogeneity of the upper soil layer was verified at the beginning of each field phase. The site characteristics are presented in Table 7.1.2.2.1-24 to Table 7.1.2.2.1-26. Soil parameters were determined at Eurofins Institut Jäger GmbH (Tübingen, Germany) from soil samples taken before application from the boundaries of the future treated subplots (SP) following segmentation according to the visually identified soil horizons.

Table 7.1.2.2.1-24: Soil characteristics of the trial sites L120555 and L120556 used to investigate the field dissipation of alpha-cypermethrin (BAS 310 I)

Trial	**S12-01863-01 (L120555)**			**S12-01863-02 (L120556)**		
Location	**Dollern, Germany (North)**			**Ölbronn-Dürrn, Germany (South)**		
Soil properties	0 - 30 cm	30 - 53 cm	53 - 90 cm	0 - 30 cm	30 – 60 cm	60 – 90 cm
Soil texture (DIN19683)	Sandy loam	Sandy loam	Sandy loam	Clay silt	Silty clay	Silty clay
Coarse sand [%]	4.0	3.9	4.5	0.73	0.43	0.24
Middle sand [%]	30.2	29.6	33.2	0.90	0.76	0.42
Fine sand [%]	25.4	28.1	29.0	0.63	0.56	0.40
Finest sand [%]	10.3	11.4	9.5	1.1	0.98	0.91
Coarse silt [%]	14.2	10.9	7.1	37.5	31.0	28.7
Middle silt [%]	5.6	5.7	2.7	27.7	25.7	25.3
Fine silt [%]	2.8	3.1	2.9	9.5	13.5	12.7
Clay [%]	7.6	7.5	11.2	22.1	27.1	31.4
Soil texture (USDA)	Sandy loam	Sandy loam	Sandy loam	Silt loam	Silt loam	Silty clay loam
sand [%]	74.0	77.3	79.6	5.0	4.3	3.2
silt [%]	18.1	14.8	8.3	73.1	68.9	65.8
clay [%]	8.0	8.0	12.2	22.0	26.8	31.1
Dry matter [%]	99.3	99.6	99.4	99.3	99.0	98.8
Total Carbon [%]	1.7	<0.3	<0.3	1.1	0.43	< 0.3
Total organic C [%]	1.7	<0.3	<0.3	1.1	0.42	<0.3
Organic matter [%]*	2.9	<0.5	<0.5	1.9	0.72	<0.5
pH [$CaCl_2$]	5.71	5.50	4.16	6.01	6.27	6.34
pH [H_2O]	5.54	5.46	4.63	5.40	5.71	5.65
CEC [meq 100 g^{-1}]	7.7	2.6	3.0	11.0	11.3	13.6
	SP1***	SP2***	SP3***	SP1***	SP2***	SP3***
MWHC [g 100 g^{-1} dry weight]	34.8	35.9	35.8	42.1	37.9	42.1
pF 2.0 [g 100 g^{-1} dry weight]**	15.3	15.7	15.8	28.3	28.7	30.9
pF 2.5 [g 100 g^{-1} dry weight]**	12.1	11.9	11.9	24.2	24.7	25.3
Dry bulk density [g cm^{-3}]	1.30	1.31	1.32	1.20	1.25	1.28

CEC = Cation exchange capacity
MWHC = Maximum water holding capacity
SP = Subplot
* Organic matter = Organic carbon x 1.724 (organic matter = C_{org})
** Water retention characteristics, soil moisture at 0.1 or 0.33 bar
*** Samples taken at 5-15 cm depth

Table 7.1.2.2.1-25: Soil characteristics of the trial sites L120557 and L120558 used to investigate the field dissipation of alpha-cypermethrin (BAS 310 I)

Trial	**S12-01863-03 (L120557)**			**S12-01863-04 (L120558)**		
Location	**Melbourne, United Kingdom**			**Letniza, Bulgaria**		
Soil properties	0 - 30 cm	30 - 90 cm		0 - 30 cm	30 – 60 cm	60 – 90 cm
Soil texture (DIN19683)	Sandy loam	Sandy loam		Silty clay	Silty clay	Silty clay
Coarse sand [%]	1.2	2.0		0.8	0.16	0.12
Middle sand [%]	12.6	17.6		1.6	0.41	0.31
Fine sand [%]	9.9	11.0		0.87	0.41	0.38
Finest sand [%]	6.9	6.7		1.7	1.5	1.6
Coarse silt [%]	22.6	20.7		28.1	26.4	25.5
Middle silt [%]	14.4	13.0		16.3	19.1	21.3
Fine silt [%]	8.5	8.5		12.9	9.0	10.0
Clay [%]	24.0	20.7		38.0	43.1	40.9
Soil texture (USDA)	Loam	Loam		Silty clay loam	Silty clay	Silty clay
sand [%]	35.2	41.7		8.0	4.9	4.8
silt [%]	40.4	35.5		54.2	53.4	53.9
clay [%]	24.5	22.8		37.8	41.5	41.4
Dry matter [%]	99.0	99.1		99.3	99.5	99.5
Total Carbon [%]	1.7	0.71		2.6	1.6	0.81
Total organic C [%]	1.6	<0.3		2.4	1.0	0.74
Organic matter [%]*	2.8	<0.5		4.1	1.7	1.3
pH [$CaCl_2$]	6.74	6.94		6.96	7.13	7.28
pH [H_2O]	6.30	7.14		6.88	6.93	7.03
CEC [meq 100 g^{-1}]	12.1	8.1		25.3	24.5	24.5
	SP1***	SP2***	SP3***	SP1***	SP2***	SP3***
MWHC [g 100 g^{-1} dry weight]	31.7	27.8	24.9	32.0	43.6	45.8
pF 2.0 [g 100 g^{-1} dry weight]**	22.2	20.4	19.1	27.2	29.1	33.4
pF 2.5 [g 100 g^{-1} dry weight]**	20.7	19.1	18.2	25.3	26.2	27.2
Dry bulk density [g cm^{-3}]	1.30	1.34	1.57	1.36	1.13	1.03

CEC = Cation exchange capacity
MWHC = Maximum water holding capacity
SP = Subplot
* Organic matter = Organic carbon x 1.724 (organic matter = Corg)
** Water retention characteristics, soil moisture at 0.1 or 0.33 bar
*** Samples taken at 5-15 cm depth

Table 7.1.2.2.1-26: Soil characteristics of the trial sites L120559 and L120560 used to investigate the field dissipation of alpha-cypermethrin (BAS 310 I)

Trial	**S12-01863-05 (L120559)**				**S12-01863-06 (L120560)**		
Location	**Almansa, Spain**				**Barry d'Islemade, France (South)**		
Soil properties	0 - 10 cm	10 - 40 cm	40 – 60 cm	60 –90 cm	0 - 30cm	30 - 60cm	60 - 90cm
Soil texture (DIN19683)	Sandy loam	Sandy loam	Sandy loam	Sandy loam	sandy loam	sandy clay loam	sandy clay loam
Coarse sand [%]	15.0	16.5	14.3	14.4	1.5	1.9	1.6
Middle sand [%]	28.3	28.6	29.0	23.1	25.2	24.9	31.0
Fine sand [%]	14.2	13.8	15.4	12.6	14.2	11.9	12.6
Finest sand [%]	5.0	4.6	5.3	6.4	6.9	4.9	4.6
Coarse silt [%]	7.1	7.0	5.3	4.4	12.4	10.8	8.2
Middle silt [%]	6.6	5.9	4.9	7.1	10.7	10.1	7.2
Fine silt [%]	6.2	5.8	6.3	9.8	9.1	7.1	6.1
Clay [%]	17.8	17.8	19.7	22.4	20.1	28.5	28.8
Soil texture (USDA)	Sandy loam	Sandy loam	Sandy loam	Sandy clay loam	Sandy clay loam	Sandy clay loam	Sandy clay loam
sand [%]	65.5	66.9	69.9	59.1	51.0	46.7	53.3
silt [%]	15.5	15.5	11.1	18.5	27.5	23.9	17.6
clay [%]	19.0	17.6	19.0	22.4	21.5	29.4	29.2
Dry matter [%]	99.3	99.3	99.4	99.4	99.4	98.9	98.9
Total Carbon [%]	5.4	4.9	5.7	8.4	0.56	<0.3	<0.3
Total organic C [%]	1.4	1.0	0.85	0.82	0.52	<0.3	<0.3
Organic matter [%]*	2.4	1.7	1.5	1.4	0.90	<0.5	<0.5
pH [$CaCl_2$]	7.47	7.63	7.66	7.64	5.39	5.82	6.05
pH [H_2O]	7.64	7.78	7.82	7.56	4.95	5.32	5.64
CEC [meq 100 g^{-1}]	9.9	8.7	8.4	11.3	7.3	10.3	11.0
	SP1***	SP2***	SP3***		SP1***	SP2***	SP3***
MWHC [g 100 g^{-1} dry weight]	28.8	29.0	30.6		29.2	27.2	25.8
pF 2.0 [g 100 g^{-1} dry weight]**	12.6	11.9	13.1		15.6	16.1	16.5
pF 2.5 [g 100 g^{-1} dry weight]**	10.0	9.7	10.5		12.7	14.0	15.0
Dry bulk density [g cm^{-3}]	1.40	1.43	1.27		1.46	1.91	1.83

CEC = Cation exchange capacity
MWHC = Maximum water holding capacity
SP = Subplot
* Organic matter = Organic carbon x 1.724 (organic matter = Corg)
** Water retention characteristics, soil moisture at 0.1 or 0.33 bar
*** Samples taken at 5-15 cm depth

The selected field sites represented typical regions of agricultural practice with soils representative for growing wheat that had been under cultivation for several years. The sites were generally flat without any significant slope except the site in Spain, which was flat with a slope of approximately 2.6%. The field sites have been without any pasture, trees, or vine since 2009. Before commencement of the first sampling, the soil at each trial site was prepared as fine and firm seedbed and rolled if considered necessary, but then was left fallow.

No product containing the active substance of the test item had been used on the test plots in the last three years.

B. STUDY DESIGN

1. Experimental treatments

The trial area at each site was divided into two plots, one untreated control plot (U1: size: 30 – 90 m^2) and one treated plot (treatment 2: size: 288 – 414 m^2). The treated plot consisted of three equal sized subplots SP1, SP2, and SP3 that were assigned for replicates.

The product, formulated as microemulsion (ME), was broadcast applied to bare soil in a single application at a nominal rate of 60 g a.s. ha^{-1} using a target water volume of 400 L ha^{-1}. Applications were conducted between early June and late July 2012 using calibrated boom sprayers. Treated subplots were three-fold replicated with subplot size ranging from 96 to 138 m^2. For each treated replicate, a separate spray mixture was prepared and the test item was applied to each subplot individually. Each spray mixture was visually checked for homogeneity and small aliquots of the spray mixture were taken before and after application of each individual subplot for later analysis.

The actual application rates determined by quantifying the amount of spray discharged ranged from 59 to 63 g a.s. ha^{-1} averaged over the three replicates of each treated plot. In addition, the dose was verified by means of sampling Petri dishes filled with fine untreated soil from the trial site (approximately 50 g per dish, sieved to 3.2 – 1 mm). The Petri dishes with an inner diameter of 10.8 cm were placed on the treated plot (ten in each subplot) before application. On completion of the application, the Petri dishes were closed with a lid, sealed with adhesive tape, stored chilled after collection, and placed in freezer storage within 5 hours after application. Further details of application are presented in Table 7.1.2.2.1-27 below.

Table 7.1.2.2.1-27: Application rates of plots treated with BAS 310 55 I (ME)

Trial Country	Application Method	No. of Applications	Subplot (m^2)	Application rate per treated subplot				Application date
				nominal [g a.s. ha^{-1}]	actual* [g a.s. ha^{-1}]	Dose verification** [g a.s. ha^{-1}]	Dose verification** % of nominal	
S12-01863-01 (L120555) Germany (North)	broadcast spray to bare soil	1	SP1 (96)	60	56	59	98	07 Jun 2012
			SP2 (96)	60	62	60	100	
			SP3 (96)	60	61	62	103	
			Average	**60**	**60**	**60**	**100**	
S12-01863-02 (L120556) Germany (South)	broadcast spray to bare soil	1	SP1 (96)	60	58	76	127	19 Jun 2012
			SP2 (96)	60	59	76	127	
			SP3 (96)	60	59	78	130	
			Average	**60**	**59**	**76**	**128**	
S12-01863-03 (L120557) UK	broadcast spray to bare soil	1	SP1 (96)	60	63	66	110	26 Jul 2012
			SP2 (96)	60	62	71	118	
			SP3 (96)	60	64	74	123	
			Average	**60**	**63**	**70**	**117**	
S12-01863-4 (L120558) Bulgaria	broadcast spray to bare soil	1	SP1 (96)	60	61	58	97	21 Jun 2012
			SP2 (96)	60	62	61	102	
			SP3 (96)	60	63	59	98	
			Average	**60**	**62**	**59**	**99**	
S12-01863-05 (L120559) Spain	broadcast spray to bare soil	1	SP1 (99)	60	61	53	88	14 Jun 2012
			SP2 (99)	60	63	61	102	
			SP3 (99)	60	62	78	130	
			Average	**60**	**62**	**64**	**106**	
S12-01863-06 (L120560) France (South)	broadcast spray to bare soil	1	SP1 (138)	60	64	61	102	07 Jun 2012
			SP2 (138)	60	61	60	100	
			SP3 (138)	60	61	60	100	
			Average	**60**	**62**	**60**	**101**	

* Determined by calculation of the applied spray volume on the plot based on analysed a.s. density and content
** Determined by means of 10 Petri dishes filled with soil

Immediately after application of the test item and before subsequent residue soil core sampling, the control plot and the treated replicates were covered with a thin layer of sand to protect the applied product from surface processes like photolysis or volatilization, and to exclude any potential impact on the degradation of the test item caused by any of these processes. The application of sand was conducted manually or using spades until complete coverage of the soil surface was reached. Fine or medium grained sand was used. The thickness of the sand layer necessary for complete coverage of the soil was at least around 3 – 5 mm (range around: 3 - 15 mm).

Thickness and homogeneity of the sand cover were controlled and maintained at least until the field received a total precipitation (rain and irrigation) of ≥ 10 mm. Since the sand cover remained intact until then, it was not necessary to renew it. Within the time period of up to 28 days after application, the individual fields received a total precipitation (rain and irrigation) of ≥ 10 mm (Germany (North): 16.2 mm within 9 days, Germany (South): 16.8 mm within 3 days, United Kingdom: 12.8 mm within 28 days, Bulgaria: 48.4 mm within 2 days, Spain: 12.7 mm within 14 days, and France: 15.8 mm after 1 day).

No tillage or fertilization was performed during the course of the study from first to last sampling and no crops were grown throughout any of the trials. The plots were kept generally free of weeds via the application of glyphosate.

Rainfall was supplemented with irrigation at sites in Germany (North; 130.7 mm), Germany (South; 57.9 mm), United Kingdom (93.3 mm), Bulgaria (408.0 mm), Spain (170.4 mm), and France (South; 49.6 mm).

Actual weather data are based on records of appropriate weather stations located on-site. Monthly summary results on temperature, precipitation and irrigation are presented in Table 7.1.2.2.1-28.

Table 7.1.2.2.1-28: Summary of climatic conditions at field trial sites used to investigate the dissipation of alpha-cypermethrin

Trial	S12-01863-01 (L120555)			S12-01863-02 (L120556)			S12-01863-03 (L120557)		
Location	Dollern			Ölbronn-Dürrn			Melbourne		
	Germany (North)			Germany (South)			United Kingdom		
Climatic conditions	T_{mean} Air [°C]	Prec. [mm]	Irrigation (treated subplots) [mm]	T_{mean} Air [°C]	Prec. [mm]	Irrigation (treated subplots)	T_{mean} Air [°C]	Prec. [mm]	Irrigation (treated subplots) [mm]
Month		Σ	Σ		Σ	Σ		Σ	Σ
Jun 2012	15.4	82.6	0.0	19.1	38.2	0	-	-	-
Jul 2012	16.8	88.0	0.0	18.2	105.0	0	15.3	0	0.0
Aug 2012	17.8	35.4	25.8	19.7	23.6	38.4	16.2	23.6	0.0
Sep 2012	13.5	14.0	12.3	14.3	70.0	19.5	12.7	24.8	0.0
Oct 2012	9.3	63.0	7.5	9.2	55.0	0.0	9.3	62.6	0.0
Nov 2012	5.6	41.2	23.0	5.9	107.0	0.0	6.6	106.4	0.0
Dec 2012	1.7	63.8	0.0	2.9	89.8	0.0	4.9	134.2	0.0
Jan 2013	0.9	87.2	0.0	1.2	16.2	0.0	3.7	36.6	0.0
Feb 2013	0.3	31.6	0.0	-0.6	46.2	0.0	3	31.8	0.0
Mar 2013	-0.5	3.2	0.0	2.2	25.8	0.0	2.4	60.8	0.0
Apr 2013	7.3	29.2	31.7	9.1	50.6	0.0	7.6	12	39.7
May 2013	12	165.2	9.5	11.3	158.8	0.0	10.7	76.8	0.0
Jun 2013	14.1	1.8	0.0	17.3	82.8	0.0	14	54	30.5
Jul 2013	-	-	-	-	-	-	18.2	27.8	23.1

Trial	S12-01863-04 (L120558)			S12-01863-05 (L120559)			S12-01863-06 (L120560)		
Location	Letniza			Almansa			Barry d' Islemade		
	Bulgaria			Spain			France (South)		
Climatic conditions	T_{mean} Air [°C]	Prec. [mm]	Irrigation (treated subplots) [mm]	T_{mean} Air [°C]	Prec. [mm]	Irrigation [mm]	T_{mean} Air [°C]	Prec. [mm]	Irrigation (treated subplots) [mm]
Month		Σ	Σ		Σ	Σ		Σ	Σ
Jun 2012	24.1	90.4	-	25	3.1	9.6	20.2	64.0	0.0
Jul 2012	26.6	13.2	71.5	24.6	2.4	4.9	20.4	53.2	11.9
Aug 2012	24.6	78.4	22.0	26.6	1.5	11.5	22.7	86.2	0.0
Sep 2012	19.8	22.8	69.0	20.3	38.6	47.5	18.2	19.6	19.3
Oct 2012	14.2	52.4	89.0	16.1	55.6	13.1	14.5	35.2	18.4
Nov 2012	7.4	11.8	11.5	10.6	215.7	0.0	8.9	65.0	0.0
Dec 2012	-1.7	56.6	9.5	7.3	23.9	8.7	6.5	104.0	0.0
Jan 2013	-1.4	20.4	8.0	7.5	13.1	10.2	4.8	152.2	0.0
Feb 2013	3.9	45.2	16.0	7.1	64.6	15.1	4.5	69.0	0.0
Mar 2013	6.3	39.8	39.0	10.3	61.9	0.0	8.9	169.6	0.0
Apr 2013	13.8	55.8	0.0	12.4	79.7	20.8	11.6	77.4	0.0
May 2013	19.1	95.0	56.0	14.8	5.9	29.0	12.9	142.0	0.0
Jun 2013	19.0	62.8	16.5	16.4	0.0	0.0	16.9	13.0	0.0

Weather data refer to time period from start of trial (day of application) until end of trial (day of last sampling).

Historical (long-term) weather data on precipitation and average air temperature from at least 10 years (exception trial Bulgaria (L120558): 9 years) were taken from official weather stations located nearby (5.3 - 30 km distance to trial site). The historical and actual data (mean air temperature and precipitation), each averaged over the complete duration of the individual trials are presented in Table 7.1.2.2.1-29.

The actual air temperature recorded at the field sites during the study period was similar to the historic values, except for significantly lower than average temperatures in Spain. Whereas the sites in Spain and France (South) received more rain during the study period compared to the historical values, rainfall was less than the historic values in Germany (North) and Bulgaria. Due to additional irrigation, the total water input at the trial sites during the study was at least 107% of the historical average rainfall, which is considered sufficient to allow the cultivation of crops like wheat.

Table 7.1.2.2.1-29: Summary of historical and actual weather data at field trial sites corresponding to the entire trial duration (1 year)

Trial Country	T_{mean} Air [°C] (average over trial period)		Precipitation [mm] (sum over trial period)		Irrigation [mm]	Sum of actual precipitation and irrigation [mm]	% of historic precipitation
	Historic*	Actual	Historic*	Actual			
L120555 Germany (North)	8.6	8.8	735.0	706.2	109.8	816.0	112
L120556 Germany (South)	9.6	10.0	789.0	869.0	57.9	926.9	117
L120557 United Kingdom	10.1	9.6	607.6	651.4	93.3	744.7	123
L120558 Bulgaria	12.8	13.5	610.7	644.6	408.5	1053.1	172
L120559 Spain	21.4	15.3	326.5	566.0	170.4	736.4	204
L120560 France (South)	13.5	13.1	736.2	1050.4	49.6	1100.0	140

* At least over nine years
**Calculated for the respective parts of the months from day of application until day of last sampling

2. Sampling

Replicate soil residue specimens (8 cores per treated subplot and 10 or 15 per control plot (exception trial Southern Germany (L120556): 15 or 8 cores per control plot) were taken at intervals up to 368 DAT and down to a maximum soil depth of 50 cm. At day 0, immediately after application, collection of Petri dishes, and sand cover, the treated subplots were sampled down to 10 cm only. From 3 DAT the sampling depth was 50 cm. The detailed sampling intervals are presented in Table 7.1.2.2.1-30.

Table 7.1.2.2.1-30: Summary of sampling intervals of residue soil samples at each field trial site

Trial	Country	Sampling intervals [days after treatment]
S12-01863-01 L120555	Germany (North)	-0, 0, 4, 8, 13, 32, 57, 91, 138, 181, 258, 309, 368
S12-01863-02 L120556	Germany (South)	-1, 0, 3, 8, 15, 30, 66, 94, 141, 176, 239/ 240, 294, 367
S12-01863-03 L120557	United Kingdom	-1, 0, 4, 7, 14, 27, 53, 92, 132, 182, 250, 302, 365
S12-01863-04 L120558	Bulgaria	-1, 0, 3, 7, 14, 30, 60, 90, 135, 180, 240, 300, 360
S12-01863-05 L120559	Spain	-3/6, 0, 4, 7, 14, 29, 61, 85, 132, 180, 243, 292, 355
S12-01863-06 L120560	France (South)	0, 4, 8, 14, 27, 56, 97, 140, 175, 238, 306, 368

Untreated specimens were sampled from the control plot on two occasions, between zero and three days before application down to a depth of 50 cm, and after about one year to a depth of 10 cm with the exception of trial Spain (L120559), where the double samples of the first sampling of untreated specimen were taken 9 days after the corresponding main samples (6 DAT). The specimens were taken within the dedicated subsubplot of the untreated plot each time and pooled according to soil depth. The 15 cores collected at the first sampling interval were taken using a soil corer equipped with a plastic or acetate tube liner of 4.3 to 5.0 cm diameter. The cores taken after about one year were collected with a metal tube or a metal tube in tube system of minimum 7.9 and maximum 10.0 cm diameter.

Treated soil specimens were taken at 8 different spots within the randomly assigned subsubplot from each of the three treated subplots SP1 to SP3 and pooled according to subplot, timing, and depth. All soil specimens from 0-10 cm depth collected from the treated subplots were taken separately using a metal tube or a metal tube in tube system of minimum 7.9 and maximum 10.0 cm diameter which left a hole contained by a metal tube. Alternatively, samples were taken by pressing the metal tube described above into the ground and collecting the soil with a spoon or similar device. Soil specimens deeper than 10 cm were collected through the centre of the excavation hole contained by the metal tube, using a soil corer fitted with a plastic or acetate tube liner of diameter 4.3 to 5.0 cm. Sampling of the 10 – 50 cm cores was conducted in one piece.

In addition to the main sampling described above, a second complete sampling (double sampling) was carried out for back-up purposes. The double samples of the 10 - 50 cm layers were not sectioned into 10 cm segments but directly put into the freezers at the field test sites or at the analytical test site. Deviating from this procedure, one double sample from trial France (L120560) was cut into segments of 0 - 30 cm, 30 - 40 cm, and 40 - 50 cm, as well as double samples from trial United Kingdom (L120557) and Bulgaria (L120558), which were sectioned into 10 cm segments.

All soil specimens intended for residue analysis were stored at about -18°C within a maximum of 8.5 hours after sampling and remained frozen through storage, shipping, and processing until final analysis. Sample processing was conducted in frozen state in a mill together with dry ice.

Shipment verification specimens were prepared to demonstrate stability of the residues in soil during storage and through any shipping processes. The samples were prepared at three occasions by fortification of soil with 0.15 mg kg^{-1} alpha-cypermethrin and were subsequently handled in the same manner as the actual residue samples. The analytical results demonstrated no significant losses from the shipping verification samples. The average recovery of the sum of cypermethrin cis isomers was 100% (range: 90 - 131%) across all trials.

3. Analytical procedure

Field soil specimens, as well as Spray broth, Petri dish, and shipping verification specimens were analyzed for cypermethrin isomers cis-1, cis-2, trans-3, and trans-4, and its metabolites 3-PBA and DCVA isomers cis and trans according to the analytical method R0034/01 [*DIAMADUROS, K., DOWNS, C., MALINSKY, D. (2013) Method for the Quantitation of the Diastereomeric Forms of BAS 311 I (Reg. 127266) and its Metabolites 3-Phenoxybenzoic Acid (Reg. No. 130213) and DCVA (Cis and Trans Isomers, Reg. No. 180011) in soil by LC-MS/MS*] provided by BASF.

The analytical method of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4 involved extraction of the soil with 0.1% formic acid in acetonitrile, evaporation to dryness and reconstitution in 0.1% formic acid in acetonitrile/water (1:1, v/v) and final determination of the analytes by HPLC-MS/MS.

The analytical method of 3-PBA and DCVA isomers cis and trans involved two extraction of the soil with acetonitrile/water (70:30, v/v), evaporation to dryness of an aliquot of the extracts and reconstitution in 0.1% formic acid in methanol/water (20:80, v/v) and final determination of the analytes by HPLC-MS/MS.

The limit of quantification (LOQ) was 0.001 mg kg^{-1} for each individual analyte related to wet soil. The limit of detection (LOD) was set at 0.0003 mg kg^{-1} (30% of LOQ).

Analysis of field soil specimens originating from the treated plots was conducted down to a depth until at least two consecutive soil segments were free of quantifiable residues (< LOQ of 0.001 mg kg^{-1}). Analysis was performed up to a maximum of 368 DAT.

The methods used were validated during the current study according to guideline SANCO/3029/99 rev. 4. In addition, untreated control and fortified samples were analyzed within each analytical sample set.

4. Storage stability experiments

Storage stability of alpha-cypermethrin and its metabolites 3-PBA and DCVA (cis and trans isomers) in frozen soil was investigated in a separate study *[CA 7.1.2.2.1/5, BASF DocID 2014/1000723]* with soils originating from the individual trial sites of the present terrestrial field dissipation study.

5. Calculation of degradation times

No calculation of degradation times is provided in the study report. A detailed kinetic evaluation of the degradation behavior of alpha-cypermethrin and its metabolites in the six European field soils is presented in a separate modeling report [*CA 7.1.2.2.1/5, STUDENROTH S., PAPE L., 2014, BASF DocID 2014/1159508*].

II. RESULTS AND DISCUSSION

1. Spray broth concentration and application verification

Spray mixtures as well as application verification samples (Petri dishes) were analyzed for cypermethrin isomers cis-1, cis-2, trans-3, and trans-4. Due to the low detections and the resulting negligible influence, cypermethrin trans isomers are not summarized below, but shown in the analytical phase report.

Spray broth homogeneity was confirmed by visual check for all trials. Spray mixtures were sampled before and after application of each subplot. Analyzed concentrations of the sum of cypermethrin cis isomers averaged for the individual trial sites were in the range of 0.134 to 0.157 g L^{-1} corresponding to 89 - 105% of the target concentration of 0.150 g L^{-1}. The analytical results were not corrected for procedural recoveries and confirm the integrity of the test item used in the trials.

Application verification was conducted by means of petri dishes filled with fine untreated soil from the trial site. Residue levels of the sum of cypermethrin isomers cis-1 and cis-2 achieved on extraction and analysis of the application monitors were converted into residue rates (in g a.s. ha^{-1}) taking into account the area of the Petri dishes (91.6 cm^2). As a result, the obtained rates for the individual trials ranged from 53 to 78 g a.s. ha^{-1} representing 88 - 130% of the target application rate. The applied amount determined via the application monitors in these trials is in agreement with the nominal value of 60 g ha^{-1}, and the results from spray broth analysis.

2. Residues in field soil samples

Untreated soil specimens (control samples) of the respective soil depths from each trial were analyzed for residues of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4 and its metabolites 3-PBA and DCVA isomers cis and trans. No residues above LOD of any analyte were detected in any of the control samples proving that there were no interferences of the untreated soil material with the analytical procedures used. Procedural recovery experiments performed with untreated field soil specimens spiked with cypermethrin isomers cis-1, cis-2, trans-3, and trans-4, and its metabolites 3-PBA and DCVA isomers cis and trans reference items at concentration levels of 0.001, 0.01, and 0.5 mg kg^{-1} (related to wet soil - the highest spiking level was used only for parent compound fortifications), yielded mean recovery rates for the individual analytes between 91 and 98%, confirming the validity of the analytical method used in this study. Detailed results are summarized in Table 7.1.2.2.1-31.

Table 7.1.2.2.1-31: Method procedural recoveries

Analyte	Fortification level [mg kg^{-1}]	n	Mean recovery ± RSD [%]
Cypermethrin isomer cis-1	0.001	74	98 ± 10
	0.01	86	97 ± 9
	0.5	77	98 ± 5
	All fortification levels	**237**	**98 ± 8**
Cypermethrin isomer cis-2	0.001	74	99 ± 12
	0.01	86	97 ± 7
	0.5	77	97 ± 5
	All fortification levels	**237**	**98 ± 9**
Cypermethrin isomer trans-3	0.001	74	100 ± 11
	0.01	86	96 ± 10
	0.5	77	97 ± 6
	All fortification levels	**237**	**97 ± 9**
Cypermethrin isomer trans-4	0.001	74	101 ± 8
	0.01	86	96 ± 8
	0.5	77	94 ± 8
	All fortification levels	**237**	**97 ± 8**
Sum of cypermethrin isomers	0.004	74	100 ± 10
	0.04	86	97 ± 8
	2.0	77	96 ± 5
	All fortification levels	**237**	**97 ± 8**
DCVA isomer cis	0.001	70	89 ± 14
	0.01	59	92 ± 13
	All fortification levels	**129**	**91 ± 14**
DCVA isomer trans	0.001	70	94 ± 14
	0.01	59	94 ± 13
	All fortification levels	**129**	**94 ± 14**
Sum of DCVA isomers [a]	0.002	70	92 ± 13
	0.02	59	93 ± 13
	All fortification levels	**129**	**92 ± 13**
3-PBA	0.001	70	92 ± 14
	0.01	59	92 ± 16
	All fortification levels	**129**	**92 ± 15**

RSD = Relative standard deviation [%]
Remark: Differences between fortification levels are due to Isomer ratio in reference items. The calculation of the recovery is related to the nominal fortification level.
[a] All Isomers are included, each individual by 50%

These data prove that the analytical method applied was able to accurately determine residues of cypermethrin isomers cis-1, cis-2, trans-3, and trans-4, and its metabolites 3-PBA and DCVA isomers cis and trans in soil samples down to a concentration of 0.001 mg kg^{-1} for each analyte.

Field soil specimens from the treated plots were analyzed down to a depth until at least two consecutive soil segments were free of quantifiable residues (< LOQ of 0.001 mg kg^{-1}, maximum depth of 50 cm). If samples were analyzed in duplicate, the individual numbers were averaged to produce a mean for the respective soil sample. If one of the values was below the LOQ, it was averaged as half of LOQ, here however related to dry soil. In general, decision weather residues are below or above LOQ are taken on wet soil basis. Individual concentrations of isomers are then converted to dry soil. In case isomers are summarized (cis-1 and cis-2), wet soil related values were added. If one value is below LOQ (selection on wet soil basis), but above LOD, a value of half of the LOQ (0.0005 mg kg^{-1} wet weight related) is converted to dry soil and is taken for calculation; values below LOD are taken as 0.0 mg kg^{-1}. This means, that LOQ for sum of two isomers (cis-1 and cis-2, as well as for DCVA cis and trans) is 0.002 mg kg^{-1}, and LOD for sum of two isomers is 0.0006 mg kg^{-1}.

All residue values presented are related to the dry weight of the soil and were not corrected for procedural recoveries. The obtained residue rates in mg kg^{-1} were summed up for all depths between 0 and 50 cm analyzed.

The analytical average results of cypermethrin isomer cis-2 (alpha-cypermethrin) and the sum of cypermethrin isomers cis-1 and cis-2 were converted from mg kg^{-1} to µg kg^{-1} for an improved readability and are summarized in Table 7.1.2.2.1-32 and Table 7.1.2.2.1-33. Trans isomers are not added to the sum of cis isomers due to their limited detections and as a result, of their negligible influence.

Table 7.1.2.2.1-32: Total residues of cis-2 cypermethrin under field conditions in soil calculated to µg kg^{-1} and summed up for all depths analyzed

Trial Country	L120555 Dollern, Germany (North)			L120556 Ölbronn-Dürrn, Germany (South)			
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	28	42	31	0	37	12	36
4	38	39	43	3	28	31	25
8	28	45	41	8	18	23	24
13	32	39	30	15	10	15	18
32	25	25	25	30	6	10	7
57	14	20	13	66	0	3	4
91	10	17	9	94	2	0	0
138	12	10	8	141	0	0	0
181	12	10	10	176	0	0	0
258	8	10	9	240	0	0	0
309	9	8	7	294	0	0	0
368	3	4	5	367	0	0	0
Trial Country	**L120557 Melbourne, United Kingdom**			**L120558 Letniza, Bulgaria**			
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	44	30	34	0	51	51	33
4	27	53	44	3	35	36	30
7	16	36	27	7	26	23	16
14	15	30	24	14	23	18	18
27	11	19	17	30	8	9	9
53	12	11	18	60	2	3	2
92	10*	11	14	90	3	3	2
132	12*	9	10*	135	0	2	1
182	9*	7	10*	180	3	2	2
250	7*	8	12*	240	0	2	2
302	10*	9	11*	300	2	1	2
365	4	5	6	360	0	0	0
Trial Country	**L120559 Almansa, Spain**			**L120560 Barry d' Islemade, France (South)**			
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	34	45	35	0	41**	49*	46*
4	24	23	19	4	23	24	28
7	20	24	24	8	17	23	17
14	14	16	10	14	15	14	14
29	12	13	11	27	7	6	7
61	4	6	6	56	2	2	2
85	2	4	4	97	1	1	0
132	0	1	1	140	0	0	0
180	0	0	0	175	0	0	0
243	0	0	0	238	0	0	0
292	0	0	0	306	0	0	0
355	0	0	0	368	0	0	0

DAT = Days after treatment
LOQ (limit of quantification): 1 µg kg^{-1}; LOD (limit of detection): 0.3 µg kg^{-1}
Residue values < LOQ (wet soil related) are reported as 0 µg kg^{-1}
* Mean value of multiple determinations of each, mean and double samples
** Mean value of determinations of each, mean and double sample

Table 7.1.2.2.1-33: Sum of cis-1 + cis-2 cypermethrin residues under field conditions in soil calculated to µg kg^{-1} and summed up for all depths analyzed

Trial Country	L120555 Dollern, Germany (North)			L120556 Ölbronn-Dürrn, Germany (South)			
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	28	42	32	0	37	12	37
4	38	39	43	3	28	31	26
8	28	46	41	8	18	24	24
13	32	39	30	15	10	16	19
32	26	25	25	30	6	10	7
57	14	20	13	66	0	3	4
91	10	17	9	94	2	0	0
138	12	10	8	141	0	0	0
181	12	10	10	176	0	0	0
258	8	10	9	239	0	0	0
309	9	8	7	294	0	0	0
368	3	4	5	367	0	0	0
Trial Country	**L120557 Melbourne, United Kingdom**			**L120558 Letniza, Bulgaria**			
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	46	30	34	0	54	54	36
4	28	54	45	3	40	42	35
7	16	37	27	7	30	27	19
14	15	31	25	14	27	21	23
27	12	20	17	30	11	12	12
53	12	11	18	60	3	3	3
92	10*	11	14	90	3	3	2
132	12*	9	10*	135	0	3	0
182	9*	7	10*	180	4	2	2
250	7*	8	12*	240	0	3	3
302	10*	9	12*	300	3	0	2
365	4	5	6	360	0	0	0
Trial Country	**L120559 Almansa, Spain**			**L120560 Barry d' Islemade, France (South)**			
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	34	47	36	0	42**	49*	47*
4	25	26	20	4	23	25	28
7	21	30	29	8	17	23	18
14	20	22	15	14	16	14	14
29	15	19	15	27	7	6	7
61	6	11	9	56	2	2	2
85	3	6	7	97	0	0	0
132	0	0	0	140	0	0	0
180	0	0	0	175	0	0	0
243	0	0	0	238	0	0	0
292	0	0	0	306	0	0	0
355	0	0	0	368	0	0	0

DAT = Days after treatment
LOQ (limit of quantification): 2 µg kg^{-1}; LOD (limit of detection): 0.6 µg kg^{-1}
Residue values < LOQ (wet soil related) are reported as 0 µg kg^{-1}
* Mean value of multiple determinations of each, mean and double samples
** Mean value of determinations of each, mean and double sample

As is evident from the analytical data, cypermethrin isomers degraded at all six European field sites. The total amount of the sum of cypermethrin isomers cis-1 and cis-2 residues detected in the soil profiles decreased from an average of 39 µg kg^{-1} at 0 DAT to an average of 8 µg kg^{-1} (range: 0 – 20 µg kg^{-1}) at 60 DAT. The residues decreased to an average of 2 µg kg^{-1} at the end of the study, ranging from 0 to 6 µg kg^{-1}.

Considering the distribution of cypermethrin isomer residues in the soil profiles, the main proportion was always measured in the top 0 - 10 cm soil layer and only small amounts of the compound were detected in the 10 – 20 cm layer (≤ 7 µg kg^{-1}). No residues above the LOQ were detected below 20 cm in any sample.

Altogether, it can be concluded that cypermethrin isomers do not show any significant tendency to move into deeper soil layers indicating low potential for cypermethrin isomer residues to leach to groundwater.

Residues of the metabolites 3-PBA and DCVA (cis isomer only) were only found in three out of six field sites: L120558 (Bulgaria), L120559 (Spain), and L120560 (France). Residues above LOQ were exclusively found between 14 and 85 DAT in the top 0 - 10 cm soil layer at L120558 and L120559.

The analytical results of the metabolites cis-DCVA and 3-PBA in field trials L120558 and L120559 are shown in Table 7.1.2.2.1-34.

Table 7.1.2.2.1-34: Total residues of cis-DCVA and 3-PBA under field conditions in soil calculated to µg kg^{-1} and summed up for all depths analyzed (trials L120558, L120559, L120560)

Trial Country	L120558, Letniza, Bulgaria					
	Cis-DCVA			3-PBA		
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	0	0	0	0	0	0
3	0	0	0	0	1.5	0
7	0	0	0	0	0	0
14	3.3	1.8	3.0	2.4	1.7	2.3
30	1.7	2.0	2.3	2.2	2.3	2.5
60	0	0	0	0	0	0
90	0	0	0	0	0	0
135	0	0	0	0	0	0
180	0	0	0	0	0	0
240	0	0	0	0	0	0
300	0	0	0	0	0	0
360	0	0	0	0	0	0
Trial Country	**L120559, Almansa, Spain**					
	Cis-DCVA			3-PBA		
DAT	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]	Subplot 1 [µg kg^{-1}]	Subplot 2 [µg kg^{-1}]	Subplot 3 [µg kg^{-1}]
0	0	0	0	0	0	0
4	0	0	0	0	0	0
7	0	0	0	0	0	0
14	0	0	0	1.4	1.2	1.0
29	1.6	1.8	1.3	2.1	1.8	1.6
61	1.6	2.3	2.7	2.5	2.8	3.0
85	0	0	0	1.0	1.5	2.3
132	0	0	0	0	0	0
180	0	0	0	0	0	0
243	0	0	0	0	0	0
292	0	0	0	0	0	0
355	0	0	0	0	0	0

DAT = Days after treatment
LOQ (limit of quantification): 1 µg kg^{-1}; LOD (limit of detection): 0.3 µg kg^{-1}
Residue values < LOQ (wet soil related) are reported as 0 µg kg^{-1}
* Mean value of multiple determinations of each, mean and double samples
** Mean value of determinations of each, mean and double sample

3. Shipment verification specimens

Shipping verification specimens spiked with alpha-cypermethrin were analyzed to check the stability of the residues in soil during storage at the test site and during any shipping processes. Concentrations of alpha-cypermethrin were not corrected for procedural recoveries.

The analytical results demonstrated no significant losses from the shipping verification samples. The average amount of the sum of alpha-cypermethrin cis isomers from the spiked shipping verification samples was 100% (range: 90 - 131%) across all trials. It was concluded that alpha-cypermethrin was stable in all soils under the storage and shipping conditions used.

4. Time of storage

The maximum period any soil sample from the present field soil dissipation study was stored from the time of sampling to extraction was 727 days for the cypermethrin isomers and ~~836~~ 708 days for the metabolites. Petri dish specimens were stored for up to 611 days after application. The maximum storage period of the spray broth samples was 355 days. Shipment verification specimens were stored for a maximum of 611 days between spiking and extraction.

III. CONCLUSION

Alpha-Cypermethrin degraded under field conditions in soil at all six European field sites. Instantly after application, cis-1 isomer could be additionally detected at 0 DAT at low rates, [redacted] The total amount of the sum of cypermethrin isomers cis-1 and cis-2 residues detected in the soil profiles decreased from an average of 39 µg kg^{-1} (range: 12 – 54 µg kg^{-1}) at 0 DAT to an average of 8 µg kg^{-1} (range: 0 – 20 µg kg^{-1}) after 60 days. Until the end of the study at about 360 DAT, residues decreased further to concentrations equal or below LOQ at four of the six trials, which equals to about 0 µg kg^{-1}. The two trials left with residues above LOQ showed mean rates of 5 µg kg^{-1} (range: 3 – 6 µg kg^{-1}) at the end of the study.

DT_{50} values either for alpha-cypermethrin and sum of both cis isomers and were subject of separate modeling reports.

Amounts of trans-3 and trans-4 isomers occurred only in two of the six trials rarely at very low amounts.

Cypermethrin isomer residues were exclusively detected in the upper 20 cm of the soils. No residues above the LOQ were detected below 20 cm in any sample. Altogether, it can be concluded that cypermethrin isomer does not show any significant tendency to move into deeper soil layers indicating very low potential for cypermethrin isomer residues to leach to groundwater.

Residues of the metabolites 3-PBA and DCVA (cis isomer only) above LOQ were only found in three out of six field sites (Bulgaria, Spain, and France). Residues above LOQ were exclusively found at trial sites Bulgaria and Spain.

Metabolite DCVA isomer cis was detected in small amounts reaching maximum amounts of 3.3 µg kg^{-1} (Bulgaria; 14 DAT) and 2.7 µg kg^{-1} (Spain; 60 DAT) on average for the three subplots. Thereafter, residues declined again and were no longer detected in amounts > LOQ after 132 days at the latest.

Metabolite 3-PBA was detected above LOQ in small amounts reaching maximum amounts of 2.5 µg kg^{-1} (Bulgaria; 30 DAT) and 3.0 µg kg^{-1} (Spain; 60 DAT) on average for the three subplots. Thereafter, residues declined again and were no longer detected in amounts > LOQ after 85 days at the latest.

Metabolites DCVA isomer cis and 3-PBA were exclusively found in the top 0 - 10 cm soil layer of the three field sites. No residues above the LOQ were observed in deeper soil layers in any sample at any site.

Calculation of maximum occurrences of DCVA and 3-PBA from the EU field dissipation study

The metabolites DCVA and 3-PBA occurred in relevant amounts in the EU field dissipation study with alpha-cypermethrin [*CA 7.1.2.2.1/4, BASF DocID 2014/1161559*]. In the following, the derivation of the maximum occurrences of the metabolites in the EU field dissipation study is described.

For the EU study, the residue data of the metabolites reported in µg kg^{-1} were converted into µg p.e. kg^{-1}, considering a molar correction factor of 0.502 for DCVA and 0.514 for 3-PBA. For alpha-cypermethrin, the sum of cis-1 and cis-2 isomer and for DCVA the cis isomer was considered. The respective trans isomers were not included due to their low detection and therefore negligible influence.

Alpha-cypermethrin was applied at one application. Maximum occurrences were calculated for the trials L120558 (Bulgaria) and L120559 (Spain), considering the maximum amount of metabolite residues (mean of three plots) after application and the initial amount of parent residues (mean of three plots). The results are summarized in Table 7.1.2.2.1-35.

Table 7.1.2.2.1-35: Maximum occurrence of alpha-cypermethrin metabolites in EU field dissipation study

Metabolite	Trial	Maximum amount of metabolite		Initial amount of parent	Maximum occurrence
		[µg kg^{-1}] [a]	[µg p.e. kg^{-1}] [b]	[µg kg^{-1}] [a]	[%]
DCVA	L120558	2.7	5.4	47.9	11.2
	L120559	2.4	4.7	38.9	12.1
3-PBA	L120558	2.3	4.5	47.9	9.5
	L120559	3.1	6.0	38.9	15.5

DAT = Days after first treatment

[a] Mean of three plots

[b] Metabolite concentrations were converted into parent equivalents (p.e.) considering a molar correction factor of 0.502 for DCVA and 0.514 for 3-PBA.

Report:	CA 7.1.2.2.1/6 Studenroth S., Pape L., 2014b Kinetic evaluation of a field dissipation study with BAS 310 I – alpha-cypermethrin conducted in 2012 and 2013: Determination of modeling endpoints according to FOCUS 2014/1159508
Guidelines:	FOCUS Degradation Kinetics (2006) Sanco/10058/2005 version 2.0 (November 2011) EFSA Guidance to obtain DegT50 values in soil (2010)
GLP:	no

Executive Summary

The dissipation behavior of the insecticide BAS 310 I – alpha-cypermethrin in soil has been investigated in a field dissipation study including six field trials located in Germany (two trials), United Kingdom, Spain, Bulgaria and France [*CA 7.1.2.2.1/4, BASF DocID 2014/1161559*]. The purpose of this evaluation was to analyze the degradation kinetics of alpha-cypermethrin and its metabolites DCVA and 3-PBA observed in the six soils according to the current guidance of the FOCUS workgroup on degradation kinetics in order to derive normalized modeling endpoints.

The field trials were situated in different regions of Europe (Germany, United Kingdom, Bulgaria, Spain and France), considering a range of different soils and climatic conditions.

As the study design was tailored to exclude surface processes, kinetic evaluation was performed according to FOCUS kinetics as recommended by *EFSA (2014)*.

Prior to kinetic evaluation, sampling intervals of the field studies were normalized to reference conditions (20°C, pF2) by time-step normalization. The kinetic evaluation was performed on the normalized dataset. Modeling endpoints were derived based on a visual and statistical assessment. Kinetic evaluation of the time-step normalized dataset (20°C, pF2) resulted in normalized field half-lives ($DegT_{50}$) for alpha-cypermethrin between 11.7 and 114.1 days, for DCVA between 16.0 and 32.4 days and for 3-PBA between 17.1 and 35.8 days.

I. MATERIAL AND METHODS

The kinetic evaluation was conducted for six field trials with alpha-cypermethrin from the data of one field dissipation study, which can be found in *CA 7.1.2.2.1/4* [*BASF DocID 2014/1161559*]. The trials were situated in different regions of Europe (Germany (DE1 and DE2), United Kingdom (UK), Bulgaria (BG), Spain (ES) and France (FR)) considering a range of different soils and climatic conditions. Detailed soil characteristics in each trial are reported in the cited study.

Alpha-cypermethrin was applied in June 2012 in the formulation BAS 310 55 I (ME) to three replicate plots per trial to bare soil at an intended application rate of 60 g a.s. ha^{-1} using a calibrated boom sprayer. Immediately after application and before subsequent soil sampling all plots were covered with a thin layer (at least 3-5 mm) of sand and maintained at least until the field received a total precipitation (rain and irrigation) of ≥10 mm to protect the applied product from surface processes like photolysis or volatilization, and to exclude any potential impact on the degradation of the test item caused by any of these processes.

The study duration was up to 368 days after application. Soil samples were taken at 12 sampling dates after application. Replicate soil specimens from the treated subplots were taken down to a maximum soil depth of 50 cm.

The samples were extracted with 0.1% formic acid in acetonitrile and extracts were analyzed for alpha-cypermethrin isomers (cis-1, cis-2, trans-3 and trans-4) by means of HPLC-MS/MS. For analysis of the metabolites DVCA (CIS and TRANS isomers) and 3-PBA the soil was extracted two times with acetonitrile/water (70:30, v/v) and extracts were analyzed by means of HPLC-MS/MS. For each analyte the limit of quantification (LOQ) was 0.001 mg kg^{-1} and the limit of detection (LOD) was set to 0.0003 mg kg^{-1}.

Alpha-cypermethrin residues were almost exclusively found in the top 0-10 cm soil layer, with only small amounts of the compound sporadically detected in the 10-20 cm layer (≤0.0054 mg kg^{-1}). No residues above the LOQ were detected below 20 cm in any sample.

The metabolites DCVA and 3-PBA were detected in the top 0-20 cm soil layer at three sites (Bulgaria, Spain and France) whereby residues above LOQ were only found in the top 0-10 cm soil layer at the sites of Bulgaria and Spain.

Kinetic modeling

As processes had been excluded by covering the soil with sand in the field study, kinetic evaluation was performed according to FOCUS kinetics as recommended by EFSA [*EFSA (2014): Guidance Document for evaluating laboratory and field dissipation studies to obtain $DegT_{50}$ values of active substances of plant protection products and transformation products of these active substances in soil. EFSA Journal 2014;12(5):3662, 37 pp. doi: 10.2903/j.efsa.2014.3662*].

The software package KinGUI version 2.2014.224.1704 was used for parameter fitting. The error tolerance and the number of iterations of the optimization tool (IRLS) were set to the default values of 1×10^{-6} and 100, respectively.

Data preparation for kinetic evaluation was based on raw data from laboratory. For the parent substance, the sum of residues of the alpha-cypermethrin isomers cis-1 and cis-2 was considered. The sums were calculated by adding individual values above LOQ (0.001 mg kg^{-1}, related to dry weight of the soil samples); if one individual value was below LOQ but above LOD (0.0003 mg kg^{-1}, related to dry weight of the soil samples), a value of 0.0005 mg kg^{-1} (half of LOQ) was taken for calculation; values below LOD were taken as 0.0 mg kg^{-1}. For the sum of the two isomers a LOQ of 0.002 mg kg^{-1} and a LOD 0.0006 mg kg^{-1} (two times the individual LOQ or LOD) was considered for further processing of the parent data.

For the trials in Bulgaria and Spain the metabolites 3-PBA and the cis isomer of DCVA were considered for the evaluation. The trans isomers of alpha-cypermethrin and DCVA were not included due to their low detection and therefore negligible influence.

Datasets were prepared for kinetic evaluation as follows:

- Values below LOQ were treated as recommended by the FOCUS workgroup [*FOCUS (2006)*]. Accordingly, values between LOQ and LOD were set to the mean of LOQ and LOD (parent: 0.0013 mg kg^{-1}, metabolites: 0.0007 mg kg) and values below LOD were set to half of LOD (parent: 0.0003 mg kg^{-1}, metabolites: 0.0002 mg kg^{-1}).

- For each sampling point, the concentration of a compound in the single soil layer given in mg kg^{-1} was transformed to its residue given in g ha^{-1} considering the height of the respective layer and the respective undisturbed soil bulk density for each sample as given in the study report. The total residues in the sampled subplot were calculated as the sum of residues of the single soil layers.

- For the metabolites, the residues in g ha^{-1} were transformed into parent equivalents in g a.s. ha^{-1}.

The measured data as well as resulting datasets submitted to kinetic analysis are provided in the original evaluation report.

Kinetic models included in the evaluations

For each data set, the kinetic models proposed by FOCUS Kinetics were tested. The recommended kinetic models, i.e. single-first order (SFO), First-order multi-compartment (FOMC), Hockey Stick (HS) and Double first-order in parallel (DFOP), were applied to the alpha-cypermethrin data.

A kinetic model is considered appropriate if the residuals are randomly distributed around zero, the χ^2 error indicates a sufficient quality of the fit (e.g. value is ideally <15 % but may be higher if the visual fit represents the degradation behavior well) and the estimated degradation parameters differ significantly from zero.

The metabolites were subsequently added to the appropriate kinetic model for the parent considering the SFO kinetic model. Simultaneous formation of the degradation products from the parent was assumed for the kinetic evaluation.

In order to derive modeling endpoints, kinetic evaluation was performed on the time-step normalized data set.

Normalization procedure

Evaluation of the suitability of field dissipation data for normalization was performed according to the evaluation criteria for normalization compiled by the Dutch regulatory authority (CTGB criteria).

The time-step normalization procedure was carried out for all field trials by reducing or increasing day lengths depending on soil temperature and moisture by means of correction factors (f_{temp} and f_{moist}) identical to those used in most regulatory leaching models. Daily soil moisture and soil temperature values were calculated by FOCUS-PEARL 4.4.4 using weather data available from on-site weather stations and soil characteristics reported in the original study report.

Temperature correction factors (f_{temp}) were determined according to Equation Equation 7.1.2.2.1-1 c to account for differences between actual daily soil temperatures as calculated by PEARL and a reference temperature of 20°C using the Q_{10} approach and considering a Q_{10} value of 2.58.

Moisture correction factors (f_{moist}) were determined according to Equation 7.1.2.2.1-1 d to account for differences between actual daily soil moisture as calculated by PEARL and the reference soil moisture at field capacity (pF 2).

The normalized day lengths were derived according to Equation 7.1.2.2.1-1 a and normalized sampling days (D_{norm}) after application were calculated by cumulatively summing up normalized day lengths according to Equation 7.1.2.2.1-1 b.

Equation 7.1.2.2.1-1:Calculation of normalized day length based on combination of soil moisture and soil temperature correction factors

a)
$$D_{norm} = D * f_{temp} * f_{moisture}$$

b)
$$t_i = \sum_{t=1}^{i-1} D_{norm}$$

with:

t_i =	Time from application till sampling at day i	[d]
D_{norm} =	Normalized day length (20°C, pF2)	[d]
i =	Time span between application and sampling	[d]

c)
$$f_{temp} = \begin{cases} Q_{10}^{\frac{Tact-Tref}{10}} & \text{for } T_{act} > 0°C \\ 0 & \text{for } T_{act} \leq 0°C \end{cases}$$

d)
$$f_{moist} = \begin{cases} \left(\frac{\theta_{act}}{\theta_{ref}}\right)^B & \text{for } \theta_{ref} > \theta_{act} \\ 1 & \text{for } \theta_{ref} \leq \theta_{act} \end{cases}$$

with:

D_{norm} =	normalized day length (temperature and moisture corrected)	[days]
D =	actual day length (1 d)	[days]
f_{temp} =	temperature correction factor	[-]
f_{moist} =	moisture correction factor	[-]
T_{act} =	actual soil temperature	[C°]
T_{ref} =	reference temperature (20°C)	[C°]
Q_{10}=	factor of increase of degradation rate with an increase in temperature of 10°C (Q_{10} = 2.58)	[-]
θ_{act} =	actual soil moisture	[m^3 m^{-3}]
θ_{ref} =	reference soil moisture at pF2	[m^3 m^{-3}]
B =	exponent of the moisture response function, B = 0.7	[-]

Table 7.1.3.1.2-1 shows the field sampling dates for the trial locations and the normalized (20°C, pF2) day lengths based on soil moisture and soil temperature data as simulated by FOCUS-PEARL 4.4.4.

Table 7.1.2.2.1-36: Time-step normalized (temperature and moisture) sampling days

Dollern, Germany (L120555, DE1)		Ölbronn-Dürrn, Germany (L120556, DE2)		Melbourne, United Kingdom (L120557, UK)		Letniza, Bulgaria (L120558, BG)		Almansa, Spain (L120559, ES)		Barry d'Islemade, France (L120560, FR)	
DAT	**D_{norm}**	**DAT**	**D_{norm}**	**DAT**	**D_{norm}**	**DAT**	**D_{norm}**	**DAT**	**D_{norm}**	**DAT**	**D_{norm}**
0	0	0	0	0	0	0	0	0	0	0	0
4	2.4	3	3.0	4	2.8	3	4.9	4	4.6	4	3.2
8	4.5	8	6.3	7	4.7	7	10.4	7	8.5	8	6.2
13	7.7	15	13.3	14	9.3	14	19.7	14	18.9	14	13.2
32	22.6	30	25.9	27	19.7	30	48.9	29	42.0	27	28.5
57	41.7	66	60.5	53	35.1	60	96.4	61	91.4	56	59.6
91	69.5	94	80.0	92	50.7	90	131.8	85	126.0	97	113.5
138	93.3	141	99.4	132	61.7	135	164.4	132	173.3	140	147.0
181	104.8	176	106.1	182	72.4	180	175.3	180	194.5	175	160.7
258	117.6	240	115.8	250	86.2	240	182.0	243	215.9	238	177.4
309	122.8	294	123.0	302	104.7	300	202.0	292	234.9	306	199.0
368	150.5	367	162.4	365	145.7	360	250.4	355	272.1	368	234.0

DAT = days after treatment
D_{norm} = normalized day (20°C, pF2)

II. RESULTS AND DISCUSSION

The SFO model was appropriate to derive modeling endpoints for alpha-cypermethrin and its metabolites DCVA and 3-PBA for field trials L120555 (DE1), L120556 (DE2), L120558 (BG) and L120559 (ES). HS model was appropriate to derive modeling endpoints for alpha-cypermethrin from field trial L120557 (UK) and FOMC model was used to derive modeling endpoints from field trial L120560 (FR). A summary of the adequate $DegT_{50}$ values, as well as of the formation fractions for the metabolites, is given in Table 7.1.2.2.1-37 to Table 7.1.2.2.1-39.

Table 7.1.2.2.1-37: Summary of modeling endpoints of alpha-cypermethrin

Field trial	Soil type (USDA)	Best-fit kinetic model	χ^2	$DegT_{50}$ [d]
L120555 (DE1)	Sandy loam	SFO	11.0	46.1
L120556 (DE2)	Silt loam	SFO	15.5	16.5
L120557 (UK)	Loam	HS	15.3	114.1 [a]
L120558 (BG)	Silty clay loam	SFO	13.1	24.6
L120559 (ES)	Sandy loam	SFO	4.4	42.8
L120560 (FR)	Sandy clay loam	FOMC	3.5	11.7 [b]

[a] Calculated as $DT_{50} = \ln(2)/k2$
[b] Calculated as $DT_{50} = DT_{90}/3.32$

Table 7.1.2.2.1-38: Summary of modeling endpoints of DCVA

Field trial	Soil type (USDA)	Kinetic model	χ^2	$DegT_{50}$ [d]	Formation fraction [-]
L120558 (BG)	Silty clay loam	SFO	34.3	16.0	0.371
L120559 (ES)	Sandy loam	SFO	34.5	32.4	0.326

Table 7.1.2.2.1-39: Summary of modeling endpoints of 3-PBA

Field trial	Soil type (USDA)	Kinetic model	χ^2	$DegT_{50}$ [d]	Formation fraction [-]
L120558 (BG)	Silty clay loam	SFO	25.5	17.1	0.364
L120559 (ES)	Sandy loam	SFO	25.4	35.8	0.437

III. CONCLUSION

Kinetic evaluation of six field trials with alpha-cypermethrin was conducted in order to derive reliable normalized modeling endpoints according to the current guidance of the FOCUS workgroup on degradation kinetics. The SFO model was appropriate to derive modeling endpoints for alpha-cypermethrin and its metabolites DCVA and 3-PBA for field trials L120555 (DE1), L120556 (DE2), L120558 (BG) and L120559 (ES). HS model was appropriate to derive modeling endpoints for alpha-cypermethrin from field trial L120557 (UK) and FOMC model was used to derive modeling endpoints from field trial L120560 (FR).

Kinetic evaluation of the time-step normalized dataset (20°C, pF2) resulted in normalized field half-lives ($DegT_{50}$) for alpha-cypermethrin between 11.7 and 114.1 days, for DCVA between 16.0 and 32.4 days and for 3-PBA between 17.1 and 35.8 days.

Summary of maximum occurences of alpha-cypermethrin metabolites and degradation rates for alpha-cypermethrin and its metabolites in field dissipation studies

Table 7.1.2.2.1-40: Summary table on maximum occurrence of alpha-cypermethrin metabolites in field dissipation studies

Metabolite	BASF DocID	Trial	Maximum occurrence [%] [a]
DCVA	2013/7002604	Louisiana Oklahoma New York California	3.6 [b] 6.8 [b] 4.9 [c] 10.9 [b]
	2014/1161559	L120558 L120559	11.2 12.1
3-PBA	2013/7002604	Louisiana Oklahoma New York California	14.5 [d] 8.8 [b] 7.5 [c] 28.6 [b]
	2014/1161559	L120558 L120559	9.5 15.5

[a] Percent of max. parent residues
[b] Maximum reached after 3rd application
[c] Maximum reached after 2nd application
[d] Maximum reached after 1st application

Table 7.1.2.2.1-41: Summary table on trigger endpoints of alpha-cypermethrin obtained in the US field soil dissipation study

BASF DocID	Trial site	pH ($CaCl_2$) [a]	Org. C [a] [%]	Best-fit DT_{50} / DT_{90} [d]	Method of calculation
2013/7002604 [b]	Louisiana	6.2	0.9	5.9 / 19.6	SFO
	Oklahoma	6.7	0.6	6.3 / 20.9	SFO
	New York	7.1	0.8	3.4 / 27.9	FOMC
	California	6.8	0.7	4.0 / 13.4	SFO

[a] In 0 - 15 cm soil depth

Table 7.1.2.2.1-42: Summary table on modeling endpoints of alpha-cypermethrin obtained in the EU field soil dissipation study

BASF DocID	Trial site	pH ($CaCl_2$) [a]	Org. C [a] [%]	Modeling DT_{50} 20°C, pF2 [d]	Method of calculation
2014/1161559 [b]	L120555 (DE1)	5.71	1.7	46.1	SFO
2014/1159507 [b]	L120556 (DE2)	6.01	1.1	16.5	SFO
	L120557 (UK)	6.74	1.6	114.1 [c]	HS
	L120558 (BG)	6.96	2.4	24.6	SFO
	L120559 (ES)	7.47	1.4	42.8	SFO
	L120560 (FR)	5.39	0.5	11.7 [d]	FOMC

[a] In 0 – 30 cm soil depth, L120559 (ES) 0 – 10 cm
[b] Covered field soil study; only to derive modeling endpoints in the soil matrix, excluding surface loss processes [*EFSA (2010)*]
[c] Calculated as ln(2)/k2
[d] Calculated as $DT_{50} = DT_{90}/3.32$

Table 7.1.2.2.1-43: Summary table on modeling endpoints of alpha-cypermethrin metabolites in EU field soil dissipation study

Metabolite	BASF DocID	Trial site	pH ($CaCl_2$) [a]	Org. C [a] [%]	Method of calculation [c]	Modeling DT_{50} [c] SFO, 20°C, pF2 [d]	Formation fraction
DCVA	2014/1161559 [b]	L120558 (BG)	6.96	2.4	SFO	16.0	0.371
	2014/1159507 [b]	L120559 (ES)	7.47	1.4	SFO	32.4	0.326
3-PBA	2014/1161559 [b]	L120558 (BG)	6.96	2.4	SFO	17.1	0.364
	2014/1159507 [b]	L120559 (ES)	7.47	1.4	SFO	35.8	0.437

[a] In 0 – 30 cm soil depth
[b] Covered field soil study; only to derive modeling endpoints in the soil matrix, excluding surface loss processes [*EFSA (2010)*]
[c] SFO kinetics for the parent

CA 7.1.2.2.2 Soil accumulation studies

Due to the fast degradation of alpha-cypermethrin no accumulation study in soil is required.

CA 7.1.3 Adsorption and desorption in soil

CA 7.1.3.1 Adsorption and desorption

Two adsorption/desorption studies were reviewed in the submission of alpha-cypermethrin (see Table 7.1.3.1-1):

1. Stevens J.K.B. (1981) Cypermethrin: Adsorption and desorption in soil
Study performed with cypermethrin in one soil, not following current guidance. The study result can only be interpreted in a qualitative way.

2. Hill A.D. (1993) [Benzyl- ^{14}C] WL85871 (FASTAC): Adsorption desorption in three soils
The current endpoints were derived from this study. The study has the following deficiencies: Only three soils were used. Questions about the solubility of alpha-cypermethrin could not be solved. It seems that the highest concentration used in the study exceeded the solubility of the compound. A very strong decline in sorption was observed for the highest concentration. In the determination of the Freundlich exponent only four data points were measured covering approximately a factor of 10. The isotherms did not show a good fit.

Therefore a new study was conducted with alpha-cypermethrin in five soils. Due to the low solubility of the compound it was decided to give up an attempt to determine the Freundlich exponent. This new study (Heinz N. 2014/1162672) was used to propose a new Koc value for risk assessment.

In addition a study with DCVA was conducted (Malinski D. S., 2004/5000486) and is presented under CA 7.1.3.1.2/1.

Table 7.1.3.1-1: List of peer reviewed soil adsorption/desorption studies performed with (alpha-) cypermethrin or cypermethrin metabolite 3-PBA

DocID	Parent compound	Soil	Application rate	Soil/solution ration	Incubation period [hours]	Remark
CY-620-006	Cypermethrin	Loamy sand	0.01, 0.025, 0.04, 0.1 µg mL^{-1}	1:50	54	Stevens 1981
AL-620-011	Alpha-cypermethrin	Sand Sandy loam Silty clay	0.25, 075, 1.0, 4.0 µg L^{-1}	1:100	22	Hill 1993
2002/5004760	3-PBA	Silty loam Sandy loam Silty loam Loamy sand	0.05, 0.1, 0.5, 1, 5.0 µg mL^{-1}	1:1	48	Holman 2002

CA 7.1.3.1.1 Adsorption and desorption of the active substance

Report: CA 7.1.3.1.1/1
Heinz N., 2014a
Determination of Adsorption and Desorption Behavior of BAS 310 I (Alpha-Cypermethrin) in 5 Soils (OECD Guideline 106)
2014/1162672
Guidelines: OECD 106
GLP: yes
(certified by Landesanstalt fuer Umwelt, Messungen und Naturschutz, Baden-Württemberg, Karlsruhe, Germany)

Executive Summary

In laboratory experiments the adsorption behavior of alpha-cypermethrin (BAS 310 I) was investigated on five European soils. The five tested soils covered a range of pH ($CaCl_2$) from 5.4 to 7.4, a range of organic carbon content from 0.67 to 1.72%, and three different USDA textural classes: two loamy fine sands, two sandy loams, and one loam. Due to the low water solubility of alpha-cypermethrin, adsorption was only tested for one concentration level (3 ng mL^{-1}). Hence, no adsorption or desorption isotherm was determined. The application solution was prepared in acetone with a nominal concentration of 10 µg mL^{-1} of the test item. The ratio of soil versus test solution was 1/100, and the measurements were performed at the adsorption equilibrium time of 2 h.

Sorption of alpha-cypermethrin to soil proceeded fast, expressed by about 95% of the initially applied amount of the test item adsorbed after an equilibration time of 2 h. Hence, almost the entire applied amount (≥ 95%) of alpha-cypermethrin was accounted to the fraction sorbed to the soil phase. Calculated mean distribution coefficients (K_d) ranged from 2366 mL g^{-1} (LUFA 2.3) to 4362 mL g^{-1} (LUFA 2.2). Distribution coefficients normalized to the fraction of organic carbon in the soils (K_{OC}) ranged from 228622 mL g^{-1} (Bruch West) to 353100 mL g^{-1} (LUFA 2.3).

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item: alpha-cypermethrin (BAS 310 I)
Reg. No.: 4078193
Batch No.: L80-24
Chemical name (IUPAC): Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Chemical purity: 99.4%
Molecular weight: 416.3 g mol^{-1}

2. Soils

The study was conducted with five different soils originating from Europe. The physico-chemical properties of the soils are provided in Table 7.1.3.1.1-1.

Table 7.1.3.1.1-1: Characterization of soils used to determine the adsorption behavior of alpha-cypermethrin

Soil designation Origin	**LUFA 2.2 Germany**	**LUFA 2.3 Germany**	**Bruch West Germany**	**Li10 Germany**	**Fiorentino Poggio Renatico 1 Italy**
Textural class (DIN 4220)	Loamy sand (Sl2)	Silty sand (Su3)	Loamy sand (Sl3)	Loamy sand (Sl2)	Loamy sand
Soil texture [%], (ISO 11277)					
Sand	85.0	62.9	66.7	81.5	41.7
Silt	10.0	29.5	22.2	13.3	41.6
Clay	5.0	7.6	11.1	5.2	16.7
Textural class (USDA scheme)	Loamy fine sand	Sandy loam	Sandy loam	Loamy fine sand	Loam
Soil texture [%], (USDA)					
Sand	87.4	64.1	69.9	83.4	49.4
Silt	7.7	28.3	18.9	11.4	33.9
Clay	5.0	7.6	11.1	5.2	16.7
Organic carbon [%] (ISO 10694)	1.72	0.67	1.62	0.95	1.07
Cation exchange capacity [$cmol^+ kg^{-1}$]	4.2	6.2	11.6	7.6	11.2
pH ($CaCl_2$)	5.8	5.4	7.1	6.3	7.4
pH (water)	6.4	6.1	8.0	7.0	8.3
Max. water holding capacity [g $100g^{-1}$ dry soil]	33.1	23.9	25.5	24.4	29.7
Bulk density [g L^{-1}]	1253	1353	1382	1358	1403

B. STUDY DESIGN

1. Experimental conditions

Preliminary tests: adsorption kinetics and soil / solution ratio

A preliminary experiment was run with two soils (LUFA 2.2 and Fiorentino Poggio Renatico 1) to determine the time needed to establish equilibrium conditions and to find the optimal soil / solution ratio for the adsorption tests. The experiments were run with a soil / solution ratio of 1/100. The test was performed using 30 µL application solution (10 µg mL^{-1} alpha-cypermethrin in acetone). The vials were protected from light and shaken at 19 - 20°C for 2, 4, 6 and 24 h. The soil / solution suspension was then centrifuged and decanted. An aliquot of the supernatant was sampled and the concentration of test item in the $CaCl_2$ solution was determined by gas chromatography coupled with mass spectrometry (GC-MS).

The parallel method was used with determination of the amount adsorbed (direct method) via solvent extraction of the soil pellets. All experiments were performed in duplicates.

Results of this first trial revealed that the adsorption equilibrium was reached fast (≤ 2 h). The soil / solution ratio of 1/100 led to significant adsorption of about 95% of the initially applied amount of the test item.

Stability and adsorption of the test substance

Due to the low solubility of the test item in water (maximal 3 µg L^{-1}) and a high adsorption to soil, no adsorption or desorption isotherms could be determined in this study. Hence, the stability test, which is typically performed during the pre-tests, corresponds to the adsorption test.

The adsorption kinetics test was performed as described for the preliminary test with the three remaining soils (Bruch West, Li10, and LUFA 2.3), but with 2 h equilibration time only.

Stability of alpha-cypermethrin in aqueous $CaCl_2$ solution was demonstrated by acceptable results for adsorption controls obtained by GC-MS analyses.

Instability of alpha-cypermethrin in soil/$CaCl_2$ solution was demonstrated by decreasing mass balances in adsorption kinetic tests, indicating degradation or formation of bound residues.

Control samples with only the test item in aqueous 0.01 M $CaCl_2$ solution (adsorption controls) were used to elucidate a potential adsorption of the test item to the surface of the test vessels.

In addition, experiments were conducted in order to estimate the adsorption to test vessel surface in presence of soil. Therefore, two experiments were performed in parallel, each one in duplicate. 1.0 g of soil was agitated with 100 mL solution for 6 h. Afterwards, phases were separated by centrifugation and decantation. One sample of the soil pellet was extracted with solvent directly in the test vessel. From the second soil sample, the soil pellet was removed as good as possible and transferred directly into extraction solvent present in another vessel equivalent to the test vessel. The difference in detected amounts of the test item between the two samples was used to estimate the surface adsorption in the test vessels.

2. Description of analytical procedures

The parallel method was performed, including the determination of the amount of the test item in the aqueous phase, as well as the determination of the amount adsorbed to soil via solvent extraction.

Principle of the methods:

To analyze alpha-cypermethrin in the aqueous phase, soil suspensions were centrifuged (5 min, 4000 rpm), decanted, and adjusted to pH 5 with formic acid. Supernatants were extracted two times with 10 mL portions of cyclohexane by mechanical shaking. The organic layers were combined and 0.2 mL of an internal standard solution (10 mg mL^{-1} lambda-cyhalothrin in toluene) was added. The extract was concentrated to the final volume of 0.2 mL and subsequently analyzed by GC-MS (with negative chemical ionization: NCI). The method has a limit of quantification (LOQ) of 0.03 ng mL^{-1} in $CaCl_2$ solution, for soil 3 ng g^{-1}. The limit of detection (LOD) in $CaCl_2$ solution is < 0.0002 ng mL^{-1} (< 0.2 pg mL^{-1}), for soil < 0.2 ng g^{-1} (< 200 pg g^{-1}).

For soil analysis, soil samples (1.0 g) were extracted with 10 mL acetonitrile/water (7:3, v/v) on a mechanical shaker (10 min) followed by sonication (5 min) in an ultrasonic bath. After extraction, the tube was centrifuged at 4000 rpm (5 min). The extraction was repeated two times with each 10 mL acetonitrile containing 0.1% formic acid. The three extracts were combined. A 5.0 mL aliquot from the extract and 5 mL of water were partitioned with 30 mL of cyclohexane by shaking 30 min on a horizontal shaker. After centrifugation (1 min, 4000 rpm) the organic layer was separated and 0.5 mL of the internal standard solution (10 mg mL^{-1} lambda-cyhalothrin in toluene) was added. The extract was concentrated to a final volume of 0.5 mL for subsequent GC-MS analysis (with negative chemical ionization: NCI).

To account for the amount of test item adsorbed to the test vessel surface in the presence of soil, glass centrifuge vials (used for adsorption kinetics experiments) were extracted two times with 10 mL of ethyl acetate on a horizontal shaker (10 min) and sonication (5 min) in an ultrasonic bath. The combined organic layer was spiked with 0.2 mL of the internal standard. After concentration to the final volume of 0.2 mL, samples were analyzed by GC-MS (with negative chemical ionization: NCI).

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The analytical methods for the determination of the test item in the aqueous phase, as well as for the determination in soil extracts were validated within this study. Both methods revealed acceptable average recoveries (71 - 96%) and relative standard deviations (RSD; ≤ 20%) for the dose levels of 15.0 and 150 ng mL^{-1} for $CaCl_2$ solution, as well as 3.0 and 300 ng g^{-1} for soil, respectively.

In the adsorption test, the average recovery values ($CaCl_2$ solution plus soil extract) ranged from 82 to 88%. For soil LUFA 2.3, the average recovery was low, exhibiting 54% of the initially applied amount of the test item, and was accounted for degradation or formation of bound residues.

B. FINDINGS

Based on the results obtained, the applied analytical methods are considered valid for the determination of alpha-cypermethrin in $CaCl_2$ solution and in soil with a validated LOQ of 0.03 ng mL^{-1} and 3 ng g^{-1}, respectively.

Results of the adsorption equilibrium test carried out with alpha-cypermethrin indicated that the adsorption equilibrium was reached within 2 h.

Results of control samples (adsorption controls), where the test item was applied to $CaCl_2$ solution incubated without soil, revealed that alpha-cypermethrin was not sorbed to the wall of the test vessels in the absence of soil (average recovery 92%; RSD ≤ 11%).

In addition, experiments with soil were conducted in order to estimate the adsorption to test vessel surface in presence of soil. As a result, the soil samples extracted in the experimental vessels show, soil related, slightly higher concentrations than the comparable soil samples, where the soil was removed and extracted out of the experimental vessel. However, considering this variability over all soils and comparing it against relative standard variation percentages of recovery rates during the course of soil residue analysis, these differences are of no significance.

Sorption of alpha-cypermethrin to soil proceeded fast. This was expressed by about 95% of the initially applied amount of the test item adsorbed to the soil phase after an equilibration time of 2 h. Hence, almost the entire applied amount (≥ 95%) of alpha-cypermethrin was accounted to the fraction sorbed to the soil phase. Calculated mean distribution coefficients (K_d) ranged from 2366 mL g^{-1} (LUFA 2.3) to 4362 mL g^{-1} (LUFA 2.2). Distribution coefficients normalized to the fraction of organic carbon in the soils (K_{OC}) ranged from 228622 mL g^{-1} (Bruch West) to 353100 mL g^{-1} (LUFA 2.3). Calculated K_d as well as K_{OC} values are presented in Table 7.1.3.1.1-2.

Table 7.1.3.1.1-2: Measured equilibrium concentrations and calculated distribution coefficients K_d and K_{OC} of alpha-cypermethrin

Soil	Soil Type (USDA)	Org. C [%]	pH ($CaCl_2$)	Adsorption	
				K_d [mL g^{-1}]	K_{OC} [mL g^{-1}]
LUFA 2.3	loamy fine sand	0.67	5.4	2366	353100
LUFA 2.2	sandy loam	1.72	5.8	4362	253632
Bruch West	sandy loam	1.62	7.1	3704	228622
Li10	loamy fine sand	0.95	6.3	3181	334804
Fiorentino Poggio Renatico 1	loam	1.07	7.4	2927	273519

III. CONCLUSION

The adsorption behavior of alpha-cypermethrin was determined on five European soils. The soils covered a range of pH from 5.4 to 7.4 and a range of organic carbon content from 0.67 to 1.72%. The soils were classified by USDA scheme into 3 different textural classes: two loamy fine sands, two sandy loams, and one loam.

The test item was fast and nearly completely adsorbed to the soil phase. This was expressed by high distribution coefficients with K_d values of 2366 to 4362 mL g^{-1}, as well as high K_{OC} values of 228622 to 353100 mL g^{-1}.

CA 7.1.3.1.2 Adsorption and desorption of metabolites, breakdown and reaction products

One adsorption/desorption study performed with alpha-cypermethrin metabolite 3-PBA was submitted and evaluated previously.

Holman J. C. 2002
^{14}C-CL 206128 (Metabolite of BAS 310 I, alphacypermethrin): Adsorption/Desorption on Soils
BASF Doc ID: AL-620-015
GLP: yes

Summary
The objective of this study was to determine the adsorption/desorption behavior of the alpha-cypermethrin (BAS 310 I) metabolite CL 206128 (3-PBA) on four soils (two silty loam soils, a sandy loam soil, and a loamy sand soil) collected in Germany,

The soil adsorption/desorption study was conducted at 20°C using the batch equilibrium method specified in OECD Guideline 106. ^{14}C-labeled 3-PBA was applied at nominal concentrations of approximately 0.05, 0.1, 0.5, 1, and 5 mg L^{-1} to 0.01 M $CaCl_2$ solution. The definitive tests were conducted in duplicate at a 1:1 soil : solution ratio and a sorption/desorption equilibrium time of 48 hours.

^{14}C-3-PBA was moderately adsorbed to all of the soils tested. The soil organic carbon normalized Freundlich adsorption coefficients (K_{OC}) values for 3-PBA ranged from 46 to 91, with an average K_{OC} of 73. Freundlich adsorption coefficients (K_{Fads}) ranged from 0.897 to 2.076 with 1/n values ranging from 0.70 to 0.86. Desorption coefficients (K_{Fdes}) ranged from 1.159 to 1.836 with 1/n values ranging from 0.58 to 0.79. A strong concentration dependence on adsorption was observed.

Report: CA 7.1.3.1.2/1
Malinsky D.S., 2005a
Adsorption/desorption of BAS 310 I metabolite (CL 912554) on soil
2004/5000486
Guidelines: EPA 163-1, EEC 91/414
GLP: yes
(certified by United States Environmental Protection Agency)

Executive Summary

The adsorption/desorption potential of CL 912554 (DCVA) was determined in five European soils, which included two loams, two loamy sands and one silt. The adsorption/desorption of ^{14}C-CL 912554 was determined using the batch equilibrium method at CL 912554 concentrations of approximately 0.250, 0.502, 1.01 and 2.10 µg a.s. g^{-1} of soil. The soils from all treatments were extracted with acetonitrile after the desorption phase of the study. The mass balance for the test soils ranged from 90 to 95% of the total applied radioactivity (TAR).

The % TAR of CL 912554 sorbed by the Bonnut, Chateauroux, Thoree les Pins, Pithiviers and Ploudalmezeau soils were 44.37, 28.07, 45.38, 18.63 and 27.76, respectively. Adsorption (K_{ads}) and desorption coefficients (K_{des}) were determined using the Freundlich equation. The K_{ads} values for the Bonnut, Chateauroux, Thoree les Pins, Pithiviers and Ploudalmezeau soils were 1.718, 0.743, 2.223, 0.514 and 0.711, respectively. There is a good correlation between the % TAR sorbed by the soils and the calculated K_{ads} values. The higher % TAR sorbed values correlated with higher K_{ads} values. The organic carbon normalized adsorption constants $K_{f,oc}$ derived from the K_{ads} values for the Bonnut, Chateauroux, Thoree les Pins, Pithiviers and Ploudalmezeau soils were 191, 57, 318, 37, and 47, respectively. These values indicate that CL 912554 will have medium mobility in Chateauroux, Pithiviers and Ploudalmezeau soils and low mobility in Bonnut and Thoree les Pins soils.
The K_{des} values (cycle 1) for the Bonnut, Chateauroux, Thoree les Pins, Pithiviers and Ploudalmezeau soils were 1.07, 1.10, 1.02, 1.03 and 1.09, respectively. All K_{des} values obtained for desorption cycle 2 were fairly similar to the values observed for desorption cycle 1.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item:	CL 912554 (DCVA)
Reg. No.:	4080830
Batch No.:	812-0101
Chemical name (IUPAC):	cis-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylic acid
Chemical purity:	99.9%
Molecular weight:	209.07 g mol^{-1}
Position of radiolabel:	cyclopropyl-1-^{14}C
Specific radioactivity:	2.68 MBq mg^{-1}
Radiochemical purity:	99.8% (by Radio-HPLC)

2. Soils

The study was conducted with five different soils originating from Europe. The physico-chemical properties of the soils are provided in Table 7.1.3.1.2-1. All soil characterization data was archived with BASF Study 131210 [*SINGH, MAHATAM, TROLLINGER, JEWEL (2003): Adsorption of ^{14}C-BAS 712 H and ^{14}C-4110778 (Metabolite of BAS 712 H) on soil, BASF DocID 2003/5000136*].

Table 7.1.3.1.2-1: Characterization of soils used to determine the adsorption/desorption behavior of CL 912554 (DCVA; alpha-cypermethrin metabolite)

Soil designation Origin	Bonnut France	Chateauroux France	Thoree les pins France	Pithiviers France	Ploudalmezeau France
Textural class	Sandy Loam	Silt Loam	Sandy Loam	Clay Loam	Clay Silt
Soil texture [%]					
Sand	59.3	12.6	78.0	11.1	13.5
Silt	31.2	63.4	12.8	48.9	74.8
Clay	9.5	24.0	9.2	40.0	11.7
Organic carbon [%]	0.9	1.3	0.7	1.4	1.5
pH	5.5	6.5	4.4	7.1	6.4
CEC [cmol$^+$ kg^{-1}]	7.5	17.0	7.5	28.8	12.0
MWHC [g/100g dry soil]	40.3	37.8	25.7	43.1	34.7

B. STUDY DESIGN

1. Experimental conditions

Preliminary tests: recovery and stability of the test item

The recovery/stability experiment was attempted several times due to the instability of ^{14}C-CL 912554 (DCVA) when exposed to light. One test solution was applied (uncovered) to deionized water to check if $CaCl_2$ contributed to the degradation of ^{14}C-CL 912554. For each test solution 125 µL of a diluted stock solution was added to 20 mL of 0.01M $CaCl_2$ solution. The concentration of ^{14}C-CL 912554 in each test solution was approximately 1.0 µg mL^{-1}. Immediately after treatment, the test solutions were assayed by liquid scintillation counting (LSC) to determine the % TAR in the solutions. Three of the test solutions were left uncovered and one was covered with aluminum foil. The test vessels were capped and placed on a shaker maintained at 20°C in the dark. After 8 hours of shaking at 140 rpm, the samples were centrifuged at 2,000 rpm for 5 minutes.
Aliquots for HPLC analysis were removed after 8, 24 and 48 hours. After 8 hours, results of HPLC analysis indicated that ^{14}C-CL 912554 in two of the three uncovered test vessels was completely degraded. In the remaining uncovered test solution ^{14}C-CL 912554 was degraded entirely after 24 h. In the $CaCl_2$ solution sample which was covered with aluminum foil) no degradation of ^{14}C-CL 912554 could be detected. The recovery remained above 101% TAR for the duration of the experiment in the covered vessel. These results indicate that ^{14}C-CL 912554 is not adsorbed to the vessel and is stable in 0.01M $CaCl_2$ solution for at least 48 hours (in the dark).

Preliminary tests: adsorption kinetics

To determine the duration to reach adsorption equilibrium a test solution was prepared by adding 1.5 mL of a diluted stock solution to 250 mL of 0.01M $CaCl_2$ solution. The concentration of ^{14}C-CL 912554 was 1.0 µg CL 912554 per mL.
The adsorption equilibrium experiment was conducted with all five soils in duplicate. Approximately 10 g of each soil was weighed into a test vessel and 20 mL of the test solution was added. The concentration of ^{14}C-CL 912554 applied to all soils was about 2.0 µg g^{-1} soil (dry weight equivalent). The test vessels were capped, covered with foil, and shaken at 140 rpm and 20°C in the dark. The % TAR of ^{14}C-CL 912554 adsorbed to the soils was determined after 4, 8, 24 and 48 hours of shaking. At the respective time interval, the soil phase and the aqueous phase were separated by centrifugation and the supernatants were radio-assayed by LSC to estimate the amount of radioactivity (% TAR) present in the supernatants.

Results of the LSC analysis of the supernatants indicated that adsorption equilibrium was reached after 48 hours. The test item was identified in the soil suspensions incubated for 24 hours by radio-HPLC.

Adsorption experiments

Three adsorption experiments were conducted. The first adsorption experiment was conducted with all five soils at concentrations of the test item of about 0.250, 0.502, 1.01 and 2.10 µg g^{-1} soil. The experiment was conducted in duplicate for each applied concentration and for a period of 48 hours.
Four test solutions were prepared with four different concentrations. The respective test solutions were prepared in 0.01 M $CaCl_2$ solution. The actual concentration was determined by LSC.
Approximately 10 g aliquots of each soil type were weighed into 40 vessels (5 soils, 4 concentrations, duplicates). To each test vessel containing the test soils 20 mL aliquots of the respective test solutions was added representing a soil to solution ratio of 1:2. After application of the respective test solution, the test vessels were capped, covered in foil, and shaken at 140 rpm and 20±1°C in the dark. After 48 hours shaking, the soil suspensions were centrifuged and aliquots of the supernatants were radio-assayed by LSC to determine the % TAR in the supernatants. The supernatants were stored in a refrigerator for later gravimetrically measurement.
Aliquots from the high dose sample supernatants were analyzed by HPLC to determine the nature of the radioactivity.

Desorption experiments

Desorption experiments were conducted for all five soils and four concentrations. After the adsorption step 20 mL of 0.01 M $CaCl_2$ was added to centrifuged and decanted soil residues. The samples were shaken again at 140 rpm and 20±1°C in the dark. After 24 hours shaking, the samples were centrifuged and aliquots were assayed by LSC to estimate the radioactivity present in the desorption supernatants. A second desorption cycle with fresh 0.01 M $CaCl_2$ solution was performed applying the same conditions. The supernatants of both desorption cycles were stored in a refrigerator for later gravimetrically measurement.
After the two desorption cycles, soil residues were extracted with 20 mL acetonitrile by shaking at 140 rpm and 20±1°C in the dark. After 24 hours shaking, the samples were centrifuged and aliquots of the acetonitrile supernatants were analyzed by LSC. Supernatant volumes had to be redetermined due to evaporation losses during refrigerator storage.
The residual soil samples were air-dried at room temperature under a fume hood. The soil residues were homogenized and aliquots were analyzed by combustion. The evolved $^{14}CO_2$ was trapped in a liquid scintillation cocktail and analyzed by LSC to determine the radioactivity remaining in soils after the adsorption and desorption experiments.

2. Description of analytical procedures

After centrifugation of the soil suspensions, the supernatant of all samples (pretests, adsorption and desorption experiment) were analyzed by LSC. In addition, aliquots of the soil suspension comprising the highest concentration of the test item (2.10 µg g^{-1} soil) were analyzed by radio-HPLC.

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The mass balance for the five soils used in the study ranged from 90 to 95% TAR comprising a relative standard deviation of < 4%.

B. FINDINGS

Adsorption data for the five European soils are presented in Table 7.1.3.1.2-2. Adsorption coefficients K_{ads} for Bonnut, Chateauroux, Thoree les Pins, Pithiviers, and Ploudalmezeau soil were 1.718, 0.743, 2.223, 0.514 and 0.711, respectively. There is good positive linear correlation between the % TAR adsorbed to the soils and the K_{ads} values obtained by the Freundlich equation. The most adsorptive soils for ^{14}C-CL 912554 (DCVA) were Thoree les Pins and Bonnut with 45.38% TAR and 44.37% TAR, respectively. The soils featuring a lower pH value exhibited higher % TAR adsorption values. Organic carbon normalized adsorption coefficients (K_{OC}) for Bonnut, Chateauroux, Thoree les Pins, Pithiviers and Ploudalmezeau were 329.1, 98.5, 547.5, 63.3 and 81.7, respectively. These K_{OC} values indicate that CL 912554 will have medium mobility in Chateauroux, Pithiviers and Ploudalmezeau soils and low mobility in Bonnut and Thoree les Pins soils.

Desorption coefficients (K_{des}; cycle 1) for the five soil ranged from 1.02 to 1.10 (Table 7.1.3.1.2-3). All K_{des} values obtained for desorption cycle 2 were fairly similar to the values observed for desorption cycle 1. R_{des1} values, expressing the percentage of desorbed ^{14}C-CL 912554 during 24 h of shaking, were about 0.16 and 0.00 for Chateauroux and Pithiviers soil, respectively (Table 7.1.3.1.2-4). Results indicate limited desorption of CL 912554 from these soil types. Bonnut and Thoree les Pins had R_{des1} values of 0.43 and 0.31, respectively, which indicate high desorption possibility of CL 912554 to these soils. The R_{des1} value for Ploudalmezeau was 0.24, which suggest moderate desorption of adsorbed CL 912554 from this soil.

The acetonitrile extracts from Bonnut, Chateauroux, Thoree les Pins, Pithiviers, and Ploudalmezeau soil contained an average of 13.16, 8.21, 22.54, 5.76, and 5.72% TAR, respectively. The % TAR still bound to Bonnut, Chateauroux, Thoree les Pins, Pithiviers and Ploudalmezeau soil accounted for 9.62, 17.12, 8.40, 13.88, and 12.19% TAR, respectively. These results demonstrate that some amounts of CL 912554 bound to the soils cannot be desorbed. The highly adsorptive soils (Bonnut and Thoree les Pins) exhibited smaller amounts of non-extractable CL 912554 compared to the moderately adsorptive soils (Chateauroux, Pithiviers and Ploudalmezeau).

Table 7.1.3.1.2-2: Soil adsorption data of CL 912554 (DCVA)

Soil	Intercept (log K_{ads})	K_{ads}	Slope (1/n)	R^2
Bonnut	0.235	1.718	0.836	0.9997
Chateauroux	-0.129	0.743	0.692	0.9948
Thoree les Pins	0.347	2.223	0.889	0.9969
Pithiviers	-0.289	0.514	0.754	0.9784
Ploudalmezeau	-0.148	0.711	0.728	0.9826

Table 7.1.3.1.2-3: Soil desorption data of CL 912554 (DCVA)

Soil	Intercept (log $_{des}$)		K_{des}		Slope (1/n)		R^2	
	cycle 1	cycle 2	cycle 1	cycle 2	cycle 1	cycle 2	cycle 1	cycle 2
Bonnut	0.03030	0.01279	1.07	1.03	2.68630	3.98187	0.99712	0.99230
Chateauroux	0.04112	0.04029	1.10	1.10	2.31446	5.73467	0.98762	0.98889
Thoree les Pins	0.00965	0.00439	1.02	1.01	3.69108	4.76941	0.99688	0.99455
Pithiviers	0.01408	0.02007	1.03	1.05	2.08476	4.90491	0.97878	0.97486
Ploudalmezeau	0.03544	0.01979	1.09	1.05	2.13107	5.86968	0.96734	0.93884

Table 7.1.3.1.2-4: R_{des} and $K_{f.oc}$ values derived from soil adsorption/desorption data of CL 912554 (DCVA)

Soil	R_{des1}	R_{des2}	$K_{f,oc}$ [a]
Bonnut	0.43	0.44	191
Chateauroux	0.16	0.23	57
Thoree les Pins	0.31	0.39	318
Pithiviers	0.00	0.30	37
Ploudalmezeau	0.24	0.27	47

[a] Recalulated values; $K_{f,oc}$ from study were calculated erroneously.

III. CONCLUSION

The adsorption/desorption behavior of CL 912554 (DCVA) was determined in five European soils, which included two loams, two loamy sands and one silt. These soils covered a pH range from 4.4 to 7.1 and a range of organic carbon content from 0.7% to 1.5%.

The $K_{f,oc}$ values for CL 912554 were 191 and 318 for both sandy loam (Bonnut and Thoree les Pins) soils. These results suggest that CL 912554 is moderately adsorbed to sandy loam soils types. The $K_{f,oc}$ values for the loam (Chateauroux, Pithiviers, and Ploudalmezeau) soils ranged from about 37 to 57, which suggest that CL 91254 is slightly adsorbed to these soil types.

R_{des1} values for Chateauroux and Pithiviers soils were about 0.16 and 0.00, respectively. These results indicate limited desorption of CL 912554 from these soil types. Bonnut and Thoree les Pins had R_{des1} values of 0.43 and 0.31, respectively, which indicate a comparatively high desorption capacity of CL 912554 from these soils. The R_{des1} value for Ploudalmezeau was 0.24, which suggest moderate desorption of adsorbed CL 912554 from this soil.

Report:	CA 7.1.3.1.2/2 Walter W., 2014a Determination of Adsorption Behavior of M310I017 cis (metabolite of BAS 310 I, Alpha-Cypermethrin) in 5 Soils (OECD Guideline 106) 2014/1315294
Guidelines:	OECD Guideline 106
GLP:	yes (certified by Landesanstalt fuer Umwelt, Messungen und Naturschutz, Baden-Württemberg, Karlsruhe, Germany)

Executive Summary

In laboratory experiments the adsorption behavior of alpha-cypermethrin metabolite M310I017 cis (containing trans isomers) was investigated on five European soils. The five tested soils covered a range of pH ($CaCl_2$) from 5.4 to 7.4, a range of organic carbon content from 0.67 to 1.72%, and three different USDA textural classes: two loamy fine sands, two sandy loams, and one loam. Due to the low water solubility of M310I017 (cis/trans), adsorption was only tested for one concentration level (25 ng mL^{-1}). Hence, no adsorption or desorption isotherm were determined. The ratio of soil versus test solution was 1/50, and the measurements were performed at the adsorption equilibrium time of 4 h.

Sorption of M310I017 cis to soil proceeded fast, expressed by ≥ 96% of the initially applied amount of the test item adsorbed after an equilibration time of 4 h. Hence, almost the entire applied amount of the test item was accounted to the fraction sorbed to the soil phase. Calculated mean distribution coefficients (K_d) ranged from 1738 mL g^{-1} (LUFA 2.3) to 3914 mL g^{-1} (Fiorentino Poggio Renatico 1). Distribution coefficients normalized to the fraction of organic carbon in the soils (K_{OC}) ranged from 139148 mL g^{-1} (LUFA 2.2) to 365806 mL g^{-1} (Fiorentino Poggio Renatico 1).

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item:	M310I017 cis (metabolite of alpha-cypermethrin)
Reg. No.:	6009306
Batch No.:	L82-159
Chemical name (IUPAC):	cyano[3-(4-hydroxyphenoxy)phenyl]methyl (1RS,3RS)-3-(2,2-dichloroethenyl)-2,2-dimethylcyclopropanecarboxylate
Chemical purity:	48.5%
Molecular weight:	432.3 g mol^{-1}

Beside cis isomer metabolites (containing cis-1 and cis-2 isomers), the test item contained an equivalent portion of [redacted] Finally, the sum of all of them was tested.

2. Soils

The study was conducted with five different soils originating from Europe. The physico-chemical properties of the soils are provided in Table 7.1.3.1.2-5.

Table 7.1.3.1.2-5: Characterization of soils used to determine the adsorption behavior of M310I017 cis

Soil designation Origin	**LUFA 2.2 Germany**	**LUFA 2.3 Germany**	**Bruch West Germany**	**Li10 Germany**	**Fiorentino Poggio Renatico 1 Italy**
Textural class (DIN 4220)	Loamy sand (Sl2)	Silty sand (Su3)	Loamy sand (Sl3)	Loamy sand (Sl2)	Loamy sand
Soil texture [%], (ISO 11277)					
Sand	85.0	62.9	66.7	81.5	41.7
Silt	10.0	29.5	22.2	13.3	41.6
Clay	5.0	7.6	11.1	5.2	16.7
Textural class (USDA scheme)	Loamy fine sand	Sandy loam	Sandy loam	Loamy fine sand	Loam
Soil texture [%], (USDA)					
Sand	87.4	64.1	69.9	83.4	49.4
Silt	7.7	28.3	18.9	11.4	33.9
Clay	5.0	7.6	11.1	5.2	16.7
Organic carbon [%] (ISO 10694)	1.72	0.67	1.62	0.95	1.07
Cation exchange capacity [$cmol^+ kg^{-1}$]	4.2	6.2	11.6	7.6	11.2
pH ($CaCl_2$)	5.8	5.4	7.1	6.3	7.4
pH (water)	6.4	6.1	8.0	7.0	8.3
Max. water holding capacity [$g\ 100g^{-1}$ dry soil]	33.1	23.9	25.5	24.4	29.7
Bulk density [$g\ L^{-1}$]	1253	1353	1382	1358	1403

B. STUDY DESIGN

1. Experimental conditions

Preliminary tests: Determination of solubility, adsorption kinetics, and soil/solution ratio

The solubility of the test item (M310I017 cis/trans) in water was tested in four experiments applying the column elution method (OECD 105) with demineralized water and demineralized water containing 0.1% acetonitrile at flow rates of 0.2 and 0.4 mL min^{-1}. The solubility in water was determined to be of about 72 µg mL^{-1} at 0.2 mL min^{-1} flow rate at 20°C and slightly higher when containing 0.1% acetonitrile.

Adsorption was assumed to be rather high and the analytes stability appeared to be questionable during equilibration time of 24 h. Therefore, both the direct (i.e. determination in 0.01 M $CaCl_2$ aqueous phase and in soil by appropriate solvent extraction) and the parallel (i.e. individual soil/solution samples) method procedures were used.

A preliminary experiment was run with two soils (LUFA 2.2 and Fiorentino Poggio Renatico 1) to determine the time needed to establish equilibrium conditions and to find the optimal soil/solution ratio for the adsorption tests. The experiments were run with a soil/solution ratio of 1/50, using 1 g soil, 50 mL 0.01 M $CaCl_2$ solution in PE centrifuge vials. After equilibration, soil suspensions were treated with about 1250 ng of the test item, corresponding to an initial concentration of about 25 ng mL^{-1}. Per soil type, a total of 10 soil samples were treated (duplicates per equilibration time), plus blank soil samples without dosing for blank controls. The samples were shaken at 20 - 21°C for 2, 4, 6 and 24 h. The soil / solution suspension was then centrifuged and decanted. An aliquot of the supernatant was sampled and the concentration of the test item in the $CaCl_2$ solution was determined by high performance liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS). A complete mass balance was obtained by the extraction of the soil pellets.

The amount of the test item in the aqueous phase was too low to be measured in the preliminary adsorption tests. The test item adsorbed to a very high extend on soil and the surface of fresh centrifugation tubes used for the long high speed centrifugation (30 min at 10000 rpm) of the aqueous phase.

The percentage of adsorption to soil was stable with prolonged equilibration time (≥ 95% of the initially applied amount of the test item), reaching the plateaus after 2 h (LUFA 2.2) and 4 h (Fiorentino Poggio Renatico 1) of equilibration time. Hence, further experiments were conducted with an equilibration time of 4 h.

Stability and adsorption of the test substance

The K_d values obtained with the direct method were all > 1500 mL g^{-1}, thus indicating that adsorption/desorption isotherm experiments would lead to inaccurate results. Therefore, these experiments were not performed.

The adsorption test was performed as described for the preliminary test in duplicates for all soils (including the soils used in the pre-tests), but with 4 h equilibration time only. It was possible to determine a concentration of the test tem in the soil water phase by modifying the centrifugation procedures.

Stability of the test item in aqueous $CaCl_2$ solution was demonstrated by acceptable results for adsorption controls obtained by LC-MS/MS analyses.

Control samples (dose and adsorption controls) with only test item in aqueous 0.01 M $CaCl_2$ solution were used to elucidate a potential adsorption of the test item to the surface of the test vessels. Dose controls were analyzed right after adding solution to soil; adsorption controls were analyzed after termination of agitation together with the samples.

In addition, experiments were conducted in order to estimate the adsorption to test vessel surface in presence of soil. Therefore, two experiments were performed in parallel, each one in duplicate. 1.0 g of soil was agitated with 50 mL solution for 6 h. Afterwards, phases were separated by centrifugation and decantation. One sample of the soil pellet was extracted with solvent directly in the test vessel. From the second soil sample, the soil pellet was removed as good as possible and transferred directly into extraction solvent present in another vessel equivalent to the test vessel. The difference in detected amounts of the test item between the two samples was used to estimate the surface adsorption in the test vessels.

2. Description of analytical procedures

The parallel method was performed, including the determination of the amount of the test item in the aqueous phase (direct method), as well as the determination of the amount adsorbed to soil via solvent extraction.

Principle of the methods:

To analyze M310I017 cis in the aqueous phase, soil suspensions were transferred to glass centrifuge tubes, and separated by centrifugation (5 min, 4000 rpm). Aliquots of the supernatants were centrifuged for 30 min at 15000 rpm to remove soil particles with a diameter of > 0.2 µm. Aliquots were diluted with methanol/water 8/2 (v/v) for LC-MS/MS analysis.

In the main adsorption study, the test vessel was additionally rinsed with the supernatant after centrifugation of the soil suspensions (to transfer the remaining soil particles) to account for amounts of the test item adsorbed to the walls of the test vessels. Afterwards, the rinse was added to the centrifuge tube and was centrifuged once again to separate soil and soil water phase. As the test item adsorbed completely on the surface of the centrifugation tubes during the long high speed centrifugation of the pre-tests, this step was left out in the main adsorption experiment.

For soil analysis, soil samples (1.0 g) were extracted on a mechanical shaker (30 min) with 5 mL acetonitrile/water (1:1, v/v) containing 0.1% formic acid, followed by sonication (10 min). After extraction, the tube was centrifuged at 4000 rpm (5 min). The extraction was repeated two more times with each 5 mL acetonitrile containing 0.1% formic acid. The three extracts were combined and diluted to 20 mL with 0.1% formic acid in water. An aliquot from the pooled extract was further diluted with methanol/water 8/2 (v/v) and analyzed by LC-MS/MS.

To account for the amount of test item adsorbed to the test vessel surface in the presence of soil (main adsorption experiment), glass centrifuge vials (used for adsorption kinetics experiments) were extracted with 20 mL acetonitrile/water (1/1, v/v) on a horizontal shaker (30 min) and by sonication (10 min). An aliquot was diluted with methanol/water 8/2 (v/v) and analyzed by LC-MS/MS.

The soil extraction method, as well as the method for the determination of the test item in the aqueous phase were pre-validated using the soils LUFA 2.2 and Fiorentino Poggio Renatico 1 at two fortification levels (i.e. 25 and 2500 ng g^{-1} (soil) and 0.25 and 25 ng mL^{-1} (aqueous phase), respectively). During the experiments the soil extraction method was concurrently validated for all five soils at two fortification levels (i.e. 12.5 and 1250 ng g^{-1}).

The limit of quantification (LOQ) of the analytical methods were 0.25 µg mL^{-1} (aqueous phase) and 12.5 ng mL^{-1} (soil), respectively. Corresponding limits of detection (LOD) were 0.02 µg mL^{-1} and 0.9 ng mL^{-1} (< 10% of LOQ), respectively.

II. RESULTS AND DISCUSSION

A. MASS BALANCE

In the adsorption experiment, the average recovery values after soil extraction ranged from 95% to 107% for all soils, with the exception of soil Fiorentino Poggio Renatico 1 with only 75% recovery on average after 4 h equilibration time. This indicated degradation and/or formation of bound residues with prolonged equilibration time for this soil.

B. FINDINGS

Pre-validation of the soil extraction method, as well as the method for the determination of the test item in the aqueous phase resulted in overall average recoveries of 108% (relative standard deviation RSD: 4%) for the soil extraction and of 82% (RSD: 20%) for the method applied for the determination in the aqueous phase. During the experiments, the soil extraction method was concurrently validated for all five soils at two fortification levels (i.e. 12.5 and 1250 ng g^{-1}), resulting in an overall average recovery of 100% (RSD: 13%).

Results of the adsorption equilibrium test carried out with alpha-cypermethrin metabolite M310I017 cis indicated that the adsorption equilibrium was reached within 4 h.

Control specimens with only the test item in aqueous 0.01 M $CaCl_2$ solution (dose controls, diluted directly after fortifying and adsorption controls, diluted after appropriate equilibration time) revealed significant adsorption on the surface of the test vessels when soil is not present in the system.

Results of control samples, where the test item was applied to $CaCl_2$ solution incubated with soil revealed that adsorption on the surface of the test vessel when soil is present in the system is negligible.

Sorption of the test item to soil proceeded fast. This was expressed by ≥ 96% of the initially applied amount of the test item adsorbed to the soil phase after an equilibration time of 4 h. Hence, almost the entire applied amount of the test item was accounted to the fraction sorbed to the soil phase. Calculated mean distribution coefficients (K_d) ranged from 1738 mL g^{-1} (LUFA 2.3) to 3914 mL g^{-1} (Fiorentino Poggio Renatico 1). Distribution coefficients normalized to the fraction of organic carbon in the soils (K_{OC}) ranged from 139148 mL g^{-1} (LUFA 2.2) to 365806 mL g^{-1} (Fiorentino Poggio Renatico 1). Calculated K_d as well as K_{OC} values are presented in Table 7.1.3.1.2-6.

Table 7.1.3.1.2-6: Measured equilibrium concentrations and calculated distribution coefficients K_d and K_{OC} of M310I017 cis

Soil	Soil Type (USDA)	Org. C [%]	pH ($CaCl_2$)	Adsorption	
				K_d [mL g^{-1}]	K_{OC} [mL g^{-1}]
LUFA 2.2	sandy loam	1.72	5.8	2393	139148
LUFA 2.3	loamy fine sand	0.67	5.4	1738	259437
Bruch West	sandy loam	1.62	7.1	2819	174041
Li10	loamy fine sand	0.95	6.3	2956	311179
Fiorentino Poggio Renatico 1	loam	1.07	7.4	3914	365806

III. CONCLUSION

The adsorption behavior of alpha-cypermethrin metabolite M310I017 cis (Reg. No. 6009306; containing trans isomers) was determined on five European soils. The soils covered a range of pH from 5.4 to 7.4 and a range of organic carbon content from 0.67 to 1.72%. The soils were classified by USDA scheme into 3 different textural classes: two loamy fine sands, two sandy loams, and one loam.

The test item was fast and nearly completely adsorbed to the soil phase. This was expressed by high distribution coefficients with K_d values of 1738 to 3914 mL g^{-1}, as well as high K_{OC} values of 139148 to 365806 mL g^{-1}. Due to the high K_d values obtained for all soils and the low water solubility of the test item, no adsorption/desorption experiments were conducted.

CA 7.1.3.2 Aged sorption

No studies to investigate aged sorption were conducted with alpha-cypermethrin.

CA 7.1.4 Mobility in soil

CA 7.1.4.1 Column leaching studies

The following studies were submitted and peer-reviewed (See Table 7.1.4.1-1).

Table 7.1.4.1-1: List of peer-reviewed column leaching studies performed with cypermethrin and cypermethrin metabolites

DocID	Parent compound	Soil	Application rate [mg kg^{-1}]	Incubation temperature	Incubation period [days]	Remark
Cy-905-040	Cypermethrin DCVA PBA PBAld	Silty clay Silty clay loam Loamy sand	0.012	n/a	Extraction after leachate percolation	Kaufman 1981 Publ.
CY-620-001	Cypermethrin	Sandy clay loam	18.9	n/a	28	Standen 1977
CY-620-002	Cypermethrin	Sandy loam	0.134	n/a	28	Jackson 1977
Cy-620-005	Cypermethrin	Clay loam Loamy sand Coarse sand Fen peat	0.846 1.269	25°C	21	Stevens 1980

n/a = not available

CA 7.1.4.1.1 Column leaching of the active substance

1. Standen M.E. (1977) The leaching and the degradation of the insecticide WL 43467 when applied as sheep-dip solution to soil
Study performed with formulation ingredients from a veterinary medicine study. Not useful to derive endpoints.

2. Jackson C. (1977) The leaching of WL 434567 through laboratory soil columns
Poorly documented study that shows that cypermethrin has no tendency to leach through the soil columns. Not useful to derive endpoints.

3. Stevens J.E.B. et al (1980) Cypermethrin: Mobility of cypermethrin and its degradation products in soil columns
The mobility of aged cypermethrin residues was investigated in four soils. After applying 67 cm of simulated rain less than 2 % of the applied radioactivity was detected in the leachates.

CA 7.1.4.1.2 Column leaching of metabolites, breakdown and reaction products

1. Kaufman D.D.et al. (1981) Movement of cypermethrin, decamethrin, permethrin, and their degradation products in soil (Publication)
Study focused on decametrin, but also the studies the leaching behaviour of the common metabolites DCVA, 3-PBA and 3-PBald. As expected the metabolites show mobility in soil, while the active substances do not move into deeper layers of soil. Study not useful to derive endpoints.

CA 7.1.4.2 Lysimeter studies

Due to the high sorption of the active substance to soil, no leaching studies were performed.

CA 7.1.4.3 Field leaching studies

Due to the high sorption of the active substance to soil, no field leaching studies were performed.

CA 7.2 Fate and behaviour in water and sediment

CA 7.2.1 Route and rate of degradation in aquatic systems (chemical and photochemical degradation)

A range of hydrolysis studies have been performed with cypermethrin or alpha-cypermethrin (see Table 7.2.1-1). The cypermethrin studies are listed for historical reasons and can be replaced by studies performed with alpha-cypermethrin. Data generated with alpha-cypermethrin and submitted in the previous Annex-I review gave a clear picture of the fate of alpha-cypermethrin in aquatic systems

Table 7.2.1-1: List of hydrolytic and photochemical degradation studies in aquatic systems performed with (alpha-) cypermethrin

DocID	Parent compound	pH	Application rate [mg L-1]	Incubation temperature	Incubation period [hours]	Remark
CY-905-038	Cypermethrin	6.3 8.7 8.3	0.05	room temperature	10	Takahashi 1985 Publ.
CY-905-039	(1R,cis,αRS)-cypermethrin; (1R,trans,αRS)-cypermethrin	3 (dest. water) 7 (dest. water) 11 (dest. water) 8 (sea water) 8 (river water)	n/a	25°C	n/a	Takahashi 1985 Publ.
CY-630-001	Cypermethrin	4.8 7.0 9.3	1.0	outdoor	32 (days)	Day 1980
AL-322-001	Alpha-cypermethrin	5 7 9	0.5-0.6	60, 70, 80°C 50, 60, 70°C 40, 60, 70°C	280, 188, 95 141, 117, 125 1, 0.9, 0.9	Salisbury 1984
AL-322-002	Alpha-cypermethrin	4 7 7 7 9 9	0.020-0.021 0.0011	50°C 50°C 60°C 75°C 25°C 50°C	10 10 11 4 11 24	v. Dijk 1993
AL-630-009	Alpha-cypermethrin	7.1	0.00185	25°C	3.2 24	Fisk 1994
AL-324-003	Alpha-cypermethrin	5	0.002	22°C	28	Concha 2001

n/a = not available

Studies performed with cypermethrin. Not considered in evaluation

1. Takahashi N. et al. (1985) Photodegradation of the pyrethroid insecticide cypermethrin in water and on soil surface (Publication)
The photolytic degradation of cis- and trans isomers of cypermethrin was investigated in water. The samples were exposed to natural sunlight. Rapid degradation was observed in distilled water which could be enhanced further with acetone. Multiple degradation compounds could be identified. No guideline applied.

2. Takahashi N. et al. (1985) Hydrolysis of the pyrethroid insecticide cypermethrin in aqueous media (Publication)
Mechanistic investigation into the hydrolysis of cis- and trans isomers of cypermethrin under different pH values. The cleavage of the ester bond is faster than the hydration of the CN-group.
At pH lower than 7 cypermethrin is stable in sterile water whereas the hydrolytic degradation increases with higher pH values. No guideline applied.

3. Day S. R. et al. (1980) 14C Cypermethrin: Aqueous photodegradation in sunlight
Cypermethrin in sterile water solutions containing acetonitrile as co-solvent were exposed to natural sunlight (outdoor conditions) for 32 days. Only 10% degradation was observed at the end of the study. Dark controls showed no significant degradation. No guidelines applied.

CA 7.2.1.1 Hydrolytic degradation

Two hydrolysis studies performed with alpha-cypermethrin were submitted and evaluated previously.

Salisbury K., Weaver R. C. and Lagner E.J. 1984
The hydrolysis of FASTAC (WL85871)
BASF Doc ID: AL-322-001
GLP: no

Summary

The hydrolysis of alpha-cypermethrin at pH 5, 7 and 9 has been measured in aqueous buffer solutions containing 1% acetone. The experiments were conducted at elevated temperatures:
pH 5 at 60, 70 and 80°C, pH 7 and pH 9 at 50, 60 and 70°C. The sampling time was adjusted to the differing rate of degradation, but for at least 7 sampling points. Calculation of the degradation half- life was performed by linear regression ln Co/Ct versus time and recalculated for 22°C by using the Arrhenius equation. Half-lives at 22°C were 162 days, 46 days and 2.9 hour at pH 5, pH 7 and pH 9 respectively. The main degradation products (which were not quantified) were PBA and DCVA.
In a second experiment the epimerization of alpha-cypermethrin was investigated at ambient temperature at pH 9. Complete epimerization was found in 30 minutes. At elevated temperature and lower pH tome for epimerization increased, but was in any case faster than hydrolytic cleavage of the molecule.

Van Dijk A. 1993
Hydrolysis determination of 14C Alpha-cypermethrin at different pH values
BASF Doc ID: AL-322-002
GLP yes

Summary

In the present study, the rate (DT50) of hydrolysis of alpha-cypermethrin was determined in aqueous buffer solutions at pH 4, 7, and 9 at 25°C. Alpha-cypermethrin was incubated under sterile conditions and nitrogen at a concentration of 0.020 – 0.021 µg mL-1 at pH 4 and 7 and temperatures of ~~30~~ 50°C (both pH values), 60°C, and 75°C (only pH 7). At pH 9, 14C-alpha-cypermethrin was incubated at a concentration of 0.0011 µg mL-1 at 25°C and 50°C.
Except for one outlier (89.5% of the total applied radioactivity (TAR)), radioactivity was completely recovered in the organic phase, ranging from 96.2 to 100.0% TAR.
In the aqueous solution at pH4, exclusively the parent compound was found until 10 days of incubation at 50°C, indicating that alpha-cypermethrin was stable against hydrolysis at acidic pH.
At pH 7, alpha-cypermethrin was hydrolyzed with half-lives of 5.3 and 2.0 days at 60°C and 75°C, respectively. At pH 9, alpha-cypermethrin was hydrolyzed with half-lives of 3.5 and 3.0 hours at 25°C and 50°C, respectively.
Besides the parent compound, almost exclusively the hydrolysis product 3-PBAld occurred throughout all incubations. At pH 7, 3-PBAld was detected in maximum amounts of 21.6% (50°C), 71.7% (60°C), and 61.5% (75°C). At pH 9, maximum 88.4% (25°C) and 92.1% TAR (50°C) were measured. A more polar and unknown hydrolysis product occurred only in small amounts below 10% TAR at pH 7 and below 5% TAR at pH 9.
In conclusion, after applying data analyses via the Arrhenius equation at pH 7, half-lives of 101 and 67 days were calculated at 20°C and 25°C for alpha-cypermethrin, respectively. At pH 9 the corresponding half-lives of alpha-cypermethrin were 7.3 and 3.5 days, respectively.

To support the studies two additional hydrolysis studies were conducted and are submitted under CA 7.2.1.1/1 and CA 7.2.1.1/2.

Report: CA 7.2.1.1/1
Hassink J., 2005a
Hydrolysis of Alpha-Cypermethrin (TGAI batch COD-000165)
2005/1016375
Guidelines: EPA 161-1, SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995), EEC 94/37, EEC 91/414
GLP: yes
(certified by Landesamt fuer Umweltschutz und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

A hydrolysis study was conducted for technical alpha-cypermethrin, TGAI batch COD-000165. Therefore, the test item was incubated in sterile buffer solution at 25°C over a study period of 30 days at pH 4, 5, 7 and 9. The study was designed to investigate the degradation rate of the test item in aqueous solutions to meet the requirements of both the EU and EPA guidelines.

The test item is stable in acidic media (pH 4), but the decline at pH 5 to 70% of the initial applied test item after 30 days and at pH 7 to 66% after 30 days indicate slow hydrolysis of alpha-cypermethrin with increasing pH. At pH 9 a significant decline to 66% of the initial applied test item after one day and to 10% after 16 days confirm the rapid hydrolysis of alpha-cypermethrin under alkaline conditions (DT50 pH: 2.9 h [SALISBURY, WEAVER, LANGNER (1984): THE HYDROLYSIS OF FASTAC (WL85871), BASF DocID AL-322-001]).

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item: alpha-cypermethrin
Reg. No.: 4078193
Batch No.: COD-000165 (TGAI)
Chemical name (IUPAC): racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-isomer
Purity: 98.8%
Molecular weight: 832.60 g mol-1 (2 x 416.30 g mol-1)
Molecular formula: 2 C22H19Cl2NO3

2. Test system

All buffer solutions were prepared with commercially available buffers (Titrisol, Merck, Darmstadt, Germany) according to the manufacturer's instructions. Thereafter, the solutions were diluted by the factor 10 to avoid interactions with the test item. The following buffer solutions were used:

pH 4: Titrisol 1.09884 (citrate – HCl)
pH 5: Titrisol 1.09885 (citrate – NaOH)
pH 7: Titrisol 1.09887 (phosphate)
pH 9: Titrisol 1.09889 (boric acid/KCl – NaOH)

B. STUDY DESIGN

1. Experimental conditions

To test the hydrolysis at different pH values, different volumes of an application solution, containing 0.5 µg mL-1 of the test item dissolved in methanol/water (50/50, v/v), were pipetted into 500 mL of the respective sterilized buffer. 0.35 mL (pH 4 and 5), 2.00 mL (pH 7), and 2.25 mL (pH 9) were diluted in the respective buffer solution reaching a nominal concentration of 0.35 (pH 4 and 5), 2.00 (pH 7), and 2.25 µg L-1 (pH 9), respectively.

Subsets of 50 mL were used for each hydrolysis experiment. The sterile samples were stored in a climatic chamber at the required temperature of 25°C for 30 days. Sterility of the solutions was checked at each sampling time for the respective sample prior to analysis. The pH was checked for each sample after analysis.

2. Sampling

The sampling was performed 0, 1, 2, 7, 10, 16, 21 and 30 days after treatment.

3. Description of analytical procedures

Samples were extracted three times with 30 mL ethyl acetate. The extracts were combined and evaporated to dryness. The residue was dissolved in 50 mL methanol/water (50/50, v/v) and subsets of 20 µL were analyzed by high performance liquid chromatography/tandem mass spectrometry (HPLC-MS/MS).

II. RESULTS AND DISCUSSION

A. FINDINGS

Recoveries of 92-104% (pH 4), 70-111% (pH 5), 66-104% (pH 7), and 6-66% (pH 9) of the initial applied amount of the test item were obtained in the test systems during the testing period of 30 days. The decline at pH 5 to 70% after 30 days and at pH 7 to 66% after 30 days indicate slow hydrolysis of alpha-cypermethrin with increasing pH. Rapid hydrolysis occurred under alkaline conditions, i.e. only 66% of the initial applied amount was present after one day, about 10% were detected in the test system after 16 days. Results are given in Table 7.2.1.1-1.

Table 7.2.1.1-1: Hydrolysis of alpha-cypermethrin (TGAI batch COD-000165) at pH 4-9 and 25°C

DAT	pH 4		pH 5		pH 7		pH 9	
	[µg L-	[%]*	[µg L-	[%]*	[µg L-	[%]*	[µg L-	[%]*
0	0.347	100.0	0.350	100.0	1.52	100.0	2.50	100.0
1	0.346	99.7	0.332	94.9	1.48	97.4	1.66	66.4
2	0.331	95.4	0.387	110.6	1.18	77.6**	1.30	52.0
7	0.359	103.5	0.358	102.3	1.45	95.4	0.791	31.6
10	0.329	94.8	0.359	102.6	1.58	103.9	0.557	22.3
16	0.320	92.2	0.329	94.0	1.43	94.1	0.244	9.8
21	0.332	95.7	0.278	79.4	1.08	71.1	0.287	11.5
30	0.352	101.4	0.244	69.7	1.01	66.4	0.159	6.4

* % of initial applied test item, concentration of day 0 set to 100%
** outlier, result not plausible

B. ESTIMATION OF HALF-LIVES

No estimation of half-lives was done since the results on the hydrolysis of technical alpha-cypermethrin confirm the pH dependency determined previously with the pure test item (DT50 pH 9: 2.9 h [BASF DocID AL-322-001].

III. CONCLUSION

Technical alpha-cypermethrin (TGAI batch COD-000165) is stable in acidic media (pH 4), but the rate of hydrolysis increases with increasing pH. At pH 9 a significant decline to 66% of the initial applied amount of test item after one day and to 10% after 16 days confirm the rapid hydrolysis of alpha-cypermethrin under alkaline conditions (DT50 pH 9: 2.9 h [BASF DocID AL-322-001]).

Report:	CA 7.2.1.1/2 Hassink J., 2005b Hydrolysis of Alpha-Cypermethrin (TGAI batch COD-000166) 2005/1016376
Guidelines:	EPA 161-1, SETAC Procedures for assessing the environmental fate and ecotoxicity for pesticides (March 1995), EEC 94/37, EEC 91/414
GLP:	yes (certified by Landesamt fuer Umweltschutz und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

A hydrolysis study was conducted for technical alpha-cypermethrin, TGAI batch COD-000166. Therefore, the test item was incubated in sterile buffer solution at 25°C over a study period of 30 days at pH 4, 5, 7 and 9. The study was designed to investigate the degradation rate of the test item in aqueous solutions to meet the requirements of both the EU and EPA guidelines.

The test item is stable in acidic media (pH 4), but the decline at pH 7 to 80% after 21 days indicate slow hydrolysis of alpha-cypermethrin with increasing pH. At pH 9 a significant decline to 66% of the initial applied amount of test item after one day and to 1-3% after 16 days confirm the rapid hydrolysis of alpha-cypermethrin under alkaline conditions (DT50 pH: 2.9 h [BASF DocID AL-322-001]).

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Test item: alpha-cypermethrin
Reg. No.: 4078193
Batch No.: COD-000166 (TGAI)
Chemical name (IUPAC): racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-isomer
Purity: 99.3%
Molecular weight: 832.60 g mol-1 (2 x 416.30 g mol-1)
Molecular formula: 2 C22H19Cl2NO3

2. Test system

All buffer solutions were prepared with commercially available buffers (Titrisol, Merck, Darmstadt, Germany) according to the manufacturer's instructions. Thereafter, the solutions were diluted by the factor 10 to avoid interactions with the test item. The following buffer solutions were used in dilution 1:9, buffer : bidest. water:

pH 4: Titrisol 1.09884 (citrate – HCl)
pH 5: Titrisol 1.09885 (citrate – NaOH)
pH 7: Titrisol 1.09887 (phosphate)
pH 9: Titrisol 1.09889 (boric acid/KCl – NaOH)

B. STUDY DESIGN

1. Experimental conditions

To test the hydrolysis at different pH values, different volumes of an application solution, containing 0.5 µg mL-1 of the test item dissolved in methanol/water (50/50, v/v), were pipetted into 500 mL of the respective sterilized buffer. 0.35 mL (pH 4 and 5), 2.00 mL (pH 7), and 2.25 mL (pH 9) were diluted in the respective buffer solution reaching a nominal concentration of 0.35 (pH 4 and 5), 2.00 (pH 7), and 2.25 µg L-1 (pH 9), respectively.

Subsets of 50 mL were used for each hydrolysis experiment. The sterile samples were stored in a climatic chamber at the required temperature of 25°C for 30 days. Sterility of the solutions was checked at each sampling time for the respective sample prior to analysis. The pH was checked for each sample after analysis.

2. Sampling

The sampling was performed 0, 1, 2, 7, 10, 16, 21 and 30 days after treatment.

3. Description of analytical procedures

Samples were extracted three times with 30 mL ethyl acetate. The extracts were combined and evaporated to dryness. The residue was dissolved in 50 mL methanol/water (50/50, v/v) and subsets of 20 µL were analyzed by high performance liquid chromatography/tandem mass spectrometry (HPLC-MS/MS).

II. RESULTS AND DISCUSSION

A. FINDINGS

Recoveries of 97-123% (pH 4), 93-117% (pH 5), 80-113% (pH 7), and 1-66% (pH 9) of the initial applied amount of the test item were obtained in the test systems during the testing period of 30 days. The decline at pH 7 to 80% after 21 days indicates slow hydrolysis of alpha-cypermethrin with increasing pH. Rapid hydrolysis occurred under alkaline conditions, i.e. only 66% of the initial applied amount was present after one day, about 1-3% were detected in the test system after 16 days. Results are given in Table 7.2.1.1-2.

Table 7.2.1.1-2: Hydrolysis of alpha-cypermethrin (TGAI batch COD-000166) at pH 4-9 and 25°C

DAT	pH 4		pH 5		pH 7		pH 9	
	[µg L-1]	[%]*	[µg L-1]	[%]*	[µg L-1]	[%]*	[µg L-1]	[%]*
0	0.271	100.0	0.158	100.0	1.51	100.0	1.78	100.0
1	0.332	122.5	0.163	103.2	1.49	98.7	1.18	66.3
2	0.278	102.6	0.162	102.5	1.43	94.7	0.722	40.6
7	0.305	112.5	0.151	95.6	1.71	113.2	0.538	30.2
10	0.264	97.4	0.157	99.4	1.44	95.4	0.228	12.8
16	0.293	108.1	0.0847	53.6**	1.34	88.7	0.0422	2.4
21	0.317	117.0	0.147	93.0	1.20	79.5	0.0133	0.7
30	0.319	117.7	0.185	117.1	1.23	81.5	0.0497	2.8

*% of initial applied test item, concentration of day 0 set to 100%
** outlier, result not plausible

B. ESTIMATION OF HALF-LIVES

No estimation of half-lives was done since the results on the hydrolysis of technical alpha-cypermethrin confirm the pH dependency determined previously with the pure test item (DT50 pH 5: 162 d; DT50 pH 7: 46 d, DT50 pH 9: 2.9 h [BASF Doc ID AL-322-001].

III. CONCLUSION

Technical alpha-cypermethrin (TGAI batch COD-000166) is stable in acidic media (pH 4), but the rate of hydrolysis increases with increasing pH. At pH 9 a significant decline to 66% of the initial applied amount of test item after one day and to 1-3% after 16 days confirm the rapid hydrolysis of alpha-cypermethrin under alkaline conditions (DT50 pH 9: 2.9 h [BASF Doc ID AL-322-001]).

The RMS BE has asked for additional kinetic evaluations on 27. Oct. 2016. The results are presented and discussed below.

Report:	CA 7.2.1.1/3 Anonymous, 2016 a Response to RMS BE - Request concerning kinetic re-evaluations – Alpha-cypermethrin 2016/1324158
Guidelines:	none
GLP:	no

Hydrolysis study by Van Dijk (1993), BASF DocID: AL-322-002

RMS: The Notifier is requested to perform a comprehensive kinetic assessment according to FOCUS of the hydrolysis study by Van Dijk, 1993.

BASF response: A kinetic evaluation was performed according to FOCUS kinetics guidance document (2006) for the data from Van Dijk (1993). Only the SFO model was applied for the evaluation. The results are summarized in Table 7.2.2.1-3 and the corresponding Kingui fits are presented in Appendix 5 in the original response document. The trigger endpoints (DT_{50}: 28.7, 5.7 and 2.2 d for 50, 60 and 75°C at pH 7; DT_{50}: 3.3 and 0.1 d for 25 and 50°C at pH 9) are comparable to those derived in the original study (DT_{50}: 27.0, 5.3 and 2.0 d for 50, 60 and 75°C at pH 7; DT_{50}: 3.5 and 0.1 d for 25 and 50°C at pH 9). For this reason and taking into account that kinetic evaluation according to FOCUS is not formally required for hydrolysis studies, the endpoints of the original study are considered still valid.

Remark: In the dossier summary erroneously an incubation temperature of 30°C for pH 5 and 7 was reported. The correct incubation temperature was 50°C.

Table 7.2.1.1-3: Statistical and visual assessment of kinetic models for hydrolysis of alpha-cypermethrin under different pH and temperatures

pH	Temperature [°C]	Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
7	50	SFO	0.5	k < 0.01	good	28.7	95.3
	60	SFO	4.1	k < 0.01	good	5.7	19.0
	75	SFO	2.3	k < 0.01	good	2.2	7.2
9	25	SFO	2.4	k < 0.01	good	3.3	11.0
	50	SFO	5.5	k < 0.01	good	0.1 [a]	0.4 [a]

[a] Recalculated based on values in hours.

Hydrolysis studies by Hassink (2005a & 2005b), BASF DocID: 2005/1016375, 2005/1016376

RMS: The Notifier is requested to perform a comprehensive kinetic assessment according to FOCUS (A kinetic assessment performed with the excel sheets recommended by FOCUS were provided by email, but it is not a comprehensive kinetic assessment since t-test results, output graphs, etc. are missing)

BASF response: A kinetic evaluation was performed according to FOCUS kinetics guidance document (2006) for the data from Hassink (2005a) and Hassink (2005b). Only the SFO model was applied for the evaluation. The results are summarized in Table 7.2.2.1-4 and Table 7.2.2.1-5 and the corresponding Kingui fits are presented in Appendix 6 in the original response document.

Table 7.2.1.1-4: Statistical and visual assessment of kinetic models for hydrolysis of technical alpha-cypermethrin (TGAI batch COD-000165)

pH	Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
5	SFO	5.6	k: < 0.01	acceptable	60.4	200.5
7	SFO	6.5	k: < 0.05	acceptable	54.2	180.1
9	SFO	16.8	k: < 0.01	acceptable	4.5	15.0

Table 7.2.1.1-5: Statistical and visual assessment of kinetic models for hydrolysis of technical alpha-cypermethrin TGAI batch COD-000166)

pH	Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	$DegT_{50}$ [d]	$DegT_{90}$ [d]
7	SFO	5.9	k: < 0.05	acceptable	85.3	283.4
9	SFO	20.8	k: < 0.01	acceptable	2.9	9.5

CA 7.2.1.2 Direct photochemical degradation

Fisk P.R., (1994)
Alpha-cypermethrin (FASTAC): Photodegradation in water (preliminary experiment) including a comparison with esfenvalerate
BASF Doc ID: AL-630-009
GLP yes

Summary
Unlabeled alpha-cypermethrin was dissolved in aqueous buffer solution and exposed to artificial sunlight. The study was not performed to guidelines nor has reliable results.

Concha M., Zhixing Y. and Beigel C. 2001
BAS 310 I (Alpha-cypermethrin): Aqueous photolysis
BASF Doc ID: AL-324-003
GLP: yes

Summary
In the present study, [benzyl-14C-] and [cyclopropane-14C-] alpha-cypermethrin were exposed to artificial sunlight in sterile aqueous pH 5 buffer solution at 0.002 ppm and 22°C for up to 15 and 28 days. Volatiles were trapped in ethylene glycol and two 10% KOH solutions. Samples were extracted after acidification (conc. HCl) with ethyl acetate and analyzed by liquid scintillation counting (LSC) and high performance liquid chromatography (HPLC).
The mass balances ranged from 90.0 to 113.2% of the total applied radioactivity (TAR) for the irradiated samples.
14C-alpha-cypermethrin degraded rapidly in the light exposed samples and represented 52.5% and 33.6% TAR in the benzyl- and cyclopropane-labeled samples, respectively, after 2 days of exposure. By the end, detected 14C-alpha-cypermethrin amounts decreased below the detection limit (both labels).
Main metabolites with the benzyl-labeled test item were 3-PBAld (maximum 15.9% TAR at day 2) and 3-PBA (maximum 22.5% TAR at day 4), both decreasing towards the end of the study period. Unextracted radioactivity increased to 26% TAR at the end of the study. Several other more polar metabolites occurred only in small amounts below 8% TAR. 21.4% TAR were mineralized, less than 3% TAR was recovered in the traps for organic volatiles at study end.
The main metabolites observed with the cyclopropane-labeled test item was CL 901649 (mixture of cis and trans), which reached a maximum of 43.7% TAR at day 8, declining to 34.8% TAR by day 28. Unextracted radioactivity amounted to maximum 34.8% TAR at day 8, decreasing to 10.3% TAR by the end of the study. Another metabolite (CL 1500788) was detected in maximum amounts of 11.4% TAR in one replicate of day 8, but was detected only in small amounts in the other replicate. In day 15 and day 28 samples, amounts were below the detection limit. 7.7% TAR were mineralized by the end of the study, 1.0% TAR was recovered in the traps for organic volatiles.
Alpha-cypermethrin did not degrade significantly in the dark control samples; therefore, degradation of alpha-cypermethrin in the irradiated samples can be attributed to photochemical reactions.

Calculated first-order DT50 and DT90 values of alpha-cypermethrin were 2.2 and 7.3 days, (benzyl label), respectively, and 1.2 and 4.1 days (cyclopropane label), respectively. Based on 8.4 hours of artificial light irradiation (1 solar day), DT50 and DT90 values of alpha-cypermethrin were 6.3 and 20.9 days, (benzyl label), respectively, and 3.4 and 11.7 days (cyclopropane label), respectively.

CA 7.2.1.3 Indirect photochemical degradation

No data were generated. Not triggered as per OECD Draft Guidance Document "Phototransformation of Chemicals in Water".

CA 7.2.2 Route and rate of biological degradation in aquatic systems

CA 7.2.2.1 "Ready biodegradability"

No new data were generated.

Table 7.2.2.1-1: List of "ready biodegradability" studies in aquatic systems performed with (alpha-)cypermethrin

DocID	Parent compound	Species	Application rate [mg L-1]	Incubation temperature	Incubation period [days]	Remark
AL-690-001	Alpha-cypermethrin	Pseudomonas fluorescens	2.9 - 100.0	20±1°C	28	Stone 1983
Hund et al., 1990	Cypermethrin	n/a	10 20	n/a	33	Submitted for Mitchell Cotts

n/a = not available

Stone C.M. and Watkinson R.J. 1983
WL85871: An assessment of ready biodegradability
BASF DocID: AL-690-001
GLP: yes

Summary
Alpha-cypermethrin was tested for ready biodegradability in 3 tests, a "Closed Bottle", a "Modified Sturm Test" and a growth inhibition test with Pseudomonas fluorescens. In the closed bottle test, no oxygen depletion could be observed after 28 days. In the modified Sturm Test no CO2 was evolved after 28 days. The growth of Pseudomonas fluorescens was not inhibited.
Conclusion: Alpha-cypermethrin is not ready biodegradable.

No further tests were performed as higher tier studies are available.

CA 7.2.2.2 Aerobic mineralisation in surface water

Report: CA 7.2.2.2/1
Lewis C.J., 2014a
14C-Alphacypermethrin (BAS 310 I): Aerobic Mineralisation in Surface Water
2014/1031017
Guidelines: OECD Guideline 309
GLP: yes
(certified by the Department of Health of the Government of the United Kingdom, United Kingdom)

Executive Summary

The purpose of this study was to determine the mineralization and degradation rates of the pyrethroid insecticide alpha-cypermethrin (BAS 310 I) in an aquatic system under dark conditions. The study was performed according to OECD guideline 309 (Aerobic mineralization in surface water – Simulation biodegradation test). The pelagic test system was chosen for this study.

The test was performed at concentration levels of 0.5 µg L-1 alpha-cypermethrin (low concentration) and 3.5 µg L-1 (high concentration), using two differently 14C-labeled test items (cyclopropyl and benzyl label). Sterilized samples were tested for each label of the higher concentration. The test vessels were attached to a flow-through system for continuous aeration and incubated at a temperature of 20 ± 2°C in the dark. Samples for the experiment were taken at 0, 1, 3, 7, 14, 22, 35 and 59 days after treatment.

After adding formic acid to the water samples, liquid/liquid partition extraction was performed using dichloromethane (DCM). The amount and nature of radioactivity in DCM layers and selected aqueous phases (phase remaining after DCM partition) was determined by liquid scintillation counting (LSC) and high performance liquid chromatography (HPLC), as well as thin layer chromatography (TLC), both coupled with radioactivity detection. Volatiles were trapped in 2M sodium hydroxide and also analyzed by LSC.

Selected samples were analyzed by chiral HPLC to investigate the individual degradation behavior of the two isomers forming alpha-cypermethrin, a racemate of the two cis-2 isomers present in cypermethrin (a racemic mixture of a total of eight isomers). At all times after 0 DAT, approximately equal levels of the two cis-1 and cis-2 isomers were detected. This shows that isomerization of cis-2 to cis-1 isomers occurred and that the four isomers appeared to be in equilibrium.

The cyclopropyl and benzyl labeled test items were significantly degraded in the natural water environment provided in this test. After 59 days, 2.3 to 11.4% TAR was recovered as unchanged active substance in viable units and 5.5 to 12.1% TAR in sterilized units. There was extensive mineralization of the benzyl ring system (34 - 39% TAR) and lower mineralization in the cyclopropyl ring system (4 - 11% TAR) in viable units but ≤ 1% TAR in sterilized units. Degradation was therefore biotic and abiotic and different routes of degradation were followed.

After treatment with [cyclopropyl-14C] alpha-cypermethrin, material balance at 0 DAT and 59 DAT ranged from 93.7 to 98.1% TAR for the mean of the two replicates in the viable test vessels, and 94.3 to 98.6% TAR for the sterilized vessels. Corresponding values obtained after treatment with [benzyl-14C] alpha-cypermethrin were 82.5 and 100.6% TAR for the viable test vessels and 86.1 and 94.8% TAR for the sterilized vessels. Recovery of radioactivity at sampling times between 0 and 59 DAT decreased to < 90% on several occasions and this was attributed to problems with the analysis caused by the occurrence of biofilm.

Biofilms formed during the incubation and adsorbed radioactivity from the solutions causing problems with mass balance. Although extra radioactivity was extracted from biofilm for some samples, not all samples had mass balance values > 90%.

Major metabolites in viable test units were 3-phenoxybenzoic acid (3-PBA), dichlorovinylcarboxylic acid (DCVA) and carbon dioxide. Maximum levels of 3-PBA (mean of two replicates) were 56.1% (benzyl label), maximum levels of DCVA were 77.7% (cyclopropyl label) and maximum levels of carbon dioxide were 37.6% (benzyl label). 3-Phenoxybenzaldehyde (3-PBAld) was detected in sterilized systems but not in non-sterilized systems (maximum levels were 8.5%, benzyl label). DCVA did not degrade during the study, but 3-PBA reached a maximum after 35 days and then slightly decreased again. 3-PBAld reached a maximum of 8.5% TAR at 14 DAT in the sterilized system and then decreased to < 1% TAR at 59 DAT. One unidentified degradation product (Unk 309-1) was present after 3 days at a maximum of 5.6% TAR in the cyclopropyl label low application rate group and decreased again. Since it exceeded 5% TAR only at one single sampling interval, it was not further identified.

Kinetic analysis and calculations of DegT50 and DegT90 values were performed following the recommendations of the FOCUS Kinetics workgroup on derivation of aquatic persistence endpoints. The analysis was done by a non-linear regression method (IRLS) using the software package KinGUI2. The evaluation of the results for high and low test concentrations for both labels showed that degradation of alpha-cypermethrin in the test systems was best described by DFOP kinetics and reliable degradation endpoints could be derived from all test systems ranging from 2.3 to 3.4 days (DegT50) and from 36.5 to 86.8 days (DegT90). For the metabolite DCVA, no reliable endpoints could be derived from any of the test systems treated with the cyclopropyl-labeled alpha-cypermethrin while for the metabolite 3-PBA reliable endpoints could be derived from the test system with benzyl-labeled alpha-cypermethrin applied at the high dose with 45.2 days and 150.2 days for DegT50 and DegT90, respectively.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

BAS-Code: BAS 310 I (alpha-cypermethrin)
Chemical name: Racemate of (S)-cyano-3-phenoxybenzyl (1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-cyano-3-phenoxybenzyl (1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate
Molecular formula: C22H19Cl2NO3
Molar mass: 416.3 g mol-1 (unlabeled)

Label 1 (cyclopropane label)
Label: cyclopropane-1-14C
Batch No.: 986-2101
Specific radioactivity: 4.71 MBq mg-1
Radiochemical purity: 96.9%
Chemical purity: 91.8%

Label 2 (benzyl label)
Label: benzylring-U-14C
Batch No.: 775-0601
Specific radioactivity: 4.74 MBq mg-1
Radiochemical purity: 99.5%
Chemical purity: 96.1%

2. Test system

Water and small amounts of sediment were collected from The Lake at Studley Royal (Ripon, United Kingdom). The physico-chemical properties of the system are summarized in Table 7.2.2.2-1.

Prior to use the sediment and water were stored together in the dark at 4 ± 2°C with free access to air. Water was filtered through a 0.1 mm sieve and sediment was passed through a 2 mm sieve.

Table 7.2.2.2-1: Characterization of the water/sediment system

Designation Origin		Fountains Abbey The Lake, Studley Royal, Ripon, UK	
Water			
Temperature	[°C]	8.3	
pH water	-	7.92	
Oxygen concentration	[mg L-1]	9.55	
Redox potential (Eh)	[mV]	186.5	
Hardness	[mmol L-1]	138	
Total organic carbon	[mg L-1]	5.84	
Total N	[mg L-1]	0.000938	
Total P	[mg L-1]	0.25	
Sediment			
Textural class		UK Particle Size Distribution	USDA
Sand	[%]	60	63
Silt	[%]	29	26
Clay	[%]	11	11
Soil type	-	Sandy loam	Sandy loam
pH (H2O)	-	7.6	
pH (CaCl2)	-	7.1	
Redox potential (Eh)	[mV]	-144.6	
Organic carbon	[%]	4.0	

B. STUDY DESIGN

1. Experimental conditions

A total of 104 test vessels was prepared: 20 test vessels for each radiolabel (cyclopropyl and benzyl) and each nominal concentration (0.5 and 3.5 µg L-1), 10 vessels for the sterile incubation (both labels; 3.5 µg L-1), 2 vessels as system control with sodium [14C]-benzoate and 2 vessels with sodium [14C]-benzoate plus treatment solvent. The vessels were filled with about 100 mL water. Appropriate amounts of the respective application solutions were pipetted dropwise to the water surface to achieve a nominal application rate of 0.5 µg L-1 or 3.5 µg L-1, respectively.

The systems were incubated at 20 ± 2°C in a metabolism apparatus (incubator) with a flow system providing a continuous flow of fresh air. Each test vessel was connected to a set of volatile traps. The first volatile trap was empty to act as a security trap and the following two traps contained trapping solutions (2x NaOH) for the 14C-volatiles to be expected. Test vessels containing sterile water were also aerated, however, the air stream was led through sterile filters to avoid contamination of the test system by airborne germs.

2. Sampling

Samples, including the sterile groups, were taken at 0, 1, 3, 7, 14, 22, 35 and 59 days after treatment (DAT). For sampling, the flasks were removed from the rigs and the temperature, O2 content, pH, and redox potential of the water was measured in a representative treated sample.

3. Description of analytical procedures

Water
Formic acid was added to each sample. Liquid-liquid partition extraction was performed using dichloromethane (DCM). The radioactivity in DCM layer as well as the water phase was measured by liquid scintillation counting (LSC). After evaporation and reconstitution in acetonitrile, aliquots of the DCM extracts were analyzed by high performance liquid chromatography (HPLC; higher application rate) and TLC (low application rate). Selected samples were analyzed by chiral HPLC to investigate the individual degradation behavior of the two isomers forming alpha-cypermethrin.

Recovery of radioactivity using the method described above was variable and sometimes lower than 90%. Two possibilities were investigated to account for the loss. The first was that radioactivity could be associated with a biofilm that was visible on the incubation vessels after washing with DCM and was present in the DCM phases as precipitates or floating debris. The second was that radioactivity was present as dissolved carbon dioxide that was lost during the acidification and partitioning process.

After washing the incubation vessels with DCM as described for the original method, acetone was added to each empty test vessel and sonicated. Radioactivity was determined by LSC. This procedure was performed at 14 DAT (except controls), and later times, but could not be performed at earlier times because the original incubation vessels were no longer available.

Volatiles
After collecting the traps attached to each test vessel being sampled (all, except the control vessels), formic acid was added and the units were re-attached to the incubation system with fresh 2M sodium hydroxide traps. Air was bubbled through each acidified unit. The units were stirred during the process. After 1 hour, any radioactivity recovered in the sodium hydroxide traps was determined by LSC. This procedure was used on units at 22 DAT onwards (except at 35 and 59 DAT), and it was limited to units treated with the benzyl label.

4. Calculation of the degradation/dissipation rates

Kinetic evaluation was performed in order to derive aquatic persistence endpoints. The kinetic analysis was carried out following the recommendations of the FOCUS work group on degradation kinetics [FOCUS (2006): “Guidance Document on Estimating Persistence and Degradation Kinetics from Environmental Fate Studies on Pesticides in EU Registration” Report of the FOCUS Work Group on Degradation Kinetics, EC Document Reference Sanco/10058/2005 version 1.0 (November 2011), 436pp.] in order to derive persistence endpoints.

The software package KinGUII (version 2.2014.224.1704) was used for parameter fitting [SCHÄFER, D., MIKOLASCH, M., RAINBIRD, P., HARVEY, B. (2007) KinGUI: A new kinetic software tool for evaluations according to FOCUS Degradation Kinetics. In: Del Re, A.A.M. et al. (Eds.): Proceedings of the XIII Symposium on Pesticide Chemistry, Piacenza, 2007, p. 916-923., WITT, J., GAO, Z., MEYER, H. (2014) KinGUII, Version 2.2014.224.1704 Bayer CropScience AG]. The error tolerance and the number of iterations of the optimization tool were set to 0.00001 and 100, respectively.

For each data set, the kinetic models proposed by the FOCUS Kinetics guidance document were tested in order to identify the best-fit model. The recommended kinetic models, i.e. the single first order kinetics (SFO), the Gustafson-Holden model (FOMC), and bi-exponential (DFOP) kinetics are already implemented in KinGUI. The Goodness-of-fit was evaluated by visual assessment, $\chi 2$ minimum error, and type-I-error rate (t-test) [for details see Chapter 6.3 in FOCUS (2006)].

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The distribution of radioactivity in the different extracts of the water system treated with 14C-alpha-cypermethrin (cyclopropyl and benzyl label) is presented in Table 7.2.2.2-2 and Table 7.2.2.2-3.

The applied mass of test item per test vessel containing 100 mL of water was 0.34 µg (high concentration) and 0.05 µg (low concentration).

The material balance for the pelagic test ranged from 82.8 – 100.8% of the total applied radioactivity (TAR) in Lake water treated with the cyclopropyl-14C-labeled test item and from 82.5 – 100.6% TAR in Lake water treated with the benzyl-14C-labeled test item. In the sterile vessels, the material balance ranged from 90.5 to 98.6 TAR (cyclopropyl label) and from 84.0 to 95.4% TAR (benzyl label), respectively.

Recovery of radioactivity at sampling times between 0 and 59 days after treatment (DAT) decreased to < 90% on several occasions. Low mass balance results generally occurred when high levels of radioactivity were associated with biofilm.

Mineralization of alpha-cypermethrin to CO2 occurred from the benzene ring system (up to 37.6% TAR) and to a much lesser extent from the cyclopropyl ring system (up to 10.9% TAR). Carbon dioxide was identified by complete precipitation of the radioactivity from trapping solutions after adding barium chloride solution to selected samples.

Table 7.2.2.2-2: Material balance and distribution of radioactivity after application of [cyclopropyl-^{14}C] alpha-cypermethrin to lake water (non-sterile and sterile)

Days after treatment (DAT)	Percent of total applied radioactivity [% TAR]						
	DCM Extract	Aqueous Extract	Dissolved CO_2 in the water phase	Acetone extract of flask	Biofilm acetone extract	Total in Volatile Traps**	Material Balance
Low concentration (0.5 µg L^{-1})*							
0	96.8	n.d.	n.a.	n.a.	n.a.	n.a.	96.8
1	86.4	2.8	n.a.	n.a.	2.0	1.2	92.5
3	91.3	n.d.	n.a.	n.a.	n.a.	1.2	92.5
7	88.3	n.d.	n.a.	n.a.	1.8	1.4	90.6
14	79.9	2.2	n.a.	4.7	4.6	2.1	93.4
22	61.2	n.d.	n.d.	18.3	2.2	3.0	83.6
35	76.0	6.0	n.a.	8.0	n.a.	10.9	100.8
59	74.9	8.1	n.a.	5.9	n.a.	4.9	93.7
High concentration (3.4 µg L^{-1})*							
0	94.1	4.0	n.a.	n.a.	n.a.	n.a.	98.1
1	89.6	3.9	n.a.	n.a.	n.a.	0.8	94.3
3	80.9	2.6	n.a.	n.a.	2.0	0.6	85.9
7	76.1	4.2	n.a.	n.a.	1.3	1.3	82.8
14	75.5	3.7	n.a.	8.2	5.3	1.8	91.7
22	61.9	3.9	0.1	22.7	1.0	2.7	91.7
35	76.8	6.4	n.a.	10.7	n.a.	2.2	96.0
59	77.2	8.5	n.a.	4.9	n.a.	3.5	94.0
Sterilized lake water (3.4 µg L^{-1})							
0	85.8	12.8	n.a.	n.a.	n.a.	n.a.	98.6
1	88.6	8.1	n.a.	n.a.	n.a.	n.d.	96.7
3	90.9	4.0	n.a.	n.a.	n.a.	n.d.	94.9
7	89.8	4.7	n.a.	n.a.	n.a.	n.d.	94.5
14	85.6	4.6	n.a.	0.3	n.a.	n.d.	90.5
22	64.2	15.1	n.d.	10.6	n.d.	1.1	91.0
35	82.7	9.8	n.a.	2.3	n.a.	0.3	95.1
59	84.7	8.5	n.a.	0.9	n.a.	0.2	94.3

TAR = Total applied radioactivity
n.a. = Not applicable
n.d. = Not detected
* Mean of two replicates
** Due to the nature of the trapping solutions and testing in other incubation groups, this may be presumed to be CO_2

Table 7.2.2.2-2: Material balance and distribution of radioactivity after application of [benzyl-U-^{14}C] alpha-cypermethrin to lake water (non-sterile and sterile)

Days after treatment (DAT)	Percent of total applied radioactivity [% TAR]						
	DCM Extract	Aqueous Extract	Dissolved CO_2 in the water phase	Acetone extract of flask	Biofilm acetone extract	Total in Volatile Traps**	Material Balance
Low concentration (0.5 µg L^{-1})*							
0	93.5	n.d.	n.a.	n.a.	n.a.	n.a.	93.5
1	89.4	n.d.	n.a.	n.a.	n.a.	0.6	90.0
3	90.8	n.d.	n.a.	n.a.	n.a.	n.d.	90.8
7	76.4	n.d.	n.a.	n.a.	0.7	1.9	78.9
14	66.0	n.d.	n.a.	5.2	12.7	2.2	86.0
22	28.0	n.d.	2.4	41.6	2.8	13.7	88.4
35	48.7	6.2	2.1	15.3	1.3	19.2	92.0
59	35.7	9.4	1.5	13.8	n.a.	32.8	93.1
High concentration (3.4 µg L^{-1})*							
0	96.4	4.3	n.a.	n.a.	n.a.	n.a.	100.6
1	95.9	1.4	n.a.	n.a.	n.a.	0.0	97.3
3	91.1	n.d.	n.a.	n.a.	n.a.	0.2	91.3
7	89.9	1.5	n.a.	n.a.	n.a.	1.1	92.4
14	71.1	3.5	n.a.	5.5	6.7	3.1	89.9
22	55.8	4.4	1.4	22.8	1.1	5.3	90.7
35	51.8	5.4	2.8	15.6	0.5	11.4	87.1
59	31.6	6.9	1.6	5.0	n.a.	37.6	82.5
Sterilized lake water (3.4 µg L^{-1})							
0	79.4	15.4	n.a.	n.a.	n.a.	n.a.	94.8
1	80.2	14.8	n.a.	n.a.	0.4	n.d.	95.4
3	89.7	1.5	n.a.	n.a.	n.a.	n.d.	91.2
7	81.5	3.3	n.a.	n.a.	1.1	n.d.	85.9
14	71.3	13.7	n.a.	1.2	0.4	n.d.	86.6
22	81.6	n.d.	n.d.	7.2	0.2	1.0	90.0
35	77.0	4.3	n.d.	1.8	0.7	0.2	84.0
59	79.7	4.7	n.d.	1.3	n.a.	0.4	86.1

TAR = Total applied radioactivity
n.a. = Not applicable
n.d. = Not detected
* Mean of two replicates
** Due to the nature of the trapping solutions and testing in other incubation groups, this may be presumed to be CO_2

B. TRANSFORMATION OF PARENT COMPOUND

Characterization and identification of residues in water extracts

Water
Cypermethrin (no separation into individual isomers) and its metabolites (DCVA, 3-PBA, 3-PBAld) from the high and low application rate groups were identified by HPLC and by TLC, respectively. The summary of radio-TLC and radio-HPLC analysis of the water phases (DCM extracts) and selected aqueous phases (phase remaining after DCM partition) is presented in Table 7.2.2.2-4 to Table 7.2.2.2-7.

Significant degradation of cypermethrin occurred forming DCVA, 3-PBA and 3-PBAld (sterilized lake water only).

DCVA was a major metabolite produced from the cyclopropane ring system. The metabolite generally increased throughout the incubation period comprising 74.1 to 77.7% TAR at 59 DAT. The metabolite also increased in the sterilized incubation group reaching a similar level at 59 DAT (77.5% TAR).

3-PBA was a major metabolite produced from the benzene ring system. The metabolite was present at up to 56.1% TAR (35 DAT). It did not continuously increase during the incubation period. It was present at generally higher levels in the sterilized test system (maximum 69.8% at 22 DAT). In viable incubation groups at the end of the test (59 DAT), levels were 31.5 to 32.5% TAR but in the sterilized incubation group levels were 64.8% TAR.

3-PBAld was only a major metabolite from the benzene ring system in sterilized lake water. Levels increased to 8.5% TAR at 14 DAT before decreasing to < 1% TAR by 59 DAT.

An unidentified metabolite (Unk 309-1) was present at up to 5.6% TAR in the cyclopropyl label low application rate group and at lesser concentrations in other treatment groups. Since it exceeded 5% TAR only at one single sampling interval, it was regarded as non-relevant and it was not further characterized. There were no other metabolites detected that were present at > 10% TAR at any time or that were present at ≥ 5% TAR at two consecutive sampling intervals.

Concentration of samples for chromatography resulted in occasional large losses of radioactivity because some of the biofilm re-precipitated from extracts used for chromatography, removing some of the radioactivity with it. A check was made that the radioactivity lost from solution was equivalent in nature to the radioactivity remaining in solution.

Sterilized samples

The main difference between results from sterilized incubation groups and viable incubation groups was that there was ≤ 1% TAR mineralized in sterile groups but up to 37.6% mineralized from viable groups. Mineralization can therefore be considered to be a biotic process. Hydrolysis occurred in the sterilized system and therefore the pH of the water was favorable for hydrolysis. DCVA formed under abiotic processes (i.e. hydrolysis) but did not mineralize in the viable samples.

The other differences concerned the metabolites present in the water. Hydrolysis to 3-PBAld was only detected in sterilized samples. In viable samples the metabolite was not detected and the hydrolysis proceeded through to 3-PBA. It can be concluded that 3-PBAld is not stable under biotic conditions.

3-PBA was present at generally higher levels in sterilized samples compared to viable samples. It is concluded that 3-PBA was the precursor to mineralization.

Table 7.2.2.2-3: Metabolite overview for the water phase (DCM extract) after application of [cyclopropyl-^{14}C] alpha-cypermethrin to lake water

Days after treatment (DAT)	Percent of total applied radioactivity [% TAR]					
	CYP	DCVA	Unk 309-1	Other Unknowns	Unresolved Background	Total
Low concentration (0.5 µg L^{-1})*						
0	95.6	n.d.	n.d.	0.6	0.6	96.8
1	70.5	13.6	1.7	n.d.	0.6	86.4
3	40.9	41.0	5.6	n.d.	3.8	91.3
7	30.3	49.7	3.6	n.d.	5.6	89.2
14	33.7	52.9	1.2	1.1	0.2	89.2
22	16.1	45.2	1.4	17.1	0.7	80.6
35	11.3	70.3	1.5	n.d.	0.9	84.0
59	2.8	77.7	n.d.	n.d.	0.3	80.8
High concentration (3.4 µg L^{-1})*						
0	91.3	n.d.	n.d.	2.2	0.6	94.1
1	69.3	16.2	n.d.	3.5	0.5	89.6
3	47.5	30.2	n.d.	4.2	1.0	82.8
7	27.5	44.4	n.d.	4.7	0.7	77.4
14	31.5	49.1	n.d.	4.7	1.1	86.3
22	19.0	60.1	n.d.	5.6	0.4	85.1
35	10.1	74.3	n.d.	2.6	0.5	87.5
59	2.3	74.1	n.d.	5.0	0.6	82.0
Sterilized lake water (3.4 µg L^{-1})						
0	85.4	n.d.	n.d.	0.4	0.1	85.8
1	81.7	6.8	n.d.	n.d.	0.1	88.6
3	79.6	10.6	n.d.	n.d.	0.7	90.9
7	69.7	17.0	n.d.	3.0	0.2	89.8
14	63.3	20.3	n.d.	1.4	0.9	85.9
22	26.8	45.9	n.d.	1.8	0.3	74.8
35	23.5	58.8	n.d.	2.6	0.1	85.0
59	5.5	77.5	n.d.	2.0	0.6	85.6

TAR = Total applied radioactivity
Unk = Unknown
CYP = Cypermethrin (not separated into individual isomers)
n.d. = Not detected
* Mean of two replicates

Table 7.2.2.2-4: Metabolite overview for the water phase (DCM extract) after application of [benzyl-U-^{14}C] alpha-cypermethrin to lake water

Days after treatment (DAT)	Percent of applied radioactivity [% TAR]						
	CYP	3-PBA	3-PBAld	Unk 309-1	Total Unk*	Unresolved Background	Total
Low concentration (0.5 µg L^{-1})**							
0*	80.0	1.1	n.d.	9.7	1.5	1.2	93.5
1	70.5	16.7	n.d.	1.4	0.0	0.8	89.4
3	40.5	40.0	n.d.	5.2	0.7	4.4	90.8
7	36.2	27.8	n.d.	4.8	4.3	3.9	77.1
14	38.3	38.3	n.d.	3.8	1.9	1.4	83.9
22	38.4	24.9	n.d.	3.9	4.2	0.9	72.4
35	21.4	39.8	n.d.	2.9	0.0	0.4	64.6
59	11.4	31.5	n.d.	3.0	3.2	0.4	49.5
High concentration (3.4 µg L^{-1})***							
0	95.3	n.d.	n.d.	n.d.	0.7	0.3	96.4
1	75.9	16.3	n.d.	n.d.	3.5	0.2	95.9
3	50.8	33.8	n.d.	n.d.	6.1	0.5	91.1
7	33.0	52.9	n.d.	n.d.	3.4	0.6	89.9
14	32.4	48.5	n.d.	n.d.	2.0	0.4	83.3
22	20.1	48.1	n.d.	n.d.	11.3	0.1	79.6
35	7.4	56.1	n.d.	n.d.	3.6	0.5	67.6
59	3.3	32.5	n.d.	n.d.	0.5	0.2	36.5
Sterilized lake water (3.4 µg L^{-1})							
0	77.5	n.d.	n.d.	n.d.	1.4	0.5	79.4
1	76.0	n.d.	4.0	n.d.	n.d.	0.3	80.2
3	80.5	0.9	7.6	n.d.	n.d.	0.7	89.7
7	67.9	6.4	8.1	n.d.	n.d.	0.1	82.6
14	37.5	24.5	8.5	n.d.	2.2	0.1	72.9
22	15.9	69.8	1.7	n.d.	1.1	0.5	89.0
35	29.2	47.6	1.0	n.d.	1.4	0.3	79.5
59	12.1	64.8	n.d.	n.d.	3.7	0.3	81.0

TAR = Total applied radioactivity
CYP = Cypermethrin (not separated into individual isomers)
Unk = Unknown
n.d. = Not detected
* Total Unk is the sum of all other peaks in the chromatogram.
** Mean of two replicates
*** Result of one replicate considered incorrect due to contamination of sample.

Table 7.2.2.2-5: Metabolite overview for the water phase (post DCM extraction) after application of [cyclopropyl-^{14}C] alpha-cypermethrin to lake water (non-sterile and sterile)

Days after treatment (DAT)	Percent of applied radioactivity (TLC system 2) [% TAR]			
	CYP	Unknowns	Unresolved Background	Total
High concentration (3.4 µg L^{-1})				
35	1.8	5.4	n.d.	7.2
35	1.4	4.0	n.d.	5.5
Sterilized lake water (3.4 µg L^{-1})				
0	11.7	0.9	0.2	12.8
1	6.5	1.5	0.1	8.1
22	12.0	2.8	0.3	15.1
35	5.9	3.7	0.2	9.8

TAR = Total applied radioactivity
CYP = Cypermethrin (not separated into individual isomers)
Samples containing > 5% TAR were analyzed.
n.d. = Not detected

Table 7.2.2.2-6: Metabolite overview for the water phase (post DCM extraction) after application of [benzyl-U-^{14}C] alpha-cypermethrin to lake water (non-sterile and sterile)

Days after treatment (DAT)	Percent of applied radioactivity (TLC system 2) [% TAR]			
	CYP	Total Unknowns	Unresolved Background	Total
High concentration (3.4 µg L^{-1})				
35	n.d.	6.6	0.1	6.7
Sterilized lake water (3.4 µg L^{-1})				
0	14.8	n.d.	0.6	15.4
1	14.4	n.d.	0.4	14.8

TAR = Total applied radioactivity
CYP = Cypermethrin (not separated into individual isomers)
n.d. = Not detected

Enantiomer specific analysis

In addition to the quantification of the parent, enantiomer-specific analyses were performed by radio-HPLC. The amounts of each cis-1 and cis-2 isomer present in the DCM extracts of water from non-sterilized and sterilized incubation groups for each label are shown in Table 7.2.2.2-8 and Table 7.2.2.2-9.

In all incubation groups, the cis-2 isomers of alpha-cypermethrin formed cis-1 isomers within three days after treatment. After this time all four cis isomers of cypermethrin decreased at the same rate and each isomer comprised a similar proportion of the total radioactivity present.

The isomerization occurred in the sterilized samples to the same extent as in the non-sterilized samples and therefore it was assumed to be a chemical rather than a microbial process.

Table 7.2.2.2-7: Isomer overview for the water phase (DCM extract) after application of [cyclopropyl-^{14}C] alpha-cypermethrin to lake water (non-sterile and sterile; chiral HPLC)

Days after treatment (DAT)	Percent of applied radioactivity [% TAR]						
	DCVA	Cis 2 α-S	Cis 1 α-S	Cis 2 α-R	Cis 1 α-R	Other ^{14}C*	Total
High concentration (3.4 µg L^{-1})**							
0	1.9	40.5	4.4	39.5	3.5	4.3	94.1
3	32.5	12.5	11.5	11.7	8.3	6.2	82.8
7	47.2	7.2	7.5	7.0	3.6	4.7	77.3
14	51.3	7.1	10.5	5.7	6.6	5.1	86.3
Sterilized lake water (3.4 µg L^{-1})							
0	n.d.	37.2	3.9	39.0	4.4	1.3	85.8
3	14.4	18.2	19.5	16.7	20.9	1.5	90.9
7	18.4	16.0	17.8	15.8	19.1	2.8	89.8
14	23.6	14.7	14.5	12.3	19.8	1.0	85.9

TAR = Total applied radioactivity
n.d. = Not detected
* Mean of two replicates
** Other radioactivity includes other peaks and unresolved background

Table 7.2.2.2-8: Isomer overview for the water phase (DCM extract) after application of [benzyl-U-^{14}C] alpha-cypermethrin to lake water (non-sterile and sterile; chiral HPLC)

Days after treatment (DAT)	Percent of applied radioactivity [% TAR]							
	3-PBA	3-PBAld	Cis-2 α-S	Cis-1 α-S	Cis-2 α-R	Cis-1 α-R	Other ^{14}C*	Total
High concentration (3.4 µg L^{-1})**								
0	1.6	n.d.	43.9	n.d.	43.6	4.1	3.1	96.4
3***	31.3	2.1	15.9	11.8	16.4	9.9	5.6	92.9
7	53.2	n.d.	8.0	9.8	7.1	5.6	6.3	89.9
14	48.8	n.d.	7.6	8.3	8.3	6.6	2.6	82.2
Sterilized lake water (3.4 µg L^{-1})								
0	n.d.	n.d.	35.6	2.7	34.6	2.4	4.2	79.4
3	n.d.	9.2	17.6	20.8	17.7	22.1	2.3	89.7
7	8.7	9.5	15.4	17.4	13.0	15.9	2.7	82.6
14	16.2	8.3	8.4	8.2	8.1	9.5	14.1	72.9

TAR = Total applied radioactivity
n.d. = Not detected
* Other radioactivity includes other peaks and unresolved background
** Mean of two replicates
*** One replicate only

Control samples
Recovery of radioactivity from the reference vessels at the end of the test was 96.5% for units treated with sodium [14C]-benzoate alone and 96.6% for units treated with sodium [14C]-benzoate and acetonitrile.

Sodium benzoate extensively mineralized to CO2 with 83.5% to 86.2% mineralization at 14 DAT and 93.2 to 94.5% mineralization at 59 DAT. The system was therefore viable. The control vessels treated with [14C]-sodium benzoate showed that the test system was microbially active both without and with the addition of acetonitrile. The total recoveries of trapped volatile radioactivity after 59 days were 94.5 and 93.2% TAR and the material balances were 96.5 and 96.6% TAR for the samples without and with acetonitrile, respectively.

The reason why mass balance was not obtained is probably related to adsorption to or incorporation into biofilm. Similar difficulties were experienced in obtaining mass balance results from units treated with alpha-cypermethrin. At 0 DAT (samples treated with acetonitrile) the mass balance was 100.3% showing that there was no error on application or sampling technique. The mass balance results of 96.5% and 96.6% at the end of the 59 day incubation also show that there was no error on application.

Degradation rates

The degradation of alpha-cypermethrin was evaluated according to the recommendations of the FOCUS workgroup on degradation kinetics [FOCUS (2006)]. A summary of the DegT50 and DegT90 values of alpha-cypermethrin and its metabolites DCVA and 3-PBA derived as persistence endpoints are given in Table 7.2.2.2-10 and Table 7.2.2.2-11.

The evaluation of the results for high and low test concentrations for both labels showed that degradation of alpha-cypermethrin in the test systems was best described by DFOP kinetics and reliable degradation endpoints could be derived from all test systems. For the metabolite DCVA, no reliable endpoints could be derived from any of the test systems treated with the cyclopropyl-labeled alpha-cypermethrin while for the metabolite 3-PBA reliable endpoints could be derived from the test system with benzyl-labeled alpha-cypermethrin applied at the high dose. As 3-PBAld was only present in sterilized samples but not in viable samples, no reliable endpoints could be derived.

Table 7.2.2.2-9 Summary of the kinetic evaluation of alpha-cypermethrin

Test system	Kinetic model	χ^2 error	Best-fit endpoints	
			$DegT_{50}$ [d]	$DegT_{90}$ [d]
Low test concentration, cyclopropyl label	DFOP	9.9	3.1	43.0
Low test concentration, benzyl label	DFOP	9.8	2.3	86.8
High test concentration, cyclopropyl label	DFOP	9.5	3.4	36.5
High test concentration, benzyl label	DFOP	6.5	3.3	36.8

Table 7.2.2.2-10: Summary of the kinetic evaluation of alpha-cypermethrin metabolites DCVA and 3-PBA

Test system	Compound	Kinetic model	χ^2 error	Best-fit endpoints	
				$DegT_{50}$ [d]	$DegT_{90}$ [d]
Low test concentration, cyclopropyl label	DCVA	SFO [a]	5.2	no reliable endpoints derived	
High test concentration, cyclopropyl label	DCVA	SFO [a]	6.8	no reliable endpoints derived	
Low test concentration, benzyl label	3-PBA	SFO [a]	15.6	no reliable endpoints derived	
High test concentration, benzyl label	3-PBA	SFO [a]	9.4	45.2	150.2

[a] DFOP kinetics for parent

III. CONCLUSION

Alpha-cypermethrin was significantly degraded in the natural water environment provided in this test. After 59 days, only 2.3 to 11.4% TAR of unchanged parent remained. There was significant mineralization of the benzyl ring system (34 - 39% TAR) and less mineralization of the cyclopropyl ring system (4 - 11% TAR). Two major metabolites were formed, DCVA (maximum 77.7% TAR at 59 DAT) and 3-PBA (56.1% TAR at 35 DAT). Furthermore, 3-PBAld was present in sterilized samples (maximum 8.5% TAR at 14 DAT) but not in viable samples which indicates that it is not stable under biotic conditions. An unidentified compound was present at up to 5.6% TAR (3 DAT) but it occurred only on single sampling events and was therefore not further identified. There were no other compounds present at ≥ 10% TAR or ≥ 5% TAR at two consecutive sampling times.

In all incubation groups, the cis-2 isomers of alpha-cypermethrin formed cis-1 isomers within 3 DAT. After this time all four cis isomers of cypermethrin decreased at the same rate and each isomer comprised a similar proportion of the total radioactivity present. The isomerization occurred in the sterilized samples to the same extent as in the non-sterilized samples and therefore it was assumed to be a chemical rather than a microbial process.

The evaluation of the results for high and low test concentrations for both labels showed that degradation of alpha-cypermethrin in the test systems was best described by DFOP kinetics and reliable degradation endpoints could be derived from all test systems ranging from 2.3 to 3.4 days (DegT50) and from 36.5 to 86.8 days (DegT90). For the metabolite DCVA, no reliable endpoints could be derived from any of the test systems treated with the cyclopropyl-labelled alpha-cypermethrin while for the metabolite 3-PBA reliable endpoints could be derived from the test system with benzyl-labeled alpha-cypermethrin applied at the high dose with 45.2 days and 150.2 days for DegT50 and DegT90, respectively.

The RMS BE has asked for additional kinetic evaluations on 07. Dec. 2016. The results are presented and discussed below.

Report:	CA 7.2.2.2/2 Anonmyous, 2016 a Response to RMS BE. Request concerning kinetic re-evaluation of Lewis 2014a – Alpha-Cypermethrin 2016/1344967
Guidelines:	none
GLP:	no

RMS: RMS notes that the Notifier performed a separate kinetic assessment for each label. In peer review, it is common practice to handle residuals from the same soil with different radiolabelled positions as replicate values. Please Notifier consider that residue data from two tested labels from the same soil need to be processed as replicates for the active substance and its metabolites.

BASF response: A kinetic evaluation was performed for the parent substance by processing the residue data of the two tested labels as replicates. The results are summarized in Table 7.2.2.2-11 and Table 7.2.2.2-12 and the corresponding Kingui fits are presented in the Appendix of the original response document. The trigger endpoints (low test concentration: $DegT_{50}$ = 2.4 d, $DegT_{90}$ = 63.1 d, DFOP model, high test concentration: $DegT_{50}$ = 2.8 d, $DegT_{90}$ = 38.2 d, DFOP model) are comparable with those derived by performing a separate kinetic assessment for each label (low test concentration: geomean $DegT_{50}$ = 2.7 d, geomean $DegT_{90}$ = 61.1 d, high test concentration: geomean $DegT_{50}$ = 3.3 d, geomean $DegT_{90}$ = 36.6 d). Therefore, the endpoints presented in the dossier are considered adequate.

The metabolites are formed label-specific (DCVA from cyclopropyl-label, 3-PBA from benzyl-label). Therefore, an evaluation with the different labels handled as replicates cannot be performed for the metabolites.

Table 7.2.2.2-11: Statistical and visual assessment of kinetic models for aerobic mineralisation of BAS 310 I in surface water, low test concentration

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	Trigger endpoints	
				$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	25.8	k: < 0.01	acceptable	12.4	41.2
FOMC	12.9	β: < 0.1	acceptable	2.9	174.7
DFOP	9.1	k1: < 0.01 k2: < 0.01 g: < 0.01	good	2.4	63.1

Table 7.2.2.2-12: Statistical and visual assessment of kinetic models for aerobic mineralisation of BAS 310 I in surface water, high test concentration

Kinetic model	χ^2 [%]	p (t-test)	Visual assessment	Trigger endpoints	
				$DegT_{50}$ [d]	$DegT_{90}$ [d]
SFO	20.6	k: < 0.01	poor	6.7	22.4
FOMC	9.7	β: < 0.01	acceptable	3.1	50.9
DFOP	6.8	k1: < 0.01 k2: < 0.01 g: < 0.01	good	2.8	38.2

CA 7.2.2.3 Water/sediment studies

Numerous studies were performed with cypermethrin or alpha-cypermethrin to address the behaviour in water /sediment systems. An overview can be found in Table 7.2.2.3-1. Sufficient data are available on alpha-cypermethrin, so that studies on cypermethrin can be disregarded for the assessment. The latest studies from Mamouni (1993) and Voelkl (1993) were re-evaluated according to FOCUS kinetics and are re-submitted and discussed here for reasons of completeness.

Table 7.2.2.3-1: List of water/sediment studies performed with (alpha-) cypermethrin

DocID	Parent compound	Test system	Application rate	Incubation temperature	Incubation period [days]	Remark
CY-630-002	Cypermethrin	River sediment of 3 tributaries (Thames) Pond sediment	0.093 µg L^{-1} water	16±1°C	34	Rapley 1981
AL-690-009	Cypermethrin	application on arable field (cotton) close to a pond	10 x 0.3 mg kg^{-1} sediment	outside temperature	n/a	Hadfield 1993
AL-630-007	Alpha-cypermethrin	Pond water/pond sediment (Headcorn, Kent, UK)	2.4 µg L^{-1} water	outside temperature	202	Dutton 1987
AL-630-008	Alpha-cypermethrin	2 freshwater ponds (Headcorn, Kent, UK)	0.04 mg kg^{-1} sediment	outside temperature	65	Pearson 1990
AL-630-011	Alpha-cypermethrin	River water/ sediment (Rhine) Pond water/ sediment	14.0 µg L^{-1} water	20±2°C	105	Mamouni 1663
AL-630-012	Alpha-cypermethrin	River water/ sediment (Rhine) Pond water/ sediment	14.0µg/L^{-1} water	20±2°C	105	Völkl 1993
Agnihotri et al., 1986						Submitted for Mitchell Cotts
Klöppel et al., 1993						Submitted for Mitchell Cotts

n/a = not available

Studies performed with cypermethrin. Not used for assessment

1. Rapley J.H. et al. (1981): Cypermethrin: Degradation in river and pond waters and sediments
Three river and a pond water and the corresponding sediments were incubated with cypermethrin labelled in the cyclopropyl and the benzene ring for up to one year at 16°C. Some incubation vessels were aerated, other were left undisturbed. Redox potentials ranged from approximately
-200 mV to + 500 mV. Cypermethrin was rapidly degraded (50% in less than two weeks). The main degradation products were cis and trans DCVA, PBA and PBAld. PBA and PBAld were further degraded. The highest aldehyde concentration was detected in a system with low redox potential. For a kinetic evaluation of cypermethrin sampling intervals were too large.For this reason and because the study was performed with cypermethrin this study will not be included into the assessment. The studies of Mamouni (1993) and Völkl (1993) will be used.

2. Hadfield S.T. et al. (1993) Pyrethroid residues in sediment and water samples from mesocosm and farm pond studies of simulated accidental aquatic exposure (Publication)
No regulatory relevant trial design. Not considered.

Trials performed with alpha-cypermethrin not used for evaluation

1. Dutton A.J. et al. (1988): An outdoor tank experiment to study the fate of FASTAC in the aquatic environment
Alpha-cypermethrin labelled in the cyclopropyl and the benzyl ring was applied to outdoor tanks containing pond water and sediment. The systems were sampled up to 202 days and water and sediment were analysed for radioactivity and chromatographed to identify metabolites. Alpha-cypermethrin rapidly dissipated from the water within 2 to 4 days. The main metabolites formed were DCVA and PBA. The study was not performed according to a guideline and quantification is questionable as results were reported as percentage of extracted radioactivity. Due to the outdoor environment a balance could not be obtained.

2. Pearson N (1990): The fate of FASTAC in experimental ponds
Similar experimental set-up as Dutton (1988), more biological testing of sensitive species. The study was not performed according to a guideline and quantification is questionable as results were reported as parent equivalent in different water layers and sediment. Due to the outdoor environment a balance could not be obtained.

The studies of Mamouni and Voelkl were considered as best description of the behaviour of alpha-cypermethrin in water/sediment systems. As a new kinetic evaluation was performed, the studies are re-submitted and summarized in detail.

Report: CA 7.2.2.3/1
Mamouni A., 1993a
Fastac (benzyl-14C): Degradation and metabolism in aquatic systems
AL-630-011
Guidelines: BBA IV 5-1
GLP: yes
(certified by Eidgenoessisches Departement des Inneren, Bern, Schweiz)

Executive Summary

The degradation of benzyl-14C-alpha-cypermethrin (FASTAC) in two aerobic water/sediment systems was investigated: the Rhine River and the Pond aquatic systems. Test vessels were treated with benzyl-14C labeled alpha-cypermethrin with a concentration corresponding to a field application rate of about 42 g a.s. ha-1 when assuming that the a.s. is homogeneously distributed in a natural water of 30 cm depth.

The test vessels were connected to an aeration system at a temperature of 20 ± 2°C in a dark room. Samples were taken directly after the treatment and 6 hours, 24 hours and 2, 7, 14, 30, 61 and 105 days after the application. Water and sediment extracts were analyzed by thin-layer-chromatography. The amount of non-extractable residues was determined by combustion and liquid scintillation counting (LSC). Volatiles were trapped in appropriate trapping solutions and also analyzed by LSC. Metabolite identification was done by co-chromatography with reference substances.

High mineralization was observed in both systems. 14CO2 amounted to 41.1% and 54.6% of the total applied radioactivity (TAR) in the Rhine and Pond system, respectively. No volatile radioactivity (< 0.1% TAR) was trapped in the ethylene glycol solutions of either system.

The disappearance of the parent compound from the water phase by hydrolysis and mineralization and adsorption to sediment was rapid in both systems, only 1.6% and 8.0% TAR remained in water after 7 days in the Rhine River and Pond aquatic systems, respectively.

Alpha-cypermethrin was degraded more rapidly in the Pond sediment than in the Rhine river sediment, because of the higher microbial activity in the Pond system. Five times as much radioactivity of the test item was present in the Rhine River sediment than in the Pond sediment after 30 days of incubation time. At the end of the incubation period, 12.9% and 2.0% TAR was present as parent compound in the Rhine and Pond sediments, respectively.

The test item was degraded to a minimum of ten radioactive fractions. M3 (WL 44607; 3-PBA) was the main radioactive fraction but was rapidly further degraded to CO2. Benzyl-14C-alpha-cypermethrin was degraded by hydrolysis at the ester bound linkage leading to the formation of WL 44607 (maximum amounts of 21.7% TAR and 23.1% TAR -referring to the whole system), which was further degraded to CO2. The remaining metabolites (all unknown except radioactive fraction M2, which was characterized as WL 46114) were found in very low amounts and did not exceed 7.9% TAR. The low amount of WL 46114 revealed that ring hydroxylation is a minor route of degradation of benzyl-14C-alpha-cypermethrin in sediment and indicates that the latter was further degraded by microorganisms.

Part of the radioactivity was converted into non-extractable residues amounting to 18.9% TAR (Rhine) and 21.2% TAR (Pond), respectively, at the end of the study.

Overall, the results of this study showed that benzyl-14C-alpha-cypermethrin was rapidly eliminated from both aquatic systems by rapid hydrolysis, microbial degradation, and mineralization.

A kinetic analysis of the data of the summarized study is presented in section CA 7.2.2.3/3 [SACHERS, S. (2014b): Kinetic evaluation of degradation of BAS 310 I - alpha-cypermethrin in water/sediment systems: Determination of trigger and modeling endpoints according to FOCUS, BASF DocID 2014/1159506].

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Internal code: BAS 310 I
Reg.No.: 4078193
CAS Number: 67375-30-8
Chemical name: 1:1 mixture of (S)-alpha-cyano-3-phenoxybenzyl-(1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-alpha-cyano-3-phenoxybenzyl-(1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethyl cyclopropanecarboxylate
Molecular formula: C22H19Cl2NO3
Molecular mass: 418.9 g mol-1
Label: benzyl-14C
Specific activity: 191.7 µCi mg-1
Radiochemical purity: >99% (checked by RCC using TLC in at least two different solvent systems)

2. Test system

Two different test systems were applied:

System I Rhine-river water + Rhine sediment (Rhine, Mumpf/Switzerland)
System II Pond water + Pond sediment (Pond, Rheinfelden [AG]/Switzerland)

The physico-chemical properties of the systems are summarized in Table 7.2.2.3-2.

Table 7.2.2.3-2:: Characterization of the water/sediment system

Water							
Origin		**River**			**Pond**		
		Study before start		**end**	**Study before start**		**end**
		sampling site	**at RCC**		**sampling site**	**at RCC**	
Temperature surface 5 cm above sediment	[°C]	3.2 3.5		n.d.	0.7 1.0		n.d.
pH surface 5 cm above sediment		8.2 8.2		8.8	7.2 7.2		8.2
Redox potential 5 cm above sediment	[mV]	264 240		210	80 55		207
Oxygen content	[mg L^{-1}]	12.2 11.8		5.5	1.3 1.2		4.9
NO_3-N NO_2-N NH_4-N N-total	[mg L^{-1}]	0.78 0.02 <0.78 max. 1.58		0.85 0.01 0.02 0.88	0.16 0.03 0.78 0.97		1.90 0.02 0.02 1.94
P as ortho-phosphorous		0.12		0.17	0.05		0.05
Total organic carbon	[mg C L^{-1}]		9.3	9.3		9.3	14.8
Hardness	[°dH]	14.0		33.0	22.0		23.0
Sediment							
Origin		**River**			**Pond**		
		Study before start		**end**	**Study before start**		**end**
		sampling site	**at RCC**		**sampling site**	**at RCC**	
pH (KCl) Redox potential	 [mV]	 -20	7.8	n.d. -146	 -70	7.1	n.d. -46
N-total (Kjeldahl)	[g kg^{-1} sed.]		0.68	n.d.		1.65	n.d.
P-total	[g kg^{-1} sed.]		0.41	n.d.		0.69	n.d.
Total organic carbon content	[g C 100 g^{-1} dry soil]		0.9	n.d.		5.4	n.d.
Biomass	[mg microbial C 100 g^{-1} soil]		51.1	11.3		480	124.5
Dry mass	[kg dry soil kg^{-1} fresh sediment]		0.65	0.64		0.30	0.28
Particle size distribution (USDA) clay (< 2 µm) silt (2-50 µm) sand (<50 µm)	 [%] [%] [%]		loamy sand 3.9 6.0 90.1	 n.d.		sandy clay 25.4 21.0 53.6	 n.d.

River and Pond water were analyzed for several pesticides (lindane, heptachlor, malation, DTT, Dieldrin, PCBs), none was detected.

B. STUDY DESIGN

1. Experimental conditions

The study was performed in an open gas-flow-system in 1000 mL all-glass metabolism flasks containing about 275 g and 190 g of sediment (to obtain a 2.5 cm sediment layer) for the Rhine and Pond aquatic systems, respectively, and 550 mL of water. These amounts were obtained by assuming a height of the water column of about 6 cm and a 2.5 cm thick sediment layer. The system was ventilated with moistened CO2-free air at a flow rate of about 60 mL min-1 with simultaneous gentle agitation of the water column by means of a suspended magnetic stirrer. The out coming air was passed through a trapping system of 2N NaOH (50 mL) and ethylene glycol (50 mL).

The test systems were acclimatized for about two weeks before application. The samples were incubated at 20 ± 2°C in a dark room (temperature controlled). During incubation, the water was gently stirred from the top without disturbing the sediments.

Aliquots of 400 µL containing 7.73 µg of benzyl-14C-alpha-cypermethrin were applied drop-wise to each flask. This corresponds to target concentration of 14 µg L-1 and a field application rate of about 42 g active substance ha-1 assuming that the test item is homogeneously distributed in the top 30 cm of the water column. Reserve as well as control flasks were included in the study.

The two untreated controls were used to determine the biomass at the end of the study.

2. Sampling

Water and sediment samples were taken in duplicates for analysis directly after the treatment and 6 hours, 24 hours and 2, 7, 14, 30, 61 and 105 days after the application. The reported results are thus, the means of both samples. The water was sampled down to a depth of 10-30 cm and the sediment was sampled from the top 5-10 cm of each system. This sampling site was located 1-2 m from firm land.

Volatile traps
Sodium hydroxide solutions were exchanged at each sampling interval and additionally on days 44, 72 and 89. Analyses of these solutions were performed accordingly. Ethylene glycol solutions were counted for after each sampling and replaced only at day 7, 44, 57, 72 and 89, due to the low amount of volatile substances detected so far (< 0.l% of the total applied radioactivity (TAR) on each sampling interval).

3. Analytical procedures

Water phase
After separation from sediment, the radioactivity in the water was determined directly by LSC. Thereafter, the water samples were partitioned with organic solvent at pH 4 using acetic acid, or otherwise stored deep-frozen at about -20°C until required for further analyses. For each sampling interval, about 300 mL water was partitioned one to three times with ethyl acetate (100 mL) (except samples after 6 hours of incubation chloroform was used). The extracts were combined then further concentrated in a Rotavap. The concentrates were submitted to TLC-analyses.

Sediment phase
The sediment samples were submitted to the following extractions:
1. - acetonitrile, one to three times (except on day 0, acetone was used 3 times).
2. - acetonitrile/water (7/3, v/v), once (on days 0.25, l, 2, 28 and 29)
   - acetonitrile/water/hydrochloric acid (50/50/1), on days 61 and 105
   - acetone, once (on days 14 and 30)
3. Soxhlet with methanol (from day 14 onward)
   The extracts mentioned above per sampling interval were combined and concentrated by evaporation of acetonitri1e and acetone in a Rotavap.

The residual radioactivity remaining in the sediment after all these extractions was then determined by combustion.

Characterization of bound residues
An additional fractionation of organic matter was carried out with the sediment residues after exhaustive extraction from interval 30 days. This fractionation was performed by extraction of the soil residues with 0.5 M NaOH solution. Thereafter, the humic acids were precipitated by decreasing the pH to about 1. Centrifugation permits separation of the fulvic acids which remain in the liquid phase. The humin fraction remains unextracted together with the clay mineral and aluminum oxides.

Volatiles
Radioactivity in the volatile trapping solutions was determined by LSC.

4. Kinetic modeling

The kinetic analysis was carried out according to previous requirements in the summarized study. A new kinetic analysis following the recommendations of the FOCUS work group on degradation kinetics [FOCUS (2006)] will be provided in section CA 7.2.2.3/3 [BASF DocID 2014/1159506].

II. RESULTS AND DISCUSSION

A. MASS BALANCE

The distribution of radioactivity to the different compartments of the water/sediment system treated with the test item is presented in Table 7.2.2.3-3 and Table 7.2.2.3-4, respectively.
Total mean recoveries were 90.1% TAR for the Rhine aquatic system and 91.9% TAR for the Pond river aquatic system, resulting from both phases, sediment and water, and volatile substances as described below (mean over all replicates).

The amount of radioactivity recovered from day 7 until the end of the study declined with time. This decrease is clearly attributed to loss of 14CO2 from the system, due to the high degree of total mineralization observed. (Additional experiments demonstrating the loss of 14CO2 during the acidification and partitioning of the water phase are described in the study report. Furthermore, loss of 14CO2 probably happened during measurement of pH, O2, and redox potential after sampling. The high amounts of 14CO2 loss neither influence the calculation of the degradation rate of benzyl-14C-alpha-cypermethrin nor the amounts of metabolites found.)

Water phase
The amount of radioactivity in the Rhine River water decreased rapidly from day 0 (49.9% TAR) to day 105 (1.9% TAR). Similarly, the radioactivity was rapidly eliminated from the Pond water decreasing from 61.2% TAR at day 0 to 0.9% TAR at the end of the incubation period.

Sediment phase
The amount of radioactivity in the sediment immediately after treatment amounted to 50.0% and 36.9% TAR in the Rhine and the Pond system, respectively. The radioactivity reached a maximum of 64.1% and 62.7% TAR on day 2, thereafter it decreased slowly and represented 41.4 and 29.9% TAR at day 105 in the Rhine and the Pond system, respectively.
In the Rhine system the amount of non-extractable radioactivity increased from 1.6% TAR after 6 hours of incubation time to a maximum of 23.2% TAR on day 61. Thereafter, until day 105, an apparent plateau value was observed (= 18.9% TAR on day 105).
In the Pond system the non-extracted radioactivity was higher than for the Rhine system on day 30 and can be explained by the higher amounts of organic carbon present in Pond sediment. For time intervals 6 hours, 30 and 105 days, the corresponding values were 0.9, 37.3, and 21.2% TAR, respectively.

Volatiles
The mineralization of benzyl-14C-alpha-cypermethrin was high in both water/sediment systems (Table 7.2.2.3-3 and Table 7.2.2.3-4). Thus the levels of 14CO2 amounted to 38.0% TAR after 61 days in the Rhine system and only 24.9% TAR were recovered on day 105. In the Pond system, levels of 14CO2 accounted for 49.0 and 53.1% TAR after 61 and 105 days, respectively.
The amount of other volatile compounds was negligible in both systems since it did not exceed 0.1% TAR during the experiment.

Table 7.2.2.3-3: Balance of the applied radioactivity in the Rhine water/sediment system treated with benzyl-^{14}C-alpha-cypermethrin [% TAR] (mean of two replicates)

Incubation time in days	Radioactivity in water*	Total radioactivity in sediment	Extractables** (sediment)	Non-Extractables (sediment)	Volatile compounds	^{14}C-CO_2	Total radioactivity
0	49.9	50.0	49.3	0.7	n.d.	n.d.	99.9
0.25	40.2	56.7	55.2	1.6	n.d.	n.d.	96.9
1	37.0	59.7	57.8	2.0	<0.1	0.1	96.8
2	32.4	64.1	60.8	3.3	<0.1	0.2	96.7
7	24.8	63.2	55.1	8.3	<0.1	1.7	89.7
14	18.1	61.8	42.3	19.4	<0.1	6.5	86.3
30	5.5	59.9	39.4	20.5	<0.1	21.5	86.9
61	1.8	50.3	27.1	23.2	<0.1	38.0	90.0
105	1.9	41.4	22.6	18.9	<0.1	24.9	68.1

TAR = Total applied radioactivity
n.d. = Not determined
* Radioactivity determined directly in water
** Includes Sohxlet extraction, for sampling intervals 14 to 105 days.

Table 7.2.2.3-4: Balance of the applied radioactivity in the Pond water/sediment system treated with benzyl-^{14}C-alpha-cypermethrin [% TAR] (mean of two replicates)

Incubation time in days	Radioactivity in water	Total radioactivity in sediment	Extractables (sediment)	Non-Extractables (sediment)	Volatile compounds	^{14}C-CO_2	Total radioactivity
0	61.2	36.9	36.0	0.9	n.d.	n.d.	98.0
0.25	50.4	47.8	46.9	0.9	n.d.	n.d.	98.2
1	45.4	46.1	44.0	2.1	<0.1	0.1	91.6
2	41.7	53.9	49.1	4.9	<0.1	0.2	95.8
7	31.2	59.9	45.1	14.8	<0.1	3.2	94.3
14	19.5	62.7	37.3	25.5	<0.1	11.2	93.4
30	10.7	50.2	12.9	37.3	<0.1	26.1	86.9
61	0.8	35.4	9.8	25.6	<0.1	49.0	85.1
105	0.9	29.9	8.7	21.2	<0.1	53.1	83.9

TAR = Total applied radioactivity
n.d. = Not determined
* Radioactivity determined directly in water
** Includes Sohxlet extraction, for sampling intervals 14 to 105 days.

Extractability of radioactivity from water

The extracted radioactivity by partitioning with ethyl acetate at pH 4 decreased with increasing incubation time, because of the decrease in the amount of radioactivity in water. It represented 49.8% and 61.0% of the total applied radioactivity on day 0 in the Rhine and in the Pond system, respectively. Thereafter, it decreased to 1.0% and 1.7% on day 30 in the Rhine and in the Pond system, respectively.
Between 77 and 100% of the radioactivity in water were recovered by partitioning with ethyl acetate at days 0-7. On days 14 and 30 this amounts were lower (16 to 60% of radioactivity) due to loss of 14CO2 during partitioning. In %TAR this loss of radioactivity amounted to 6.9 and 3.3% in the Rhine system at day 14 and 30 and to 6.7 and 8.2% in the Pond system.
The remaining amount of radioactivity in the aqueous phase was very low and ranged from 0.9% TAR on day 0 to a maximum of 4.3% TAR on day 7 in the Rhine system and 0.2% on day 0 to 5.4% 6 hours after the application in the Pond system.
Analysis of the water phases on day 61 and 105 was not performed, because only very low amounts of the total applied radioactivity (maximum of 1.9% on day 105 in the Rhine system and 2.0% on day 61 in the Pond system) were present in water.

B. TRANSFORMATION OF PARENT COMPOUND

Characterization and identification of residues in water and sediment extracts

An overview of active substance and metabolites for the water samples and sediment extracts is presented in Table 7.2.2.3-5 and Table 7.2.2.3-6.
The radioactive fractions found in water had a similar chromatographic behavior as those found in sediment extracts. Hence, only one notation was used for the same metabolite found in water or sediment for the Rhine and the Pond system, respectively.

Table 7.2.2.3-5: Metabolite overview for water and sediment after application of benzyl-^{14}C-alpha-cypermethrin to the Rhine system [% TAR] (mean of two replicates)

Incubation time [days]		Radioactive fraction							
		P	**M1**	**M2**	**M3** [a]	**M4**	**M5**	**M6**	**M8**
0	water	49.9	-	-	-	-	-	-	-
	sediment	*49.3*	-	-	-	-	-	-	-
	total	**99.2**	**-**	**-**	**-**	**-**	**-**	**-**	**-**
0.25	water	39.0	1.2	-	-	-	-	-	-
	sediment	*53.7*	-	-	-	-	-	*1.5*	-
	total	**92.7**	**1.2**	**-**	**-**	**-**	**-**	**1.5**	**-**
1	water	34.0	3.0	-	-	-	-	-	-
	sediment	*52.5*	-	-	-	-	-	*5.3*	-
	total	**86.5**	**3.0**	**-**	**-**	**-**	**-**	**5.3**	**-**
2	water	15.9	1.0	1.4	11.6	0.4	1.7	0.4	-
	sediment	*54.9*	-	-	-	-	*2.0*	*3.8*	-
	total	**70.8**	**1.0**	**1.4**	**11.6**	**0.4**	**3.8**	**4.3**	**-**
7	water	1.6	2.5	2.6	17.3	0.8	-	-	-
	sediment	*36.5*	*2.3*	-	*4.4*	*4.4*	*5.8*	*1.7*	-
	total	**38.1**	**4.8**	**2.6**	**21.7**	**5.2**	**5.8**	**1.7**	**-**
14	water	1.4	3.7	-	11.9	-	0.8	0.2	-
	sediment	*27.0*	*2.7*	-	*3.9*	*3.2*	*3.0*	*2.6*	-
	total	**28.4**	**6.4**	**-**	**15.8**	**3.2**	**3.8**	**2.8**	**-**
30	water	n.d.	3.7	-	1.8	-	-	-	-
	sediment	*21.7*	*3.4*	-	*1.4*	*4.1*	*4.9*	*4.0*	-
	total	**21.7**	**7.1**	**-**	**3.2**	**4.1**	**4.9**	**4.0**	**-**
61	water	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.
	sediment	*14.6*	*1.9*	-	-	*1.9*	*3.2*	*3.1*	*2.3*
	total	**14.6**	**1.9**	**-**	**-**	**1.9**	**3.2**	**3.1**	**2.3**
105	water	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.
	sediment	*12.9*	*1.0*	-	*0.2*	*2.3*	*1.9*	*3.1*	*1.0*
	total	**12.9**	**1.0**	**-**	**0.2**	**2.3**	**1.9**	**3.1**	**1.0**

TAR = Total applied radioactivity
P = Parent compound
- = Not detected
n.p. = Not performed
[a] 3-PBA

Table 7.2.2.3-6: Metabolite overview for water and sediment after application of benzyl-^{14}C-alpha-cypermethrin to the Pond system [% TAR] (mean of two replicates)

Incubation time [days]		Radioactive fraction										
		P	**M1**	**M2**	**M3** [a]	**M4**	**M5**	**M6**	**M7**	**M8**	**M9**	**M10**
0	water	61.2	-	-	-	-	-	-	-	-	-	-
	sediment	34.9	-	-	-	-	-	*1.0*	-	-	-	-
	total	**96.1**	-	-	-	-	-	**1.0**	-	-	-	-
0.25	water	50.4	-	-	-	-	-	-	-	-	-	n.p.
	sediment	45.0	-	-	-	-	-	*1.9*	-	-	-	*0.5*
	total	**95.4**	-	-	-	-	-	**1.9**	-	-	-	**0.5**
1	water	42.6	2.8	-	-	-	-	-	-	-	-	-
	sediment	30.5	-	-	-	-	-	-	-	-	-	-
	total	**73.1**	**2.8**	-	-	-	-	-	-	-	-	-
2	water	28.8	0.5	-	9.6	0.7	1.2	0.9	-	-	-	-
	sediment	46.8	-	-	-	-	*1.2*	*1.0*	-	-	-	-
	total	**75.6**	**0.5**	**-**	**9.6**	**0.7**	**2.4**	**1.9**	-	-	-	-
7	water	8.0	1.1	1.2	18.0	-	3.0	-	-	-	0.8	-
	sediment	27.9	-	-	*5.1*	-	*4.9*	*4.1*	-	-	-	-
	total	**35.9**	**1.1**	**1.2**	**23.1**	**-**	**7.9**	**4.1**	-	-	-	-
14	water	-	1.7	-	16.2	-	0.3	1.3	-	-	-	-
	sediment	19.8	-	-	*4.4*	*5.0*	*5.3*	*2.8*	-	-	-	-
	total	**19.8**	**1.7**	**-**	**20.6**	**5.0**	**5.6**	**4.0**	**-**	**-**	**-**	**-**
30	water	-	3.5	2.0	2.3	-	0.5	1.0	0.6	-	-	-
	sediment	4.1	*3.4*	-	*2.0*	-	*1.6*	*1.8*	-	-	-	-
	total	**4.1**	**6.9**	**2.0**	**4.3**	**-**	**2.1**	**2.8**	**0.6**	**-**	**-**	**-**
61	water	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.
	sediment	4.0	*0.7*	*0.7*	*0.5*	*1.4*	*0.4*	*1.3*	*0.3*	*0.4*	-	-
	total	**4.0**	**0.7**	**0.7**	**0.5**	**1.4**	**0.4**	**1.3**	**0.3**	**0.4**	**-**	**-**
105	water	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.	n.p.
	sediment	2.0	*0.8*	*0.5*	*0.9*	*1.3*	*0.9*	*1.0*	*0.2*	*0.3*	*0.5*	*0.3*
	total	**2.0**	**0.8**	**0.5**	**0.9**	**1.3**	**0.9**	**1.0**	**0.2**	**0.3**	**0.5**	**0.3**

TAR = Total applied radioactivity
P = Parent compound
- = Not detected
n.p. = Not performed
[a] 3-PBA

Water phase

The amount of benzyl-14C-alpha-cypermethrin (P) in the Rhine River water decreased rapidly from 49.9% TAR at day 0 to 1.4% TAR at day 14. Thereafter, no parent compound was detected.

Besides parent compound, mainly one metabolite (M3) identified as WL 44607 (3-PBA), was found at maximum amounts of 17.3% TAR. Radioactive fraction M2, characterized as WL 46114, was detected in low amounts (≤ 2.6% TAR). In addition, four other minor metabolites (all unknown) were detected in the Rhine River water, but their concentrations did not exceed 3.7% TAR.

Similarly, the test item disappeared rapidly from the water in the Pond system, from 61.2% TAR at day 0 to 8.0% TAR at day 7. Thereafter, no parent compound was detected in water extracts.
The parent compound was degraded to a maximum of eight metabolites during the incubation period. The main degradation product was metabolite M3, identified as WL 44607 (3-PBA), reaching a maximum of 18.0% TAR on day 7. Radioactive fraction M2 (WL 46114) was detected only on day 7 and day 30 and amounted to 1.2 and 2.0% TAR, respectively. Other radioactive fractions (all unknown) were found at maximal amounts of 3.5% TAR.

Sediment phase
The amount of benzyl-14C-alpha-cypermethrin in Rhine sediment increased from 49.3% TAR on day 0 to 54.9% TAR on day 2. Thereafter, the extracted amount decreased to 12.9% TAR (day 105).
Apart from parent compound, six metabolites were detected. The compound M3 (WL 44607,; 3-PBA) reached a maximum of 4.4% TAR at day 7. Other radioactive fractions (M1, M4, M5, M6, M8) reached maximum amounts of 3.4, 4.4, 5.8, 5.3, and 2.3% TAR, respectively.

In the Pond sediment, the extracted amount of the test item increased from 34.9% TAR on day 0 to 46.8% TAR after 2 days. Thereafter, the amount decreased to 2.0% TAR by day 105.
A minimum of ten metabolites were found as well. Metabolite M3 represented a maximum of 5.1% TAR on day 7 and was identified as WL 44607 (3-PBA). Metabolite fractions M1, M4, M5, and M6 were detected at maximum amounts of 3.4, 5.0, 5.3, and 4.1% TAR, respectively. Other radioactive fractions (M2, M7, M5, M9, M10) were detected sporadically, not exceeding 0.7% TAR.

A proposed route of degradation of alpha-cypermethrin in water/sediment systems is given in Figure 7.2.2.3-1.

Characterization of non-extractable residues

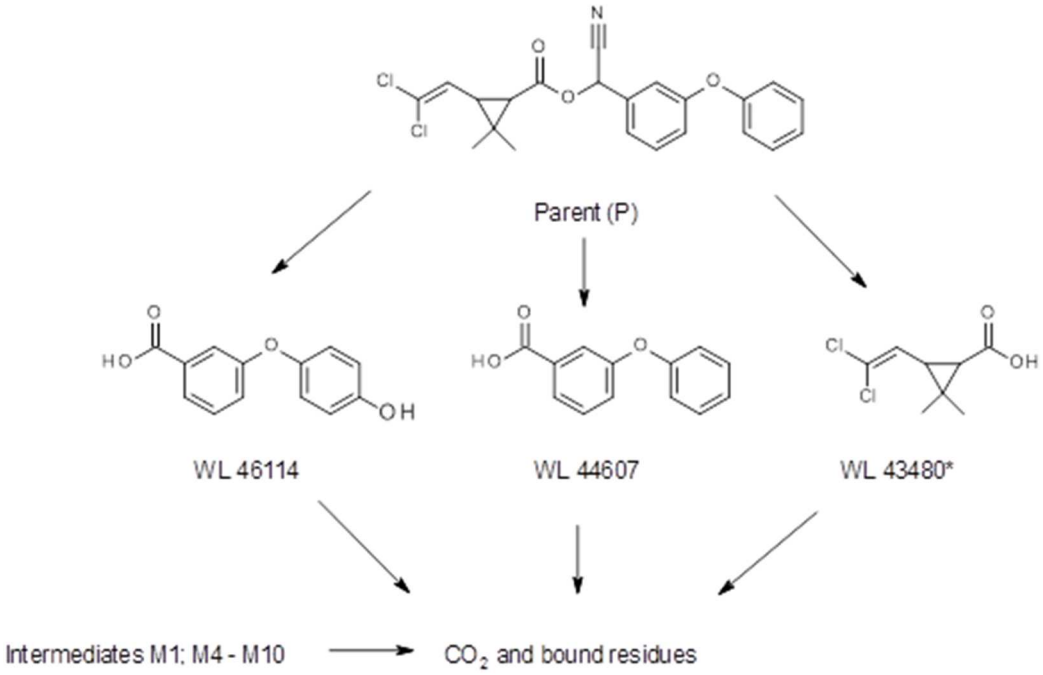
After sediment extraction on day 30 and 105 of incubation, 1-2 sediment samples per aquatic system were monitored for the radioactivity bound to humic acids, fulvic acids, and humin fractions. The distribution of radioactivity (in % TAR) is presented in Table 7.2.2.3-7.

Table 7.2.2.3-7: Distribution of radioactivity between fulvic acids, humic acids, and humins after application of benzyl-^{14}C-alpha-cypermethrin to water/sediment systems [% TAR]

DAT	Sample	NER	Fulvic acids	Humic acids	Humins
Rhine system					
30	A	22.4	5.8	7.2	9.4
30	B	18.6	5.1	6.0	7.5
105	B	20.0	6.0	7.9	6.1
Pond system					
30	A	37.2	4.2	6.8	26.2
30	B	37.3	4.0	5.5	27.7
105	A	21.7	2.3	16.4	2.9

TAR = Total applied radioactivity
DAT = Days after treatment
NER = Non-extractable radioactivity

Figure 7.2.2.3-1: Proposed route of degradation of alpha-cypermethrin in water/sediment systems



* detected in study AL-630-012 [VOELKL, S. (1993): 14C-ALPHACYPERMETHRIN (CYCLOPROPYL—1-14C): DEGRADATION AND METABOLISM, BASF DocID 1993/7002022]

Dissipation and degradation rates

The calculation of dissipation and degradation rates in the water/sediment system according to the recommendations of the FOCUS work group on degradation kinetics [FOCUS (2006)] are subject of a separate report provided in section CA 7.2.2.3/3 [BASF DocID 2014/1159506].

III. CONCLUSION

Results revealed that the test item was rapidly eliminated from both aquatic systems at 20°C ± 2°C in the dark. It quickly dissipated in the water phase by hydrolysis, mineralization, and adsorption to sediment. Only 1.6 and 8.0% TAR remained in water after 7 days in the Rhine River and Pond aquatic systems, respectively.
The minera1ization of benzyl-14C-alpha-cypermethrin was very high and amounted to more than 38 and 53% TAR in Rhine River and Pond system, respectively.
The test item in the sediment system was degraded relatively fast as well – at the end of the study the amount of parent compound left amounted to 12.9 and 2.0% TAR of the in the Rhine and the Pond system, respectively.
Non-extractable residues were found at the end of the study in amounts of 18.9% TAR (Rhine) and 21.2% TAR (Pond).

Report: CA 7.2.2.3/2
Voelkl S., 1993a
14C-Alphacypermethrin (cyclopropyl-1-14C): Degradation and metabolism in aquatic systems
AL-630-012
Guidelines: BBA IV 5-1
GLP: yes
(certified by Eidgenoessisches Departement des Inneren, Bern, Schweiz)

Executive Summary

The degradation of cyclopropyl-1-14C-alpha-cypermethrin in two aerobic water/sediment systems was investigated: one Rhine River and one Pond aquatic system.

Test vessels were treated with cyclopropyl-1-14C-alpha-cypermethrin with an initial concentration of 0.014 µg L-1 corresponding to a field application rate of about 42 g a.s. ha-1 when assuming that the a.s. is homogeneously distributed in a natural water of 30 cm depth. The test vessels were connected to an aeration system at a temperature of 20 ± 2°C in a dark room. Samples were taken directly after the treatment and 6 hours, 24 hours and 2, 7, 14, 30, 62 and 105 days after application. Water and sediment extracts were analyzed by thin-layer-chromatography. The amount of non-extractable residues was determined by combustion and liquid scintillation counting (LSC). Volatiles were trapped in appropriate trapping solutions and also analyzed by LSC. Metabolite identification was done by co-chromatography with reference substances.

After a lag period of about 30 to 60 days, the parent molecule was rapidly mineralized, resulting in high amounts of 14C-CO2. The mean amount of 14C-CO2 accounted for 33.2% of the total applied radioactivity (TAR) after 105 days in the Rhine system and to 40.0% TAR in the Pond system. The amount of other volatile compounds was very low ≤ 0.2% TAR).

The amount of the test item detected in the water phase of the Rhine system decreased from 48.2% TAR on day 0 to 0.8% TAR after 14 days. Thereafter, the parent molecule was no longer detected. The corresponding values obtained for the water in the Pond system were 49.5% and 2.7% TAR, on day 0 and after 7 days, respectively. After 14 days, the parent molecule was not detectable in the water phase.

Analysis of the water phase revealed that, besides the parent molecule, up to ten and five radioactive fractions in the Rhine system and Pond system were present, respectively.

One major radioactive fraction, designated as RW1 and PW1, was found in the Rhine and Pond system. The chromatographic behavior of this fraction matched with that of the reference WL 43480 (DCVA). This fraction ranged from 2.4% to 29.4% TAR for the Rhine system. The corresponding values for the Pond system ranged from 3.2% to 47.3% TAR.

Radioactive fractions RW7 and RW9, detected in the Rhine water, reached maximum amounts of 8.2% and 11.2% TAR after 105 days of incubation. Radioactive fractions RW2 - RW6, RW8, and RW10 did not exceed 4.9% TAR during the whole incubation time.

One radioactive fraction (PW3), detected in the Pond water, reached a maximum amount of 8.9% TAR at one sampling occasion. The other radioactive fractions present did not exceed an amount of 3.4% TAR.

Extraction of the soil sediments showed an increasing amount of radioactivity from 41.0% TAR on day 0 to a maximum amount of 59.0% TAR on day 14. Afterwards, the amount decreased to 26.1% TAR until 105 days of incubation for the Rhine system. The corresponding values obtained for the Pond system were 42.6% TAR on day 0, 52.6% TAR after 7 days, and 8.5% TAR after 105 days, respectively.

Analysis of the extractable radioactivity showed that besides the parent molecule up to six and seven radioactive fractions for Rhine and Pond system were present, respectively.

Radioactive fractions RS1 and PS1 represented the main metabolite, characterized as reference WL 43480 (DCVA). They were detected from day 1 onward and occurred for a maximum amount of 9.5% and 19.3% TAR in the Rhine and Pond system, respectively. One radioactive fraction in the Rhine sediment, designated as RS2, reached a maximum amount of 10.3% TAR. Other radioactive fractions were present at lower percentages, accounting for a maximum of 5.6% TAR.

The results obtained indicated, that the elimination of test item from the total aquatic system was dependent from the organic carbon content of the sediments (Rhine system: 0.9 g C x100 g-1 dry soil and Pond system 5.4 g C x100 g-1 dry soil). Thus, in the Rhine system 22.6% TAR were present in water at the end of the study and 8.9% TAR in the Pond system. Furthermore, higher amounts of 14C-CO2 (see above) and bound residues were formed in the Pond system.
Bound residues were formed in amounts of 37.1% TAR in the Pond system compared to 16.2% TAR in the Rhine system.

From these results and with respect to the study design, it could be stated that the elimination of cyclopropyl-1-14C-alpha-cypermethrin from the water phase proceeded mainly via rapid degradation to polar metabolites. After a lag period of 30 to 60 days, mineralization and adsorption to the humin, humic, and fulvic acid fractions in the sediments took place.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Material

Internal code: BAS 310 I
Reg. No.: 4078193
CAS Number: 67375-30-8
Chemical name: 1:1 mixture of (S)-alpha-cyano-3-phenoxybenzyl-(1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (R)-alpha-cyano-3-phenoxybenzyl-(1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethyl cyclopropanecarboxylate
Molecular formula: C22H19Cl2NO3
Molecular mass: 418.0 g mol-1
Label: cyclopropyl-1-14C
Specific activity: 134 mCi g-1
Radiochemical purity: 99% (checked by RCC using TLC in at least two solvent systems)

2. Test system

Two different test systems were applied:

System I Rhine-river water + Rhine sediment (Rhine, Mumpf/ Switzerland)
System II Pond water + Pond sediment (Pond, “Judenweiher”/Switzerland)

The physico-chemical properties of the systems are summarized in Table 7.2.2.3-8.

Table 7.2.2.3-8: Characterization of the water/ sediment system

Water							
Origin		**River**			**Pond**		
		Study before start		**end**	**Study before start**		**end**
		sampling site	**at RCC**		**sampling site**	**at RCC**	
Temperature surface 5 cm above sediment	[°C]	3.2 3.5		n.d.	0.7 1.0		n.d.
pH surface 5 cm above sediment		8.2 8.2		7.5	7.2 7.2		8.2
Redox potential 5 cm above sediment	[mV]	264 240		201	80 55		188
Oxygen content	[mg L^{-1}]	12.2 11.8		5.4	1.3 1.2		6.0
NO_3-N NO_2-N NH_4-N N-total	[mg L^{-1}]	0.78 0.02 <0.78 max. 1.58		0.12 0.01 0.01 0.14	0.16 0.03 0.78 0.97		0.41 0.01 0.02 0.44
P as ortho-phosphorous		0.12		0.38	0.05		0.09
Total organic carbon	[mg C L^{-1}]		3.0	16.4		9.3	18.6
Hardness	[°dH]	14.0		30.0	22.0		23.0
Sediment							
Origin		**River**			**Pond**		
		Study before start		**end**	**Study before start**		**end**
		sampling site	**at RCC**		**sampling site**	**at RCC**	
pH (KCl) Redox potential	 [mV]	 -20	7.8	n.d. -248	 -10	7.1	n.d. -400
N-total (Kjeldahl)	[g kg^{-1} sed.]		0.68	n.d.		1.65	n.d.
P-total	[g kg^{-1} sed.]		0.41	n.d.		0.69	n.d.
Total organic carbon content	[g C 100 g^{-1} dry soil]		0.9	n.d.		5.4	n.d.
Cation exchange capacity	[mVal N 100 g^{-1} dry soil]		6.6	n.d.		39.9	n.d.
Biomass	[mg microbial C 100 g^{-1} soil]		51	47.2		480	247.1
Dry mass	[kg dry soil kg^{-1} fresh sediment]		0.66	0.66		0.30	0.30
Particle size distribution (USDA) clay (< 2 µm) silt (2-50 µm) sand (<50 µm)	 [%] [%] [%]		loamy sand 3.9 15.2 80.9	 n.d.		sandy clay 25.4 31.8 42.8	 n.d.

B. STUDY DESIGN

1. Experimental conditions

The study was performed in an open gas-flow-system in 1000 mL all-glass metabolism flasks containing about 275 g and 190 g of sediment for the Rhine and Pond aquatic systems, respectively, and 550 mL of water. The system was ventilated with moistened CO2-free air with simultaneous gentle agitation of the water column by means of a suspended magnetic stirrer. The out coming air was passed through a trapping system of two bottles of 2N NaOH (50 mL) and one bottle of ethylene glycol (50 mL).

The water/sediment systems were incubated at 20 ± 2°C in the dark until an equi1ibrium based on measurable variables was reached. For this purpose, the oxygen concentration, pH and redox potential of selected samples were measured after the colloidal particles had settled down. One day before the application of the test article, the oxygen concentration, pH and redox potential of all samples were determined.

Aliquots of 390 µL containing 7.8 µg of cyclopropyl-14C-alpha-cypermethrin in acetone were applied drop-wise to each flask. Reserve as well as control flasks were included in the study. By applying the maximum recommended field rate of 42 g cyclopropyl-1-14C-alpha-cypermethrin ha-1 and assuming that the test item is homogeneously distributed in the top 30 cm of the water column a target concentration of 14 µg L-1 was achieved.

The microbial biomass of the sediments was determined at the start of the incubation period and at the end of the experiment in the control samples.

2. Sampling

Samples of water and sediment were taken for analysis directly after the treatment and 6, 24, 48 hours and 7, 14, 30, 62 and 105 days after application. For each sampling day, duplicates from each system, Rhine and Pond, were analyzed. The water was sampled down to a depth of 10-30 cm and the sediment was sampled from the top 5-10 cm of each system. This sampling site was located 1-2 m from firm land.

Volatile traps
The absorption solutions were replaced on each sampling interval or about every two weeks and monitored for 14C-CO2 and volatiles.

3. Analytical procedures

Water phase
After separation from sediment, the radioactivity in the water was determined directly by LSC.
Aliquots of up to 300 mL of the water phase were submitted to partitioning with ethyl acetate for two times. In order to obtain a balance, aliquots of the water phases after extraction as well as the radioactivity extracted with ethyl acetate were sampled and submitted to LSC.
The radioactivity remaining in the water phase after extraction was designated as unresolved radioactivity (Table 7.2.2.3-11 and Table 7.2.2.3-13).

Sediment phase
The sediment samples were submitted to the following extractions:
1. - 1-4 times with acetonitrile. The acetonitrile extracts were combined and concentrated under reduced pressure and submitted to TLC-analysis
2. For samples obtained on days 30, 62 and 105, a Soxhlet extraction of the sediments was performed with acetonitrile for about 16 hours. The Soxhlet samples were not analyzed due to their low radioactivity level.

After extraction, aliquots of the sediment samples were submitted to combustion in order to determine the amount of non-extractable radioactivity.

Characterization of bound residues
Organic matter fractionation was performed on sediment samples from sampling day 105 after extraction. Only one sample from the river and Pond sediments per interval was used. This fractionation was performed by extraction of the soil sample with 0.5 M NaOH solution, thereafter, the humic acids were precipitated by decreasing the pH to about 1. Centrifugation permits separation of the fulvic acids which remain in the liquid phase. The humin fraction remains unextracted together with the clay mineral and aluminum oxides.

Volatiles
Radioactivity in the volatile trapping solutions was determined by LSC.

4. Kinetic modeling

The kinetic analysis was carried out according to previous requirements in the summarized study. A new kinetic analysis following the recommendations of the FOCUS work group on degradation kinetics [FOCUS (2006)] will be provided in section CA 7.2.2.3/3 [BASF DocID 2014/1159506].

II. RESULTS AND DISCUSSION

A. MASS BALANCE

Total mean recoveries were 94.3% TAR for water/sediment system I (Rhine; Table 7.2.2.3-9) and 100.8% TAR for system II (Pond; Table 7.2.2.3-10), respectively. These recoveries result from hydrosoil, water and volatile substances, as described below (mean over all replicates).

Water phase
The level of radioactivity in the Rhine-river water decreased from 53.7% TAR at day 0 to 22.6% TAR after 105 days (Table 7.2.2.3-9). The radioactivity level in Pond detected at day 0 amounted to 55.5% TAR and decreased to 8.9% TAR at day 105 (Table 7.2.2.3-10).

Sediment phase
The extractable amount of radioactivity (ERR) in the Rhine-river sediment amounted to 41.0% TAR at day 0 and increased to 73.7% TAR at day 2. Thereafter, ERR decreased to 26.1% TAR at day 105. The non-extractable radioactivity was low until day 14, ranging from 1.9% TAR at day 0 to 6.1% TAR after 14 days (Table 7.2.2.3-9). Thereafter, the non-extractable amount accounted for 16.9% and 16.2% TAR after 60 and 105 days, respectively.

The extractable amount of radioactivity in the Pond system increased from 42.6% on day 0 to a maximum amount of 57.4% TAR on day 2. Until the end of incubation, it decreased to 8.5%. The non-extractable radioactivity was low until day 7, increasing from 2.3% at day 0 to 7.4% TAR. Thereafter, it increased to 37.1% TAR after 105 days of incubation (Table 7.2.2.3-10).

Volatiles
Mineralization of cyclopropyl-1-14C-alpha-cypermethrin was significant in the Rhine system. After a lag period of 60 days, the mineralization reached 33.2% TAR on day 10, (Table 7.2.2.3-9). The amount of other volatile compounds was very low (≤ 0.2% TAR).

Slightly lower amounts of 14C-CO2 were found within the first 60 days after incubation in the Pond system. After a lag period of 60 days, 8.1% TAR were determined to be 14C-CO2 on day 60, increasing to 40.0% TAR after 105 days, respectively. The volatile radioactivity trapped in ethylene glycol did not exceed 0.2% TAR (Table 7.2.2.3-10).

Table 7.2.2.3-9: Balance of the applied radioactivity in the Rhine water/sediment system treated with cyclopropyl-1-^{14}C-alpha-cypermethrin [% TAR] (mean of two replicates)

Incubation time in days	Radio-activity in water	Soxhlet extract from sediment	"Cold" extractables from sediment	Non-extractables (sediment)	Total radio-activity in sediment	Volatile compounds	^{14}C-CO_2	Total radio-activity
0	53.7	n.d.	41.0	1.9	42.8	n.d.	n.d.	96.5
0.25	25.1	3.1	47.0	0.3	50.4	<0.1	<0.1	75.4
1	33.2	n.d.	56.3	1.6	57.9	<0.1	<0.1	91.1
2	27.8	n.d.	73.7	2.9	76.6	<0.1	<0.1	104.4
7	28.4	0.0	57.0	3.9	60.9	<0.1	0.2	89.6
14	38.2	0.0	59.0	6.1	65.1	<0.1	0.6	104.0
30	42.1	2.2	38.8	13.8	54.7	<0.1	3.9	100.7
60	22.2	2.1	31.0	16.9	50.0	<0.1	16.1	88.3
105	22.6	0.4	25.7	16.2	42.4	0.2	33.2	98.4

TAR = Total applied radioactivity
n.d. = Not determined

Table 7.2.2.3-10: Balance or the applied radioactivity in the Pond water/sediment system treated with cyclopropyl-1-^{14}C-alpha-cypermethrin [% TAR] (mean of two replicates)

Incubation time in days	Radio-activity in water	Soxhlet extract from sediment	"Cold" extractables from sediment	Non-extractables (sediment)	Total radio-activity in sediment	Volatile compounds	^{14}C-CO_2	Total radio-activity
0	55.4	n.d.	42.6	2.3	44.9	n.d.	n.d.	100.3
0.25	35.3	3.0	47.1	0.2	50.3	<0.1	<0.1	85.5
1	54.3	n.d.	50.3	2.6	52.9	<0.1	<0.1	107.1
2	47.0	n.d.	57.4	3.0	60.4	<0.1	<0.1	107.3
7	48.3	n.d.	52.6	7.4	60.0	<0.1	0.1	108.3
14	52.4	n.d.	45.4	10.3	55.7	0.1	0.2	108.2
30	53.1	4.0	28.4	15.4	43.9	<0.1	2.2	103.2
60	31.6	3.2	27.9	21.6	49.5	<0.1	8.1	92.4
105	8.9	0.2	8.3	37.1	45.4	0.2	40.0	94.7

TAR = Total applied radioactivity
n.d. = Not determined

B. TRANSFORMATION OF PARENT COMPOUND

Characterization and identification of residues in water and sediment extracts

An overview of active substance and metabolites for the water samples and sediment extracts is presented in Table 7.2.2.3-11 to Table 7.2.2.3-14.

Table 7.2.2.3-11: Pattern of metabolites in organic extracts of water samples of the aquatic system I (Rhine) after various time intervals [% TAR] (mean of two replicates)

Incubation time [days]	Radioactive fraction												
	P	RW1	RW2	RW3	RW4	RW5	RW6	RW7	RW8	RW9	RW10	un-resolved[a]	Total
0	48.2	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	5.5	53.7
0.25	18.1	2.4	n.d.	0.9	0.9	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	2.8	25.1
1	18.2	7.1	1.3	2.2	2.2	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	2.2	33.2
2	3.5	16.6	1.7	n.d.	2.2	n.d.	n.d.	n.d.	n.d.	n.d.	0.9	2.9	27.9
7	0.7	22.5	n.d.	n.d.	2.3	n.d.	0.6	0.5	n.d.	0.3	0.7	0.8	28.4
14	0.8	29.4	n.d.	n.d.	2.3	n.d.	1.7	n.d.	n.d.	n.d.	1.9	2.1	38.2
30	n.d.	26.9	n.d.	n.d.	2.6	2	4.9	n.d.	n.d.	4.1	n.d.	1.6	42.1
60	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	7.5	n.d.	10.4	n.d.	4.3	22.2
105	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	8.2	0.5	11.2	2.8	n.d.	22.6

TAR = Total applied radioactivity
P = Parent compound
n.d.= Not detected
RW1 = WL 43480 (DCVA)
[a] Radioactivity remaining in the water phase after extraction

Table 7.2.2.3-12: Pattern of metabolites in extracts of sediment samples of the aquatic system I (Rhine) after various time intervals [% TAR] (mean of two replicates)

Incubation time [days]	Radioactive fraction							
	P	RS1 WL 43480	RS2	RS3	RS4	RS5	RS6	Total
0	37.0	1.5	2.5	n.d.	n.d.	n.d.	n.d.	41.0
0.25	43.5	3.4	n.d.	n.d.	n.d.	n.d.	n.d.	47.0
1	48.0	0.9	7.4	n.d.	n.d.	n.d.	n.d.	56.3
2	61.8	1.6	10.3	n.d.	n.d.	n.d.	n.d.	73.1
7	41.6	6.3	9.2	n.d.	n.d.	n.d.	0.0	57.0
14	41.2	9.5	2.8	5.6	n.d.	n.d.	0.0	59.0
30	20.4	9.0	n.d.	5.3	1.2	0.9	2.1	34.8
60	19.2	3.3	1.4	1.9	1.7	2.2	1.3	31.0
105	17.0	1.8	2.2	n.d.	1.2	2.3	1.2	26.7

TAR = Total applied radioactivity
P = Parent compound
RS1 = WL 43480 (DCVA)
n.d. = Not detected

Table 7.2.2.3-13: Pattern of metabolites in organic extracts of water samples of the aquatic system II (Pond) after various time intervals [% TAR] (mean of two replicates)

Incubation time [days]	Radioactive fraction							
	P	PW1 WL 43480	PW2	PW3	PW4	PW5	unresolved [b]	Total
0	49.5	n.d.	n.d.	n.d.	n.d.	n.d.	6.0	55.5
0.25	26.6	3.2	1.6	n.d.	n.d.	n.d.	4.0	35.3
1	33.1	9.5	2.7	2.6	n.d.	n.d.	6.3	54.3
2	15.8	19.3	3.4	3.6	n.d.	n.d.	4.9	47.0
7	2.7	40.3	2.6	1.6	n.d.	n.d.	1.1	48.3
14	n.d.	47.3	2.6	1.6	n.d.	n.d.	0.8	52.3
30	n.d.	41.9	2.9	7.6	n.d.	n.d.	0.8	53.1
60	n.d.	17.9	2.0	8.9	1.0	n.d.	1.8	31.6
105	n.d.	n.d.	n.d.	4.7	2.9	1.4	n.d.	8.9

TAR = Total applied radioactivity
P = Parent compound
PW1 = WL 43480 (DCVA)
n.d.= Not detected
[b] Radioactivity remaining in the water phase after extraction

Table 7.2.2.3-14: Pattern of metabolites in extracts of sediment samples of the aquatic system II (Pond) after various time intervals [% TAR] (mean of two replicates)

Incubation time [days]	Radioactive fraction								
	P	PS1 WL 43480	PS2	PS3	PS4	PS5	PS6	PS7	Total
0	40.8	1.8	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	42.6
0.25	47.1	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	47.1
1	43.3	3.1	3.9	n.d.	n.d.	n.d.	n.d.	n.d.	50.3
2	51.9	0.8	4.7	n.d.	n.d.	n.d.	n.d.	n.d.	57.4
7	32.9	9.4	4.1	n.d.	6.2	n.d.	n.d.	n.d.	52.6
14	16.9	19.3[a]	2.4	n.d.	6.6	n.d.	n.d.	n.d.	45.4
30	4.7	16.8	n.d.	0.7	2.4	1.7	0.5	1.6	28.4
60	5.6	15.7	n.d.	0.6	1.0	2.9	1.1	1.0	27.9
105	1.6	1.4	n.d.	0.4	0.5	1.8	0.9	1.8	8.4

TAR = Total applied radioactivity
P = Parent compound
PS1 = WL 43480 (DCVA)
n.d.= Not detected
[a] Erroneously reported as 19.5% TAR

A proposed route of degradation of alpha-cypermethrin in water/sediment systems is given in section CA 7.2.2.3/1 *[MAMOUNI, A. (1993): FASTAC (BENZYL-14C): DEGRADATION AND METABOLISM IN AQUATIC SYSTEMS, BASF DocID 19937002021].*

Water phase

The amount of parent substance decreased from 48.2% TAR at day 0 to 0.7% TAR at day 7. At day 14, the parent molecule was only detected for one replicate. Besides the parent compound, up to ten metabolite fractions (RW1 - RW10) were found in the Rhine river water. RW1 represents the main metabolite fraction. Its chromatographic behavior matched with that of reference compound WL 43480 (DCVA). It could be detected unti1 day 30, starting from 2.4% TAR after 6 hours to its maximum of 29.4% TAR on day 14 and ending at 26.9% TAR after 30 days of incubation. Radioactive fractions RW2, RW3, RW4 RW5, RW6, RW8 and RW10 were detected at a maximum amount of 4.9% TAR (0.0007 mg parent equivalents kg-1).

Radioactive fractions RW7 and RW9 were the main radioactive fractions after 60 days and 105 days of incubation. They reached a maximum amount of 8.2% and 11.2% TAR after 105 days of incubation, respectively. The metabolite fractions RW9 and RW7 remained unknown. Its chromatographic behavior did not match with that of reference compound WL 47133. The very low Rf values of metabolites RW7 and RW9 as well as their increase after the dissipation of DCVA allow the conclusion that they are oxidation products of DCVA, most likely hydroxylated at one of the methyl groups (RW7, M310I003) and further oxidized to a dicarboxylic acid (RW9).

The radioactivity that was not extractable from the water phase by partitioning was designated as unresolved radioactivity. It did not exceed 5.5% TAR.

The amount of parent substance in the Pond water decreased continuously from 49.5% TAR at day 0 to 2.7% TAR after 7 days of incubation.
Besides the parent molecule, up to five radioactive fractions, designated as PW1 to PW5, were detected in the Pond water. PW1 represented the main radioactive. The chromatographic behavior of this fraction matched with that of the reference compound WL 43480 (DCVA). Its amount increased from 3.2% TAR after 6 hours to its maximum amount of 47.3% TAR after 14 days of incubation. Thereafter, it decreased to 17.9% TAR after 60 days of incubation.
Radioactive fraction PW2 was present at oscillating values. The amount ranged between 1.6% TAR after 6 hours of incubation and 3.4% TAR after 48 hours of incubation time.
Radioactive fraction PW3 occurred from day 1 onward also at oscillating amounts. The amount detected ranged from 2.6% to 8.9% TAR. This fraction shows a similar chromatographic behavior as fraction RW9 detected in water of the Rhine system.
PW4 and PW5 represented highly polar fractions not exceeding 2.9% TAR.
The amount of radioactivity which was not extracted by partitioning was designated as unresolved radioactivity. Its amount did not exceed 6.3% TAR.

Sediment phase
Besides the parent molecule, up to six radioactive fractions (RS1 - RS6) were detected in the Rhine sediment. The amount of the parent molecule increased from 37.0% TAR immediately after application to a maximum amount of 61.8% TAR on day 2 and decreased thereafter to 17.0% TAR on day 105.
Radioactive fraction RS1 represented the main metabolite in the sediment of the Rhine system. Its chromatographic behavior matched with that of reference WL 43480 (DCVA). This fraction was detected first immediately after application with 1.5% TAR and amounted to 9.0% TAR after 30 days of incubation. After 105 days of incubation it accounted for 1.8% TAR.
Radioactive fraction RS2 was present from day 0 onward until day 14 and on day 60 and 105. The amount detected for RS2 started from 2.5% TAR on day 0 to a maximum amount of 10.3% after 2 days of incubation. Thereafter, its amount decreased to 2.8% and 2.2% TAR after 14 and 105 days of incubation.
Radioactive fractions RS3 to RS6 were detected irregularly. They did not exceed 5.6% TAR.

Beside the parent molecule, up to seven radioactive (PS1 -PS7) fractions were detected in the Pond sediment. The amount of the parent molecule increased from 40.8% TAR on day 0 to a maximum amount of 51.9% TAR after 2 days of incubation. Thereafter, the amount decreased to 1.6% TAR after 105 days of incubation.
Radioactive fraction PS1 was the main metabolite in the Pond system and matched with reference compound WL 43480 (DCVA). This fraction amounted from 1.8% TAR on day 0 to 19.3% TAR on day 14 and decreased to 1.4% TAR on day 105.
Radioactive fractions PS2 to PS7 did not exceed 6.6% TAR.

Characterization of non-extractable residues

After sediment extraction on day 105 of incubation, one sediment sample per aquatic system was monitored for the radioactivity bound to humic acids, fulvic acids, and humin fractions. The distribution of radioactivity (in % TAR) is presented in Table 7.2.2.3-15.

Table 7.2.2.3-15: Distribution of radioactivity between fulvic acids, humic acids, and humins after application of cyclopropyl-1-^{14}C-alpha-cypermethrin to water/sediment systems [% TAR]

DAT	Sample	NER	Fulvic acids	Humic acids	Humins
105	River sediment, sample A	16.6	7.4	5.4	3.8
105	Pond sediment, sample A	40.6	6.9	23.6	10.1

TAR = Total applied radioactivity
DAT = Days after treatment
NER = Non-extractable radioactivity

Dissipation and degradation rates

The calculation of dissipation and degradation rates in the water/sediment system according to the recommendations of the FOCUS work group on degradation kinetics [FOCUS (2006)] are subject of a separate report provided in section CA 7.2.2.3/3 [BASF DocID 2014/1159506].

III. CONCLUSION

The present study showed that cyclopropyl-1-14C-alpha-cypermethrin was rapidly eliminated from both aquatic systems at 20°C ± 2°C in the dark.
The disappearance of the parent compound from the water phase by hydrolysis and mineralization and adsorption into sediment was relatively rapid in both systems, only 0.7% and 2.7% TAR remained in water after 7 days in the Rhine River and Pond aquatic systems, respectively.
For both systems, several radioactive fractions were detected. One radioactive fraction designated as RW1 and PW1 in the water phase of the Rhine and Pond as well as RS1 and PS1 occurring in the sediments of the systems represented the main metabolite of cyclopropyl-1-14C-alpha-cypermethrin, which was rapidly further degraded. The chromatographic behavior of these fractions matched with that of the reference compound WL 43480 (DCVA). The maximum amount detected for this fraction was 29.4% TAR in the Rhine water and 47.3% TAR in the Pond water.
The minera1ization of cyclopropyl-1-14C-alpha-cypermethrin was very high and amounted to more than 33 and 40% TAR in Rhine-river and Pond system, respectively.

The rate of degradation in sediment was higher in the Pond system because of the higher microbial activity. Higher adsorption did not prevent further mineralization.

Report: CA 7.2.2.3/3
Sachers S., 2014c
Kinetic evaluation of degradation of BAS 310 I - alpha-cypermethrin in water/sediment systems: Determination of trigger and modeling endpoints according to FOCUS
2014/1159506
Guidelines: none
GLP: no

Executive Summary

The aim of the study was to evaluate the dissipation and degradation kinetics of alpha-cypermethrin and its metabolites 3-PBA, DCVA, RS2, and RW9/RS5 in two aerobic water/sediment systems with two different labels [*BASF DocID 1993/7002021 (AL-630-011) and BASF DocID 1993/7002022 (AL-630-012)]* and to derive trigger and modeling endpoints according to the recommendations of FOCUS kinetics.

In the two laboratory studies, the degradation of alpha-cypermethrin was investigated over a period of up to 105 days in two water/sediment systems called System I and II (a river and a pond located in Switzerland). Two different radio-labels of the active substance were used in the studies. The experimental data were evaluated using single first order (SFO), first-order multi-compartment (FOMC), double first-order in parallel (DFOP) and hockey stick (HS) kinetic models at the evaluation levels P-I, P-II, and M-I (dissipation and degradation).

The evaluation at Level P-I resulted in reliable trigger and modeling half-lives, except for the sediment phase of System I (cyclopropyl label). DegT50 values for the whole system ranged from 3.0 to 4.7 days as trigger endpoints and from 5.4 to 1000 days (default value) as modeling endpoints. The evaluation at Level P-II did not result in reliable fits for any of the evaluated water/sediment systems.

Dissipation half-lives for the metabolites at Level M-I for the whole system derived as trigger as well as modeling endpoints ranged from 10.2 to 12.5 days for 3-PBA and from 23.2 to 35.3 days for DCVA. Degradation half-lives for the metabolites 3-PBA and DCVA at Level M-I for the whole system derived as trigger as well as modeling endpoints ranged from 7.6 to 8.5 days and from 25.5 to 30.3 days, respectively. No dissipation and degradation endpoints could be derived for metabolite RS2, for metabolite RW9/RS5, only a formation fraction could be calculated.

I. MATERIAL AND METHODS

Kinetic evaluation of the dissipation and degradation behavior of alpha-cypermethrin and its metabolites 3-PBA, DCVA, RS2, and RW9/RS5 was conducted for two aerobic water/sediment studies in the dark with two natural aerobic aquatic systems. The studies are summarized in section *CA 7.2.2.3/1 [BASF DocID 1993/7002021 (AL-630-011)*] and section *CA 7.2.2.3/2 [BASF DocID 1993/7002022 (AL-630-012*)]. The water/sediment systems were taken from the river Rhine (System I) and pond Judenweiher (System II), both located in the area close to Rheinfelden, Switzerland. Kinetic evaluation was performed in order to derive degradation and dissipation parameters as triggers for additional work (trigger endpoints) and for modeling purposes (modeling endpoints) according to the recommendation of the FOCUS workgroup on degradation kinetics [*FOCUS (2006*)].

Two radio-labels of the active substance, referred to as benzyl- [*BASF DocID 1993/7002021 (AL-630-011)*] and cyclopropyl-label [*BASF DocID 1993/7002022 (AL-630-012)*] were used. The test vessels were treated at a rate corresponding to a field application rate of about 42 g a.s. ha-1. The test system was incubated for up to 105 days at 20°C in the dark.

Kinetic modeling

The appropriate kinetic model was identified considering the procedures and kinetic models proposed by the FOCUS workgroup on degradation kinetics [FOCUS (2006)]. According to FOCUS, degradation and dissipation endpoints were derived for use as triggers for future work and for use as modeling inputs.

Kinetic models included in the assessment

Kinetic evaluation at Level P-I (one-compartment approach) was performed for alpha-cypermethrin (both labels) degradation in the total system as well as dissipation from the water and sediment phase of the test systems.

At Level P-II (two-compartment approach: water and sediment), the kinetic analysis considered the degradation in water and sediment and the partitioning between both phases.

Kinetic evaluation at Level M-I dissipation (one-compartment approach) was performed for the metabolites 3-PBA (benzyl label) and DCVA (cyclopropyl label) which were observed in both compartments and the metabolite RS2 which was only observed in the sediment phase of the cyclopropyl label. For DCVA for the water phase of System I, where no decline of the residues was observed, no kinetic evaluation at Level M-I dissipation was performed. The metabolite RW9/RS5 was not included at this level as there was no decline of the residues observed during the study. Estimation of persistence as well as modeling endpoints was based on metabolite decline from the maximum observed concentration in the total system and in the water and sediment phase.

In addition, the degradation of the metabolites 3-PBA, DCVA, and RW9/RS5 in the total system was assessed at Level M-I degradation by kinetic evaluation of the complete degradation pathway with a combined fit of parent and metabolites. 3-PBA and DCVA were assumed to be formed directly from alpha-cypermethrin while RW9/RS5 was assumed to be formed from DCVA. The metabolite RS2 was not included at this level as only its concentration in the sediment was reported and there was no information available on its possible occurrence in the water phase.

As the purpose of the study was to derive modeling and trigger endpoints, all four kinetic models proposed by FOCUS were used during the evaluation (SFO, FOMC, DFOP and HS). Details on the models are given in the FOCUS Kinetics guidance [*FOCUS (2006*)].

At Level P-I and M-I dissipation, trigger endpoints were derived from the kinetic models that provided the best fit to the measured data, generally indicated by the lowest $\chi 2$ - error. Modeling endpoints were derived preferably from the SFO model. If the SFO model was not appropriate, pragmatic procedures were used to derive conservative pseudo-SFO degradation rates from the appropriate bi-phasic model.

The appropriateness of a distinct kinetic model to describe degradation can be tested with the following checks recommended by FOCUS [*FOCUS (2006*)]:

Visual assessment of goodness-of-fit
Estimation of the error percentage at which the $\chi 2$ test is passed [*Equation 6-2 in FOCUS (2006)]*
t-test to evaluate whether estimated degradation parameters differ from zero.

A kinetic model is considered appropriate if the residuals are randomly distributed, the $\chi 2$ - error value is low (ideally below 15% but larger values may be acceptable if the visual fit is acceptable) and the t-test for the degradation parameters is passed at 10% error level.

Data handling

At Level P-I and P-II of the analysis as well as at Level M-I degradation, the kinetic evaluation started on the day of treatment (i.e. 0 DAT). The initial concentration of the parent substance in the total system or in water was set to the material balance recovered at 0 DAT as recommended by FOCUS [*FOCUS (2006)*]. Accordingly, the initial concentration in the sediment phase was assumed to be zero at Level P-II.

The assessment of dissipation in sediment at Level P-I and in all compartments at Level M-I dissipation only requires kinetics to be fitted to the corresponding decline data, starting from the maximum observed concentration in the compartment. The dissipation of the respective compound was thus evaluated starting at the day of maximum occurrence that was defined as 0 days after maximum concentration (0 DAMC). All later time points were adjusted accordingly as days after maximum concentrations (DAMC).

Values below the quantification (LOQ) or detection limit (LOD) for parent compound and degradation products were treated as recommended by FOCUS [*FOCUS (2006)*]. The LOD in water and sediment determined in study conducted with the cyclopropyl-labeled test item [*BASF DocID 1993/7002022 (AL-630-012)*] amounted to 0.07 ppb which corresponds to 0.5% TAR. The LOD retrieved from the study conducted with the benzyl-labeled test item [*BASF DocID 1993/7002021 (AL-630-011)]* was given as 0.05 ppb which corresponds to 0.4% TAR.

Software for kinetic evaluation

The software package KinGUII (version 2.2014.224.1704) was used for parameter fitting [BASF DocID 2007/1062781; WITT, J., GAO, Z., MEYER, H. (2014)].

Experimental data

The experimental data of alpha-cypermethrin and its metabolites 3-PBA, DCVA, RS2, and RW9/RS5 used as model input values for the kinetic evaluations are given in Table 7.2.2.3-16 and Table 7.2.2.3-17 (benzyl label [*BASF DocID 1993/7002021 (AL-630-011)*]) and Table 7.2.2.3-18 and Table 7.2.2.3-19 ([*BASF DocID 1993/7002022 (AL-630-012*)]).

Table 7.2.2.3-16: Experimental data of alpha-cypermethrin (benzyl label) and 3-PBA in System I (Rhine) used for kinetic evaluation

DAT [d]	Alpha-cypermethrin residues [% TAR]			3-PBA residues [% TAR]		
	Total system	Water	Sediment	Total system	Water	Sediment
0	98.9[a]	98.9[a]	0[b] / 51.1	0[c]	<LOD	<LOD
0	100.8[a]	100.8[a]	0[b] / 47.4	0[c]	<LOD	<LOD
0.25	89.6	36.1	53.5	<LOD	<LOD	<LOD
0.25	95.8	41.8	54.0	<LOD	<LOD	<LOD
1	86.7	35.2	51.5	0.2[d]	<LOD	<LOD
1	86.1	32.7	53.4	0.2[d]	<LOD	<LOD
2	70.4	15.0	**55.4**	10.7	10.7	<LOD
2	71.1	16.7	**54.4**	12.4	12.4	<LOD
7	40.2	1.6	38.6	**22.0**	**17.4**	**4.6**
7	35.8	1.5	34.3	**21.5**	**17.3**	**4.2**
14	31.5	2.9	28.6	8.0	8.0	0.2[d]
14	25.4	0.2[d]	25.4	23.7	15.9	7.8
30	23.8	0.2[d]	23.8	2.1	0.8	1.3
30	19.5	<LOD	19.5	4.3	2.8	1.5
61	16.8	n.a.[e]	16.8	0.2[d]	0.2[d,e]	0.2[d]
61	12.4	n.a. [e]	12.4	0.2[d]	0.2[d,e]	0.2[d]
105	15.4	n.a. [e]	15.4	0.5	n.a. [e]	0.5
105	10.5	n.a. [e]	10.5	<LOD	n.a. [e]	<LOD

DAT = Days after treatment
TAR = Total applied radioactivity
n.a.= Not analyzed
Bold numbers: peak concentration considered for single-compartment evaluation; previous values were omitted; sampling dates were adjusted accordingly.
[a] Set to material balance.
[b] Set to zero for kinetic evaluation at Level P-II.
[c] Set to zero for kinetic evaluation at Level M-I degradation.
[d] Set to ½ LOD (LOD = 0.4% TAR).
[e] Samples were not processed further as the radioactivity recovered in the water phase from day 61 and 105 was very low (max. of 2.0% on day 61).

Table 7.2.2.3-17: Experimental data of alpha-cypermethrin (benzyl label) and 3-PBA in System II (Judenweiher) used for kinetic evaluation

DAT [d]	Alpha-cypermethrin residues [% TAR]			3-PBA residues [% TAR]		
	Total system	Water	Sediment	Total system	Water	Sediment
0	96.0[a]	96.0[a]	0[b] / 40.0	0[c]	<LOD	<LOD
0	100.0[a]	100.0[a]	0[b] / 29.9	0[c]	<LOD	<LOD
0.25	95.7	51.3	44.4	<LOD	<LOD	<LOD
0.25	95.0	49.4	45.6	<LOD	<LOD	<LOD
1	78.1	42.9	35.2	0.2[d]	<LOD	<LOD
1	68.2	42.4	25.8	0.2[d]	<LOD	<LOD
2	73.2	31.0	**42.2**	8.1	8.1	<LOD
2	78.1	26.6	**51.5**	11.1	11.1	<LOD
7	33.5	7.0	26.5	**27.7**	**21.1**	**6.6**
7	38.3	9.0	29.3	**18.5**	**14.8**	**3.7**
14	23.1	0.2[d]	23.1	20.6	15.3	5.3
14	16.6	0.2[d]	16.6	20.6	17.1	3.5
30	3.4	<LOD	3.4	3.4	1.7	1.7
30	4.8	<LOD	4.8	5.3	2.9	2.4
61	6.2	n.a.[e]	6.2	0.2[d]	0.2[d,e]	0.2[d]
61	1.9	n.a.[e]	1.9	1.0	0.2[d,e]	1.0
105	2.4	n.a.[e]	2.4	1.2	n.a.[e]	1.2
105	1.6	n.a.[e]	1.6	0.6	n.a.[e]	0.6

DAT = Days after treatment
TAR = Total applied radioactivity
n.a. = Not analyzed
Bold numbers: peak concentration considered for single-compartment evaluation; previous values were omitted; sampling dates were adjusted accordingly.
[a] Set to material balance.
[b] Set to zero for kinetic evaluation at Level P-II.
[c] Set to zero for kinetic evaluation at Level M-I degradation.
[d] Set to ½ LOD (LOD = 0.4% TAR).
[e] Samples were not processed further as the radioactivity recovered in the water phase from day 61 and 105 was very low (max. of 2.0% on day 61).

Table 7.2.2.3-18: Experimental data of alpha-cypermethrin (cyclopropyl label), DCVA, RW9/RS5, and RS2 in System I (Rhine) used for kinetic evaluation

DAT [d]	Alpha-cypermethrin residues [% TAR]			DCVA residues [% TAR]			RW9/RS5 residues [% TAR]			RS2 residues [% TAR]
	Total system	Water	Sediment	Total system	Water	Sediment	Total system	Water (RW9)[e]	Sediment (RS5)[e]	Total system
0	96.6[a]	96.6[a]	0[b] / 33.5	0[c] / 2.2	<LOD	2.2	0[c]	<LOD	<LOD	0[c] / 3.5
0	96.5[a]	96.5[a]	0[b] / 40.5	0[c] / 0.8	<LOD	0.8	0[c]	<LOD	<LOD	0[c] / 1.5
0.25	70.3	20.7	49.6	6.4	2.5	3.9	<LOD	<LOD	<LOD	<LOD
0.25	53.1	15.6	37.5	5.3	2.3	3.0	<LOD	<LOD	<LOD	<LOD
1	70.8	19.2	51.6	6.8	6.8	<LOD	<LOD	<LOD	<LOD	9.8
1	61.5	17.1	44.4	9.2	7.3	1.9	<LOD	<LOD	<LOD	5.0
2	62.6	3.1	**59.5**	19.9	16.8	3.1	<LOD	<LOD	<LOD	**11.6**
2	68.1	3.9	**64.2**	16.4	16.4	<LOD	0.25[d]	<LOD	<LOD	**9.0**
7	31.7	0.5	31.2	23.4	17.7	5.7	0.6	0.6	<LOD	9.9
7	52.9	0.9	52.0	34.3	27.4	6.9	0.25[d]	<LOD	<LOD	8.5
14	52.6	1.6	51.0	**33.2**	**25.9**	**7.3**	0.25[d]	<LOD	<LOD	<LOD
14	31.3	0.25[d]	31.3	**44.6**	**32.9**	**11.7**	0.25[d]	<LOD	<LOD	5.6
30	16.0	0.25[d]	16.0	32.1	25.6	6.5	6.9	5.2	1.7	<LOD
30	24.8	<LOD	24.8	39.6	28.2	11.4	3.0	3.0	<LOD	<LOD
60	24.6	<LOD	24.6	3.7	0.25[d]	3.7	12.9	10.7	2.2	1.2
60	13.7	<LOD	13.7	3.0	0.25[d]	3.0	12.4	10.1	2.3	1.5
105	23.9	<LOD	23.9	1.3	<LOD	1.3	12.7	10.6	2.1	2.0
105	10.1	<LOD	10.1	2.3	<LOD	2.3	14.4	11.8	2.6	2.5

DAT = Days after treatment
TAR = Total applied radioactivity
Bold numbers: peak concentration considered for single-compartment evaluation; previous values were omitted; sampling dates were adjusted accordingly.
[a] Set to material balance.
[b] Set to zero for kinetic evaluation at Level P-II.
[c] Set to zero for kinetic evaluation at Level M-I degradation.
[d] Set to ½ LOD (LOD = 0.5% TAR).
[e] Not evaluated, no decline observed.

Table 7.2.2.3-19: Experimental data of alpha-cypermethrin (cyclopropyl label) and DCVA in System II (Judenweiher) used for kinetic evaluation

DAT [d]	Alpha-cypermethrin residues [% TAR]			DCVA residues [% TAR]		
	Total system	Water	Sediment	Total system	Water	Sediment
0	101.1[a]	101.1	0[b] / 47.1	0[c]	<LOD	<LOD
0	99.6[a]	99.6	0[b] / 54.5	0[c] / 3.3	<LOD	3.3
0.25	65.4	27.7	37.7	3.0	3.0	<LOD
0.25	81.9	25.4	56.5	3.4	3.4	<LOD
1	81.5	37.1	44.4	8.5	6.7	1.8
1	71.3	29.1	42.2	16.5	12.3	4.2
2	68.3	17.9	**50.4**	20.5	18.9	1.6
2	67.1	13.8	**53.3**	19.7	19.7	<LOD
7	35.6	1.0	34.6	52.9	43.5	9.4
7	35.5	4.3	31.2	46.5	37.2	9.3
14	15.6	0.25[d]	15.6	**68.8**	**49.7**	**19.1**
14	18.1	0.25[d]	18.1	**64.3**	**44.9**	**19.4**
30	4.3	<LOD	4.3	57.6	42.3	15.3
30	5.0	<LOD	5.0	59.7	41.4	18.3
60	2.8	<LOD	2.8	25.4	13.4	12.0
60	8.3	<LOD	8.3	41.7	22.3	19.4
105	1.4	<LOD	1.4	0.9	0.25[d]	0.9
105	1.7	<LOD	1.7	1.8	0.25[d]	1.8

DAT = Days after treatment
TAR = Total applied radioactivity
Bold numbers: peak concentration considered for single-compartment evaluation; previous values were omitted; sampling dates were adjusted accordingly.
[a] Set to material balance.
[b] Set to zero for kinetic evaluation at Level P-II.
[c] Set to zero for kinetic evaluation at Level M-I degradation.
[d] Set to ½ LOD (LOD = 0.5% TAR).

II. RESULTS AND DISCUSSION

The datasets for each water/sediment system were analyzed considering the procedures and kinetic models proposed by the FOCUS workgroup on degradation kinetics [FOCUS (2006)]. The complete fitting procedure is given in the original study report.

Level P-I

The evaluation of both test systems for both labels at Level P-I resulted in reliable endpoints for alpha-cypermethrin for degradation in the total system as well as for dissipation in the water and sediment phase, except for the sediment phase of System I (cyclopropyl label) where no acceptable fit was obtained.

An overview of the estimated trigger and modeling endpoints for alpha-cypermethrin in both water/sediment systems is given in Table 7.2.2.3-20 and Table 7.2.2.3-21.

Table 7.2.2.3-20: Trigger endpoints for alpha-cypermethrin at Level P-I

Label, System	Total system		Water		Sediment	
	$DegT_{50}$/ $DegT_{90}$ [d]	**Kinetic model**	**DT_{50}/ DT_{90} [d]**	**Kinetic model**	**DT_{50}/ DT_{90} [d]**	**Kinetic model**
Benzyl, System I (Rhine)	4.7 / 123.5	DFOP	0.2 / 3.4	HS	12.5 / 797.0	FOMC
Benzyl, System II (Judenweiher)	4.6 / 23.6	FOMC	0.4 / 5.9	HS	8.6 / 33.8	HS
Cyclopropyl, System I (Rhine)	3.0 / >1000	FOMC	0.1 / 1.5	HS	No reliable endpoints derived	
Cyclopropyl, System II (Judenweiher)	3.3 / 26.9	FOMC	0.1 / 4.2	HS	7.5 / 31.0	HS

Table 7.2.2.3-21: Modeling endpoints for alpha-cypermethrin at Level P-I

Label, System	Total system		Water		Sediment	
	$DegT_{50}$ [d]	**Kinetic model**	**DT_{50} [d]**	**Kinetic model**	**DT_{50} [d]**	**Kinetic model**
Benzyl, System I (Rhine)	83.5	DFOP	1.0[a]	HS	95.0[b]	DFOP
Benzyl, System II (Judenweiher)	5.4	SFO	1.8[a]	HS	8.7	SFO
Cyclopropyl, System I (Rhine)	1000[c]	FOMC	0.5[a]	HS	No reliable endpoints derived	
Cyclopropyl, System II (Judenweiher)	8.1[b]	FOMC	1.3[a]	HS	7.7	SFO

[a] Calculated as $DT_{50} = DT_{90}/3.32$
[b] Calculated as $DT_{50} = \ln 2/k2$
[c] Default value as calculated $DegT_{90}$ value was >1000 days.

Level P-II

The kinetic evaluation revealed no reliable fit for any of the evaluated water/sediment systems. Consequently, no trigger or modeling endpoints were calculated.

Level M-I dissipation

Trigger and modeling endpoints at Level M-I dissipation were calculated for the metabolites 3-PBA and DCVA for the total system, the water, and sediment phase of all water/sediment systems, except for DCVA for the water phase of System I where no decline of the residues was observed. For the metabolite RS2, the kinetic evaluation at Level M-I dissipation was conducted for the sediment phase of System I (cyclopropyl label) but did not result in an acceptable fit. The metabolite RW9/RS5 was not considered for the evaluation at Level M-I dissipation as there was no decline of the residues observed. In each case, SFO was selected as the most appropriate kinetic model to derive trigger as well as modeling endpoints. The results for the metabolites 3-PBA and DCVA are summarized in Table 7.2.2.3-22 and Table 7.2.2.3-23.

Table 7.2.2.3-22: Summary of trigger and modeling endpoints for 3-PBA, Level M-I dissipation

Label, System	Trigger endpoints [a]			Modeling endpoints [a]		
	Total system	Water	Sediment	Total system	Water	Sediment
	DT_{50}/ DT_{90} [d]	DT_{50}/ DT_{90} [d]	DT_{50}/ DT_{90} [d]	DT_{50} [d]	DT_{50} [d]	DT_{50} [d]
Benzyl, System I (Rhine)	10.2 / 34.0	9.2 / 30.6	16.7 / 55.3	10.2	9.2	16.7
Benzyl, System II (Judenweiher)	12.5 / 41.4	11.4 / 38.0	19.3 / 64.1	12.5	11.4	19.3

[a] SFO selected as best-fit model; also used or derivation of modeling endpoints

Table 7.2.2.3-23: Summary of trigger and modeling endpoints for DCVA, Level M-I dissipation

Label, System	Trigger endpoints [a]			Modeling endpoints [a]		
	Total system	Water	Sediment	Total system	Water	Sediment
	DT_{50}/ DT_{90} [d]	DT_{50}/ DT_{90} [d]	DT_{50}/ DT_{90} [d]	DT_{50} [d]	DT_{50} [d]	DT_{50} [d]
Cyclopropyl, System I (Rhine)	23.2 / 77.0	n.c.	36.4 / 120.8	23.2	n.c.	36.4
Cyclopropyl, System II (Judenweiher)	35.3 / 117.3	30.5 / 101.2	50.4 / 167.4	35.3	30.5	50.4

n.c. = Not calculated (no decline observed)
[a] SFO selected as best-fit model; also used or derivation of modeling endpoints

Level M-I degradation

Trigger and modeling endpoints at Level M-I degradation for the metabolites 3-PBA, DCVA, and RW9/RS5 were derived from combined fitting of all data of parent and metabolites from the total system considering the best-fit model for the parent as derived at Level P-I and the SFO model for the metabolites. For 3-PBA and DCVA, formation from the parent was assumed as degradation pathway while RW9/RS5 was assumed to be formed from DCVA. A summary of the results for the metabolites 3-PBA, DCVA, and RW9/RS5 are shown in Table 7.2.2.3-24 to Table 7.2.2.3-26.

Table 7.2.2.3-24: Summary of trigger and modeling endpoints for 3-PBA, Level M-I degradation

Label, System	Kinetic model	Trigger endpoints		Modeling endpoints	
		$DegT_{50}$ [d]	$DegT_{90}$ [d]	$DegT_{50}$ [d]	Formation fraction
Benzyl, System I (Rhine)	SFO	8.5	28.2	8.5	0.460
Benzyl, System II (Judenweiher)	SFO	7.6	25.3	7.6	0.526

Table 7.2.2.3-25: Summary of trigger and modeling endpoints for DCVA, Level M-I degradation

Label, System	Kinetic model	Trigger endpoints		Modeling endpoints	
		$DegT_{50}$ [d]	$DegT_{90}$ [d]	$DegT_{50}$ [d]	Formation fraction
Cyclopropyl, System I (Rhine)	SFO	25.5	84.8	25.5	0.706
Cyclopropyl, System II (Judenweiher)	SFO	30.3	100.8	30.3	0.957

Table 7.2.2.3-26: Summary of trigger and modeling endpoints for RW9/RS5, Level M-I degradation

Label, System	Kinetic model	Trigger endpoints		Modeling endpoints	
		$DegT_{50}$ [d]	$DegT_{90}$ [d]	$DegT_{50}$ [d]	Formation fraction
Cyclopropyl, System I (Rhine)	SFO	n.c.[a]		n.c.[a]	0.262[b]

n.c. = Not calculated
[a] No reliable endpoints derived, no decline observed
[b] From DCVA (formation fraction of DCVA from parent: 0.706)

III. CONCLUSION

The dissipation and degradation kinetics of alpha-cypermethrin and its metabolites 3-PBA, DCVA, RS2, and RW9/RS5 in two water/sediment systems were evaluated according to the recommendations of the FOCUS workgroup on degradation kinetics [FOCUS (2006)]. The visual assessment and goodness-of-fit statistics of the respective models indicate plausible fit. Therefore, the resulting endpoints can be considered reliable.

The experimental data of alpha-cypermethrin and its metabolites 3-PBA, DCVA, RS2, and RW9/RS5 in both test systems (Systems I and II) and for both labels (benzyl and cyclopropyl) were evaluated at Level P-I, Level P-II, and Level M-I (dissipation and degradation).

The evaluation at Level P-I resulted in reliable trigger and modeling half-lives, except for the sediment phase of System I (cyclopropyl label). DegT50 values for the whole system ranged from 3.0 to 4.7 days as trigger endpoints and from 5.4 to 1000 days (default value) as modeling endpoints.

Dissipation half-lives for the metabolites at Level M-I for the whole system derived as trigger as well as modeling endpoints ranged from 10.2 to 12.5 days for 3-PBA and from 23.2 to 35.3 days for DCVA. Degradation half-lives for the metabolites 3-PBA and DCVA at Level M-I for the whole system derived as trigger as well as modeling endpoints ranged from 8.5 to 7.6 days and from 25.5 to 30.3 days, respectively. No dissipation and degradation endpoints could be derived for metabolite RS2, for metabolite RW9/RS5, only a formation fraction could be calculated.

Supplementary information on the behaviour of alpha-cypermethrin in water/sediment systems from literature

Though the studies were not performed with alpha-cypermethrin, conclusion for this compound can be drawn.

Report: CA 7.2.2.3/4
Meyer B.N. et al., 2013a
"Laboratory Degradation Rates of 11 Pyethroids under Aerobic and Anaerobic Conditions" (J. Agric Food Chem. 2013, 61, 4702-4708)
2013/1416300
Guidelines: yes
GLP: no

Executive Summary

Laboratory experiments were conducted to determine degradation rates of 11 pyrethroids (including ζ-cypermethrin and cypermethrin) in water / sediment systems incubated under aerobic and anaerobic conditions at 25 ± 1°C and darkness.

Three sediment samples originating from three different sites and with different physico-chemical properties were combined with water and tested in this study. ζ-cypermethrin and cypermethrin were applied separately in two test mixtures containing other pyrethroids at a nominal concentration of 50 µg kg-1. Water / sediment samples were incubated from zero to 100 days. Samples were extracted with acetonitrile followed by two steps of solid phase extraction (SPE). Analyses were performed by gas-chromatography/mass spectrometry (GC-MS).

Degradation rates were calculated for the overall system, and no attempt was made to distinguish between material in the solution or sediment phases.
Degradation rates appeared to follow SFO kinetics until approximately 50 – 75% of the applied pesticide was degraded. Afterwards, degradation rates slowed down. Degradation was faster under aerobic than under anaerobic conditions. Degradation of ζ-cypermethrin was slightly slower with calculated DT50 values (best fit kinetics if endpoint reached within incubation period) ranging from 4 to 13 days (DT90: 51 days - > end of study period) under aerobic conditions and 26 to 33 days under anaerobic conditions (DT90: > end of study period). DT50 values of cypermethrin ranged from 2.7 to 8.6 days (DT90: 23 - 100 days) when incubated under aerobic conditions and from 13 to 20 days when incubated under anaerobic conditions (DT90: 90 days - > end of study period). The aerobic degradation rate was the slowest for sediment 2, which possessed the highest organic carbon content, while differences in the anaerobic degradation rates were less pronounced.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test material

The eleven test items were grouped into two treatment mixtures. The isomeric analytes were placed into different test mixtures. Hence, ζ-cypermethrin (test mixture 1) and cypermethrin (test mixture 2) were applied separately to the water/sediment samples.

2. Test sites

The sediments used in this study were sediments originating from three sites in California (USA). They differed widely in their physico-chemical properties (e.g., organic carbon content and texture), shown in Table 7.2.2.3-27.

Table 7.2.2.3-27: Soil properties of soil used to investigate degradation rate of cypermethrin and ζ-cypermethrin in sediment

Soil designation origin	**Sediment 1** East of San Diego	**Sediment 2** King Island	**Sediment 3** Contra Costa
Sand [%]	90.3	20.4	49.5
Silt [%]	1.8	30.1	25.9
Clay [%]	7.9	49.5	24.6
Textural class	Sand	Clay	Sandy clay loam
Total organic carbon [%]	0.6	7.4	2.7
pH ($CaCl_2$)	6.4	5.8	6.6
Cation exchange capacity [meq/100 g dry soil]	6.0	19.0	14.8

B. STUDY DESIGN

1. Sampling and analysis

All sediments were sieved to a particle size of ≤ 2mm. Because water was not present in some of the sediments surveyed for inclusion in this study, water was not collected with the sediment. Instead, conditioned water typically used for aquatic studies in the Ecotoxicology Department at the Bayer Facilities in Stilwell, KS, was used.

150-270 mL water was added to 25-50 g sediment, depending on the respective sediment properties. Prior to treatment, a pre-incubation period (12-32 days) was used to acclimate and establish the appropriate conditions (aerobic or anaerobic) in the systems as judged by dissolved oxygen and redox. In the aerobic experiment, flasks containing the water / sediment samples were supplied with humidified air and were incubated at 25 ± 1°C and darkness. Incubation was the same for anaerobic incubated samples, but sample flasks were purged with nitrogen instead of air. Additional control samples were incubated as well under the same conditions. Test mixtures 1+2 were applied separately to the samples at a nominal concentration of 50 µg kg-1 sediment (dry weight equivalent). Incubation was performed under the above mentioned conditions. Dissolved oxygen, pH and redox potential were measured continuously. Microbiological activity was determined at day 0 and day 100 by aerobic or anaerobic plate counts.

Samples were collected in duplicates at seven time intervals (0, 3, 7, 14, 28, 60 and 100 days after treatment). For sample workup, 250 mL acetonitrile was added to the test vessels, the sample was transferred to 1 L plastic bottles, and shaken for 30 min. After vacuum filtration, solid phase extractions (SPE) were performed for sample clean-up and pre-concentration. Therefore, filtrates were extracted first with a 1 g ENVI-Carb cartridge (Sigma-Aldrich, St. Louis, MO, USA). The SPE cartridge was rinsed with acetonitrile / water (3:2, v/v) and methanol, and eluted with dichloromethane (DCM). This solution (dried and redissolved in DCM/cyclohexane [2:3, v/v]) was extracted in a second SPE step using a 1 g NH2 cartridge (Agilent Technologies, Santa Clara, CA, USA). Cartridges were rinsed again with DCM : cyclohexane (2:3, v/v). After adding an internal standard (cyfluthrin-d6), samples were concentrated to dryness, redissolved in cyclohexane and analyzed by gas-chromatography/mass spectrometry (GC-MS).

Analyses were performed using a GC-MS system (Trace GC Ultra; Thermo Scientific) and a Rtx-5MS capillary column (Restek Corporation, Bellefonte, PA, USA).

The limit of quantification (LOQ) was 5.0 ppb for all analytes/test systems, the limit of detection (LOD) ranged from 0.2 – 1.4 ppb depending on the analyte/test system.

2. Kinetic evaluations

Kinetic evaluations were performed using KinGUI 1.1using criteria specified by the FOrum for Co-ordination of pesticide fate models and their USe (FOCUS) [FOCUS (2006)], applying single first-order (SFO) and first-order in parallel (DFOP) kinetics.

Determination of the microbial activity revealed that the test systems remained microbially active until the end of the study.

The analysis method was validated by investigating five replicate test systems with freshly spiked sediment samples at fortification levels of 5 and 50 ppb (each test mixture). Average recoveries of test mixture 1 (containing ζ-cypermethrin) were between 74 and 106% of the applied amounts (relative standard deviation (RSD): 1-8%) and between 72 and 107% of the applied amounts (RSD: 1-13%) for test mixture 2 (containing cypermethrin).

Procedural recoveries of all analyzed samples ranged from 71 to 114% with an overall mean of concurrent of 95.8% (standard deviation (SD): 6.2%). Therefore, residue values were not corrected for concurrent recovery results.

Residue concentrations of ζ-cypermethrin and cypermethrin declined in all of the total water / sediment systems, but the rates of decline varied among the three tested sediments. The authors mentioned that the degradation rates appeared to follow SFO kinetics until approximately 50 – 75% of the applied pesticide was degraded. Afterwards, degradation rates slowed down. Degradation was faster under aerobic than under anaerobic conditions. Degradation of ζ-cypermethrin was slightly slower with calculated DT50 values (best fit kinetics if endpoint reached within incubation period) ranging from 4 to 13 days (DT90: 51 days - > end of study period) under aerobic conditions and 26 to 33 days under anaerobic conditions (DT90: > end of study period). DT50 values of cypermethrin ranged from 2.7 to 8.6 days (DT90: 23 – 100 days) when incubated under aerobic conditions and from 13 to 20 days when incubated under anaerobic conditions (DT90: 90 days - > end of study period). The slowest aerobic degradation rate was found for sediment 2, which possessed the highest organic carbon content. Differences in the anaerobic degradation rates were less pronounced.
First-order half-life values ranged from 5.0 to 21.6 days (ζ-cypermethrin) and 3.0 to 14.1 days (cypermethrin) under aerobic conditions, respectively, and from 32.0 to 46.3 days (ζ-cypermethrin) and 20.1 to 24.5 days (cypermethrin) under anaerobic conditions.

The authors admit that the degradation rates of ζ-cypermethrin and cypermethrin were in the range observed in previous studies conducted with single analytes, revealing that the mixture of compounds had no impact on the observed degradation rates.

III. CONCLUSION

Degradation rates for ζ-cypermethrin and cypermethrin were determined in water / sediment systems for the whole system under aerobic and anaerobic conditions. DT50 values for ζ-cypermethrin and cypermethrin ranged from 2.7 to 13 days under aerobic conditions (DT90 from 51 days to > end of study period) and from 13 to 33 days under anaerobic conditions (DT90: 90 days - > end of study period).

In this study a fast degradation following a biphasic kinetic of cypermethrins was observed. This confirms the results from the water/sediment guideline studies (Mamouni, 1993, CA 7.2.2.3/1 and Voelkl 1993, CA 7.2.2.3/2).

In the water sediment studies from Mamouni, 1993, CA 7.2.2.3/1 and Voelkl 1993, CA 7.2.2.3/2 no chiral chromatographic methods were applied to obtain information on enantioselective degradation. In the aerobic mineralization study (according to OECD 309) these techniques were applied but no enantioselectivity was observed. But in a water/sediment system the microbial activity may be higher, therefore increasing the chance of chiral degradation. The following studies from literature provide information on this topic. The study of Liu CA 7.2.2.3/5 cannot be used to derive endpoints as the test compounds were incubated with strains known to degrade pythrethroids or adapted sediment, but they provide information on enantioselectivity in biological systems.

Report: CA 7.2.2.3/5
Liu W. et al., 2004a
"Isomer Selectivity in Aquatic Toxicity and Biodegradation of Cypermethrin"
(J. Agric Food Chem. 2004, 52, 6233-6238)
2004/1040294
Guidelines: none
GLP: no

Executive Summary

Isomer selectivity of cypermethrin (CP) in its degradation was studied in a degradation experiment with three bacterial degraders, as well as in pond sediment.

In the biodegradation experiment, CP with a nominal concentration of 100 µg L-1 (pH 7) was incubated separately with three known CP degraders (*Vibrio hollisae, Burkholderia picketti, Erwinia carotovora*) in three different population densities. Extraction of CP was performed three times with ethyl acetate at different time intervals. Samples were analyzed by gas-chromatography (GC). Results revealed that the trans diastereomers was degraded preferentially (half-life: 20.6 – 30.0 h) resulting in a relative enrichment of the cis isomers (half-life: 33.5 – 57.7 h). The highest selectivity was observed for *Erwinia carotovora*. The half-life of the insecticidally active enantiomer 1R-cis-αS* was comparatively long (33.6 – 40.3 h), while the unresolved enantiomer pair 1R-trans-αS* + 1S-trans-αR, containing the other active enantiomer, appeared to degrade faster (15.6 – 18.9 h).

In the sediment incubation experiment, CP was applied to pond sediment at a nominal concentration of 1.0 mg kg-1. At different time intervals, CP was extracted three times with acetone : hexane (50:50, v/v) and analyzed by GC. Results revealed a similar selectivity during CP degradation in sediment. Hence, calculated DT50 values were longer for the cis diastereomers (56.8 and 69.3 days) than for the trans diastereomers (39.6 and 33.3 days) The calculated DT50 values of the insecticidally active enantiomer *1R-cis-αS** was among the longest with 74.5 days, whereas the active enantiomer *1R-trans-αS** was probably among the shortest with 40.5 days.

The authors concluded that the overall agreement between the biodegradation and the sediment experiment suggests that the isomer selectivity in CP degradation in sediment was biologically mediated and that isomer selectivity may occur widely in the environment.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test material

Racemically mixed cypermethrin (CP; 98%) and β-CP (86%, enriched in 1R-cis-αS* + 1S-cis-αR and 1S-trans-αR + 1R-trans-αS*) were purchased from Chem Service (West Chester, PA). The isomer-enriched formulations α-CP (96%, enriched in 1R-cis-αS* + 1S-cis-αR) and θ-CP (99%, enriched in 1Rcis-αS*) were provided by FMC (Princeton, PA).

2. Test systems

Bacterial isolates (biodegradation experiment)
Bacterial isolates were obtained by enrichment from sediment previously contaminated with bifenthrin and permethrin, collected from a nursery run-off channel. Three fast-growing bacteria strains were selected as inoculants for the biodegradation experiment.

Sediment sample (degradation experiment)
A sediment sample was collected from a sedimentation pond at a nursery site in Irvine, CA, and contained 0.65% organic carbon and 5% clay. Chemical analysis showed that the sediment was free of CP residues.

B. STUDY DESIGN

1. Experimental conditions

Biodegradation experiment
Isomer selectivity was characterized during degradation of CP by bacterial isolates under aerobic conditions.
A mineral salt solution was adjusted to pH 7.0 with NaOH solution and spiked with CP at a nominal concentration of 100 µg L-1. The initial acetone concentration was 100 µg mL-1. Aliquots were amended with 1.0 mL of enriched bacteria solution. Three CP degraders (Vibrio hollisae, Burkholderia picketti, Erwinia carotovora) were applied separately to reach the following population densities: 8.3 x 107, 2.4 x 108, and 2.5 x 108 colony-forming units per mL. All samples were continuously mixed on a mechanical shaker at room temperature. At different time intervals, triplicate samples were extracted for pesticide residues. Therefore, an aliquot of each sample was three times extracted with 50 mL ethyl acetate for 1 min. The combined extracts were filtered, concentrated and analyzed by gas-chromatography (GC). The overall recovery of the extraction procedure was > 95% for CP or CP isomers.

Degradation in sediment
Changes in the isomer composition due to degradation were further characterized in whole sediment. Wet aliquots from the sediment were immersed with deionized water, spiked with CP and incubated at room temperature (21 ± 2°C). The initial CP concentration was 1.0 mg kg-1. At different time intervals, aliquots of the sediment were removed and analyzed for concentrations of CP diastereomers as well as CP enantiomers. Therefore, the aliquots were extracted three times with acetone : hexane (50:50, v/v) for 60 Minutes. Subsequently, the samples were centrifuged (1300 x g, 20 min), followed by the decantation of the supernatant. The combined extracts were combined, dried with anhydrous sodium sulphate, concentrated, and analyzed by GC. The procedural recovery was > 90%.

2. Stereoisomer separation and analysis

GC methods described in another publication of Liu et al. [LIU, W.P., GAN, J.J. (2004) Separation and analysis of diastereomers and enantiomers of cypermethrin and cyfluthrin by gas chromatography. .J. Agric. Food Chem., 2004, Vol. 52, p. 755-761] were applied to identify CP diastereomers and enantiomers. Separation and analysis was performed using an achiral column for CP diastereomers and on a chiral column for CP enantiomers. The analyses with the achiral column resulted in four peaks, each peak representing two diastereomers. The analyses with the chiral column resulted in six peaks, representing the four cis-isomers of CP in single peaks (separation of enantiomers) and two groups of trans-isomers in two peaks. In both cases, analyses were carried out using an Agilent 6890N GC with electron capture detector (ECD).
Dissipation of CP diastereomers and cis enantiomers were fitted to a first-order decay model to estimate the rate constant k (h-1) and half-life DT50 (h). The fit was generally good, with $r2 \leq 0.91$ for the diastereomers (achiral analysis) and $r2 \leq 0.87$ for the resolved enantiomers (chiral analysis).

II. RESULTS AND DISCUSSION

Biodegradation experiment
CP diastereomers were degraded relatively rapid by the bacterial isolates with the longest half-life of about 58 h. In control samples, CP was relatively stable (< 10% loss after 112 days). In the inoculated samples, a preferentially degradation of the CP trans diastereomers was observed. Half-life values calculated from the results of chiral and achiral analyses ranged from 29.3 to 57.7 h for the cis-isomers of CP and from 15.6 to 30.0 h for the trans-isomers of CP.
The half-life of the insecticidally active enantiomer 1R-cis-αS* was comparatively long (33.6 to 40.3 h), in contrast to the other stereoisomers (29.3 to 39.6 h) for the same degrading bacteria. However, the unresolved enantiomer pair 1R-trans-αS* + 1S-trans-αR (15.6 to 18.9 h), containing the insecticidally active enantiomer, seemed to degrade faster than the enantiomer pair of the other diastereomer (16.4 to 22.9 h).

Degradation in sediment
Dissipation of CP diastereomers or resolved enantiomers followed first order kinetics with $r^2 \geq 0.98$ for the diastereomers and $r2 \geq 0.93$ for the enantiomers. Rate constants and half-lives are given in Table 7.2.2.3-28.

Consistent with the results from the biodegradation experiment, the cis diastereomers were more persistent than the trans diastereomers in the pond sediment. The half-life of 1R-cis-αS* was among the longest with 74.5 days, whereas that for 1R-trans-αS* was probably among the shortest with 40.5 days. The latter could not be clarified due to a lack of separation for the trans diastereomers.

Table 7.2.2.3-28: Half-fife (DT_{50}), and correlation coefficient r^2 for the degradation of cypermethrin diastereomers (determined by achiral GC analysis) and enantiomers (determined by chiral GC analysis) in sediment (according to Liu et al. 2004)

	Diastereomers			
	cis		**trans**	
	1R-cis-αR + 1S-cis-αS	*1R-cis-αSa + 1S-cis-αR*	*1S-trans-αS + 1R-trans-αR*	*1S-trans-αR + 1R-trans-αS*[a]
DT_{50} [d]	56.8	69.3	39.6	33.3
r^2	0.98	0.99	0.99	0.99

	Enantiomers					
	cis				**trans**	
	1R-cis-αR	*1S-cis-αS*	*1R-cis-αS*[a]	*1S-cis-αR*	*1S-trans-αS + 1R-trans-αR*	*1S-trans-αR + 1R-trans-αS*[a]
DT_{50} [d]	78.7	40.1	74.5	43.8	31.5	40.5
r^2	0.94	0.99	0.99	0.99	0.95	0.93

[a] Denotes insecticidally active enantiomers.

III. CONCLUSION

The authors concluded that the overall agreement between the biodegradation and the sediment experiment suggests that the isomer selectivity in capermethrin degradation in sediment was biologically mediated and that isomer selectivity may occur widely in the environment.

Report: CA 7.2.2.3/6
Qin S. et al., 2006a
"Enantioselective Degradation and Chiral Stability of Pyrethroids in Soil and Sediment (J. Agric Food Chem. 2006, 61, 5040-5045)
2006/1050993
Guidelines: none
GLP: no

Executive Summary

Field sediment samples were analyzed and long-term laboratory incubation experiments were conducted to better understand the effect of chirality of pyrethroide insecticides (inter alia cypermethrin (CP)) on biodegradation.

Field sediment samples were taken from the Upper Newport Bay – San Diego Creek watershed (CA, USA) in spring and summer of year 2005. Field samples were extracted three consecutive steps with acetone : methylene chloride (50;50, v/v).

Cypermethrin was only detected in field sediment samples collected in spring. Enantioselective degradation was found for these samples. The direction of enantioselectivity appeared to depend on the sampling location and environmental conditions.

Laboratory incubation experiments were conducted under aerobic and anaerobic conditions with samples derived from one soil and one sediment. In addition, the importance of the microbial community for the enantioselective degradation of CP was investigated in sterilized soil and sediment samples. Incubation samples were treated with either 1R-cis-αS-CP or 1S-cis-αR-CP and extracted after different time intervals by using a mixture of hexane and acetone (50:50, v/v). Analyses were performed with chiral and achiral gas-chromatography.

Half-life values of CP ranged from 53 to 154 days. Enantioselective degradation of cypermethrin occurred in non-sterilized soils under aerobic and under anaerobic incubation. Under both conditions, enantiomer 1R-cis-αS-CP was degraded more rapid than the 1S-cisαR-CP enantiomer. Results from the incubation experiment with sterilized soil and sediment suggested that the enantioselective degradation of CP was driven by microbial transformation.

I. MATERIAL AND METHODS

A. MATERIALS

1. Test material

An analytical standard of racemic cypermethrin (CP; 98%) purchased from Chem Service (West Chester, PA, USA) was used in this study. The isomers 1R-cis-αS-CP and 1S-cis-αR-CP were isolated by using a chiral high performance liquid chromatography (HPLC) column.

2. Test sites

Field sediment samples
Field sediment samples originated from the Upper Newport Bay – San Diego Creek watershed (CA, USA).

Sediment samples for laboratory studies
The study was performed with samples (0-10 cm) of a sandy loam soil (Arlington soil taken from a turfgrass plot at the Agricultural Experiment Station near the University of California, Riverside campus; 67% sand, 24% silt, 9% clay, 0.82% organic matter, pH 6.7), and of a sediment (sediment from San Diego Creek in Irvine, CA; 20% sand, 46% silt, 34% clay, 1.09% organic matter, pH 7.9).

B. STUDY DESIGN

1. Sampling and analysis

Field sediment samples
Field sediment samples were taken in the Upper Newport Bay – San Diego Creek watershed (CA, USA) based on accessibility and land use. Sampling was performed in 2005, one sampling in spring after the rainy season and one sampling in summer in the dry season. Samples of 100 g were homogenized, centrifuged, and the supernatant was decanted.

Aliquots of the sediment samples were mixed with anhydrous sodium sulphate to remove the excess water and extracted by three consecutive steps with acetone : methylene chloride (50;50, v/v) and sonication. After filtration, concentration, and several clean-up steps, samples were analyzed by gas-chromatography (GC).

Sediment samples for laboratory studies
Prior to the experiments, soil and sediment samples were air-dried for 24 h at room temperature, homogenized, and passed through a 2 mm sieve.

2 x 18 aliquots of 10 g of each, soil and sediment, were weight into glass vials. The soil moisture was adjusted to 60% of field holding capacity. For the sediment, around 6 mL of deionized water was added to form a 0.5 cm water layer. In each case, one sample set was incubated under aerobic and one under anaerobic conditions. An additional set of samples was prepared and sterilized by autoclavation to test whether enantioselective degradation was microbially driven. (The respective samples were autoclaved twice at 121°C for 60 min, with a 24 h interval in between.)
The two investigated CP enantiomers (1S-cis-αR-CP and 1R-cis-αS-CP) were applied individually on soil and sediment samples with an application rate of 1 mg kg-1. All spiked samples were incubated at room temperature (20 ± 1°C). Three replicate samples were removed at different time intervals, mixed with anhydrous sodium sulphate, and three-fold extracted with hexane : acetone (50:50, v/v) on a mechanical shaker. Subsequently, samples were centrifuged, concentrated, and analyzed by GC. Preliminary experiments revealed extraction recoveries of > 90%. Samples were stored at -20°C prior to analysis.

Analyses of field sediment samples as well as samples from the incubation experiment were performed on an Agilent 6890N GC system (Palo Alto, CA, USA) with an electron capture detector using either a non-chiral selective column or an enantioselective column.

II. RESULTS AND DISCUSSION

Findings

Field sediment samples

Cypermethrin was only detectable in the field sediment samples collected in spring from the Upper Newport Bay – San Diego Creek watershed. Sediment extracts were analyzed on an enantioselective column and the peak area of the separated peaks was used to calculate the relative fraction of the resolved stereoisomers. The column allowed to obtain single peaks for the four cis-isomers of CP and two peaks for the four trans-isomers of CP. Enantioselective degradation was found for all samples. The direction of enantioselectivity appeared to depend on the sampling location and environmental conditions.

Laboratory degradation experiments

For all treatments, data fitted well to the first order decay model (r^2 = 0.87-1.00). Cypermethrin enantiomers exhibited half-lives ranging from 53 to 154 days. The authors attributed the long half-lives compared to other studies to the comparatively low organic carbon content in the soil and the sediment (0.8 and 1.1% organic matter).

Enantioselective degradation of cypermethrin occurred in non-sterilized soils under aerobic and under anaerobic incubation. Half-lives under aerobic incubation were 71 (*1S-cis-αR-CP*) and 63 days (*1R-cis-αS-CP*) in soil and 85 (*1S-cis-αR-CP*) and 53 days (*1R-cis-αS-CP*) in sediment, respectively. Under anaerobic conditions, half-lives were higher with 120 (1S-cis-αR-CP) and 71 days (*1R-cis-αS-CP*) in soil and 154 (*1S-cis-αR-CP*) and 136 days (*1R-cis-αS-CP*) in sediment, respectively. Hence, the enantiomer *1R-cis-αS-CP* was degraded more rapid than the 1S-cis-αR-CP enantiomer in non-sterilized soil/sediment samples. This difference was significant at α = 0.05.
In sterilized soil/sediment samples no enantioselective degradation was observed. Half-lives under sterilized conditions were 116 (*1S-cis-αR-CP*) and 120 days (*1R-cis-αS-CP*) in soil and120 (*1S-cis-αR-CP*) and 122 days (*1R-cis-αS-CP*) in sediment. The results suggest that the enantioselective degradation of CP was driven by microbial transformation.

The degradation of CP was in part slower under anaerobic conditions than under sterilized conditions. As photodegradation is known to be important for CP degradation, the authors attributed the slower degradation under anaerobic conditions partly to the fact that photodegradation was completely inhibited under anaerobic conditions, as the samples were kept in the nitrogen filled chamber.

III. CONCLUSION

The study demonstrates that enantioselective degradation commonly occurred for cypermethrin under both field and laboratory conditions and that the selectivity was attributed to microbial transformations. However, the direction and degree of enantioselectivity were not always predictable. It appears that both stereochemistry of the chiral compound and environmental conditions influenced the direction and rate of enantioselective degradation.

Summary of maximum occurrences for alpha-cypermethrin metabolites in aquatic laboratory studies

Table 7.2.2.3-29: Maximum occurrences of the metabolites of DCVA, 3-PBA, and 3-PBAld in aquatic laboratory studies

Metabolite	BASF DocID	Study type	Test system	Max. occurrence [%]
DCVA	1993/7002022 (AL-630-012)	Water/sediment	Rhine river	38.9 [a]
			Judenweiher pond	66.6 [a]
3-PBA	1993/7002021 (AL-630-011)	Water/sediment	Rhine river	21.7 [a]
			Judenweiher pond	23.1 [a]
3-PBAld	1993/7001988 (AL-322-002)	Hydrolysis	pH 7, 50°C	21.6[b]
	2001/7001562 (AL-324-003)	Aqueous photolysis	pH 5	15.9

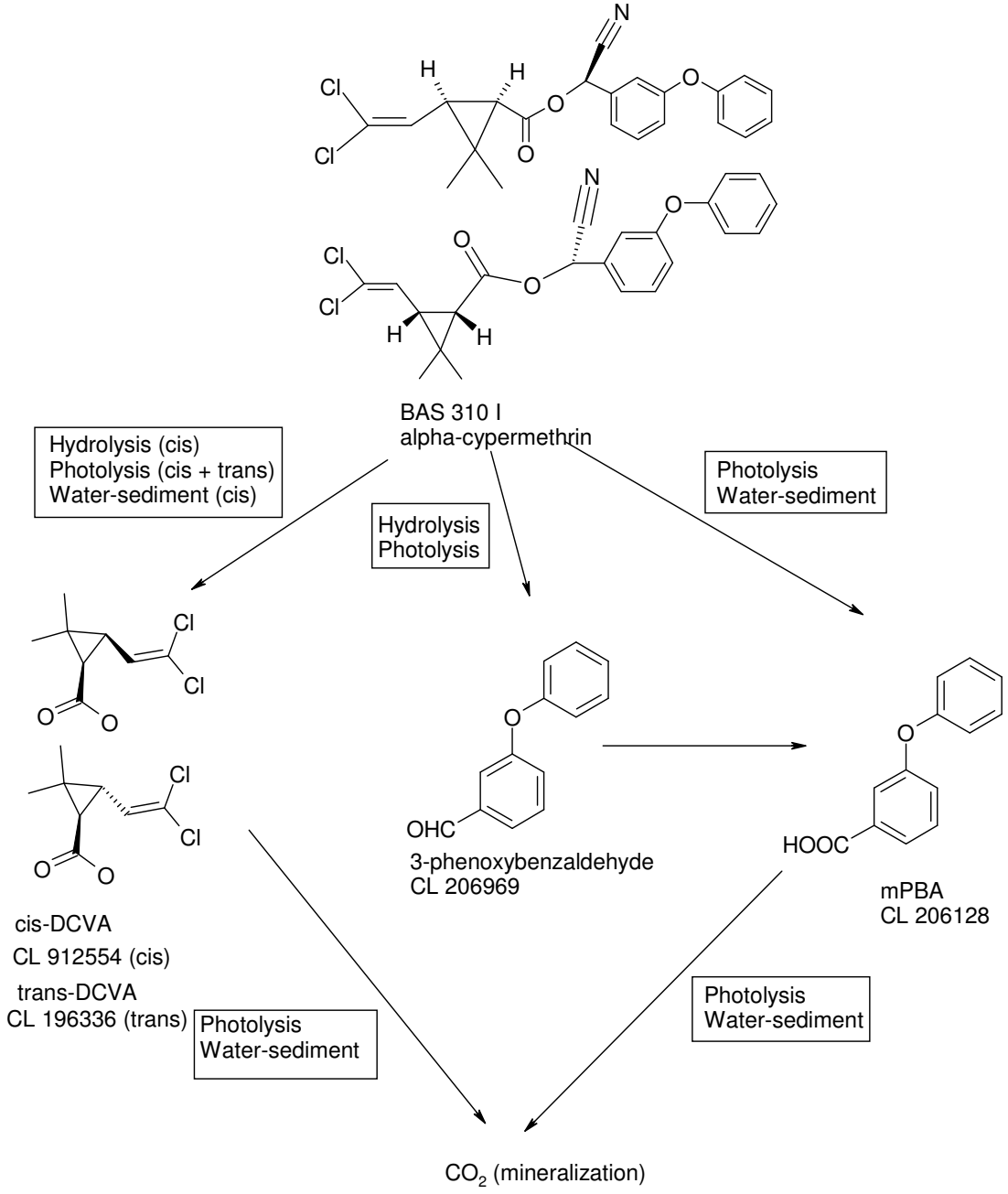
[a] Sum of water and sediment phase; mean of two replicates

[b] not relevant for risk assessment

CA 7.2.2.4 Irradiated water/sediment study

No irradiated water/sediment study was performed. Alpha-cypermethrin dissipates rapidly from the water phase due to its high sorption to sediment. Therefore light may have a limited effect on degradation, though the aqueous photolysis study show enhanced degradation of the parent compound under sunlight.

Figure 7.2.2.4-1: Route of degradation of alpha-cypermethrin in water



CA 7.2.3 Degradation in the saturated zone

Due to its low leaching potential, alpha-cypermethrin is not expected to reach deeper soil layers or saturated zones. Therefore, investigations on the degradation in the saturated zone are considered to be not necessary.

CA 7.3 Fate and behaviour in air

CA 7.3.1 Route and rate of degradation in air

Table 7.3.1-1: List of degradation studies in air performed with alpha-cypermethrin

DocID	Parent compound	Estimated parameters	Remark
Craig, 1999	Alpha-cypermethrin	Henry's law constant	
Grayson et al., 1982	Alpha-cypermethrin	Vapour pressure	
AL-324-002	Alpha-cypermethrin	Half-live values	G. Mangels 1995

The studies in Table 7.3.1-1 will no longer be used. Vapour pressure was determined by Kroehl (2014 a):

3.8 10^{-7} Pa at 20°C

and the Henry constant was calculated by Kroehl (2014 i) to be:

H = 5.3 * 10^{-2} Pa m^3 / mol

Recalculation with AOP v1.92 (Hydroxyl radicals) and AOP v1.91 (Ozone reaction)

The degradation rates for reactions of alpha-cypermethrin with OH-radicals and ozone in the atmosphere were calculated using the AOPWIN program based on ATKINSON's increment method. The SMILE notation used for alpha-cypermethrin in AOPWIN was:

CC1([C@H]([C@H]1C(=O)O[C@H](C#N)c2cccc(c2)Oc3ccccc3)C=C(Cl)Cl)C

The atmospheric degradation half-life of the substance via this reaction route was estimated to be $t_{1/2}$ = 0.499 d (5.99 h).

Based on the results of the atmospheric degradation half-life of alpha-cypermethrin ($t_{1/2}$ = 0.5 d), it can be concluded that the substance will be rapidly degraded by photochemical processes in the troposphere. Hence, due to the rapid degradation in air, it can be concluded that there is no risk of long-range transport of alpha-cypermethrin.

CA 7.3.2 Transport via air

Alpha-cypermethrin has a very low volatilization potential and is degraded very fast by photochemical processes. Consequently, there is no risk of long-range transport of alpha-cypermethrin.

CA 7.3.3 Local and global effects

No effects are expected since transport via air is very unlikely (for details see above).

CA 7.4 Definition of the residue

CA 7.4.1 Definition of the residue for risk assessment

According to the results presented in M-CA 7.1 – 7.3 the following compounds have to be considered for the environmental risk assessment:

Soil:
Alpha-cypermethrin and its aerobic soil metabolites DCVA, 3-PBA and M310I017 (Reg No. 6002320, hydroxy-alpha-cypermethrin)

The metabolites DCVA and 3-PBA were tested for ecotoxicological effects. In addition, it can be assumed that in long-term studies with alpha-cypermethrin, its metabolites will form and are thus included in the testing. Based on the obtained results it can be concluded that the risk of metabolites for soil organisms is low.

Groundwater:
Alpha-cypermethrin, its aerobic soil metabolites DCVA, 3-PBA and M310I017 (Reg No. 6002320, hydroxy-alpha-cypermethrin)

Based on the high adsorption and/or the fast degradation rates in soil, neither the parent molecule nor one of its metabolites poses any risk of leaching to groundwater. The predicted annual leachate concentrations of alpha-cypermethrin and all soil metabolites were below 0.001 µg L^{-1}.

Surface Water:
Alpha-cypermethrin, DCVA and 3-PBA, 3-PBAldehyde and M310I017 (Reg No. 6002320, hydroxy-alpha-cypermethrin)

The aquatic toxicity of DCVA, 3-PBA and 3-PBAldehyde was tested and was found to be orders of magnitude lower for the most sensitive species than parent. For M310I017 the concentrations in surface water at FOCUS step 2 are already < 0.001 µg/L and thus not relevant. Therefore, sufficient margins of safety were achieved with FOCUS surface water step 1-2 calculations and it is concluded that the risk of metabolites for aquatic organisms is low.

Sediment:
Alpha-cypermethrin, DCVA, 3-PBA, 3-PBAldehyd and M310I017 (Reg. No. 6002320)

Due to the low toxicity to aquatic arthropods, sediment toxicity testing of metabolites was not triggered. Thus, it is concluded that the risk of metabolites for sediment organisms is low.

Air:
Alpha-cypermethrin

No volatile metabolite was detected.

CA 7.4.2 Definition of the residue for monitoring

According to the results of the risk assessment the following compounds should be considered for environmental monitoring:

Soil:	Alpha-cypermethrin, DCVA and 3-PBA
Ground Water:	Alpha-cypermethrin DCVA and 3-PBA
Surface Water:	Alpha-cypermethrin DCVA and 3-PBA
Sediment:	Alpha-cypermethrin DCVA and 3-PBA
Air:	Alpha-cypermethrin (parent only)

CA 7.5 Monitoring data

According to the knowledge of the applicant, there are currently no published environmental monitoring data available for alpha-cypermethrin or its metabolites, which would provide knowledge on the environmental behaviour not covered by this dossier.

During literature search a few publications were found dealing with environmental monitoring of various pesticides in surface water, groundwater and/or sediment inside and outside of the EU (e.g. US, Argentina, Vietnam, Canada, Greece, Portugal) mentioning also cypermethrin or alpha-cypermethrin. The results showed that alpha-cypermethrin could be detected in local areas characterized by intensive insecticide use during the spraying season but also due to application of alpha-cypermethrin outside agriculture (biocidal use). Frequencies of detections as well as measured concentrations were usually very low.

Monitoring data of alpha-cypermethrin from within EU countries have been evaluated during the review process of the Water Framework Directive (WFD) Priority Substances list. Monitoring data are available from two countries, from France (2000 to 2007) and from Finland (only 2005). In sum 18695 measurements are available from river waters, however only two of them were > LOQ. A summary of the surface water monitoring data is given in the WFD alpha-cypermethrin factsheet (http://www.priority.substances.wfd.oieau.fr/pdf/alpha-Cypermethrin_%28aka_alphamethrin%29.pdf).
With the EU directive 2013/39/EU cypermethrin has been included in the list of priority substances in the field of water policy. Therefore, surface water monitoring within the monitoring program of the Water Framework Directive will include measurements of alpha-cypermethrin from Dec 2018 onwards in the EU.

BASF
The Chemical Company

Alpha-Cypermethrin

Document M-CA, Section 8

ECOTOXICOLOGICAL STUDIES ON THE ACTIVE SUBSTANCE

Compiled by:

[redacted]

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
March 20, 2015	Added Table 8.1 and a bird study summary CA 8.1.1.3	
October 09, 2015	Added the higher-tier acute Daphnia study summary 8.2.4/1. Updated Table 8.2-1	
July 10, 2017	Udated according to correspondence with the RMS; e.g. recalculation of endpoints, QSAR calculations for metabolite	BASF DocID 2017/1134417

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 8 ECOTOXICOLOGICAL STUDIES ON THE ACTIVE SUBSTANCE

CA 8.1 Effects on birds and other terrestrial vertebrates

Introduction

Table 8-1: Summary of EU agreed (EC review report, 2004) and additional toxicity studies relevant for AIR3 for the active substance alpha-cypermethrin for birds and mammals.

Test system	Test species	Reference [Author, BASF DocID]	EU agreed
Alpha-cypermethrin			
Birds			
Acute oral toxicity	*Colinus virginianus*	[redacted] 2000, AL-505-002	Yes
	Taeniopygia guttata	[redacted] 2009, 2009/1114317	No (new study)
Sub-chronic toxicity and reproduction	*Colinus japonica*	Study proprietary Gharda Chemical Ltd. Bombay[1)]	Yes
	Colinus virginianus	[redacted] 2001, AL-534-002	Yes (study submitted again to RMS, but already included in reference list of EU Review Report)
Mammals			
Acute oral toxicity	Rat	[redacted] 1993, AL-410-003	Yes
Reproductive toxicity	Rat	Study proprietary Gharda Chemical Ltd. Bombay[1)]	Yes
Developmental toxicity	Rabbit	[redacted] 1994 AL-432-004	Yes (previously not considered in ecotoxicology)
	Rat	[redacted] 1994 AL-432-002	

1) This study is not BASF proprietary, but owned by Gharda Chemical Ltd. Bombay, another notifier for the existing Annex I listing of alpha-cypermethrin. Consequently, for avian long-term risk assessments regarding alpha-cypermethrin, BASF can only refer to the BASF property reproduction study in bobwhite quails with a NOEC of 150 mg a.s./kg diet, corresponding to a NOAEL of 16 mg/kg b.w./day.

CA 8.1.1 Effect on birds

CA 8.1.1.1 Acute oral toxicity to birds

Report: CA 8.1.1.1/1
[redacted] 2009a
BAS 310 I - Acute toxicity in the Zebra finch (Taeniopygia guttata) after single oral administration (LD50)
2009/1114317
Guidelines: EPA 540/9-82-024, EPA 540/9-85-007, EPA 850.2100, EPA 712-C-96-139, EPA 71-1
GLP: yes
(certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

An avian acute oral toxicity test with the active substance BAS 310 I was conducted. The objective of the study was to evaluate the acute toxicity of the substance in Zebra finches (*Taeniopygia guttata*) and to determine the oral LD_{50} and the no observed effect level (NOEL).

The test substance was administered via a single oral dose of 500, 1000 or 2000 mg a.s./kg body weight to groups of 2-month old Zebra finches. Ten birds (5 males and 5 females) were used in each test substance group. The doses were administered in aqueous carboxy methyl cellulose (CMC). Birds that have been fasted for about 20 to 22 hours were administered the test substance. Birds of all groups received food and water *ad libitum* throughout the test. The test was terminated after 14 days.

All groups were observed for mortality, signs of clinical toxicity, impact on food consumption and body weight for 14 consecutive days post dosing.

No mortality occurred throughout the duration of the study in the control and at the dose rate of 500 mg a.s./kg body weight (b.w.). The mortality observed in dose rate of 1000 mg a.s./kg b.w. was 40% and at the highest dose rate of 2000 mg a.s./kg b.w. 70%. No substance-related impairment of feed uptake in comparison to the control was observed in any of the dose groups. No substance-related impairment of food uptake in comparison to the control was observed in any of the dose groups. No statistically significant (Dunnett test) substance-related reduction of the body weights in the male and female birds was observed on day 7 and at day 14 (sacrifice) in any of the dose groups compared to the control and thus the body weight development was not impaired compared to the control group. All birds were examined macroscopically (post-mortem) after study termination. No abnormalities were detected in surviving birds after sacrifice. One female bird in the dose group receiving 500 mg/kg body weight head a lesion at the head most likely caused by fighting.

In an acute oral toxicity test with the Zebra finch (*Taeniopygia guttata*), the LD_{50} of BAS 310 I was found to be 1360 mg a.s./kg b.w. The NOEL was < 500 mg a.s./kg b.w.

I. MATERIAL AND METHODS

Test item:	BAS 310 I (Reg. No. 4078193), batch no. COD 000595, purity: 99.2% (tolerance ± 1.0%).
Test species:	Zebra finches (*Taeniopygia guttata*), visually indistinguishable from wild birds; adults, age: approx. 2 month (before beginning of first egg-laying period); supplier: Kölle-Zoo, Ludwigshafen, Germany.
Test design:	Birds were administered single doses of 500, 1000 or 2000 mg a.s./kg body weight of the test substance BAS 310 I in 0.5% aqueous carboxy methyl cellulose by gavage into the crop in a total amount of 10 g preparation per kg body weight; 5 males and 5 females per dose group were used; observation period of 14 days; assessment of mortality and clinical signs was carried out four times on day of dosing and daily thereafter; assessment of body weight was carried out on the day of dosing and on day 7 and 14; mean food consumption (g/bird/day) was calculated from the daily food consumption per cage separately for male and female birds after dosing. Gross-pathological post-mortem examinations of all birds at study termination on day 14 after dosing.
Endpoints:	Mortality, clinical signs, feed consumption, body weight (b.w.), and gross-pathological examinations were conducted on all birds sacrificed at the termination of the definitive test. Calculation of LD_{50} and NOEL.
Test concentrations:	0 (Control), 500, 1000, and 2000 mg a.s./kg body weight (nominal).
Test conditions:	Birds fasted for about 20 h to 22 h before administration of the test substance; temperature: 20.7 °C - 23.6 °C; relative humidity: 45% - 70% (limits); photoperiod: 8 hours light : 16 hours dark, light intensity: 41 lux - 132 lux.
Analytics:	The test substance concentrations were analyzed using HPLC.
Statistics:	Descriptive statistics, Probit analyses according to Finney's method for LD50; Dunnett test for body weight data.

II. RESULTS AND DISCUSSION

Analytical measurements: The results of the analytical verification of the test substance concentration in the diet were within a range of 100% to 102% of the nominal concentrations during the test. The biological results are therefore based on the nominal values.

Biological results: No mortality occurred throughout the duration of the study in the control and at the dose rate of 500 mg a.s./kg body weight (b.w.). The mortality observed in dose rate of 1000 mg a.s./kg b.w. was 40% and at the highest dose rate of 2000 mg a.s./kg b.w. 70%. No toxic signs were observed in the control and in all test substance concentrations. No substance-related impairment of food uptake in comparison to the control was observed in any of the dose groups. No statistically significant (Dunnett test) substance-related reduction of the body weights in the male and female birds was observed on day 7 and at day 14 (sacrifice) in any of the dose groups compared to the control and thus the body weight development was not impaired compared to the control group. All birds were examined macroscopically (post-mortem) after study termination. No abnormalities were detected in surviving birds after sacrifice. One female bird in the dose group receiving 500 mg/kg body weight head a lesion at the head most likely caused by fighting. The relevant data and endpoints of the study are summarized in the table below

Acute toxicity of BAS 310 I to Zebra finch (*Taeniopygia guttata*)

	Dose rate [mg/kg b.w.]			
	0 (control)	**500**	**1000**	**2000**
Number of dead birds	0	0	4	7
Dead birds Percentage [%]	0	0	40	70 [1)]

Endpoints	**Dose [mg a.s./kg b.w.]**
Highest dose causing no substance-related mortality	500
LD_{50} (14 d)	1360
NOEL	< 500

b.w. = body weight

1) Erroneously one male bird was assigned to the cage with female birds.

2) Unexpectedly the test item caused mortality in the zebra finch in doses < 2000 mg/kg body weight. In consequence the requirement of the test guideline (EPA) to perform the test with 5 doses plus a control group in case of toxicity was not met. However, a clear dose-effect relationship could be estabilshed with the 3 doses tested in this study with partial mortality in 2 dose groups, one below and one above the LD50. Therefore the calculation of the LD50 does provide reliable and valid results. The deviation had thus no influence on the LD50 derived in this study. However, no "No observed effect level" (NOEL) could be established.

CONCLUSION

In an acute oral toxicity test with the Zebra finch (*Taeniopygia guttata*), the LD_{50} of BAS 310 I was found to be 1360 mg a.s./kg b.w. The NOEL was < 500 mg a.s./kg b.w.

CA 8.1.1.2 Short-term dietary toxicity to birds

No new study available.

CA 8.1.1.3 Sub-chronic and reproductive toxicity to birds

Report: CA 8.1.1.3/1
[redacted] 2001
Alphacypermethrin (BAS 310I) Assessment to Determine the Effects on Reproduction in Northern Bobwhite (*Colinus virginianus*). Vol. 1-2-3
Huntingdon Life Sciences. Report No.: CYD 630/003917; BASF Report No.: ETX-00-183.
AL-534-002
Guidelines: U.S. EPA Guideline Number 71-4; USEPA OPPTS 850.2300; and OECD 206.
GLP: Yes (certified by the Department of Health of the Government of the United Kingdom)

Executive Summary

Three treated groups of 20 replicates, one male and one female per replicate, were offered test diet containing 50, 150,450 ppm ai Alphacypennethrin over a period of 22 weeks (for 10 weeks prior to the start of egg production and during 12 weeks of egg production). A similar sized control group was given untreated diet only.

From the beginning of Week 11, eggs were incubated to hatching and chicks observed over fourteenday periods. Observations including mortality, clinical signs, bodyweight and food consumption were made during the study. Reproductive success was assessed by examining the following parameters: Eggs laid, eggs damaged, egg shell thickness, embryonic viability, and chick survival and growth.

Dietary administration of Alphacypermethrin to the Northern Bobwhite at dose concentrations up to 150 ppm a.s. had no effect on the health or reproductive performance of adult birds or on the health and growth of their chicks.

At 450 ppm a.s., group mean male adult bodyweights were significantly lower than the controls at termination of the study (p<0.05) and chick survival was reduced compared with the controls (p<0.05).

Food consumption of adult birds was shown to be statistically higher at 150 and 450 ppm a.s. compared with the control (p<0.05). However, this was considered to be of no adverse biological significance.

The NOEC in the avian reproduction study with Northern Bobwhite was determined to be 150 mg a.s./kg diet.

I. MATERIAL AND METHODS

Test item:	BAS 310 I: Technical grade Alphacypermethrin (also known as AC 900049); Lot number AC 12395-18; Purity 96.1% (Purity adjusted 100% while making the test diet).
Test species:	Norther Bobwhite (*Colinus virginianus*) Origin: Monkfield Nutrition, Royston, England.
Test design:	Adult birds were housed as pairs (one male and one female) in polythene coated steel wire cages. The adult birds were maintained on an 7-hour light : 17-hour dark photoperiod during acclimatisation and during the first five weeks. At the start of Week 6, the light portion of the photoperiod was increased to 16 hours light : 8 hours dark. The photoperiod remained at this interval until termination of the adult portion of the study. The total time on the treated diet was 22 weeks.
Endpoints:	Adults were observed daily for mortality, abnormal behaviour and signs of toxicity. Food consumption was monitored. Eggs were collected daily from the onset of egg production and set weekly for incubation. In addition, every other week throughout the egg laying period, eggs laid on the first day of the week were collected from every replicate (when available) for egg shell thickness measurements. During incubation, the eggs were candled to check for embryo development. All hatchlings were observed after hatching for 14 days to assess hatchling survival.
Test concentrations:	0, 50, 150, 450 mg a.s./kg diet. Dietary concentrations tested were 0 (Control), 50, 150, 450 ppm ai in the diet.
Test conditions:	Adult bobwhite quail study room: mean daily maximum temperature 22 C, mean daily minimum 20 C; mean daily relative humidity: 44%; photoperiod: 7 hours light (week 1 – 5), lengthened the photoperiod to 16 hours (week 6 – termination). Incubation of eggs: placed in a commercial storage facility: temperature: 16°C. Eggs were set every other week for incubation: temperature: 37.5°C, relative humidity: 55%; after 21 days transferred to the hatcher: temperature: 37.5°C. Juveniles: (F1 generation): Mean maximum room temperature: 25 – 29°C, mean minimum room temperature 23 - 25°C; relative humidity: 38 – 40%.
Analytics:	Alphacypermethrin in the test diets were analytically verified during the test.

II. RESULTS AND DISCUSSION

Analytical measurements:

Samples of the test diets presented to the adult birds during the test averaged 96% (50 mg a.s./kg diet), 99% (150 mg a.s./kg diet), 100% (450 mg a.s./kg diet) of nominal. No Alphacypermethrin was detected (limit of detection <1.125 ppm) in the control diet.

Biological results:

The effect of the various treatments on the reproductive parameters evaluated during the test are summarised in the Table 8.1.1.3-1.

Table 8.1.1.3-1 Effects of Alphacypermethrin (BAS 310 I) on the reproduction of Northern Bobwhite (*Colinus virginianus*).

Parameters	mg a.s./kg diet			
	Control	50	150	450
Adult				
Initial (week -0) mean bodyweight of male (g)	190.2	188.6	195.0	196.7
Terminal (week - 22) mean bodyweight of male (g)	209.1	208.1	206.7	202.4*
Initial (week -0) mean bodyweight female (g)	190.8	193.3	187.3	191.1
Terminal (week - 22) mean bodyweight of female (g)	225.3	230.6	214.0	227.4
Mean food consumption (g/bird/day)	19	21	21≠	21≠
Non treatment-related mortalities (all female)	1	2	0	3
Treatment-related mortalities	0	0	0	0
Treatment-related clinical signs	0	0	0	0
Treatment-`related macroscopic *post mortem* observation	0	0	0	0
Reproductive				
Number of eggs laid	1014	1079	1025	1163
Eggs laid per female	53.4	55.2	51.3	60.5
Cracked eggs	56	36	42	35
Eggs cracked of eggs laid (%)	5.5	3.3	4.1	3.0
Mean egg shell thickness (mm)	0.22	0.21	0.21	0.21
Eggs set	892	965	919	1029
Viable embryos	814	884	867	957
Viable embryos of eggs set (%)	91	92	94	93
Live 3-week embryos	798	828	842	935
Live 3-week embryos of viable embryos (%)	98	94	97	98
Normal hatchlings	762	751	781	897
Normal hatchlings of viable embryos (%)	94	85	90	94
Normal hatchlings of live 3-week embryos (%)	95	91	93	96
14-day old survivors	750	729	762	802
14-day old survivors of eggs laid (%)	74	68	74	69
14-day old survivors of normal hatchlings (%)	98	97	98	89*
14-day old survivors per female	39.5	37.3	38.1	41.7
Chick bodyweight at hatching (g)	6.9	7.1	7.2	6.9
Chick bodyweight at 14 days (g)	21	20	21	20

* parameter statistically lower than control (p<0.05)
≠ parameter statistically higher than control (p<0.05)

Dietary administration of Alphacypermethrin to the Northern Bobwhite at dose concentrations up to 150 ppm a.s. had no effect on the health or reproductive performance of adult birds or on the health and growth of their chicks.

At 450 ppm a.s., group mean male adult bodyweights were significantly lower than the controls at termination of the study (p<0.05) and chick survival was reduced compared with the controls (p<0.05).

Food consumption of adult birds was shown to be statistically higher at 150 and 450 ppm a.s. compared with the control (p<0.05). However, this was considered to be of no adverse biological significance.

Conclusion

The NOEC in the avian reproduction study with Northern Bobwhite was determined to be 150 mg a.s./kg diet.

CA 8.1.2 Effects on terrestrial vertebrates other than birds

CA 8.1.2.1 Acute oral toxicity to mammals

No new study available.

CA 8.1.2.2 Long-term and reproductive toxicity to mammals

No new study available.

CA 8.1.3 Effects of active substance bioconcentration in prey of birds and mammals

Bioaccumulation in terrestrial food chains is addressed in Chapter CP 10.1.

CA 8.1.4 Effects on terrestrial vertebrate wildlife (birds, mammals, reptiles and amphibians)

According to the revised data requirements under regulation 1107/2009 (Commission Regulations (EU) 283/2013 and 284/2013 for the active ingredient and the plant protection products, respectively), the risk to amphibians and reptiles shall be addressed. Nevertheless, unlike birds and mammals, toxicity tests for amphibian and reptile species are not requested. In the EU there is no guidance or validated regulatory protocols yet available neither on the type of regulatory testing necessary nor on how to conduct a risk assessment for amphibians and reptiles. In the case of alpha-cypermethrin, there are some studies in the literature on the toxicity of this active ingredient on aquatic life-stages of amphibians. For reptiles, no toxicity data of alpha-cypermethrin are available.

According to the new aquatic guidance document (EFSA, 2013) amphibians should be included in the aquatic and terrestrial risk assessment. In absence of GLP studies the assessment should be based on any existing relevant information (testing of amphibians is not recommended in the first instance, due to animal welfare reasons and the absence of standard guidelines for amphibian testing). With regard to the aquatic risk assessment several data analyses indicate that the risk assessment for aquatic organisms (and fish in particular) covers the risk assessment for aquatic phases of amphibians (Fryday and Thompson, 2012; Weltje et al., 2013). The review by Weltje et al. (2013) includes acute and chronic data for amphibians and alpha-cypermethrin (Yu et al., 2013 and Greulich and Pflugmacher, 2003); both in acute and chronic studies amphibians were less sensitive than fish. Based on these extensive data reviews it can be concluded that the acute and chronic risk to aquatic life-stages of amphibians is covered by the currently requested and conducted risk assessment for fish (for more details see chapter 10.2).

Regulatory ecotoxicological information on terrestrial amphibians is scarce in general. However, in the few cases where terrestrial stages of amphibians were exposed orally in the same study as birds and mammals the general pattern is that amphibians are less sensitive than the latter two taxa (see Table 13 in Fryday and Thompson, 2012). This suggests that the quantitative risk assessment done for birds and mammals would be conservative for the terrestrial phase of amphibians.

In the case of reptiles there is even less information available than for amphibians (see the review by Fryday and Thompson, 2009). The risk from dietary exposure can be assumed to be lower for reptiles than for birds and mammals due to the lower food intake rate (FIR) (Fryday and Thompson 2009). This is because reptiles are poikilotherm (i.e., do not maintain a constant body temperature) and as a result feeding activity will peak at warm days and will be zero during hibernation or at cold days. In contrast, birds and mammals will have to maintain a constant body temperature and, hence, will need to feed every day (Fryday & Thompson 2009).

References:

Commission Regulation (EU) No 283/2013 setting out data requirements for active substances, in accordance with Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market. Official Journal of the European Union: 1st March 2013.

Commission Regulation (EU) No 284/2013: setting out the data requirements for plant protection products, in accordance with Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market. Official Journal of the European Union: 1st March 2013.

Fryday S and Thompson H (2009a): Literature reviews on ecotoxicology of chemicals with a special focus on plant protection products. Lot 1. Exposure of reptiles to plant protection products. EFSA (CFT/EFSA/PPR/2008/01).

Fryday S and Thompson, H (2012): Toxicity of pesticides to aquatic and terrestrial life stages of amphibians and occurrence, habitat use and exposure of amphibian species in agricultural; Food and Environment research agency, UK

Weltje L., Simpson P., Gross M., Crane M., Wheeler J.R. (2013): Comparative acute and chronic sensitivity of fish and amphibians: a critical review of data. Environmental Toxicology and Chemistry, Vol. 32, No. 5, pp. 984-994

CA 8.1.5 Endocrine disrupting properties

Overall there is no convincing evidence for a potential of endocrine activity of alpha-cypermethrin *in vivo* up to dose levels not also causing significant systemic toxicity. A potential non-specific testicular toxicity has been investigated for alpha-cypermethrin in rats (BASF DocID 2014/1275120). This study showed no effects on sex organ weight parameters, no effect on sperm parameters and no indication of any effect on androgen receptor density. Therefore, it is appropriate to conclude that alpha-cypermethrin is neither endocrine active nor does it show testicular toxicity up to maximal tolerable dose levels. Further details on the rat testicular toxicity study and endocrine potential of alpha-cypermethrin can be found in section M-CA 5.8.3.
Similarly, in the available long-term bird and fish studies there were no signs of endocrine potential of alpha-cypermethrin.

CA 8.2 Effects on aquatic organisms

Since Annex I inclusion of alpha-cypermethrin (BAS 310 I), new toxicity studies on the active substance and its major metabolites have been performed and as a result there are new endpoints which are now used in the aquatic risk assessment. Summaries of these new studies are provided below. For completeness this includes some older studies, which have not been submitted during the previous Annex I inclusion process (e.g. because there is no respective data requirement in the EU). In addition, summaries are provided for peer-reviewed scientific literature that was considered to be of relevance for the aquatic risk assessment of alpha-cypermethrin.

Details on the EU agreed studies which have been already evaluated within the Annex I inclusion of alpha-cypermethrin are provided in the EU Review documents of alpha-cypermethrin (Monograph, Vol. 3, Annex B.8, September 1999; Addendum to the Monograph, Vol. 3, Annex B.9, July 2003; EU Review Report (SANCO/4335/2000-final), February 2004).

For better transparency and traceability of the active substance history, the results of all studies are summarized in Table 8.2-1 and Table 8.2-2.

Full references used within the following chapters are given at the end MCA 8.2. Document N3 contains structures and synonyms for all metabolites.

Table 8.2-1: List of studies and endpoints for aquatic organisms exposed to the active substance alpha-cypermethrin (BAS 310 I)

Organism	Endpoint	Value [µg/L] (except BCF & results of spiked sediment studies)	Reference (BASF Name / DocID)	EU agreed
Fish				
Oncorhynchus mykiss	96 h LC_{50}	**1.77**	AL-511-001 2016/1117187	yes
Pimephales promelas	34 d NOEC (ELS study)	0.03	AL-512-002	yes
Aquatic invertebrates				
Daphnia magna	48 h EC_{50}	**0.22**	AL-511-001 2016/1117187	yes
Daphnia magna	21 d NOEC	0.03	AL-523-001	yes
Sediment dwelling aquatic invertebrates				
Chironomus riparius *	48 h EC_{50}	0.0126	2009/1102214	no (new data requirement)
Chironomus riparius	28 d NOEC (spiked water) 28-d EC_{10} 28-d EC_{20}	0.024 0.028 0.057	AL-523-002	yes

Organism	Endpoint	Value [µg/L] (except BCF & results of spiked sediment studies)	Reference (BASF Name / DocID)	EU agreed
Chironomus riparius *	28 d EC_{10} (spiked sediment)	**51.4** [3)]	2011/1124187 + Amendment: 2012/1156939	no (data generated for biocide Annex I listing)
Algae [$]				
Pseudokirchneriella subcapitata	72 h E_rC_{50} / E_yC_{50}	> 1000	2002/1004851 (= AL-520-002)	yes
Pseudokirchneriella subcapitata	72 h E_rC_{50} / E_yC_{50}	**> 100**	AL-511-001	yes
Aquatic macrophytes [$]				
Lemna gibba *	7 d E_rC_{50} / E_yC_{50}	> 1.39 [5)]	2009/1108874	no (new data for US registration)
Bioconcentration				
Cyprinus carpio *	BCF (10 weeks exposure, 2 weeks depuration)	**910 L/kg** [6)]	AL-519-004	no (not included in the monograph; included for completeness)
Oncorhynchus mykiss	BCF (19 d exposure, 46 d depuration)	1204 L/kg [6)]	1981/7001062 (= AL-519-003)	yes (replaced by study conducted with alpha-cypermethrin; AL-519-004)
Oncorhynchus mykiss	BCF (22 d exposure, 51 d depuration)	1000 L/kg [6)]	1978/7000725 (=AL-519-002)	yes (replaced by study conducted with alpha-cypermethrin; AL-519-004)
Data on additional species / Higher-tier studies / Peer-reviewed literature studies				
Aquatic vertebrates				
Pimephales promelas	96 h LC_{50}	0.93 [++]	AL-512-002	yes
Rutilus rutilus caspicus **	96 h LC_{50}	0.627	2012/1367682	no (peer-reviewed scientific study; used for risk refinement)
Hypophthalmichthys molitrix **	96 h LC_{50}	0.917		
Huso huso **	96 h LC_{50}	0.952	2012/1367683	no (peer-reviewed scientific study; used for risk refinement)
Poecilia reticulata **	96 h LC_{50}	9.43	2004/1040654	no (peer-reviewed scientific study; used for risk refinement)

Organism	Endpoint	Value [µg/L] (except BCF & results of spiked sediment studies)	Reference (BASF Name / DocID)	EU agreed
Oreochromis niloticus **	96 h LC_{50}	5.99	2009/1131342	no (peer-reviewed scientific study; used for risk refinement)
Oreochromis niloticus **	96 h LC_{50}	3.42	2005/1043560	no (peer-reviewed scientific study; used for risk refinement)
Oryzias latipes *	48 h LC_{50}	12.0	AL-519-004	no (not included in the monograph; included for completeness)
Xenopus laevis **	96 h LC_{50}	30.6 (embryos) 6.9 (larvae)	2013/1417940	no (peer-reviewed scientific study)
Aquatic vertebrate SSD	HC_5	0.344	See MCP 10.2	no
Pimephales promelas *	34 d NOEC [1)] (ELS, pulse-dose study)	0.3	2009/1031203	no (data generated for risk refinement in support of EU product registrations)
Aquatic invertebrates				
Chaoborus crystallinus *	48 h EC_{50}	0.045	2009/1085205	no (data generated for risk refinement in support of EU product registrations)
Daphnia magna [+, *]	48 h EC_{50} (bioavailability study)	0.18 µg/L in M4 medium 0.515 µg/L in presence of humic acid 0.695 µg/L in presence of algae	2013/1404157	no (data generated for risk refinement in support of EU product registrations)
Daphnia magna *	21 d NOEC [2)] (pulse-dose study)	0.149	2007/1016502	no (data generated for risk refinement in support of EU product registrations)
Lumbriculus variegatus *	28 d NOEC (spiked sediment)	71.3 [3)]	2012/1205915	no (data generated for biocide Annex I listing)

Organism	Endpoint	Value [µg/L] (except BCF & results of spiked sediment studies)	Reference (BASF Name / DocID)	EU agreed
Caenorhabditis elegans *	96 h NOEC (spiked sediment)	≥ 28600 [3)]	2013/1250848	no (data generated for biocide Annex I listing)
Chironomidae (rice field trial)**	24 - 29 d NOEAEC	≈ 0.16 - 0.23	2013/1250848	no (peer-reviewed scientific study; used for risk refinement)
Skeletonema costatum *,[4)]	72 h E_rC_{50} / E_yC_{50} #	> 33.4	2009/1109079	no (new data for US registration)
Anabaena flos-aquae *	72 h E_rC_{50} / E_yC_{50} #	> 27	2009/1109080	no (new data for US registration)
Navicula pelliculosa *	72 h E_rC_{50} / E_yC_{50} #	> 70.3	2009/1109081	no (new data for US registration)
Higher tier testing on aquatic invertebrates and algae	EAC §	**0.015**	2003/1024853	yes
Outdoor mesocosm +, *	$NOEC_{mesocosm}$ $NOEAEC_{mesocosm}$ (class 3A)	0.00403 2 x 0.0170 or 1 x 0.0385	2014/1102015 + 2014/1246534 (analytical report)	no (new data generated for AIR3 renewal)
Review of 9 mesocosm studies *	$NOEC_{mesocosm}$ $NOEAEC_{mesocosm}$	0.00403 2 x 0.0170 or 1 x 0.0385	2014/1102014	no (evaluation of existing and new mesocosm data)
Chaoborus population modeling using FOCUS scenarios *	Modelling confirms population recovery within 8 wks after exposure to max 0.042 µg/L (NOEAEC)		2014/1162681	no (new data generated for AIR3 renewal)
Chaoborus population development in mesocosms *	Analysis of control data of 19 mesocosm studies: provides information on *Chaoborus* population dynamics, their life cycle, the natural variability of *Chaoborus* populations and their influence on the zooplankton community structure		2014/1102016	no (evaluation of mesocosm data)

Bold figures: Where several endpoints are available for the same group or where several endpoints are available for one study based on different effect parameters (*e.g.* for algae and macrophytes), only the relevant endpoint(s) is used in the risk assessment presented in chapter 10.2 of the MCP dossier part for Annex I renewal.

Abbreviations: ELS = early life stage; BCF= bioconcentration factor; EAC = ecologically acceptable concentration

* Study has not been submitted during the Annex I inclusion process of alpha-cypermethrin. A study summary is provided below.

** Data derived from relevant peer-reviewed scientific study (for details see chapter 8.2.8 below).

+ Study was performed with the formulation BAS 310 55 I (containing 50.0 g a.s./L, nominally).

++ Study is EU agreed predominantly for the chronic part. However, the acute study is valid as well.

In accordance to the EFSA Aquatic Guidance Document (EFSA, 2013) and OECD guideline 201 (2001) the 72 h endpoints obtained in the 96 h alga studies are considered as relevant endpoint and are presented here.

§ EAC based on effects on macroinvertebrates from a series of studies including mesocosm studies, additional single species testing and population modeling (BASF DocIDs: AL-560-031, AL-560-023, AL-560-056, 2003/1012038, AL-560-054, 2002/1013889 and 2003/1012036).

$ In accordance to the EFSA Aquatic Guidance Document (EFSA 2013) and OECD guidelines 201 (OECD, 2011) and 221 (OECD, 2006), only the EC_{50} values for the more relevant endpoint 'growth rate' (E_rC_{50}) are considered for the risk assessment for aquatic primary producers.

1) Pulse exposure study simulating realistic drift scenario with two peak concentrations at day 0 and day 7 or at day 7 and day 14. No significant effects of the test item were observed up to and including two peak concentrations of 0.3 µg/L alpha-cypermethrin.
2) Study simulating realistic exposure scenario with two peak concentrations of the test item (7 days interval) and subsequent dissipation. No ecologically relevant effects of the test item were observed up to and including two peak concentrations of 0.149 µg/L alpha-cypermethrin.
3) µg/kg dry sediment (spiked sediment study)
4) marine / saltwater species
5) based on frond number and on dry weight
6) At the time of the Annex I inclusion two fish bioconcentration studies were available with cypermethrin and rainbow trout. Since then a bioconcentration study with alpha-cypermethrin and *Cyprinus carpio* has become available and is considered to provide a more relevant BCF value, as the study was conducted with the actual active substance under evaluation.

Table 8.2-2: List of studies and endpoints for aquatic organisms exposed to the major metabolites of alpha-cypermethrin (BAS 310 I)

Organism	Endpoint	Value [µg/L]	Reference (BASF Name / DocID)	EU agreed
Metabolite: DCVA				
Fish				
Lepomis macrochirus	96 h LC_{50}	**> 102800**	2002/1004682 (= AL-570-011)	yes
Oncorhynchus mykiss	96 h LC_{50}	180000	CY-570-003 [1), 2)]	yes
Aquatic invertebrates				
Daphnia magna	48 h EC_{50}	**61900** [#]	2001/1017462 (= AL-520-001)	yes
	48 h EC_{50}	13000	CY-570-003 [1), 2)]	yes
Algae [§]				
Pseudokirchneriella subcapitata	72 h E_rC_{50} 72 h E_yC_{50}	**70000** 31600	2002/1004139	yes
	96 h E_rC_{50}	110000	CY-570-003 [1), 2)]	yes
Metabolite: 3-PBA				
Fish				
Lepomis macrochirus	96 h LC_{50}	**> 104300** [+]	2002/1004683 (= AL-570-013)	yes
Oncorhynchus mykiss	96 h LC_{50}	> 10000	CY-570-002 [1), 2), 3)]	yes
Aquatic invertebrates				
Daphnia magna	48 h EC_{50}	39000	2001/1014673 (= AL-570-008)	yes
Algae [§]				
Pseudokirchneriella subcapitata	72 h E_rC_{50} 72 h E_yC_{50}	**85000** 38100	2002/1004140	yes
Macrophytes				
Myriophyllum elatinoides **	14 d NOEC	≥ 3280 [$] (based on length & fresh weight)	2012/1367684	yes
Metabolite: 3-PBAld				
Fish				
Oncorhynchus mykiss *	96 h LC_{50}	> 2000 < 4000 (geometric mean: 2830)	1977/7000346 (= CY-570-001) [1), 3), 4)]	no (non-GLP study; used for risk assessment of metabolite)
Fish	96-h LC_{50}	1347	See MCP 10.2	No (QSAR estimate)
Aquatic invertebrates				
Daphnia magna	48 h EC_{50}	800	2002/1004857 (= AL-570-012)	yes
Daphnia magna	48 h EC_{50}	901	See MCP 10.2	No (QSAR estimate)
Algae				

Green algae	96-h EC_{50}	2050	See MCP 10.2	No (QSAR estimate)

Bold figures: Where several endpoints are available for the same group or where several endpoints are available for one study based on different effect parameters (*e.g.* for algae/macrophytes), the relevant endpoint is used in the risk assessment presented in chapter 10.2 of the MCP dossier part for Annex I renewal.

* Study has not been submitted during the Annex I inclusion process of alpha-cypermethrin. A study summary is provided below.

** Data derived from relevant peer-reviewed scientific study (for details see chapter 8.2.8 of the MCA dossier for Annex I renewal).

The higher endpoint from the study on *D. magna* (BASF DocID 2001/1017462) is considered as relevant endpoint; the lower endpoint obtained in a second study (BASF DocID CY-570-003) was not used as no analytical verification of test item concentrations was conducted in this study.

+ The higher endpoint from the study on *L. macrochirus* (BASF DocID 2002/1004683) is considered as relevant endpoint; the lower endpoint obtained in the study on *O. mykiss* (BASF DocID CY-570-002) was not used as no analytical verification of test item concentrations was conducted in this study.

§ In accordance with the new EFSA Aquatic Guidance Document (EFSA, 2013) and OECD guideline 201 (2001) only the EC_{50} values determined for the endpoint 'growth rate' (E_rC_{50}) are considered for the risk assessment for algae if both “growth rate” and “biomass” endpoints are available.

$ Concentration in overlying water or in sediment pore water

1) No analytical verification of test item concentrations was conducted.

2) Study was evaluated for Annex I inclusion of cypermethrin (see Monograph, Addenda and EU Review Report).

3) non-GLP study

4) This non-GLP study is of limited design (no analytical verification, only 4 fish per treatment). However, it is considered sufficient to judge on the acute toxicity to fish and the non-relevance of the metabolite.

CA 8.2.1 Acute toxicity to fish

The following (non-GLP) study with rainbow trout (*Salmo gairdneri*) performed with the metabolite 3-phenoxybenzaldehyde (3-PBAld) is of limited design (no analytical verification, only 4 fish per treatment). However, it is considered sufficient to judge on the acute toxicity to fish and the non-relevance of the metabolite. Thus, the study is provided in support of the aquatic risk assessment and has not been evaluated previously on EU level.

Report:	CA 8.2.1/1 [redacted] 1977a The acute toxicity to rainbow trout (Salmo gairdneri) of some compounds involved with the manufacture of pyrethroid insecticide WL 43467 CY-570-001
Guidelines:	none
GLP:	no

Executive Summary

In a static acute laboratory study, juvenile rainbow trout were exposed to nominal concentrations of 2000, 4000, 8000 and 10000 µg 3-phenoxybenzaldehyde/L (3-PBAld) in groups of 4 animals in glass aquaria containing 10 L of water. One aquarium with water served as control. Fish were observed for survival after 24, 48, 72 and 96 hours after start of exposure.

The biological results are based on nominal concentrations. After 96 hours, no mortality was observed in the control whereas 3-PBAld caused 25% mortality at a concentration of 2000 µg/L and 100% mortality was observed in fish exposed to the three highest test item concentrations of 4000, 8000 and 10000 µg/L.

In a static acute toxicity study with rainbow trout, the LC_{50} (96 h) for 3-phenoxybenzaldehyde (3-PBAld) was determined to be > 2000 and < 4000 µg/L (geometric mean: 2830 µg/L), based on nominal concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: 3-phenoxybenzaldehyde (3-PBAld; Reg. No. 4080665).

B. STUDY DESIGN

Test species: Rainbow trout fingerlings, *Salmo gairdneri* (syn. *Oncorhynchus mykiss*); body weight between 1.00 - 2.00 g; supplied by Parkwood Trout Hatcheries, Harrietsham, Kent, U.K.

Test design: Static system (96 hours); 4 fish per aquarium (10 L) and per concentration; loading: less than 1 g fish/L water; assessment of mortality after 24, 48, 72 and 96 h.

Endpoints: LC_{50} (only range could be determined because only a small amount of test compound was available).

Test concentrations: Control, 2000, 4000, 8000 and 10000 µg 3-PBAld/L (nominal).

Test conditions: 10 L glass aquaria, non-chlorinated tap water; temperature 15 - 16°C; pH 8.0 - 8.4; oxygen content > 9.0 mg/L; total hardness: approx. 250 - 270 ppm $CaCO_3$; slight aeration.

Analytics: No analytical verification of test item concentrations was conducted.

Statistics: Geometric mean calculation for determination of LC_{50}.

II. RESULTS AND DISCUSSION

Analytical measurements: No analytical verification of test item concentrations was conducted.

Biological results: After 96 hours, no mortality was observed in the control whereas 3-PBAld caused 25% mortality at a concentration of 2000 µg/L and 100% mortality was observed in fish exposed to the three highest test item concentrations of 4000, 8000 and 10000 µg/L. The results are summarized in Table 8.2.1-1.

Table 8.2.1-1: Acute toxicity (96 h) of 3-phenoxybenzaldehyde on rainbow trout (*Oncorhynchus mykiss*)

Concentration [µg/L] (nominal)	**Control**	**2000**	**4000**	**8000**	**10000**
Mortality [%]	0	25	100	100	100
Endpoints [µg/L] (nominal)					
LC_{50} (96 h)	> 2000 < 4000 (geometric mean: 2830)				

III. CONCLUSION

In a static acute toxicity study with rainbow trout, the LC_{50} (96 h) for 3-phenoxybenzaldehyde (3-PBAld) was determined to be > 2000 and < 4000 µg/L (geometric mean: 2830 µg/L), based on nominal concentrations.

From the performed literature search the following five peer-reviewed scientific studies investigating the acute toxicity of alpha-cypermethrin to several fish species were considered relevant for the aquatic risk assessment of alpha-cypermethrin. Due to missing analytical measurements in these studies, they were formally classified as being "not reliable" (Reliability Index (RI) 3). For details please see the literature search and evaluation files also provided within the submission for Annex I Renewal. Nevertheless, the studies were well performed and in general agreement with current guidelines for acute fish toxicity testing (e.g. OECD guideline 203). Furthermore, the endpoints derived from these studies are in a range expected from the available acute GLP data for fish and alpha-cypermethrin. Finally, it was considered that for vertebrates it would be preferable to use available data rather than to conduct additional vertebrate testing (animal welfare considerations). Hence, it is considered appropriate to use the results of these studies for a refined acute risk assessment for aquatic vertebrates. The data had not been used or evaluated during the previous Annex I inclusion process and thus, relevant information and results of these studies are described in the following summary.

Report:	CA 8.2.1/2 Shaluei F. et al., 2012a Evaluation of the acute toxicity of Cypermethrin and its effect on behavioral responses of Caspian roach (Rutilus rutilus caspicus) and Silver carp (Hypophthalmicthys molitrix) 2012/1367682
Guidelines:	none
GLP:	no

Report:	CA 8.2.1/3 Jahanbakhshi A. et al., 2012a Acute toxicity of Cypermethrin on the great sturgeon (Huso huso) juveniles 2012/1367683
Guidelines:	none
GLP:	no

Report:	CA 8.2.1/4 Yilmaz M. et al., 2004a Acute toxicity of Alpha-Cypermethrin to guppy (Poecilia reticulata, Pallas, 1859) 2004/1040654
Guidelines:	none
GLP:	no

Report:	CA 8.2.1/5 Sarikaya R., 2009a Investigation of acute toxicity of Alpha-Cypermethrin on adult nile tilapia (Oreochromis niloticus L.) 2009/1131342
Guidelines:	none
GLP:	no

Report:	CA 8.2.1/6 Yilmaz M., 2005a Acute toxicity of Alpha-Cypermethrin on tilapia (Oreochromis niloticus L.) larvae 2005/1043560
Guidelines:	none
GLP:	no

Executive Summary

The acute toxicity of alpha-cypermethrin to fish was investigated in four 96-hour static laboratory studies on Caspian roach (*Rutilus rutilus caspicus*) juveniles, Silver carp (*Hypophthalmichthys molitrix*) juveniles, adult male guppy (*Poecilia reticulata*) and adults and larvae of Nile tilapia (*Oreochromis niloticus*) and in one 96-hour semi-static laboratory study on Great sturgeon (*Huso huso*) juveniles. In these studies fish were exposed to a solvent control and to 5 – 9 nominal alpha-cypermethrin concentrations ranging from 0.01 to 20 µg a.s./L in groups of 7 to 15 animals in aquaria. Fish were observed for survival and symptoms of toxicity directly after start of exposure and 24, 48, 72 and 96 hours after start of exposure.

The biological results of all studies are based on nominal concentrations of the test item. After 96 hours of exposure, in all tests no mortality or toxic effects were observed in the control group.
In all studies, fish mortality increased significantly when the concentration and the time of exposure were increased. The LC_{50} (96 h) values of alpha-cypermethrin derived from these studies were between 0.627 and 9.43 µg a.s./L.
In all performed studies behavioral changes were recorded. Depending on the tested species, behavioral changes manifested at concentrations ≥ 0.5 µg a.s./L for Caspian roach, Silver carp and Great sturgeon or ≥ 8 µg a.s./L for guppy, ≥ 5 µg a.s./L for adult Nile tilapia and ≥ 1.5 µg a.s./L for Nile tilapia larvae.

In one semi-static and four static acute toxicity studies with five different fish species the LC_{50} (96 h) values of alpha-cypermethrin were between 0.627 and 9.43 µg a.s./L, based on nominal concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I; Reg. no.: 4 078 193), technical grade purities: 95% - 98%; for sources please see the respective publications.

B. STUDY DESIGN

Test species:	Caspian roach (*Rutilus rutilus caspicus*): juveniles, mean body weight: 3.5 ± 0.32 g, mean body length: 7.3 ± 0.65 cm; Silver carp (*Hypophthalmichthys molitrix*), juveniles; mean body weight: 40.68 ± 5.87 g, mean body length: 15.34 ± 3.12 cm; both purchased from by Bony Fish Propagation and Rearing Center of Sijeval (Bandar, Torkaman, Gorgan), Iran. Great sturgeon (*Huso huso*): juveniles; mean body weight: 183.20 ± 4.61 g, mean body length: 36.31 ± 1.27 cm; supplied by Shahid Marjani proliferation and culture centre for sturgeon fish, Gorgan, Iran. Guppy (*Poecilia reticulate*): adult males (no information is given on body weight / length / age of fish), supplied by a local breeder in Ankara, Turkey. Nile tilapia (*Oreochromis niloticus*): adults; mean body weight: 25.03 ± 5.35 g, mean body length: 11.05 ± 1.05 cm; supplied by the Fisheries Unit of Fisheries and Aquaculture Department of Ankara University, Ankara, Turkey. Nile tilapia (*Oreochromis niloticus*): larvae, 3 to 4 days old; mean body weight: 0.054 ± 0.001 g, mean body length: 0.79 ± 0.03 cm; supplied by A. Çağlan Karasu Benli of the Faculty of Agriculture, Ankara University, Ankara, Turkey.
Test design:	Static system (96 h) or semi-static system (96 h; with replacement of the test solution every 24 h); 5 - 9 test item concentrations plus a solvent control (acetone), 1 to 3 replicates per treatment with 7 - 15 fish per aquarium; assessment of mortality and sublethal effects directly after start of exposure and 24, 48, 72 and 96 hours after start of exposure.
Endpoints:	LC_{50}, mortality and sub-lethal effects.
Test concentrations:	Caspian roach and Silver carp: solvent control (acetone), 0.01, 0.5, 2, 4, 8 and 16 µg a.s./L. Great sturgeon: solvent control (acetone), 0.5, 1, 2, 4, 6 and 8 µg a.s./L. Guppy: solvent control (acetone), 5, 8, 10, 15 and 20 µg a.s./L. Nile Tilapia, adults: solvent control (acetone), 5, 6, 7, 8 and 10 µg a.s./L. Nile Tilapia, larvae: solvent control (acetone), 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0, 4.5 and 5.0 µg a.s./L.

Test conditions: Caspian roach and Silver carp: 120 L rectangular aquaria, test volume: 100 L; dilution water: dechlorinated tap water; temperature: 19.95 ± 1.5 °C; pH 7.88 ± 0.76; oxygen content: 8.01 ± 0.35 mg/L; hardness: 300 ± 12.25 mg $CaCO_3$/L; constant aeration; no feeding.

Great sturgeon: 500 L fiberglass tank; dilution water: tap water; temperature: 23 ± 1 °C; pH 7.89 ± 0.9; oxygen content: 7.8 ± 0.5 mg/L; hardness: 295 ± 10 mg $CaCO_3$/L; photoperiod: 14 h light : 10 h dark; constant aeration; no feeding.

Guppy: 20 L aquaria; dilution water: well water; temperature: 20 - 25 °C; pH 7.2; oxygen content: 7.5 mg/L; constant aeration; no feeding.

Nile Tilapia, adults: 20 L plastic aquaria, test volume: 20 L; dilution water: tap water; temperature: 24 ± 1 °C; pH 6.8; oxygen content: 6.9 ± 0.2 mg/L; hardness: 31.6 mg $CaCO_3$/L; alkalinity: 32 mg HCO_3/L; conductivity: 0.182 -0.197 mS/cm; constant aeration; no feeding.

Nile Tilapia, larvae: 20 L plastic aquaria, test volume: 20 L; temperature: 23 ± 1 °C; pH 7.2 ± 0.1; oxygen content: 7.6 ± 0.5 mg/L; conductivity: 0.225 ± 0.010 mS/cm.; constant aeration; no feeding.

Analytics: No analytical measurements were carried out.

Statistics: Descriptive statistics; probit analysis for calculation of LC_{50} values.

II. RESULTS AND DISCUSSION

Analytical measurements: No analytical measurements were carried out. The biological results of all studies are based on nominal concentrations of the test item.

Biological results: After 96 hours of exposure, in all tests no mortality or toxic effects were observed in the solvent control groups. In all studies, fish mortality increased significantly when the concentration and the time of exposure were increased.

In the static study with Caspian roach and Silver carp (DocID 2012/1367682) juveniles no mortality was recorded at the lowest applied test concentration of 0.01 µg a.s./L, whereas higher alpha-cypermethrin concentrations caused increased mortality in both test species. After 96 hours all fish were dead at the two highest tested concentrations of 8 and 16 µg a.s./L in the test with both fish species. The 96 h LC_{50} values were determined to be 0.627 g a.s./L for Caspian Roach and 0.917 µg a.s./L for Silver Carp. At concentrations ≥ 0.5 µg a.s./L, both fish species showed abnormal behavioral responses (*i.e.* rapid gill movement, nervous manifestations, erratic swimming, loss of equilibrium and inability to remain upright). The abnormal swimming behavior increased with increasing concentration of alpha-cypermethrin and exposure time.

Based on the results of the semi-static study with Great sturgeon juveniles (DocID 2012/1367683) a 96 h LC_{50} value of 0.952 µg a.s./L was derived for alpha-cypermethrin. Abnormal behavioral responses started 1 hour after dosing (*i.e.* rapid gill movement, nervous manifestations, erratic swimming, loss of equilibrium and inability to remain upright) and were observed in fish at all tested concentrations (*i.e.* at ≥ 0.5 µg a.s./L).

The 96 h LC_{50} value derived from the static study with adult male guppy (DocID 2004/1040654) was 9.43 µg a.s./L (DocID 2004/1040654). The changes in behavioral response started 2 h after dosing. The fish had less activity at 8 µg a.s./L compared with the control group. The behavioral changes manifested themselves starting at alpha-cypermethrin concentration of 8 µg a.s./L. Other changes were loss of balance, respiratory difficulties, become motionless, attempt to breathe from the surface, change of color in abdominal area and enlargement of the eyes.

In the acute study on adult Nile tilapia (DocID 2009/1131342), no mortality occurred in the lowest tested concentration of 5 µg a.s./L whereas 75% mortality was observed at the concentrations of 6.0 and 7.0 µg a.s./L after 96 hours of exposure. The two highest tested concentrations of 8 and 10 µg a.s./L caused 100% mortality after 96 hours of exposure. The 96-h LC_{50} value derived from the study with for Nile tilapia adults was estimated as 5.99 µg a.s./L. In addition, behavioral changes at each alpha-cypermethrin concentration were observed for the individual fish. The changes in behavioral response started 1-2 hours after dosing, depending on the concentration of toxicant. The fish started to display intense activity one hour after exposure. They left themselves to water currents and made sudden movements at a concentration of 6 µg/L. The fish gave response when tapped on the aquaria walls at a toxicant concentration of 7 µg/L. They gave no such response at higher concentrations and made movements such as somersaulting around their own axis.

Exposure of Nile tilapia larvae (DocID 2005/1043560) to alpha-cypermethrin resulted in a LC_{50} of 3.42 µg a.s./L. Changes in behavioral response started 1 - 2 hours after dosing, depending on the concentration of toxicant. The fish started to display intense activity one hour after exposure. They left themselves to water currents and made sudden movements at a concentration of 5 µg/L. The fish gave response when tapped on the aquaria walls at a toxicant concentration of 1 µg/L. They gave no such response at higher concentrations and made movements such as somersaulting around their own axis.

The endpoints obtained in the fish acute toxicity studies are summarized in Table 8.2.1-2.

Table 8.2.1-2: List of endpoints for alpha-cypermethrin derived from acute toxicity studies conducted with for five fish species

Test species, life stage	**96 h LC_{50} [µg a.s./L]**	**Reference** (BASF DocID)
Caspian roach (*Rutilus rutilus caspicus)*, juveniles	0.627 (95% confidence limits: 0.284 - 0.995)	2012/1367682
Silver carp (*Hypophthalmichthys molitrix*), juveniles	0.917 (95% confidence limits: 0.507 - 1.357)	
Great sturgeon (*Huso huso*), juveniles	0.952 (95% confidence limits: 0.693 - 1.242)	2012/1367683
Guppy (*Poecilia reticulata*), adults (male)	9.43 (95% confidence limits: 8.26 - 11.20)	2004/1040654
Nile tilapia (*Oreochromis niloticus*), adults	5.99 (95% confidence limits: 5.40 - 6.51)	2009/1131342
Nile tilapia (*Oreochromis niloticus*), larvae	3.42 (95% confidence limits: 3.11 - 3.73)	2005/1043560

III. CONCLUSION

In one semi-static and four static acute toxicity studies with five different fish species the LC_{50} (96 h) values of alpha-cypermethrin were between 0.627 and 9.43 µg a.s./L, based on nominal concentrations.

CA 8.2.2 Long-term and chronic toxicity to fish

CA 8.2.2.1 Fish early life stage toxicity test

In addition to the standard fish early life stage (ELS) study on *Pimephales promelas* which has already been evaluated during the Annex I inclusion process a new fish ELS study with pulse-dosed exposure on fathead minnow was conducted to generate further data for a risk refinement and to address potential latency of effects. The study has been used in previous EU end-use product registrations, but has not been evaluated on EU level.

Report:	CA 8.2.2.1/1 [redacted] 2009b BAS 310 I - Early life-stage test on the fathead minnow (Pimephales promelas) with pulse dose exposure 2009/1031203
Guidelines:	OECD 210, EPA 72-4 (a), EPA 850.1400, EPA 540/9-86-138
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

The chronic toxicity of alpha-cypermethrin (BAS 310 I) to fathead minnow (*Pimephales promelas*) was evaluated in a 34-day early life-stage test with pulse dose exposure under flow-through conditions. Two exposure peaks spaced by 7 days were applied to each test group. The peak concentrations declined with a half-life of one day. Two different peak concentrations of 0.15 and 0.30 µg a.s./L) were tested and the peaks were applied according to two different time schedules, to cover the most sensitive life stages. Additionally to the 4 test groups with peak exposure scheme, a dilution water control was tested. Hatchability, post-hatch survival rate, signs of toxicity and growth parameters of fathead minnow embryos were assessed at intervals throughout the study.

The results are based on nominal concentrations. The survival was not impaired in any of the concentration groups compared to the control group. The time to hatch and swim-up was similar in all test groups including the control and thus, was not affected by the test substance. No signs of test item-related toxicity or abnormalities could be observed in any of the peak exposure groups. No test item-related ecologically significant effects were observed in any of the peak exposure groups compared to the control. A tendency towards a reduced body weight was observed in test group 4 (-13% in comparison to the control). However, the ecological significance of such a deviation is considered to be low and may be easily caused by very slight deviations in the environmental conditions. *E.g.* the lowest mean body weight in the replicates of the control group (replicate 0B) is 16% lower than the highest mean body weight measured in replicate 0A. Therefore the observed decrease in body weight is not necessarily substance-related.

In an early life stage study applying a realistic pulse dose exposure pattern with two peaks of alpha-cypermethrin, no substance-related effects on fathead minnow (*Pimephales promelas*) were observed at a nominal peak exposure concentration of 0.30 µg a.s./L.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I); batch no: COD-000165; purity: 98.8% ± 1%.

B. STUDY DESIGN

Test species: Fathead minnow (*Pimephales promelas*), eggs less than 4 hours old, source: parental from Osage catfisheries Inc., Osage Beach (MO), USA.

Test design: Flow-through system (34 d); two concentrations and two peak scenarios (spaced by 7 days) plus one dilution water control; 4 replicates per treatment with 25 fertilized eggs in each. Eggs and larvae were exposed in cylindrical glass vessels and were transferred into stainless steel aquaria on day 19. The test solution flowed continuously from the mixing tank into an "udder" which splitted the test water into 4 equal parts for the 4 replicate test aquaria. On day 34 fish were sacrificed and body length and weight were determined. Daily assessment of hatch, swim-up, survival, signs of toxicity and abnormalities.

Endpoints: semi-NOEC values based on hatching rate, post-hatch survival, toxic signs, growth rates and time spans to hatch and swim-up.

Test concentrations: Control (dilution water), 4 treatment groups: 0.15 and 0.30 µg a.s./L (nominal peak concentration on day 0 and day 7 and on day 7 and day 14, respectively).

Test conditions: Test vessels: Cylindrical glass vessels: water volume: 1.7 L; stainless steel aquaria (29 x 21 x 22 cm), water volume: 9 L. Dilution water: non-chlorinated, filtered drinking water (diluted with deionized water); temperature: 23.5 - 25 °C; pH 7.7 - 8.0; oxygen content: 4.6 - 8.4 mg/L; total hardness: approx. 96 - 102 mg $CaCO_3$/L; conductivity: 258 - 260 µS; acid capacity: 2.46 - 2.5 mmol/L; light intensity: 187 - 391 lux in the cylindrical glass vessels and 98 - 208 lux in the stainless steel aquaria; photoperiod: 16 hours light : 8 hours dark; flow rates: 7.5 L/hour/ treatment group, 1.9 L/hour/test vessel. Feeding: freshly hatched Artemia nauplii starting at day 7 and commercial fish diet (TetraMin) from day 9 on. Aeration: from day 20 on.

Analytics: Analytical verification of test item concentrations was conducted using a GC-method with MS detection.

Statistics: Descriptive statistics; Dunnett`s test for weight and length data (two-sided; $\alpha = 0.01$; $\alpha = 0.05$), Fisher's exact test for survival data (one-sided; $\alpha = 0.01$; $\alpha = 0.05$), Wilcoxon-test for variability between replicates (one-sided; $\alpha = 0.05$).

II. RESULTS AND DISCUSSION

Analytical measurements: The control group was analyzed during the peak exposure dates and was free of contaminations with the test substance on days 0 and 7. However, on day 14 concentrations around the limit of quantification were determined for the control group. It was assumed that this was most likely caused by contamination of the samples. However, the contamination of the control group with the concentrations found was not expected to have any impact on the study results. The analytically determined concentration values of the test item in the test water confirmed that the nominal peak concentrations were reached or even exceeded, while the peak concentrations declined slightly faster than with the intended half-life of one day. Taking into account the properties of the test substance, which is readily adsorbed, the low test concentrations and the difficulties in the analysis, the actual exposure pattern can be regarded as close to the intended exposure scheme. Therefore, the following biological results are based on nominal concentrations.

Biological results: The survival was not impaired in any of the concentration groups compared to the control group. The time to hatch and swim-up was similar in all test groups including the control and thus, was not affected by the test substance. No signs of test item-related toxicity or abnormalities could be observed in any of the peak exposure groups. No test item-related ecologically significant effects were observed in any of the peak exposure groups compared to the control. A tendency towards a reduced body weight was observed in test group 4 (-13% in comparison to the control). However, the ecological significance of such a deviation is considered to be low and may be easily caused by very slight deviations in the environmental conditions. E.g. the lowest mean body weight in the replicates of the control group (replicate 0B) is 16% lower than the highest mean body weight measured in replicate 0A. Therefore the observed decrease in body weight is not necessarily substance-related. The results are summarized in Table 8.2.2.1-1.

Table 8.2.2.1-1: Chronic toxicity of alpha-cypermethrin to fathead minnow (*P. promelas*) in an fish early life stage test (34 d)

Peak concentration [µg/L] (nominal)	**Control**	**0.15**	**0.30**	**0.15**	**0.30**
Embryo survival until hatch [%]	94	94	94	96	97
Survival of larvae from hatch until end of swim-up (day 8) [%]	97	98	97	97	92
Survival of young fish (day 8 - 34) [%]	96	99	99	97	93
Mean larval survival (34 d) [%]	87	91	90	90	83
Start of hatch [day] End of hatch [day]	3 6	3 6	3 6	3 6	3 6
Start of swim-up End of swim-up	5 7 - 8	5 7	5 7 - 8	5 7	5 7 - 8
Symptoms	none	none	none	none	none
Mean weight (34 d) [mg]	209	216	206	191	183
% of control [1)]	100	103	99	91	87
Mean length (34 d) [cm]	2.6	2.6	2.5	2.4	2.4
% of control [1)]	100	101	99	95	94

[1)] Calculated on the basis of the individual values.

III. CONCLUSION

In an early life stage study applying a realistic pulse dose exposure pattern with two peaks of alpha-cypermethrin, no substance-related effects on fathead minnow (*Pimephales promelas*) were observed at a nominal peak exposure concentration of 0.30 µg a.s./L.

CA 8.2.2.2 Fish full life cycle test

The chronic toxicity to fish is fully addressed in two early life stage studies (one of these studies was already evaluated during the previous Annex I inclusion process; the second study was performed for risk refinement in support of EU product registrations). No additional fish full life cycle study is required and no (new) study has been conducted.

CA 8.2.2.3 Bioconcentration in fish

Two bioconcentration studies performed with *cypermethrin* were evaluated during the Annex I inclusion process of alpha-cypermethrin. However, a specific alpha-cypermethrin study was missing, which is now included. In the framework of this BCF study, an acute toxicity study was conducted using Japanese medaka (*Oryzias latipes*) which has not been evaluated previously on EU level. Thus, a summary of the bioconcentration study is provided, which also includes data and results of the acute toxicity testing.

Report: CA 8.2.2.3/1
[redacted] 1997a
Bioconcentration study of Alpha-Cypermethrin with carp
AL-519-004
Guidelines: OECD 305
GLP: yes

Executive Summary

The bioconcentration potential of alpha-cypermethrin (BAS 310 I) was investigated in common carp (*Cyprinus carpio*) in a flow-through system. Animals were exposed to the test item at a low and a high exposure level of 0.010 and 0.10 µg a.s./L, respectively, for a 73-day uptake phase. Additionally, a dilution water control was set up. Subsequently, fish were kept in dilution water for 2 weeks (depuration phase). Fish samples for determination of test item concentrations in fish were taken on weeks 2, 4, 6, 8 and 10 after start of exposure. Analysis of the test item in tissues and organs of the test fish was carried out on day 73 after test initiation. From the results of the elimination test, half-lives of the test item in fish body were determined.
In addition, a 48-hour static acute toxicity laboratory study was conducted in parallel. In this study medaka (*Oryzias latipes*) were exposed to a solvent control (acetonitrile and dispersing agent HCO-30) and to nominal concentrations of 2.0, 3.6, 6.5, 12 and 20 µg alpha-cypermethrin/L (nominal) in groups of 10 animals. Fish were observed for survival 24 and 48 hours after start of exposure.

Bioconcentration test: No toxic effects like increased mortality or changes in behavior or appearance were observed in the test item treatment organisms. The bioconcentration factors (BCFs) of the test item at both exposure levels appeared to reach steady-state plateau by week 4 of exposure. The maximum BCF values at high and low exposure levels were 579 and 910, respectively. The elimination test revealed that half-life of the test item in fish was 8.6 days at the high exposure level and 6.9 days at the low exposure level. From the result of the tissue distribution analysis, the BCFs of the test substance in tissue (of fish exposed for 10 weeks) were the highest in viscera and lowest in fillet.

Acute toxicity test: The biological results are based on nominal concentrations. After 48 hours of exposure, no mortality was observed in the solvent control and at test item concentrations of up to and including 6.5 µg a.s./L, whereas 50% mortality was observed at 12 µg a.s./L and 100% mortality occurred at the highest tested concentration of 20 µg a.s./L. The LC_{50} (48 h) of alpha-cypermethrin was determined to be 12.0 µg a.s./L.

In a flow-through bioconcentration study, common carp were exposed to ^{14}C-labeled alpha-cypermethrin at nominal concentrations of 0.010 and 0.10 µg a.s./L in water. The maximum BCF values obtained in this study during the uptake period were 910 and 579 for the nominal test item concentrations of 0.010 and 0.10 µg a.s./L, respectively. In the static acute toxicity study with medaka (*Oryzias latipes*), the LC_{50} (48 h) of alpha-cypermethrin was 12 µg a.s./L, based on nominal concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Non-radiolabelled alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193; batch no. AC10194-61, purity 96.1%); ^{14}C-labeled BAS 310 I (alpha-cypermethrin; batch-no.: AC10727-40A), specific activity: 60.8 µCi/mg, chemical purity: > 99%, radiochemical purity: 99.6%.

B. STUDY DESIGN

Test species: Bioconcentration test: Common carp (*Cyprinus carpio*), body weight: 22 - 46 g; body length: 9 - 13 cm, mean fat content: 5.0%; source: Sankyo Suisan Co, Tokyo, Japan.
Acute toxicity test: medaka (*Oryzias latipes*); body length: approx. 2 cm; body weight: approx. 0.2 g; supplied by Niikura Fishing (1217 Shimotani, Isehara, Kanagawa, Japan).

Test design: Bioconcentration test: flow-through system (73 days uptake, 2 weeks depuration); two test item treatments plus a dilution water control treatment, one test vessel per treatment with 12 fish per vessel for the control and 22 fish per vessel for the test item treatments at the start of the uptake period. Stock solutions were prepared with acetonitrile and dispersing agent HCO-30. For the depuration phase the test water in the aquaria was replaced by dilution water.
Analysis of the test item concentration in test water was conducted twice a week during the uptake period of 73 days. Fish samples for determination of test item concentrations in fish were taken on weeks 2, 4, 6, 8 and 10 after start of exposure.

	Analysis of the test item in tissues and organs of the test fish was carried out on day 73 after test initiation. After 73 days of exposure, an elimination test was carried out for 2 weeks for each exposure level. Two fish were collected from both exposure aquaria on days 4, 7 and 14 after the initiation of the elimination test. From the results of the elimination test, half-lives of the test item in fish body were determined. Acute toxicity test: Static system (48 h); 5 test item concentrations plus a control; 10 fish per aquarium; stock solutions were prepared with acetonitrile and dispersing agent HCO-30; assessment of mortality 24 hours and 96 hours after start of exposure.
Endpoints:	Bioconcentration test: bioconcentration potential (bioconcentration factor, BCF), depuration half-life. Acute toxicity test: LC_{50}, mortality.
Test concentrations:	Bioconcentration test: Water control, 0.010, 0.10 µg a.s./L (nominal). Acute toxicity test: Solvent control (0.2 µg HCO-30/L and 0.002 mL acetonitrile/L), 2.0, 3.6, 6.5, 12 and 20 µg alpha-cypermethrin/L (nominal).
Test conditions:	Bioconcentration test: 50 L glass aquaria (60 x 30 x 36 cm); water volume: approx. 45 L; flow-through system; dilution water: deionized water; temperature: 25 °C ± 2 °C; oxygen content: > 4 mg/L; turnover rate for each aquarium: 9 times/day, flow rate: 400 L/day/aquarium; population density: exposure groups: 22 fish / 45 L test water, control group: 12 fish / 45 L test water; continuous aeration, feeding: commercial fish diet (Minipet) at approx. 2.5% of the body weight once per day. Acute toxicity test: Test volume: 3 L; dilution water: dechlorinated water; temperature: 25 ± 2°C; oxygen content: > 5.4 mg/L; continuous aeration; no feeding.
Analytics:	Bioconcentration test: Determination of test item concentrations in water and fish or tissue samples was conducted by radioactivity measurements using Liquid Scintillation Counting (LSC). Acute toxicity test: not reported.
Statistics:	Bioconcentration test: calculations according to OECD test guideline 305. Acute toxicity test: Descriptive statistics; graphical determination of LC_{50}.

II. RESULTS AND DISCUSSION

Analytical results

Test item concentration in the water: For the bioconcentration test, analysis of the test item concentration in test water was conducted twice a week during the uptake period of 73 days. The average concentration of the test substance in water during the uptake period was 0.110 ± 0.0173 µg a.s./L for the high exposure level and 0.0113 ± 0.00321 µg a.s./L for the low exposure level. For the acute toxicity test, analytical verification of test item concentrations was not conducted and the following biological results are based on nominal concentrations.

*Test item concentrations in fish: T*he maximum concentrations of the test item in fish during the uptake period were 66.0 ng a.s./g and 9.65 ng a.s./g at the high and the low exposure level, respectively.

Biological results

Bioconcentration test: No toxic effects like increased mortality or changes in behavior or appearance were observed in the test item treatment organisms. The BCFs of the test item at both exposure levels appeared to reach steady-state plateau by week 4 of exposure. The maximum BCF values at high and low exposure levels were 579 and 910, respectively. The elimination test carried out after 10 weeks of exposure revealed that half-life of the test item in fish was 8.6 days at the high exposure level and 6.9 days at the low exposure level. From the result of the tissue distribution analysis, the BCF of the test substance in tissue (of fish exposed for 10 weeks) was the highest in viscera and lowest in fillet. The results are summarized in Table 8.2.2.3-1.

Table 8.2.2.3-1: Bioconcentration Factors (BCF) obtained during the uptake period

Exposure period (weeks)			**2**	**4**	**6**	**8**	**10**
High exposure level	Mean test item concentration in test water [µg a.s./L]		0.109	0.110	0.114	0.114	0.112
	BCF	Sample fish No. 1	286	466	406	458	388
		Sample fish No. 2	294	294	426	579	303
Low exposure level	Mean test item concentration in test water [µg a.s./L]		0.0085	0.0088	0.0106	0.0110	0.0113
	BCF	Sample fish No. 1	155	567	903	441	463
		Sample fish No. 2	138	474	910	422	304

Acute toxicity test: After 48 hours of exposure, no mortality was observed in the solvent control and at test item concentrations of up to and including 6.5 µg a.s./L, whereas 50% mortality was observed at 12 µg a.s./L and 100% mortality occurred at the highest tested concentration of 20 µg a.s./L. **The LC_{50} (48 h) of alpha-cypermethrin was determined to be 12.0 µg/L.** The results are summarized in Table 8.2.2.3-2.

Table 8.2.2.3-2: Acute toxicity (48 h) of alpha-cypermethrin on medaka (*Oryzias latipes*)

Concentration [µg/L] (nominal)	**Control**	**2.0**	**3.6**	**6.5**	**12**	**10**
Mortality (48 h) [%]	0	0	0	0	50	100
Endpoint [µg/L] (nominal)						
LC_{50} (48 h)	12					

III. CONCLUSION

In a flow-through bioconcentration study, common carp were exposed to ^{14}C-labeled alpha-cypermethrin at nominal concentrations of 0.010 and 0.10 µg a.s./L in water. The maximum BCF values obtained in this study during the uptake period were 910 and 579 for the nominal test item concentrations of 0.010 and 0.10 µg a.s./L, respectively.
In the static acute toxicity study with medaka (*Oryzias latipes*) the LC_{50} (48 h) of alpha-cypermethrin was 12 µg a.s./L based on nominal concentrations.

CA 8.2.3 Endocrine disrupting properties

Based on the physical, chemical and structural characteristics of the active substance alpha-cypermethrin as well as based on results of available long-term fish studies (and studies on terrestrial vertebrates; see chapter MCA-8.1.5) there is no indication of endocrine disrupting properties of this active substance. Thus, no further studies are required.

CA 8.2.4 Acute toxicity to aquatic invertebrates

CA 8.2.4.1 Acute toxicity to Daphnia magna

To study the influence of organic matter on the bioavailability and toxicity of alpha-cypermethrin to the standard sensitive arthropod species *Daphnia magna*, an acute higher tier study was conducted in the presence of humic acid and green algae (thus incorporating a contaminated food route). In parallel, a treatment series without organic material (i.e. a standard test) was run.
The study was already listed in the "Application" document submitted for the alpha-cypermethrin AIR 3 renewal process. Two previous runs of the study were invalid due to stability issues with the used test item batch as confirmed by analytical values. In 2015 a 3rd, valid, run of the study was started and is reported here.

New non-standard study conducted on the representative alpha-cypermethrin solo-formulation BAS 310 55 I. Study was submitted under the AIR3 process.

Report:	CA 8.2.4.1/1 Weltje L., Janson G.-M., 2015a The influence of humic acid and green algae on the toxicity of BAS 310 55 I to Daphnia magna STRAUS 2013/1404157
Guidelines:	OECD 202, EPA 850.1010 draft April 1996
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a static acute test, water flea neonates were exposed to BAS 310 55 I at nominal concentrations of 0.6, 2, 6 and 20 µg BAS 310 55 I/L (for the mean measured concentrations in the different test setups see below) in 4 replicates per concentration, each containing 5 daphnids. A dilution water control was tested in parallel. Three setups were run: dilution water only (M4-medium), M4-water with humic acid, and M4-water with green algae. The test item was dispersed in M4-water and diluted with the respective test water (M4-water, M4-water with humic acid, resp. M4-water with green algae) to obtain the desired test concentrations. Daphnids were observed for immobility 24 hours and 48 hours after start of exposure.

The biological results are based on mean measured concentrations of the test item. After 48 hours no immobility of daphnids exposed to BAS 310 55 I in M4-water only was observed in the control and at the lowest test item concentration of 0.40 µg/L, whereas the three highest BAS 310 55 I concentrations in M4-water showed statistically significantly reduced mobility of daphnids of 25%, 45% and 80%, respectively. Regarding the effects of BAS 310 55 I in M4-water with humic acid, no statistically significant effect on mobility was observed in the control treatment and at test

item concentrations of 0.5, 1.6 and 4.0 µg/L, whereas the highest treatment concentration of 13 µg/L showed a statistically significant reduction of mobility by 55%. Mobility of daphnids exposed to BAS 310 55 I in M4-water with green algae was not affected in the control and at the two lowest test item concentrations of 0.42 and 1.2 µg/L, while at 3.6 µg/L 5% immobility was observed. The highest treatment of 14 µg/L showed a statistically significant inhibition of 50%.

In a 48-hour static acute toxicity test with *Daphnia magna* exposed to BAS 310 55 I the lowest EC_{50} was obtained in the standard setup with M4-water and was 3.6 µg/L based on mean measured concentrations. The corresponding NOEC was 0.4 µg/L. In the test water with humic acid and green algae, the EC_{50} values were determined to be 10.3 and 13.9 µg/L, respectively. The corresponding NOEC values were 4.0 and 3.6 µg/L.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 55 I, batch no. FD-150123-0010, content of a.s.: alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193): 49.9 g analyzed (50.0 g/L nominal).

B. STUDY DESIGN

Test species: Water flea (*Daphnia magna* STRAUS), neonates from in-house culture (originally obtained from Institute National de Recherché Chimique Appliquée, France), > 2 < 24 hours old at test initiation.

Test design: Static system (48 hours), 3 parallel test setups were run: M4-water only, M4-water with humic acid and M4-water with green algae; each setup with 4 test concentrations plus a control, 4 replicates per concentration with 5 daphnids in each; assessment of immobility after 24 and 48 hours.

Endpoints: EC_{50} and NOEC based on immobility of daphnids.

Test concentrations: M4-water, M4-water with humic acid or M4-water with green algae with 0 (control), 0.6, 2, 6 and 20 µg BAS 310 55 I/L (nominal), corresponding to the following mean measured concentrations: M4-water: 0.40, 1.1, 2.9, 12.5 µg/L; M-4 water with humic acid: 0.50, 1.6, 4.0, 13 µg/L; M-4 water with algae: 0.42, 1.2, 3.6, 14 µg/L.

Test conditions: Glass vessels, test volume 50 mL, dilution water: "M4" (Elendt medium); M4-water with humic acid: humic acid sodium salt was added to obtain a total organic carbon (TOC) of 6.4 mg/L (25.6 mg humic acid/4000 mL M4-water); M4-water with green algae: M4-water was inoculated with *Desmodesmus subspicatus* to obtain a TOC of 4 mg/L (10.06 mL green algae/4000 mL M4-water); temperature: 20.8°C - 21.3°C; pH 7.90 - 8.28; oxygen content: 8.47 mg/L – 9.13 mg/L; total hardness: 2.51 mmol/L; conductivity: 670 µS/cm; photoperiod: 16 hours light : 8 hours dark; light intensity: 208 - 560 lux; no feeding and no aeration.

Analytics: Analytical verification of test item concentrations was conducted using a GC-method with MS-detection.

Statistics: Descriptive statistics; EC_{50} was calculated using probit analysis, Fisher's Exact Binomial Test with Bonferroni Correction for determination of the NOEC.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations was conducted in the controls and the test item treatments at the beginning and at the end of the test. Measured values for BAS 310 55 I (M4-water) were 44 - 84% of nominal concentrations at test initiation and 51 - 61% of nominal at test termination. Measured values for BAS 310 55 I (M4-water with humic acid) were 57 - 93% of nominal at test initiation and 73 - 80% of nominal at test termination. Measured values for BAS 310 55 I (M4-water with green algae) were 82 - 94% of nominal at test initiation and 31 - 62% of nominal at test termination. Biological results were based on mean measured concentrations.

Biological results: After 48 hours no immobility of daphnids exposed to BAS 310 55 I in standard M4-water was observed in the control and at the lowest test item concentration of 0.40 µg/L, whereas the three highest BAS 310 55 I concentrations in M4-water showed statistically significantly reduced mobility of daphnids of 25%, 45% and 80% (Fisher's Exact Binominal Test with Bonferroni Correction, $p < 0.05$), respectively. Regarding the effects of BAS 310 55 I in M4-water with humic acid, no statistically significant effect on mobility was observed in the control treatment and at test item concentrations of 0.5, 1.6 and 4.0 µg/L, whereas the highest treatment concentration of 13 µg/L showed a statistically significant reduction of mobility by 55% (Fisher's Exact Binominal Test with Bonferroni Correction, $p < 0.05$). Mobility of daphnids exposed to BAS 310 55 I in M4-water with green algae was not significantly affected in the control and at the three lowest test item concentrations, whereas the highest treatment of 14 µg/L showed statistically significant inhibition of 50% (Fisher's Exact Binominal Test with Bonferroni Correction, $p < 0.05$). For results see Table 82.4.1-1.

Table 8.2.4.1-1: Effect of BAS 310 55 I on *Daphnia magna* mobility

Setup	**Concentration [µg/L] (nominal)**	**Control**	**0.6**	**2**	**6**	**20**
M4-water	**Concentration [µg/L] (mean measured)**	**Control**	**0.40**	**1.1**	**2.9**	**12.5**
	Immobility (24 h) [%]	0	0	0	0	35*
	Immobility (48 h) [%]	0	0	25*	45*	80*
	Endpoints [µg BAS 310 55 I/L] (mean measured)					
	EC_{50} (48 h)	3.6 (95% confidence limits: 2.4 – 6.3)				
	NOEC (48 h)	0.4				
M4-water with humic acid	**Concentration [µg/L] (mean measured)**	**Control**	**0.50**	**1.6**	**4.0**	**13**
	Immobility (24 h) [%]	0	0	0	0	10
	Immobility (48 h) [%]	0	0	15	25	55*
	Endpoints [µg BAS 310 55 I/L] (mean measured)					
	EC_{50} (48 h)	10.3 (95% confidence limits: 6.1 – 30.8)				
	NOEC (48 h)	4.0				
M4-water with green algae	**Concentration [µg/L] (mean measured)**	**Control**	**0.42**	**1.2**	**3.6**	**14**
	Immobility (24 h) [%]	0	0	0	0	10
	Immobility (48 h) [%]	0	0	0	5	50*
	Endpoints [µg BAS 310 55 I/L] (mean measured)					
	EC_{50} (48 h)	13.9 (95% confidence limits: 9.5 – 30.6)				
	NOEC (48 h)	3.6				

* Statistically significantly different compared to the control (Fisher's Exact Binominal Test with Bonferroni Correction, $p < 0.05$)

III. CONCLUSION

In a 48-hour static acute toxicity test with *Daphnia magna* exposed to BAS 310 55 I the lowest EC_{50} was obtained in the standard setup with M4-water and was 3.6 µg/L based on mean measured concentrations. The corresponding NOEC was 0.4 µg/L. In the test water with humic acid and green algae, the EC_{50} values were determined to be 10.3 and 13.9 µg/L, respectively. The corresponding NOEC values were 4.0 and 3.6 µg/L.

Study Comments: 8.2.4.1/1	
Agreed Endpoint: 8.2.4.1/1	

CA 8.2.4.2 Acute toxicity to an additional aquatic invertebrate species

The following acute toxicity study with the active substance alpha-cypermethrin on larvae and pupae of the phantom midge *Chaoborus crystallinus* is provided for aquatic risk refinement and has not been evaluated previously. This study has erroneously been listed under 10.2.4.1 in the application submitted for renewal of approval.

Report: CA 8.2.4.2/1
Janson G.-M.,Weltje L., 2009b
Acute toxicity of BAS 310 I (Reg.No. 4078193) to larvae of the phantom midge Chaoborus crystallinus in a 48 hour static test
2009/1085205
Guidelines: OECD 202
GLP: yes
(certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a static acute toxicity laboratory study, larvae of the phantom midge *Chaoborus crystallinus* (Diptera, Chaoboridae) were exposed to alpha-cypermethrin (BAS 310 I) at nominal concentrations of 0 (control), 0 (solvent control), 6.25, 12.5, 25, 50 and 100 ng/L in 4 replicates per concentration, containing 5 larvae each. Additionally, a solvent control and a 50 ng/L treatment were initiated with four replicates, containing 5 pupae in each. Midge larvae and pupae were observed for immobility and other signs of toxicity 24 hours and 48 hours after start of exposure.

The biological results are based on geometric mean measured concentrations. After 24 and 48 hours of exposure statistically significant effects on larval mobility compared to the pooled control were observed at the two highest test item concentrations of 42.43 and 92.47 ng a.s./L. In the two lowest treatments and in the controls some larvae pupated (a sign for normal development). Since the pupae are mobile as well, mobility assessments were made in the same way as for larvae. At test end (48 h) 30% immobility of pupae was observed at a test item concentration of 42.43 ng a.s./L compared to 55% immobility of larvae. Thus, it was shown that pupae of *C. crystallinus* are less sensitive to alpha-cypermethrin than larvae.

In a 48-hour static acute toxicity study with *Chaoborus crystallinus* larvae the EC_{50} of alpha-cypermethrin was 44.54 ng a.s./L based on geometric mean measured concentrations. The NOEC was determined to be 24.87 ng a.s./L (geometric mean measured).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg. No. 4078193), batch no. COD-000595, purity: 99.7% (tolerance ± 1%).

B. STUDY DESIGN

Test species: *Chaoborus crystallinus* (phantom midge, Diptera, Chaoboridae); maintained in-house (non-GLP) for an acclimation period of at least 5 days (obtained from pet shop "Koelle Zoo", Ludwigshafen, Germany), body length of approx. 1.1 - 1.2 cm at test initiation.

Test design: Static system (48 hours), 5 test concentrations plus water and solvent control, 4 replicates with 5 larvae in each; additionally a solvent control and one test concentration (50 ng a.s./L) with 4 replicates, each containing 5 pupae; assessment of immobility after 24 and 48 hours.

Endpoints: NOEC, EC_{50} based on immobility.

Test concentrations: 0 (control), 0 (solvent control), 6.25, 12.5, 25, 50 and 100 ng a.s./L (nominal); corresponding to geometric mean measured concentrations of 8.31, 12.47, 24.87, 42.43 and 92.47 ng a.s./L.

Test conditions: 250 mL glass vessels, test volume 150 mL, filtered natural water from mesocosm pond; pH 7.94 - 8.14; oxygen content: 8.4 - 9.1 mg/L; total hardness: 2.23 mmol/L at test initiation; conductivity: 552 µS/cm at test initiation; temperature: 20.3 - 20.6 °C; light intensity 620 - 936 lux; photoperiod: 16 h light : 8 h dark, no feeding, no aeration.

Analytics: Analytical verification of test item concentrations was conducted using a GC/ECD method.

Statistics: Descriptive statistics; Fisher's exact test ($p < 0.05$) for determination of the NOEC; probit analysis for determination of the EC_{50} value.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentration was conducted in each concentration at the beginning and at the end of the test. Measured concentrations for alpha-cypermethrin ranged from 90% to 147% of nominal at test initiation and from 80% to 120% of nominal at test termination. The following biological results are based on geometric mean measured concentrations.

Biological results: Since there were no statistical differences between the control and the solvent control (t-test, $p < 0.05$) controls were pooled for comparison with the treatments. After 24 and 48 hours of exposure statistically significant effects on larval mobility compared to the pooled control were observed at the two highest test item concentrations of 42.43 and 92.47 ng a.s./L (Fisher's exact test, $p < 0.05$). In the two lowest treatments and in the controls some larvae pupated (a sign for normal development). Since the pupae are mobile as well, mobility assessment was made in the same way as for larvae. At test end (48 h) 30% immobility of pupae was observed at a test item concentration of 42.43 ng a.s./L compared to 55% immobility of larvae. Thus, it was shown that pupae of *C. crystallinus* are less sensitive to alpha-cypermethrin than larvae. For results see Table 8.2.4.2-1.

Table 8.2.4.2-1: Effects of alpha-cypermethrin on the mobility of *Chaoborus crystallinus*

Concentration [ng a.s./L] (geom. mean measured)	**Control**	**Solvent control**		**8.31**	**12.47**	**24.87**	**42.43**		**92.47**
Immobility (24 h) [%]	10	10	30 #	5	20	25	45 *	10 #	70 *
Immobility (48 h) [%]	20	20	35 #	15	45	40	55 *	30 #	80 *
	Endpoints [ng a.s./L] (geom. mean measured)								
EC_{50} (48 h)	44.54 (95% confidence limits: 32.12 - 66.65)								
NOEC (48 h)	24.87								

* Statistically significant different compared to the pooled control (Fisher's exact test, $p < 0.05$).
\# Immobility of pupae

III. CONCLUSION

In a 48-hour static acute toxicity study with *Chaoborus crystallinus* larvae the EC_{50} of alpha-cypermethrin was 44.54 ng a.s./L based on geometric mean measured concentrations. The NOEC was determined to be 24.87 ng a.s./L (geometric mean measured).

CA 8.2.5 Long-term and chronic toxicity to aquatic invertebrates

CA 8.2.5.1 Reproductive and development toxicity to Daphnia magna

The following chronic pulse-dose study toxicity study on *Daphnia magna* performed with the active substance alpha-cypermethrin is provided in support of the aquatic risk assessment and has not been evaluated previously on EU level.

Report:	CA 8.2.5.1/1 Bergtold M., 2007a Chronic toxicity of BAS 310 I to Daphnia magna STRAUS in a 21 day semi-static test - A time to effect study 2007/1016502
Guidelines:	OECD 211, EPA 850.1300
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a semi-static chronic toxicity laboratory study, water flea neonates (between 2 and 24 hours old) were exposed to alpha-cypermethrin (BAS 310 I) at peak concentrations of 0.0372, 0.0744, 0.1117 and 0.1489 µg a.s./L. To evaluate chronic effects after a realistic exposure scenario, a semi-static test has been conducted simulating peak concentrations after two applications of the test item (7 days interval) with subsequent dissipation. Daphnids were observed for parent mortality; body length and reproductive performance throughout the test period.

No mortality of the parent daphnids was observed in the controls and in the treatment with 0.0372 µg a.s./L peak concentration. One daphnid died at concentrations of 0.0744 µg a.s./L and 0.1117 µg a.s./L peak concentration and three daphnids died at 0.1489 µg a.s./L peak concentration. There was no significant treatment effect on mortality. First brood was observed on day 8-12 in the controls and on day 9-13 in the treatments. The number of offspring varied between 112.9 and 127.1. No significant effects on reproduction were found in the treatments compared to the pooled controls. Mean body length of the daphnids varied between 4.26 mm to 4.44 mm. Statistical analysis showed a minor effect on body length in the treatment with 0.1489 µg a.s./L peak concentrations compared to the pooled controls. The effect was judged to be ecologically not relevant.

In a 21-day semi-static chronic toxicity study with *Daphnia magna* no significant effects of the test item were observed on reproduction at the highest treatment comprising two peaks of 0.149 µg a.s./L alpha-cypermethrin (BAS 310 I), whereas a minimal (i.e. 3.2%) effect on body length occurred. As this was judged to be of no ecological relevance, the overall NOAEC is 0.149 µg a.s./L.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg.No. 4078193), batch no. COD-000595, purity: 99.7%.

B. STUDY DESIGN

Test species: Water flea (*Daphnia magna* STRAUS), neonates at test initiation between 2 and 24 hours old; from in-house culture.

Test design: Semi-static test (21 days) with renewal of test water on day 3, 5, 7, 10, 12, 14, 17 and 19; 6 treatment groups (4 test item concentrations, control, solvent control); 10 replicates with 1 daphnid in each treatment; assessment of parent mortality and reproduction throughout the test period; body length at test end.

Endpoints: Parent mortality; body length and reproductive performance.

Test concentrations: Control, solvent control, 0.0372, 0.0744, 0.1117 and 0.1489 µg a.s./L (peak concentrations).

Test conditions: M4 Elendt medium, 50 mL test volume; pH 7.92 - 8.15 (new test solutions), pH 7.74 - 8.13 (old test solutions); oxygen content: 8.38 mg/L - 9.43 mg/L (new test solutions), 7.92 mg/L - 9.43 mg/L (old test solutions); total hardness: 2.30 mmol/L - 2.60 mmol/L (old test solutions); light intensity: approx. 120 lux - 450 lux; temperature 18.9 °C - 21.1 °C (new test solutions), 19.4 °C - 21.0 °C (old test solutions); photoperiod: 16 h light : 8 h dark; feeding with algae, no aeration.

Analytics: The test item concentrations were analyzed using GC with MS-detection.

Statistics: Descriptive statistics, ANOVA followed by Bonferroni-test and William's-test for body length and reproduction data; Fisher's exact test for mortality ($\alpha = 0.05$).

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of the test item was performed in all peak concentrations at study start (day 0) and day 7 in the new test water. Recovery of the test item was in the range of 71.2% to 133.9% and 78.7% to 118.7% of the nominal concentration at day 0 and at day 7, respectively. At the other days of test water exchange the concentration was determined in the stock solutions, where the recovery was in the range of 79.6 and 111.8% of the nominal values.

Biological results: No mortality of the parent daphnids was observed in the controls and in the treatment with 0.0372 µg a.s./L peak concentration. One daphnid died at concentrations of 0.0744 µg a.s./L and 0.1117 µg a.s./L peak concentration and three daphnids died at 0.1489 µg a.s./L peak concentration. There was no significant treatment effect on survival (Fisher's exact test, $\alpha = 0.05$). First brood was observed on day 8-12 in the controls and on day 8-13 in the treatments. The number of offspring varied between 112.9 and 127.1. No significant effects on reproduction were found in the treatments compared to the pooled controls (Bonferroni-test, William's-test, $\alpha = 0.05$, replicates with dead parent daphnids were excluded from statistical evaluation). Mean body length of the daphnids varied between 4.26 to 4.44 mm. Statistical analysis showed a minor but significant effect in the treatment with 0.1489 µg a.s./L peak concentration compared to the pooled controls (Bonferroni test, Williams' test, $\alpha = 0.05$). Although significant, body length was reduced by only 3.2% and thus not considered to be of biological relevance. The results are summarized in Table 8.2.5.1-1.

Table 8.2.5.1-1: Effect (21 d) of alpha-cypermethrin (BAS 310 I) on *Daphnia magna*

Concentration [µg a.s./L] [1)]	**Control**	**Solvent control**	**0.0372**	**0.0744**	**0.1117**	**0.1489**
Parent mortality [%]	0	0	0	10	10	30
No. of offspring per parent	112.9	115.4	126.0	127.1	118.1	117.6
Day of first brood	8-11	8-12	9-11	8-12	10-11	10-13
Mean body length [mm]	4.43	4.37	4.44	4.31	4.73	4.26*

1) Peak concentration

* significant difference compared to the pooled controls (Bonferroni's, Williams' test, $\alpha = 0.05$). Body length was reduced by 3.2%.

III. CONCLUSION

In a 21-day semi-static chronic toxicity study with *Daphnia magna* no significant effects of the test item were observed on reproduction at the highest treatment comprising two peaks of 0.149 µg a.s./L alpha-cypermethrin (BAS 310 I), whereas a minimal (i.e. 3.2%) effect on body length occurred. As this was judged to be of no ecological relevance, the overall NOAEC is 0.149 µg a.s./L.

CA 8.2.5.2 Reproductive and development toxicity to an additional aquatic invertebrate species

No study required; thus, this point is not addressed *via* (new) toxicity studies.

CA 8.2.5.3 Development and emergence in Chironomus riparius

The following acute water-only toxicity study on *Chironomus riparius* larvae with the active substance alpha-cypermethrin was conducted to fulfill the data requirements for Annex I renewal and has not been evaluated previously.

Report:	CA 8.2.5.3/1 Janson G.-M. et al., 2011a Acute toxicity of Alpha-Cypermethrin (BAS 310 I) and of Cypermethrin (BAS 311 I) to the non-biting midge Chironomus riparius in a 48 hour static test 2009/1102214
Guidelines:	OECD 202
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a 48 hours static acute toxicity study, non-biting midge larvae (*Chironomus riparius*) were exposed to alpha-cypermethrin at nominal test concentrations of 0 (control), 5, 10, 20, 40 and 80 ng a.s./L in 4 replicates per concentration, containing 5 larvae each. Chironomids were observed for immobility 24 hours and 48 hours after start of exposure.

The biological results are based on mean measured concentrations of alpha-cypermethrin. After 24 h no significant effects on mobility of chironomids was observed up to and including a concentration of 40 ng a.s./L. After 48 h no significant effects on mobility of chironomids was observed at test item concentrations of 5 and 10 ng a.s./L. Sublethal effects, like lethargy of mobile larvae, were observed in the three highest test concentrations after 24 h and after 48 h in almost all tested concentrations.

In a 48 h static acute toxicity study with *Chironomus riparius* the EC_{50} of alpha-cypermethrin was determined to be 12.6 ng a.s./L based on mean measured concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg. No. 4078193), batch no. COD-000595, purity: 99.2%.

B. STUDY DESIGN

Test species: Non-biting midge (*Chironomus riparius*), first instar larvae; source: in-house culture.

Test design: Static system (48 hours), 5 test concentrations plus control, 4 replicates per concentration and control; 5 larvae per replicate; assessment of immobility after 24 h and 48 h .

Endpoints: NOEC and EC_{50} (regarding immobilization).

Test concentrations: Control, 5, 10, 20, 40 and 80 ng alpha-cypermethrin/L (nominal).

Test conditions: Glass vessels, 50 mL M4 water (Elendt medium); pH 7.93 - 8.04; oxygen content: 8.4 - 9.2 mg/L; total hardness: 2.45 mmol/L (at test initiation); conductivity: 665 µS/cm (at test initiation); water temperature: 20.9 - 21.0°C; light intensity: 600 lux; photoperiod: 16 h light : 8 h dark; no feeding, no aeration.

Analytics: The test item concentrations were analyzed using a GC-ECD method after liquid-liquid extraction of the test item into hexane.

Statistics: Descriptive statistics, Fisher´s exact test ($\alpha = 0.05$) for determination of NOEC; probit analysis for determination of EC_{50}.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations was conducted in each concentration at the beginning and the end of the test. Recoveries for alpha-cypermethrin (Reg. No. 4078193) were in a range of 58% - 80% (average 65.6%) of the nominal concentrations at test initiation and 40% - 80% (average 63.6%) at test termination. The following mean measured concentrations were derived: 4, 6.5, 14, 23.5 and 39 ng a.s./L. The biological results are based on mean measured concentrations of alpha-cypermethrin.

Biological results: BAS 310 I: After 24 h no significant effects on mobility of chironomids was observed up to and including a concentration of 40 ng alpha-cypermethrin /L. After 48 h no significant effects on mobility of chironomids was observed at test item concentrations of 5 and 10 ng a.s./L. Sublethal effects were observed after 24 h in the concentrations 20, 40 and 80 ng a.s./L, and after 48 h, in the test item concentrations 5, 10, 20 and 40 ng a.s./L; the mobile larvae appeared lethargic. The results are summarized in Table 8.2.5.3-1.

Table 8.2.5.3-1: Effects of alpha-cypermethrin on *Chironomus riparius* immobility

Concentration (nominal) [ng a.s./L]	**Control**	**5**	**10**	**20**	**40**	**80**
Concentration (mean measured) [ng a.s./L]	**Control**	**4**	**6.5**	**14**	**23.5**	**39**
Immobility (24 h) [%]	0	0	10	5	15	**45 ***
Immobility (48 h) [%]	5	10	25	**55 ***	**75 ***	**100 ***
	Endpoints [ng a.s./L] (mean measured)					
EC_{50}	12.6 (95% confidence limit: 10.0 - 15.8)					
NOEC	6.5					

* Statistically significantly different from control (Fisher's Exact Test, $\alpha < 0.05$).

III. CONCLUSION

In a 48 h static acute toxicity study with *Chironomus riparius* larvae the EC_{50} of alpha-cypermethrin was determined to be 12.6 ng a.s./L based on mean measured concentrations.

An EU agreed spiked water study on *Chironomus riparius* performed with the active substance alpha-cypermethrin is already available. The following spiked sediment toxicity study was originally conducted for the biocide Annex I listing and is provided here in support of the aquatic risk assessment and has not been evaluated previously.

Report:	CA 8.2.5.3/2 Backfisch K.,Weltje L., 2011a Chronic toxicity of Reg.No. 4078193 (BAS 310 I; Alpha-Cypermethrin) to the non-biting midge Chironomus riparius - A spiked sediment study 2011/1124187
Guidelines:	OECD 218 (2004)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Report:	CA 8.2.5.3/3 Backfisch K.,Weltje L., 2012a Amendment No. 1 - Chronic toxicity of Reg.No. 4078193 (BAS 310 I; Alpha-Cypermethrin) to the non-biting midge Chironomus riparius - a spiked sediment study 2012/1156939
Guidelines:	OECD 218 (2004)
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a 28-day static spiked sediment study, non-biting midge larvae (*Chironomus riparius*) were exposed to alpha-cypermethrin at nominal concentrations of 15, 30, 60, 120 and 240 µg a.s./kg dry sediment (corresponding to mean measured concentrations of 10.5, 22.5, 45.0, 84.5 and 148 µg a.s./kg dry sediment). Additionally, a solvent (acetone) control and a water control were set up. All test item concentrations and the water control had 4 replicates, whereas 6 replicates were tested for the solvent control. 20 larvae were added to each test vessel.

The biological results are based on mean measured concentrations of the test item. First emerged midges were observed on DAI 14 (= day after insertion of larvae). The solvent control data were used for statistical evaluation of treatment related effects. Statistically significant differences compared to the solvent control were found for the emergence rates at the two highest test item concentrations. Statistically significant effects on the development rate were observed in the three highest test item treatments.

In a 28-day static sediment test with *Chironomus riparius*, the EC_{10} values based on emergence rate and development rate were determined to be 51.4 µg a.s./kg dry sediment and 74.1 µg a.s./kg dry sediment, respectively (mean measured).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193), batch no. COD-000595, purity: 99.2%.

B. STUDY DESIGN

Test species: Non-biting midge (*Chironomus riparius*), first instar larvae, not older than 2 days at test initiation; source: in-house culture (non-GLP), originally obtained from "Zoological Institute of the J.W. Goethe University", Frankfurt am Main, Germany.

Test design: Static system (28 days); 5 test concentrations plus a solvent (acetone) control and a water control, 4 replicates per test item concentration and for the water control, 6 replicates for the solvent control; 20 larvae were added to each test vessel; assessment of emergence rate and development rate.

Endpoints: NOEC, EC_{10} and EC_{50} (regarding emergence rate and development rate).

Test concentrations: Solvent (acetone) control, water control, 15, 30, 60, 120 and 240 µg a.s./kg dry sediment (nominal), corresponding to mean measured concentrations of 10.5, 22.5, 45.0, 84.5 and 148 µg a.s./kg dry sediment.

Test conditions: 600 mL glass vessels with 100 g spiked wet artificial sediment (according to OECD 218), 400 mL M4 water (Elendt medium) corresponding to a water layer of about 7.5 - 8.0 cm; pH 7.83 - 8.29; oxygen content: 7.89 - 10.06 mg/L; total hardness: 2.8 mmol/L; conductivity: 679 µS/cm; ammonia: 0.8 mg/L at test initiation and 2.0 mg/L at test termination; water temperature: 19.7 °C - 20.9 °C; light intensity: 536 - 952 lux; photoperiod: 16 h light : 8 h dark; continuous gentle aeration; food: TetraMin (0.25 - 1.0 mg food/larva/day until DAI 24.

Analytics: Analytical verification of test item concentrations in sediment was conducted using a GC-method with MS detection. Overlaying water concentrations were measured using a GC-method with ECD detection.

Statistics: Descriptive statistics, ANOVA followed by Williams' Multiple sequential t-test procedure for determination of the NOEC based on emergence and development rate ($\alpha = 0.05$); probit analysis using linear max. likelihood regression for determination of EC_x values.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations in the overlying water, the pore water and the sediment was conducted in each concentration at the beginning and the end of the test. Recoveries in the sediment were in a range between 60.8% and 80.0% of the nominal concentrations at test initiation and 62.5% and 76.7% of nominal at test termination. Alpha-cypermethrin concentrations found in the overlaying water ranged from < 0.002 µg a.s./L (limit of quantification = 0.005 µg/L) to 0.049 µg a.s./L at test initiation and from < 0.002 to 0.036 µg a.s./L at test end. Measured pore water concentrations were between 0.031 and 0.210 µg a.s./L at the beginning of the test and between < 0.002 and 0.140 µg a.s./L at test termination. The following biological results are based on mean measured sediment concentrations.

Biological results: First emerged midges were observed on DAI 14. The solvent control data were used for statistical evaluation of treatment related effects. Statistically significant differences compared to the solvent control were found for the emergence rates at the two highest test item concentrations (Williams' Multiple sequential t-test procedure, $\alpha = 0.05$). Statistically significant effects on the development rate were observed in the three highest test item treatments (Williams' Multiple sequential t-test procedure, $\alpha = 0.05$). The results are summarized in Table 8.2.5.3-2.

Table 8.2.5.3-2: Effects of alpha-cypermethrin on emergence and development of *Chironomus riparius*

Concentration [µg a.s./kg dry sediment] (nominal)	**Control**	**Solvent control**	**15**	**30**	**60**	**120**	**240**
Concentration [µg a.s./kg dry sediment] (mean measured)	**--**	**--**	**10.5**	**22.5**	**45.0**	**84.5**	**148**
Emergence rate (ER) #	0.9125 ± 0.0479	0.9000 ± 0.0548	0.9000 ± 0.0408	0.8875 ± 0.1109	0.8125 ± 0.0479	0.7250 ± 0.1041 *	0.1250 ± 0.1190 *
Development rate per day (DR) #	0.0665 ± 0.0010	0.0672 ± 0.0016	0.0652 ± 0.0020	0.0651 ± 0.0033	0.0618 ± 0.0020 *	0.0594 ± 0.0013 *	0.0577 ± 0.0063 *
Endpoints [µg alpha-cypermethrin/kg dry sediment] (mean measured)							
$EC_{10\ emergence\ rate}$ (28 d)	51.4						
$EC_{10\ development\ rate}$ (28 d)	74.1						

Values represent mean and standard deviation from all replicates, each with 20 larvae.
* Statistically significant difference compared to the solvent control (Williams' Multiple sequential t-test procedure, $\alpha = 0.05$).

III. CONCLUSION

In a 28-day static sediment test with *Chironomus riparius*, the EC_{10} values based on emergence rate and development rate were determined to be 51.4 µg a.s./kg dry sediment and 74.1 µg a.s./kg dry sediment, respectively (mean measured).

CA 8.2.5.4 Sediment dwelling organisms

The following spiked sediment toxicity study on *Lumbriculus variegatus* conducted with the active substance alpha-cypermethrin was originally conducted for biocide Annex I listing and is provided here in support of the aquatic risk assessment and has not been evaluated previously.

Report:	CA 8.2.5.4/1 Gilberg D. et al., 2013a Alpha-Cypermethrin (BAS 310 I): A study on the chronic toxicity to the sediment dweller Lumbriculus variegatus 2012/1205915
Guidelines:	OECD 203 (1992), OECD 225 Sediment-water Lumbriculus toxicity test using spiked sediment (October 2007)
GLP:	yes (certified by Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden)

Executive Summary

In a 28-day static spiked sediment study, sediment-dwelling worms (*Lumbriculus variegatus*) were exposed to alpha-cypermethrin at nominal concentrations of 62.5, 125, 250, 500 and 1000 µg alpha-cypermethrin/kg dry sediment (corresponding to initial mean measured concentrations of 71.3, 143, 285, 570 and 1140 µg a.s./kg dry sediment). Additionally, a solvent (acetone) control was set up. All test item concentrations had 4 replicates, whereas 6 replicates were tested for the solvent control. 10 worms were added to each test vessel.

The biological results are based on initial mean measured concentrations of the test item. Concentration-dependent sediment avoidance, reduction, respectively, delay in production of fecal pellets and likely immobile worms on the sediment surface were observed at the three highest concentration levels. After 28 days of exposure, survival was not affected significantly up to and including the highest test item concentration. Since no meaningful number of dead worms (at test termination, only one worm was found to be dead, each in the test item treatments of 250 and 1000 µg a.s./kg dry sediment) and no concentration response relation was observed, no statistical evaluation was performed for the endpoint survival. Statistically significant differences compared to the solvent control were found for the parameters reproduction and total weight at the four highest test item concentrations, whereas for the parameter individual weight, statistically significant effects compared to the solvent control were observed at the three highest test item concentrations.

In a 28-day static sediment test with *Lumbriculus variegatus*, the NOEC values of alpha-cypermethrin were determined to be 71.3 µg a.s./kg dry sediment (initial mean measured) based on both reproduction and biomass and 143 µg a.s./kg dry sediment based on individual biomass (initial mean measured).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193), batch no. COD-000595, purity: 99.4%.

B. STUDY DESIGN

Test species: Sediment-dwelling worm (*Lumbriculus variegatus*), synchronized adult worms of similar size; source: in-house culture, originally obtained from "Fischfutter Etzbach", Mechernich-Bergheim, Germany.

Test design: Static system (28 days); 5 test concentrations plus a solvent (acetone) control, 4 replicates per test item concentration, 6 replicates for the solvent control; 10 worms were added to each test vessel; assessment of reproduction (worm number), total weight and mean individual weight at test termination.

Endpoints: NOEC and EC_{50} (regarding reproduction, total weight and individual weight).

Test concentrations: Solvent (acetone) control, 62.5, 125, 250, 500 and 1000 µg alpha-cypermethrin/kg dry sediment (nominal), corresponding to initial mean measured concentrations of 71.3, 143, 285, 570 and 1140 µg a.s./kg dry sediment.

Test conditions: 250 mL glass vessels with 80 g spiked wet artificial sediment (according to OECD 225), reconstituted water (according to OECD 203), approximately 175 ml overlying water per test vessel; pH 7.6 - 8.1; oxygen content: 90% to 94%; total hardness: 273.3 - 292.9 mg $CaCO_3$/L; conductivity: 614 µS/cm; ammonia: < LOQ (limit of quantification = 1.3 mg/L) - 3.29 mg/L; water temperature: 20.3 °C - 20.7 °C; light intensity: 168 - 414 lux; photoperiod: 16 h light : 8 h dark; continuous gentle aeration; feed in sediment (addition of *Urtica*-powder (0.25% on dry sediment) and cellulose (0.25% on dry sediment) before application of the test item).

Analytics: Analytical verification of test item concentrations was conducted using a GC-MS-method.

Statistics: Descriptive statistics, Welch-t test and Williams t-test for determination of the NOEC values (α = 0.05), probit analysis for determination of EC_x values.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations in the overlying water and the sediment was conducted in the test concentrations of 62.5 and 1000 µg a.s./kg dry sediment at the beginning and the end of the test. Recoveries in the sediment were in a range between 110% and 118% of the nominal concentrations at test initiation and 101% and 102% of nominal at test termination. Alpha-cypermethrin concentrations found in the overlaying water ranged from 4.0 to 63 ng a.s./L on day 0 and from 29 to 95 ng a.s./L of nominal on day 28. The following biological results are based on initial mean measured sediment concentrations.

Biological results: Concentration-dependent sediment avoidance, reduction, respectively delay in production of fecal pellets and likely immobile worms on the sediment surface were observed at the three highest concentration levels. After 28 days of exposure, survival was not affected significantly up to and including the highest test item concentration. Since no meaningful number of dead worms (at test termination, only one worm was found to be dead, each in the test item treatments of 250 and 1000 µg a.s./kg dry sediment) and no concentration response relation was observed, no statistical evaluation was performed for the endpoint survival. Statistically significant differences compared to the solvent control were found for the parameters reproduction and total weight at the four highest test item concentrations (Welch t-test, $\alpha = 0.05$), whereas for the parameter individual weight, statistically significant effects compared to the solvent control were observed at the three highest test item concentrations (Williams t-test, $\alpha = 0.05$). The results are summarized in Table 8.2.5.4-1.

Table 8.2.5.4-1: Effects of alpha-cypermethrin on reproduction and biomass of *Lumbriculus variegatus*

Concentration [µg a.s./kg dry sediment] (nominal)	**Solvent control**	**62.5**	**125**	**250**	**500**	**1000**
Concentration [µg a.s./kg dry sediment] (initial mean measured)	**--**	**71.3**	**143**	**285**	**570**	**1140**
Reproduction (worm number at 28 d) #	24.3 ± 2.34	21.3 ± 3.50	10.5 ± 1.00 *	9.8 ± 0.50 *	10.0 ± 0.00 a)	9.8 ± 0.50 *
Total weight at 28 d [mg dry weight] #	30.05 ± 3.87	26.63 ± 2.73	14.30 ± 0.98 *	7.25 ± 0.73 *	3.35 ± 0.19 *	2.28 ± 0.39 *
Individual weight at 28 d [mg dry weight per worm] #	1.247 ± 0.2156	1.285 ± 0.2868	1.375 ± 0.1915	0.748 ± 0.1180 +	0.335 ± 0.0191 +	0.234 ± 0.0413 +
	Endpoints [µg a.s./kg dry sediment] (initial mean measured)					
$NOEC_{survival}$	≥ 1140					
$NOEC_{reproduction}$ (28 d)	71.3					
$NOEC_{biomass}$ (28 d)	71.3					
$NOEC_{individual\ biomass}$ (28 d)	143					

Values represent mean and standard deviation from all replicates.
* Statistically significant difference compared to the solvent control (Welch t-test, $\alpha = 0.05$).
\+ Statistically significant difference compared to the solvent control (Williams t-test, $\alpha = 0.05$).
a) Welch t-test could not be performed

III. CONCLUSION

In a 28-day static sediment test with *Lumbriculus variegatus*, the NOEC values of alpha-cypermethrin were determined to be 71.3 µg a.s./kg dry sediment (initial mean measured) based on both reproduction and biomass and 143 µg a.s./kg dry sediment based on individual biomass (initial mean measured).

The following chronic spiked sediment toxicity study on the nematode *Caenorhabditis elegans* conducted with the active substance alpha-cypermethrin was originally conducted for biocide Annex I listing and is provided here in support of the aquatic risk assessment and has not been evaluated previously.

Report:	CA 8.2.5.4/2 Hoess S., 2013a Chronic toxicity of Alpha-Cypermethrin to Caenorhabditis elegans exposed via spiked sediment according to ISO guideline 10872 (2010) 2013/1250848
Guidelines:	ISO 10872 (2010)
GLP:	yes (certified by Landesanstalt fuer Umwelt, Messungen und Naturschutz Baden-Wuerttemberg, Karlsruhe, Germany)

Executive Summary

In a chronic 96-h static spiked sediment study, nematodes (*Caenorhabditis elegans*) were exposed to alpha-cypermethrin. Nominal test concentrations were 300, 1000, 3000, 10000 and 30000 µg a.s./kg dry sediment (corresponding to mean measured concentrations of 230, 890, 3850, 7450 and 28650 µg a.s./kg dry sediment and to initial measured concentrations of 230, 730, 4390, 7510 and 28230 µg a.s./kg dry sediment). Additionally, a solvent (acetone) control and a dilution water control were set up. All test item concentrations, the dilution water control and the solvent control had 6 replicates. To each test vessel, 10 nematodes were added.

The biological results are based on mean measured concentrations, as well as on initial measured concentrations. Nematode growth and reproduction in the solvent control did not significantly differ from values of the control, meaning that no effect of the solvent occurred. The test item showed no significant inhibitory effect on growth and reproduction of *C. elegans* compared to the solvent control at any tested concentration. Even though the mean value was decreased in the lowest treatment group, the number of offspring did not differ significantly from the solvent control and was not considered to be a treatment related effect.

In a chronic 96-h static sediment test with *Caenorhabditis elegans* the NOEC of alpha-cypermethrin for both growth and reproduction was determined to be ≥ 28600 µg a.s./kg dry sediment (≥ 17200 µg a.s./kg wet sediment) based on mean measured concentrations and ≥ 28200 µg a.s./kg dry sediment (≥ 16900 µg a.s./kg wet sediment) based on initially measured concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg. no. 4 078 193), batch no. COD-000595, purity: 99.2% ± 1%.

B. STUDY DESIGN

Test species: Nematode (*Caenorhabditis elegans*), strain N2, wild type; first stage J1 at test beginning; initial body length: 275 ± 49 µm; source: Caenorhabditis Genetic Centre, Dept of GCD, University of Minnesota, Minneapolis, USA.

Test design: Static system (96 hours); 5 test concentrations plus a solvent (acetone) control and a dilution water control, 6 replicates per test item concentration and for the control groups; 10 test organisms per replicate; assessment of growth and reproduction.

Endpoints: NOEC based on growth and reproduction.

Test concentrations: Solvent (acetone) control, dilution water control, 300, 1000, 3000, 10000 and 30000 µg a.s./kg dry sediment (nominal), corresponding to mean measured concentrations of 230, 890, 3850, 7450 and 28650 µg a.s./kg dry sediment and to initial measured concentrations of 230, 730, 4390, 7510 and 28230 µg a.s./kg dry sediment.

Test conditions: 5 mL glass vessels with 0.5 g spiked wet artificial sediment (according to ISO 10872) and 0.2 mL M9 medium; food: bacterial suspension with 11,924 FAU; temperature: 19.5 °C - 20.0 °C.

Analytics: Analytical verification of test item concentrations was conducted using a GC-method with MS detection.

Statistics: Descriptive statistics, t-test for comparison of the control data ($p < 0.05$); one-way ANOVA followed by Dunnett's test ($p < 0.05$).

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations in the sediment was conducted in each concentration at the beginning and the end of the test. Recoveries in the sediment were in a range between 73% and 146 % of the nominal concentrations at test initiation. At test termination the detected concentrations ranged from 74% to 110% of the nominal values. The following biological results are based on mean measured sediment concentrations. Additionally, results are presented based on initial measured sediment concentrations.

Biological results: Nematode growth and reproduction in the solvent control did not significantly differ from values of the control (t-test, $p > 0.05$), meaning that no effect of the solvent occurred. The test item showed no significant inhibitory effect on growth and reproduction of *C. elegans* compared to the solvent control at any tested concentration (one-way ANOVA followed by Dunnett's test; $p > 0.05$). Even though the mean value was decreased in the lowest treatment group, the number of offspring did not differ significantly from the solvent control (one-way ANOVA followed by Dunnett's test; $p > 0.05$) and was not considered to be a treatment related effect. The results are summarized in Table 8.2.5.4-2.

Table 8.2.5.4-2: Effects of alpha-cypermethrin on growth and reproduction of *Caenorhabditis elegans* after 96 hours of exposure

Concentration [µg a.s./kg dry sediment] (nominal)	**300**	**1000**	**3000**	**10000**	**30000**
Concentration [µg a.s./kg dry sediment] (mean measured)	**230**	**890**	**3850**	**7450**	**28650**
Concentration [µg a.s./kg dry sediment] (initial measured)	**230**	**730**	**4390**	**7510**	**28230**
Growth (% Inhibition compared to solvent control) [#]	-1.6	-6.5	-2.9	-5.7	-2.3
Reproduction (Offspring / test organism) (% Inhibition compared to solvent control) [#]	30.5	-9.4	-5.9	-16.0	-24.4
	Endpoints [µg a.s./kg dry sediment] (mean measured / initially measured)				
$NOEC_{growth\ /\ reproduction}$	≥ 28600 / ≥ 28200				

[#] Negative inhibition values indicate higher performance than in the solvent control

III. CONCLUSION

In a chronic 96-h static sediment test with *Caenorhabditis elegans* the NOEC of alpha-cypermethrin for both growth and reproduction was determined to be ≥ 28600 µg a.s./kg dry sediment (≥ 17200 µg a.s./kg wet sediment) based on mean measured concentrations and ≥ 28200 µg a.s./kg dry sediment (≥ 16900 µg a.s./kg wet sediment) based on initially measured concentrations.

CA 8.2.6 Effects on algal growth

CA 8.2.6.1 Effects on growth of green algae

EU agreed endpoints for the freshwater green alga *Pseudokirchneriella subcapitata* are available, thus no further studies are required.

CA 8.2.6.2 Effects on growth of an additional algal species

The following 96 hour algal study on the freshwater diatom *Navicula pelliculosa* performed with the active substance alpha-cypermethrin is not required for registration in the EU. The study was conducted for U.S. registration purposes and has not been evaluated previously on EU level. In accordance to the EFSA Aquatic Guidance Document (EFSA, 2013) and OECD guideline 201 (2001) the 72 h endpoints obtained in the 96 h study are considered as relevant endpoint for the aquatic risk assessment, therefore both the 72 h and 96 h endpoints are reported in the study summary below.

Report:	CA 8.2.6.2/1 Hoffmann F., 2009a Effect of BAS 310 I (Reg.No. 4078193) on the growth of the fresh water diatom Navicula pelliculosa - A limit test 2009/1109081
Guidelines:	OECD 201, EPA 850.5400
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a 96-hour static toxicity limit test, the effect of alpha-cypermethrin on the growth of the freshwater diatom *Navicula pelliculosa* was investigated. Due to its poor solubility in water the test substance was applied as pure centrifuged stock solution at a geometric mean measured test concentration of 70.3 µg a.s./L. Assessment of growth was conducted 24 h, 48 h, 72 h and 96 h after test initiation.

The biological results are based on the geometric mean measured concentration of the test item. No morphological effects on algae were observed in the control and at the tested treatment concentration. Furthermore, no statistically significant differences of growth and yield compared to the control were observed after exposure over 96 hours.

In a 96 hour algal toxicity limit test with *Navicula pelliculosa,* the E_rC_{50} and the E_yC_{50} for alpha-cypermethrin were both determined to be > 70.3 µg a.s./L after exposure over 72 and 96 hours, based on geometric mean measured concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I; Reg. No. 4 078 193), batch no. COD-000595; purity: 99.2 ± 1.0%.

B. STUDY DESIGN

Test species: Freshwater diatom, *Navicula pelliculosa*, strain UTEX 674, stock originally obtained from the "UTEX Culture Collection of Algae", University of Texas, Austin, USA.

Test design: Static system; test duration 96 hours; limit test: 1 test concentration plus a control; due to the poor water solubility of the test item a stock solution was prepared, constantly stirred for 24 hours and centrifuged to remove undissolved particles; the centrifuged stock solution was used for the limit test; 6 replicates for the test item and 10 replicates for the control; daily assessment of growth.

Endpoints: EC_{50} with respect to growth rate and yield after exposure over 72 hours and 96 hours.

Test concentrations: Control (dilution water), pure centrifuged stock solution: 70.3 µg alpha-cypermethrin/L (geometric mean measured).

Test conditions: 100 mL dimple flasks; test volume 60 mL; nutrient solution according to OECD 201; pH 8.1 at test initiation and pH 8.36 - 8.41 at test termination; temperature: 22 ± 1 °C; initial cell densities 1 x 10^4 cells/mL; continuous light at approx. 8000 lux; constant shaking at 135 rpm.

Analytics: Analytical verification of test item concentrations was conducted using a GC-method with MS detection.

Statistics: Descriptive statistics; Student t-test ($\alpha = 0.05$) for determination of the NOEC (72 h and 96 h) values.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations was conducted in the tested concentration at the beginning and at the end of the test. The analyzed contents of alpha-cypermethrin ranged from 44.4 to 64.3 µg a.s./L at test initiation and from 56.5 to 116 µg a.s./L at test termination. The resulting geometric mean measured concentration of alpha-cypermethrin was 70.3 µg a.s./L and is used for the following biological results.

Biological results: No morphological effects on algae were observed in the control and at the tested treatment concentration. Furthermore, no statistically significant differences of growth and yield compared to the control were observed after exposure over 96 hours (Student t-test; $\alpha = 0.05$).The effects on algal growth are summarized in Table 8.2.6.2-1.

Table 8.2.6.2-1: Effect of alpha-cypermethrin on the growth of the freshwater diatom *Navicula pelliculosa*

Concentration [µg a.s./L] (geometric mean measured)	**70.3**
Inhibition in 72 h (growth rate) *	-0.1
Inhibition in 72 h (yield) *	-0.4
Inhibition in 96 h (growth rate)	0.1
Inhibition in 96 h (yield)	0.4
Endpoints [µg alpha-cypermethrin/L] (geometric mean measured)	
E_rC_{50} / E_yC_{50} (72 h & 96 h)	> 70.3
NOE_rC / NOE_yC (96 h)	≥ 70.3

* Negative values indicate stimulated growth compared to the control.

III. CONCLUSION

In a 96 hour algae toxicity limit test with *Navicula pelliculosa,* the E_rC_{50} and the E_yC_{50} for alpha-cypermethrin were both determined to be > 70.3 µg a.s./L after exposure over 72 and 96 hours, based on geometric mean measured concentrations.

The following 96 hour algal study on the freshwater blue-green alga *Anabaena flos-aquae* performed with the active substance alpha-cypermethrin is not required for registration in the EU. The study was conducted for U.S. registration purposes and has not been evaluated previously on EU level. In accordance to the EFSA Aquatic Guidance Document (EFSA, 2013) and OECD guideline 201 (2001) the 72 h endpoints obtained in the 96 h study are considered as relevant endpoint for the aquatic risk assessment, therefore both the 72 h and 96 h endpoints are reported in the study summary below.

Report:	CA 8.2.6.2/2 Hoffmann F., 2009b Effect of BAS 310 I (Reg.No. 4078193) on the growth of the blue-green alga Anabaena flos-aquae - A limit test 2009/1109080
Guidelines:	OECD 201, EPA 850.5400
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a 96-hour static toxicity limit test, the effect of alpha-cypermethrin on the growth of the freshwater blue-green alga *Anabaena flos-aquae* was investigated. The test item concentration reflected the maximum solubility of the test item under test conditions and corresponded to a geometric mean measured test item concentration of 27.0 µg alpha-cypermethrin/L. Assessment of growth was conducted 24 h, 48 h, 72 h and 96 h after test initiation.

The biological results are based on geometric mean measured concentrations of the test item. No morphological effects on algae were observed in the control and the tested treatment concentration. Furthermore, no statistically significant differences of growth and yield compared to the control were observed after exposure over 96 hours.

In a 96 hour algae toxicity limit test with *Anabaena flos-aquae,* the E_rC_{50} and the E_yC_{50} for alpha-cypermethrin were both determined to be > 27.0 µg a.s./L after exposure over 72 and 96 hours, based on geometric mean measured concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I; Reg. No. 4 078 193), batch no. COD-000595; purity: 99.2 ± 1.0%.

B. STUDY DESIGN

Test species:	Freshwater blue-green alga, *Anabaena flos-aquae*, specification UTEX B 674, stock originally obtained from "UTEX Culture Collection of Algae", University of Texas, Austin, USA.
Test design:	Static system; test duration 96 hours; limit test: 1 test concentration plus a control; 6 replicates for the test item and 10 replicates for the control; the test item concentration reflects the maximum solubility of the test item under test conditions; daily assessment of growth.
Endpoints:	EC_{50} and NOEC with respect to growth rate and yield after exposure over 72 hours and 96 hours.
Test concentrations:	Control, pure centrifuged stock solution: 27.0 µg alpha-cypermethrin/L (geometric mean measured).
Test conditions:	100 mL dimple flasks; test volume 60 mL; nutrient solution according to OECD 201; pH 7.5 at test initiation and pH 7.24 - 7.30 at test termination; temperature: 24 ± 1 °C; initial cell densities 1 x 10^4 cells/mL; continuous light at approx.. 3500 lux; constant shaking at 170 rpm.
Analytics:	Analytical verification of test item concentrations was conducted using a GC-method with MS detection.
Statistics:	Descriptive statistics; Student t-test ($\alpha = 0.05$) for determination of the NOEC value.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of the test item concentrations was conducted in the tested concentration at the beginning and at the end of the test. The mean analyzed content of alpha-cypermethrin in the test item treatment was 28.0 µg/L at test initiation and 26.1 µg/L at test termination, resulting in a geometric mean measured concentration of 27.0 µg a.s./L. The following biological results are based on the geometric mean measured concentration.

Biological results: No morphological effects on algae were observed in the control and the tested treatment concentration. Furthermore, no statistically significant differences of growth and yield compared to the control were observed after exposure over 96 hours (Student t-test; $\alpha = 0.05$). The effects on algal growth are summarized in Table 8.2.6.2-2.

Table 8.2.6.2-2: Effect of alpha-cypermethrin on the growth of the freshwater blue-green alga *Anabaena flos-aquae*

Concentration [µg a.s./L] (geometric mean measured)	**27.0**
Inhibition after 72 h (growth rate)	0.0
Inhibition after72 h (yield) [#]	- 0.1
Inhibition after 96 h (growth rate)	0.0
Inhibition after 96 h (yield)	0.1
Endpoints [µg alpha-cypermethrin/L] (geometric mean measured)	
E_rC_{50} / E_yC_{50} (72 h & 96 h)	> 27.0
NOE_rC / NOE_yC (96 h)	≥ 27.0

[#] Negative values indicate stimulated growth compared to the control.

III. CONCLUSION

In a 96 hour algae toxicity limit test with *Anabaena flos-aquae,* the E_rC_{50} and the E_yC_{50} for alpha-cypermethrin were both determined to be > 27.0 µg a.s./L after exposure over 72 and 96 hours, based on geometric mean measured concentrations.

The following 96 hour alga study on the marine diatom *Skeletonema costatum* performed with the active substance alpha-cypermethrin is not required for registration in the EU. The study was conducted for U.S. registration purposes and has not been evaluated previously on EU level. In accordance to the EFSA Aquatic Guidance Document (EFSA, 2013) and OECD guideline 201 (2001) the 72 h endpoints obtained in the 96 h study are considered as relevant endpoint for the aquatic risk assessment, therefore both the 72 h and 96 h endpoints are reported in the study summary below.

Report: CA 8.2.6.2/3
Hoffmann F., 2009c
Effect of BAS 310 I (Reg.No. 4078193) on the growth of the marine diatom Skeletonema costatum - A limit test
2009/1109079
Guidelines: OECD 201, EPA 850.5400
GLP: yes
(certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a 96-h static toxicity limit test, the effect of alpha-cypermethrin on the growth of the marine diatom *Skeletonema costatum* was investigated. The test item concentration reflected the maximum solubility of the test item under test conditions and corresponded to a geometric mean measured test item concentration of 33.4 µg alpha-cypermethrin/L. Assessment of growth was conducted 24 h, 48 h, 72 h and 96 h after test initiation.

The biological results are based on the geometric mean measured concentration of the test item. No morphological effects on algae were observed in the control group and at the tested concentration. After 96 hours of exposure, no statistically significant differences compared to the control were observed at the tested concentration of 33.4 µg a.s./L.

In a 96-h algae toxicity limit test with *Skeletonema costatum*, the E_rC_{50} and the E_yC_{50} of alpha-cypermethrin were both determined to be > 33.4 µg a.s./L after exposure over 72 and 96 hours, based on geometric mean measured concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I; Reg. no.: 4 078 193), batch no. COD-000595; purity: 99.2% ± 1%.

B. STUDY DESIGN

Test species: Marine diatom, *Skeletonema costatum*, specification: UTEX 2308; stock obtained from "UTEX Culture collection of Algae", University of Texas, Austin, USA.

Test design: Static system (96 hours); limit test: 1 test concentration plus a control; 6 replicates for the test item and 10 replicates for the control; the test item concentration reflects the maximum solubility of the test item under test conditions; daily assessment of growth.

Endpoints: EC_{50} and NOEC with respect to growth rate and yield after exposure over 72 hours and 96 hours.

Test concentrations: Control, pure centrifuged stock solution: 33.4 µg alpha-cypermethrin/L (geometric mean measured).

Test conditions: 100 mL Erlenmeyer dimple flasks; test volume: 60 mL; nutrient solution according to OECD 201; pH 8.1 at test initiation and pH 7.79 - 7.83 at test termination; temperature: 20 ± 1 °C; initial cell densities: 1 x 10^4 cells/mL; photoperiod: 14 hours light : 10 hours dark, light intensity: about 4400 lux, continuous shaking at 60 rpm.

Analytics: Analytical verification of test item concentrations was conducted using a GC method with MS detection.

Statistics: Descriptive statistics, Student t-test (α = 0.05) for determination of the NOEC (96 h) values.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of the test item concentrations was conducted in the tested concentration at the beginning and at the end of the test. The mean analyzed content of alpha-cypermethrin in the test item treatment was 34.6 µg/L at test initiation and 32.3 µg/L at test termination, resulting in a geometric mean measured concentration of 34.4 µg a.s./L. The following biological results are based on the geometric mean measured concentration.

Biological results: No morphological effects on algae were observed in the control group and at the tested concentration. After 96 hours of exposure, no statistically significant differences compared to the control were observed at the tested concentration of 33.4 µg a.s./L (Student t-test, α = 0.05). The effects on algal growth are summarized in Table 8.2.6.2-3.

Table 8.2.6.2-3: Effect of alpha-cypermethrin on the growth of the marine diatom *Skeletonema costatum*

Concentration [µg a.s./L] (geometric mean measured)	**33.4**
Inhibition in 72 h (growth rate) [%]	- 0.4
Inhibition in 72 h (yield) [%]	- 1.3
Inhibition in 96 h (growth rate) [%]	- 0.4
Inhibition in 96 h (yield) [%]	- 1.8
Endpoints [µg alpha-cypermethrin/L] (geometric mean measured)	
E_rC_{50} / E_yC_{50} (72 h & 96 h)	> 33.4
NOE_rC / NOE_yC (96 h)	≥ 33.4

* Negative values indicate stimulated growth compared to the control.

III. CONCLUSION

In a 96-h algae toxicity limit test with *Skeletonema costatum*, the E_rC_{50} and the E_yC_{50} of alpha-cypermethrin were both determined to be > 33.4 µg a.s./L after exposure over 72 and 96 hours, based on geometric mean measured concentrations.

CA 8.2.7 Effects on aquatic macrophytes

The following toxicity study on the aquatic plant *Lemna gibba* performed with the active substance alpha-cypermethrin is not required for registration in the EU. The study was conducted due to U.S. data requirements and has not been evaluated previously on EU level.

Report:	CA 8.2.7/1 Hoffmann F., 2009d Effect of BAS 310 I (Reg.No. 4078193) on the growth of Lemna gibba - A limit test 2009/1108874
Guidelines:	OECD 221, EPA 850.4400, ASTM E 1415-91
GLP:	yes (certified by Landesamt fuer Umwelt, Wasserwirtschaft und Gewerbeaufsicht, Mainz, Germany)

Executive Summary

In a 7-day static limit test, the effect of alpha-cypermethrin on the growth of the duckweed *Lemna gibba* was investigated. The test item concentration reflected the maximum solubility of the test item under test conditions and corresponded to a geometric mean measured test item concentration of 1.39 µg alpha-cypermethrin/L. Additionally, a dilution water control was set up. Assessment of plant growth and other effects was conducted 3, 5 and 7 days after test initiation. The percentage growth inhibition in the test item treatment, relative to the control, was calculated based on growth rates and final yield for the parameters frond number and plant dry weight.

The biological results are based on the geometric mean measured concentration of the test item. The duckweed population in the control vessels showed sufficient, exponential growth. No morphological effects on the duckweed were observed in the control and at the test item concentration of 1.39 µg a.s./L. There were no statistically significant effects on the growth of *Lemna gibba* compared to the control at the tested concentration of 1.39 µg a.s./L.

In a 7-day aquatic-plant limit test with *Lemna gibba,* both the E_rC_{50} and the E_yC_{50} values of alpha-cypermethrin based on frond number and dry weight were determined to be > 1.39 µg a.s./L, based on geometric mean measured concentrations.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I; Reg. no.: 4 078 193), batch no. COD-000595; purity: 99.2%.

B. STUDY DESIGN

Test species: Duckweed (*Lemna gibba* G3); inocula: 10 days old cultures; maintained in-house; stock obtained from "ÖkoTox Moser & Pickl GbR", Stuttgart, Germany.

Test design: Static system; test duration 7 days; limit test: 1 test item concentration plus a control with 6 replicates, respectively, 2 plants with 4 fronds and 1 plant with 3 fronds, total number of fronds at test initiation: 11 per replicate; assessment of growth and other effects on days 3, 5 and 7.

Endpoints: EC_{50} with respect to growth rate and yield after exposure over 7 days.

Test concentrations: Control (dilution water) and geometric mean measured test item concentration of 1.39 µg alpha-cypermethrin/L (reflects the maximum solubility of the test item under test conditions).

Test conditions: 400 mL glass flasks, test volume: 160 mL, 20 x-AAP medium, pH 7.50 - 7.51 at test initiation and pH 8.56 - 8.63 at test termination; mean water temperature: 24.3 °C, continuous light, light intensity: about 8200 lux.

Analytics: Analytical verification of the test item was conducted using a GC-method with MS-detection.

Statistics: Descriptive statistics, standard procedures.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of the test item concentration was conducted in the control and the test item treatment at the beginning and at the end of the test. The mean analyzed content of alpha-cypermethrin in the test item treatment was 2.45 µg/L at test initiation and 0.788 µg/L at test termination, resulting in a geometric mean measured concentration of 1.39 µg/L. The following biological results are based on the geometric mean measured concentration.

Biological results: The duckweed population in the control vessels showed exponential growth, increasing from 11 fronds per vessel to an average of 142.7 fronds per vessel in the control after 7 days, corresponding to a 13.0 x multiplication. The dry weight increased from 2.0 mg to an average of 21.2 mg per vessel in the control at test termination. No morphological effects on the duckweed were observed in the control and at the test item concentration of 1.39 µg a.s./L. There were no statistically significant effects on the growth of *Lemna gibba* compared to the control at the tested concentration of 1.39 µg a.s./L. Effects on growth rate and yield are summarized in Table 8.2.7-1.

Table 8.2.7-1: Effects of alpha-cypermethrin on the growth of duckweed *Lemna gibba*

Concentration [µg a.s./L] (geometric mean measured)	**Control**	**1.39**
Inhibition after 7 d [%] (growth rate based on frond no.)	--	0.6
Inhibition after 7 d [%] (growth rate based on dry weight)	--	0.6
Inhibition after 7 d [%] (yield based on frond no.)	--	1.9
Inhibition after 7 d [%] (yield based on dry weight)	--	1.6
	Endpoints [µg a.s./L] (geometric mean measured)	
E_rC_{50} / E_yC_{50} (7 d) based on frond no. and dry weight	> 1.39	

III. CONCLUSION

In a 7-day aquatic-plant limit test with *Lemna gibba,* both the E_rC_{50} and the E_yC_{50} values of alpha-cypermethrin based on frond number and dry weight were determined to be > 1.39 µg a.s./L, based on geometric mean measured concentrations.

CA 8.2.8 Further testing on aquatic organisms

The following outdoor mesocosm study performed with the solo-formulation BAS 310 55 I (containing 50.0 g alpha-cypermethrin/L, nominally) is provided in support of the aquatic risk assessment and has not been evaluated previously on EU level.

Report:	CA 8.2.8/1 Stegger P.,Janz P., 2015a Outdoor aquatic mesocosm study with Alpha-Cypermethrin applied as solo-formulation BAS 310 55 I 2014/1102015
Guidelines:	SANCO/3268/2001 rev. 4 (final) 17 Oct 2002
GLP:	yes (certified by Hessisches Ministerium fuer Umwelt, Laendlichen Raum und Verbraucherschutz)

Report:	CA 8.2.8/2 Class T., 2014a Outdoor aquatic mesocosm study with ME solo-formulation of Alpha-Cypermethrin (BAS 310 55 I, nominal 50 g/L) - Preparation and verification of dose solutions, analysis of water and sediment samples 2014/1246534
Guidelines:	SANCO/3029/99 rev. 4 (11 July 2000)
GLP:	yes (certified by Landesamt fuer Umwelt, Messungen und Naturschutz Baden Wuerttemberg, Karlsruhe, Germany)

Executive Summary

In a 12-weeks outdoor mesocosm study, the acute and chronic effects of the active substance alpha-cypermethrin (BAS 310 I; applied as solo-formulation BAS 310 55 I) on functional and structural parameters of aquatic ecosystems were investigated under field conditions in natural pond water. Mesocosms consisted of eighteen stainless steel enclosures each containing approx. 1600 L of water with a sediment layer. The test item was applied directly into the water column at nominal initial water concentrations of 0.12, 0.6, 3.0, 15, 30 and 90 ng a.s./L (corresponding to measured peak concentrations of 0.21, 0.87, 4.03, 17.0, 38.5 and 138 ng a.s./L) with two replicate enclosures per concentration. Four untreated enclosures were used as controls. All test item concentrations were applied twice (on May 09, 2012 and May 17, 2012), except for the 30 ng a.s./L treatment which was applied only once. The second application of 15 ng a.s./L was time-shifted (on May 25, 2012).. Post-application samplings and analysis of biota and physico-chemical parameters were conducted at weekly or bi-weekly intervals until 12 weeks after treatment. The effects monitored at the ecosystem level included functional parameters measured via physical-chemical water parameters and structural parameters such as diversity of species and effects on and recovery time of the communities of macrozoobenthos, emerging insects, zooplankton, phytoplankton and macrophytes..

The effects monitored at the population level included abundance of individual populations of phytoplankton, zooplankton and macrozoobenthos and recovery time of sensitive organisms.

Analyses of alpha-cypermethrin in water samples collected 1 to 3 h after application showed that peak concentrations were between 113% and 173% of the nominal alpha-cypermethrin concentrations. Therefore, the biological results are based on mean measured peak concentrations. No effects on any of the monitored population or community were observed at the three lowest test item concentrations 0.21, 0.87 and 4.03 ng a.s./L (class 1). Pronounced effects with recovery within 8 weeks after the first application (class 3A) were found at 17.0 ng a.s./L for the community structure and the total abundance of macroinvertebrates, for *Chaoborus* sp. as well as for some zooplankton populations (namely copepodites of Diaptomidae) and at 138 ng a.s./L for populations of emerging insects (based on Chaoboridae sp.). At 38.5 ng a.s./L (single application) the observed effects were very similar to the effects found after a double application of 17.0 ng a.s./L, except that no consistent effects on the total abundance of macroinvertebrates were observed but slight effects on the zooplankton community (class 2). After twofold application at 17.0 ng a.s./L and single application at 38.5 ng a.s./L, no effects on emerging insects, phytoplankton or macrophytes were observed. Pronounced effects without recovery within the test period (class 5B) were found at 138 ng a.s./L for the community structure and the populations of macroinvertebrates (based on total abundance of *Asellus aquaticus* and *Baetidae* sp.) and zooplankton (based on the copepodites of Diaptomidae). Furthermore, at 138 ng a.s./L, slight and short term increases in abundances of phytoplankton populations (based on *Cryptomonas erosa et ovata* and *Navicula pupula*) were observed (class 2+) whereas no effects on the community of emerging insects, on the community of phytoplankton or on macrophytes were observed (class 1).

The overall NOEC of this aquatic outdoor mesocosm study is 4.03 ng a.s./L (based on mean-measured peak concentration) as no consistent effects on any of the monitored endpoints were observed up to this concentration. Two applications of 17.0 ng a.s./L and one application of 38.5 ng a.s./L are defined as the study specific effect class 3A NOEAEC, since no long lasting effects were observed at these treatment levels.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 55 I, batch no. 101198; content of a.s.: alpha-cypermethrin (BAS 310 I, Reg. No. 4078193): 51.1 g/L (nominal: 50 g/L); density: 0.994 g/cm^3.

B. STUDY DESIGN

Test system: The mesocosm site is located on the grounds of 'Institut für Gewässerschutz' MESOCOSM GmbH, Homberg (Ohm), Germany. Outdoor mesocosm study was conducted in stainless-steel enclosures each with a diameter of approximately 143 cm (surface approx. 1.60 m^2) and a depth of approximately 150 cm. The nominal depth of the water body was 100 cm ± 10%, resulting in a total volume of approximately 1600 L. 18 stainless steel enclosures were pressed into the sediment of an artificial pond (diameter of approximately 7.7 m, water volume approx. 47,000 L) 49 days before the first test item application. 15 enclosures were selected for the use in the study. Because of an amendment two further enclosures were reintroduced in the study after the first application. The artificial pond was filled with sediment and water collected from a reference lake on-site. The pond sediment was placed in the artificial pond (10 - 15 cm) onto a layer of clay (5 - 10 cm). The organic matter content of the mixed sediment was 1.9%. Washed sand was added to achieve the optimum organic carbon content. The test system was established in March 2009, so the period for equilibration of the test system was about 38 months.
The test systems contained various naturally growing macrophytes (*Ceratophyllum demersum*, *Potamogeton natans*, *Chara intermedia*, *Zannichellia palustris*) of comparable density and composition per enclosure at the start of the exposure.

Test design: Static test system. The test item was applied twice to the outdoor mesocosm community with two replicate enclosures per concentration. Due to a calculation error of the analytical laboratory for preparation of the application solutions of the higher concentrations the application of 15 ng a.s./L was time-shifted and 30 ng a.s./L was only applied once. Four untreated enclosures were used as controls The test item was applied directly into the water column on May 09, 2012 and May 17, 2012. The second application for the treatment receiving 15 ng a.s./L was on May 25, 2012. The in-life phase was terminated 12 weeks after the first application. Water and sediment samples were taken from enclosures after applications at regular intervals for residue analysis. All enclosures were monitored for water parameters (temperature, oxygen concentration, pH, conductivity and nutrients), macrophytes, phytoplankton, zooplankton, emerging insects and macrozoobenthos at weekly or bi-weekly intervals. The biological samples were collected from the enclosures at regular intervals over the course of the study by depth-integrated sampling (zooplankton, phytoplankton), under water traps and netting (macroinvertebrates), emergence traps (emerging insects).

Endpoints:	NOEC and NOEAEC plus corresponding 'effect classes'; based on functional parameters (physical-chemical water parameters such as conductivity, pH, dissolved oxygen, DOC, nitrate, phosphate and ammonium), structural parameters (diversity of species and effects on and recovery time of the communities of macrozoobenthos, emerging insects, zooplankton, phytoplankton and macrophytes) and population effects (abundance of individual populations of macrozoobenthos, zooplankton and phytoplankton, recovery time of sensitive organisms).
Test concentrations:	0 (water control), 2.33, 11.7, 58.4, 291.8, 583.6 and 1750.6 ng BAS 310 55 I/L (nominal); equivalent to 0.12, 0.6, 3.0, 15, 30 and 90 ng alpha-cypermethrin/L (corresponding to measured peak concentrations of 0.21, 0.87, 4.03, 17.0, 38.5 and 138 ng a.s./L). Second application of 15 ng a.s./L (nominal) was time-shifted and the 30 ng a.s./L treatment (nominal) was applied only once.
Test conditions:	The test was performed under outdoor conditions. Climatic conditions and water parameters were recorded throughout the conduct of the study.
Analytics:	Test water and sediment samples collected at various time points from the mesocosm enclosures were analyzed for alpha-cypermethrin concentrations using GC/MS(NCI) after liquid/liquid partition (water) and extraction/partition (sediment).
Statistics:	Descriptive statistics; multiple comparison test by Dunnett ($\alpha = 0.05$) to determine the NOEC values; diversity analysis by calculation of the Shannon-Index and Evenness; similarity analysis using Steinhaus´ and Stander´s index; calculation of Principal Response Curves (PRC) as a multivariate approach to analyze and visualize effects on the community level. The program Community Analysis V4.3 (CA) was used for the calculations of effects (NOECs, diversity indices). The PRC analysis and the Redundancy Analysis (RDA) and Principal Component Analysis (PCA) were performed using CANOCO 4.5 (Biometrics Plant Research International, Wageningen, The Netherlands). Observed effects were classified according to the scheme suggested in the EU Guidance Document on Aquatic Ecotoxicology (SANCO 2002) and the refinement suggested by de Jong et al. (2008) in order to facilitate the estimation of the study specific NOEC and NOEAEC.

II. RESULTS AND DISCUSSION

Analytical measurements:

Analyses of alpha-cypermethrin in water samples collected 1 to 3 hours after application showed that peak concentrations were between 113% and 173% of the nominal alpha-cypermethrin concentrations. Therefore, the biological results are based on the mean measured peak concentrations. Alpha-cypermethrin dissipated fast from the water phase with a DT_{50} of ca. 24 h. The concentration of alpha-cypermethrin in sediment samples of control and treatments was below the LoQ (0.1 µg a.s./kg wet weight), indicating the decline of alpha-cypermethrin in the water phase is probably due to hydrolytic degradation and not to adsorption and sedimentation.

Biological results:

Functional parameters

All physical and chemical parameters (*e.g.* pH 8.71 - 10.05; conductivity: 183 - 255 µS/cm at 50 cm below water surface) were within the range of mesotrophic natural pond water during the study. Water temperatures ranged from around 11.5 °C (April) to around 21 °C (July) during the study. No treatment related effects on pH, oxygen levels or conductivity were observed (Dunnett`s test, $\alpha = 0.05$). Furthermore, there were no apparent treatment related effects in the measured nutrient parameters like water hardness (usually 2.6 - 5.7°dH with a trend to decrease to values around 3°dH at the end of the study), nitrate (always < 0.4 mg/L), and ammonium (always < 0.1 mg/L). The total phosphate concentration of the water was usually around or below 0.2 mg/L with a general trend to increase at the end of the study. The maximum concentration found was 1.0 mg/L.

Structural parameters

Macroinvertebrates

In total, 34 taxa respectively stages were identified in the 176 macroinvertebrates samples. The macroinvertebrates were dominated by the isopod *Asellus aquaticus*, mayflies (Ephemeroptera, mostly Baetidae), the phantom midge *Chaoborus* and flatworms of the order Tricladida.

No adverse direct or indirect effects of the test item were found either on the community or on the population level of the macroinvertebrates at the three lowest treatment levels of 0.21, 0.87 and 4.03 ng a.s./L (class 1). Pronounced effects on the community structure, the total abundance of macroinvertebrates and the *Chaoborus* population were demonstrated at 17.0 ng a.s./L. Since these endpoints recovered within 8 weeks after the first application, the effects at 17.0 ng a.s./L were classified as class 3A. At 138 ng a.s./L no full recovery regarding the community structure, the total abundance and the populations of *Asellus aquaticus* and Baetidae / Sum Ephemeroptera took place during the study. Thus, the effects at the highest concentration are considered to be class 5B effects.

After a single application of 38.5 ng a.s./L effects of class 3A (community structure and *Chaoborus* sp.) were observed.

Emerging insects
In total, 16 taxa of eight different orders were found in the 223 samples of the emerging insects. Since no treatment related effects on the community and the population level of emerging insects were found at 0.21, 0.87, 4.03, 17.0 (twofold application) and at 38.5 ng a.s./L (single application), effect class 1 can be assigned to all of these treatment levels.
At 138 ng a.s./L no effects were observed on community structure and total abundance. For the population of Chaoboridae a pronounced decrease with recovery within 8 weeks after the first application was found at 138 ng a.s./L, resulting in a classification to effect class 3A.

Zooplankton
Species of three classes (Insecta, Crustacea and Rotatoria) were present in the 272 zooplankton samples. 13 different taxa respectively life stages (nauplia, copepodites and adults of copepods) of crustaceans were identified and 17 different taxa of rotifers.
No direct or indirect effects on zooplankton community or at the population level were observed up to a concentration of 4.03 ng a.s./L (class 1). At 17.0 ng a.s./L no effects on the community, but slight (*Chydorus sphaericus*) respectively pronounced effects with recovery within 8 weeks after the first application (Diaptomidae copepodites) on the zooplankton populations were found. Based on the effects on Diaptomidae copepodites these effects are classified as class 3A.
At 138 ng a.s./L no effects on the number of taxa or the Shannon index, but short term effects on total abundance and diversity (Evenness) were observed (class 2). Pronounced long term effects on the community structure were indicated by similarity and ordination analyses (PCA). As full recovery was not proven for two consecutive sampling dates, these effects are considered as class 5B effects. In addition, the effects on the zooplankton populations were also rated as class 5B effects based on Diaptomidae copepodite.
At 38.5 ng a.s./L (single application) slight temporary effects on the zooplankton community (Stander`s index) were observed (class 2), but pronounced short term effects on Diaptomidae were demonstrated. Thus, the effects on the zooplankton populations are assigned as class 3A.

Phytoplankton
The analysis with inverted microscopy of the 160 phytoplankton samples resulted in 140 taxa belonging to nine different classes.
Regarding the phytoplankton community and populations no direct or indirect effects were observed after twofold application at up to 17.0 ng a.s./L and after single application at 38.5 ng a.s./L (class 1).
At the highest treatment level (138 ng a.s./L) no effects on the community structure or total abundance were observed (class 1), but slight short term, positive, effects on the populations of *Cryptomonas erosa + ovata* and *Navicula pupula* occurred on two consecutive samplings days at 138 ng a.s./L, resulting in a class 2 effect.

Macrophytes
The following macrophyte species were found: *Ceratophyllum demersum*, *Potamogeton natans* and *Zannichellia palustris*. The algae *Chara intermedia* and filamentous algae were also considered within the functional group of macrophytes. The percentage of coverage of all macrophytes and of each species was comparable at all treatment levels. No unusual observations were made.

A summary of the effect classes for the different types of endpoints and the concentrations tested is provided in Table 8.2.8-1 below. The classes on the taxon level are based on the most sensitive taxon found in the community.

Table 8.2.8-1: Summary of effect classes # in the outdoor mesocosm study with alpha-cypermethrin

Endpoint	Mean-measured peak concentration [ng a.s./L] *					
	0.21	0.87	4.03	17.0	138	38.5
Macroinvertebrates						
Community structure	1	1	1	3A	5B	3A
Abundance of populations	1	1	1	3A	5B	3A
Emerging insects						
Community structure	1	1	1	1	1	1
Abundance of populations	1	1	1	1	3A	1
Zooplankton						
Community structure	1	1	1	1	5B	2
Abundance of populations	1	1	1	3A	5B	3A
Phytoplankton						
Community structure	1	1	1	1	1	1
Abundance of populations	1	1	1	1	2+	1
Macrophytes						
Coverage	1	1	1	1	1	1
Study			NOEC	NOEAEC		NOEAEC

* Note: 0.22, 0.87, 4.03, 17.0, and 138 ng a.s./L were applied twice, 38.5 ng a.s./l was applied once.

Classes according to the Guidance Document on Aquatic Ecotoxicology (SANCO 2002) and de Jong et al. (2008): 1 = effects could not be demonstrated; 2 = slight and/or temporary effects on single sampling dates; 3A = pronounced short-term effect with recovery within 8 weeks after first application or total period of effects < 8 weeks; 5B = pronounced effects without full recovery within the study. A '+' indicates an increase of abundance.

III. CONCLUSION

The overall NOEC of this aquatic outdoor mesocosm study is 4.03 ng a.s./L (based on mean-measured peak concentration) as no consistent effects on any of the monitored endpoints were observed up to this concentration. Two applications of 17.0 ng a.s./L or one application of 38.5 ng a.s./L are defined as the study specific effect class 3A NOEAEC, since no long lasting effects were observed at these treatment levels.

The following summary of a Review of available mesocosm studies conducted with alpha-cypermethrin is provided in support of the aquatic risk assessment and has not been evaluated previously on EU level.

Report: CA 8.2.8/3
Janz P., 2015a
A critical review of available mesocosm studies with Alpha-Cypermethrin
2014/1102014
Guidelines: none
GLP: no

Executive Summary

In the period 1997 to 2012, nine aquatic field studies (*i.e.* mesocosms) applying different exposure regimes were conducted at three different sites across Germany. In this review each of these studies was evaluated using a unified set of criteria. This chapter brings together all results in a weight of evidence approach, considering the consistency in results and reliability of the studies.
The results and final conclusions of the evaluated mesocosm studies appeared highly consistent (see Table 8.2.8-2). The effects after one, two and three applications of alpha-cypermethrin are summarized in the following and appropriate endpoints are proposed.

One application
In studies dealing with effects after a single application of alpha-cypermethrin no effects (class 1) on any of the monitored endpoints were found up to a concentration of 3 ng a.s./L. In the single application study with a study specific NOEC of 0.6 ng a.s./L (Dawo 2005a), 3 ng a.s./L was not applied and 15 ng a.s./L was the next higher treatment level. **Thus, 3 ng a.s./L is considered as a fully supportable overall $NOEC_{mesocosm}$ for alpha-cypermethrin after a single application.** Further, this value needs to be compared with (and adjusted to) the two application NOEC, as the latter cannot be higher.

Huber et al. 2000a, Huber et al. 2000b, Grünwald 2003, Dawo 2005a and Stegger and Janz 2014 showed that no direct long-lasting effects (class 3A) occurred after a single application of 12, 15, 15, 15 or 38.5 ng a.s./L. Thus, in consideration of the concentration in the next higher treatment level (*i.e.* 60 or 75 ng a.s./L) and the reliability of 1 for the Stegger and Janz (2014) study, **38.5 ng a.s./L is regarded as an overall and consistent $NOEAEC_{mesocosm}$ for alpha-cypermethrin after a single application.**

At the treatment level of 12, 15 or 38.5 ng a.s./L the effects on macroinvertebrates in the studies were comparable. In each study the phantom midge *Chaoborus* was the most sensitive organism. The effects on the *Chaoborus* populations as well as the impacts on the macroinvertebrate communities were rated as class 3A effects in each study. The effects on zooplankton at these concentrations differed slightly between the studies. Class 3A effects on the zooplankton community were observed in two studies (Huber et al. 2000b and Dawo 2005a). Class 2 effects on the zooplankton community were found in three studies (Huber et al. 2000a, Grünwald 2003, Stegger and Janz 2014). Changes in the zooplankton community structures were mainly indirect effects and due to lower abundance of *Chaoborus*, resulting in reduced predation pressure. The most sensitive group of organisms belonging to the zooplankton community was Copepoda. Indirect long lasting effects (class 5A+) were revealed at 15 ng a.s./L in Dawo 2005a. This isolated observation was not considered to disagree with the above mentioned overall $NOEAEC_{mesocosm}$ since the effect was indirect, not ecologically relevant (had no influence on any other endpoint) and the effect duration could not be confirmed in any of the other evaluated studies (even after multiple applications of alpha-cypermethrin they did not last longer than 8 weeks, Huber et al. 2001 and Dawo 2005b). No effects on copepods at 15 ng a.s./L were found in Huber et al. (2000a and 2000b) and Grünwald (2003; note, the classification of effects on zooplankton populations into class 3A was based on *Chaoborus* in this study). Class 3A effects on copepods were also described by Stegger and Janz (2014). The impacts on phytoplankton, the food source of zooplankton, were comparable in all studies. Effects on the phytoplankton community were shown in none of the studies at 12, 15 or 38.5 ng a.s./L. However, at these concentrations short term effects (class 2) on phytoplankton populations were found in Huber et al. (2000b), Dawo (2005a) and Stegger and Janz (2014), while no effect on phytoplankton populations were revealed in Grünwald (2003). Huber et al. (2000a) did not analyse phytoplankton at the population level.

Class 5 effects were always measured at the next higher tested treatment level above 12, 15 and 38.5 ng a.s./L, respectively. These concentrations were 60 (Huber et al. 2000a), 75 (Huber et al. 2000b, Grünwald 2003 and Dawo 2005) and two applications of 138 ng a.s./L (Stegger and Janz 2014).

Two applications

No effects (class 1) on any of the monitored endpoints were demonstrated after a double application of alpha-cypermethrin up to a concentration of 4.03 ng a.s./L in Stegger and Janz (2014). A reliability index of 1 was assigned to this study. No $NOEC_{mesocosm}$ was found in the study of Sevim 2012, in which only one concentration (20 ng a.s./L) was tested. Jordan (2012) and Sippenhauer (2012) determined a $NOEC_{mesocosm}$ of 0.6 ng a.s./L. However, the findings of this study are not reliable (Ri = 3, low acceptability). Therefore, the derivation of an overall $NOEC_{mesocosm}$ is based on the $NOEC_{mesocosm}$ specified in the reliable study by Stegger and Janz (2014). Thus, **4.03 ng a.s./L is considered as a fully supportable overall $NOEC_{mesocosm}$ for alpha-cypermethrin after two applications with an interval of approximately one week**.
This value would then also be applicable for a single application.

In the reliable study conducted by Stegger and Janz (2014) the study specific $NOEAEC_{mesocosm}$ was determined as 17 ng a.s./L. The effects after a double application of the nominal concentration of 20 ng a.s./L described by Sevim 2012 were also rated as class 3A effects. However, as the effects in Sevim (2012) cannot be related to measured concentrations and consequently the reliability of this study is rated as 3 (not reliable), the test concentration of 20 ng a.s./L is not considered as a fully supportable $NOEAEC_{mesocosm}$. The same applies for the $NOEAEC_{mesocosm}$ of 135 ng a.s./L as derived from Jordan (2012) and Sippenhauer (2012). Consequently, **17 ng a.s./L is regarded as a fully supportable overall $NOEAEC_{mesocosm}$ for alpha-cypermethrin after two applications with an interval of approximately one week.**

The effects described by Stegger and Janz (2014) after two applications of 17 ng a.s./L with a one week interval were very similar to those found after one application of 12, 15 and 38.5 ng a.s./L. Class 3A effects were observed for the macroinvertebrate community, for *Chaoborus* as well as for zooplankton populations, namely copepodites. In contrast to the results of the single application studies, at 17 ng a.s./L no effects on the zooplankton community were described by Stegger and Janz (2014). *Chaoborus*, the species which caused the indirect effects on the zooplankton community, was by far less dominant (15.5% of macroinvertebrates) in the test system used by Stegger and Janz (2014) compared to the ponds in the studies performed at TU Munich (e.g. 68% of macroinvertebrates in Dawo 2005a). As a consequence, the influence of *Chaoborus* on the zooplankton community was clearly much weaker in Stegger and Janz (2014). This is probably the reason why no indirect effects on the zooplankton community were shown at 17 ng a.s./L in Stegger and Janz (2014). No effects on emerging insects, phytoplankton or macrophytes were found by Stegger and Janz (2014) at 17 ng a.s./L. In the non reliable study performed by Sevim (2012) which focused on the population dynamics of *Chaoborus*, class 3A effects were observed after two applications of 20 ng a.s./L at an interval of two weeks. In the non-reliable study of Jordan (2012) and Sippenhauer (2012) the effects were classified as 3A up to and including a concentration of 135 ng a.s./L.
In Stegger and Janz (2014) class 5 effects on macroinvertebrates and zooplankton were measured at the next higher treatment level (138 ng a.s./L).

Three applications

Both studies with a triple exposure regime (Huber et al. 2001 and Dawo 2005b) were scientifically not reliable (Ri 3) due to lacking analytical verification of nominal concentrations. Therefore, the results of these studies should be interpreted with care and a fully supportable overall $NOEC_{mesocosm}$ and $NOEAEC_{mesocsm}$ could not be defined with reasonable certainty for three applications. However, the initial effects during the first days after application in both studies were perfectly in line with the other mesocosm studies (Huber et al. 2000a and 2000b, Grünwald 2003 and Stegger and Janz 2014), providing some evidence that the target concentrations were achieved. Huber et al. 2001 (three applications at an interval of two weeks) as well as Dawo 2005b (three applications at an interval of three weeks) determined 0.6 ng a.s./L to be the study specific NOEC. Since 3 ng a.s./L was not applied in these studies, it remains unclear, if this concentration is also a valid NOEC after triple application of alpha-cypermethrin.

In both studies pronounced effects with recovery within 8 weeks after the last application (class 3B) were detected for *Chaoborus* and the macroinvertebrate communities at 15 ng a.s./L. (changes in the macroinvertebrate community were mainly due to the effects on *Chaoborus*). The effects on *Chaoborus* and the macroinvertebrate community changed from class 3A (recovery within 8 weeks after the first application) to class 3B (recovery within 8 weeks after the last application), when 15 ng a.s./L of alpha-cypermethrin was applied three times instead of one or two times with a one week interval. As a consequence, *Chaoborus* populations and the macroinvertebrate communities should have recovered within four weeks and two weeks after the last application, respectively, for these effects to be classified into class 3A. Obviously, this time frame is too short for complete recovery of these endpoints after exposure to 15 ng a.s./L.

As a result of the longer lasting effects on *Chaoborus*, the indirect effects on zooplankton were stronger and also longer lasting after three applications compared to one or two applications. The effects on the zooplankton community in Huber et al. (2001) were classified as 3A and in Dawo (2005b) as 3B. Furthermore, the indirect effects on zooplankton populations were rated as class 3B based on planktonic arthropods and copepods in Huber et al. (2001) and on copepods in Dawo (2005b), respectively. Effects on the phytoplankton community were not found in these studies. Dawo (2005b) described slight effects on phytoplankton populations.

Table 8.2.8-2: Effect class concentrations [ng a.s./L] of the most sensitive endpoints in aquatic outdoor mesocosm studies with alpha-cypermethrin.

The effect classes are expressed in terms of nominal concentrations and measured mean peak concentrations in Stegger and Janz (2014). Ri = Reliability index (determined according to de Jong et al. 2008, 1 = reliable, 2 = less reliable, 3 = not reliable)

Exposure regime	Effect class 1	Effect class 2	Effect class 3A	Effect class 3B	Effect class 5	Reference	Ri
Single	3	-	12		60	Huber et al. 2000a	2
Single	3	-	15		75	Huber et al. 2000b	1
Single			15		75	Grünwald 2003	2
Single	0.6	-	15*		75	Dawo 2005a	3
Single			38.5			Stegger and Janz 2014	1
Double (two week interval)			20			Sevim 2012	3
Double (one week interval)	4.03	-	17	-	138	Stegger and Janz 2014	1
Double (one week interval)	0.6				135	Jordan (2012), Sippenhauer (2012)	3#
Triple (at intervals of two weeks)	0.6	-	-	15	375	Huber et al. 2001	3
Triple (at intervals of three weeks)	0.6	-	-	15	75	Dawo 2005b	3

* except an indirect effect on Copepoda: class 5A+
# low acceptability

The phantom midge *Chaoborus*

The phantom midge *Chaoborus* was the most sensitive organism in all evaluated mesocosm studies. Therefore, the effects on this genus are discussed in detail in this section.

As mentioned above, up to a concentration of 4.03 ng a.s./L no effects on *Chaoborus* populations were observed in the evaluated mesocosm studies. Pronounced effects on *Chaoborus* at 12, 15, 20 or 38.5 ng a.s/L were described in each evaluated mesocosm study. Considering only the period after the last application, it appears, that it makes no difference for the effect duration, how often alpha-cypermethrin was applied in the weeks before the last application (if alpha-cypermethrin is applied at an approximately one week interval). In the single application studies the effects on *Chaoborus* lasted four weeks (Huber et al. 2000b and Grünwald 2003), five weeks (Huber 2000a and Dawo 2005a) and six weeks (Stegger and Janz 2014) after an application of 12, 15 or 38.5 ng a.s./L, respectively. In the two applications study the effects on *Chaoborus* lasted five weeks after the second application of 17 ng a.s./L (Stegger and Janz 2014). The less reliable studies conducted by Sevim (2012) and Jordan (2012) and Sippenhauer (2012) confirm this observation. Three applications of 15 ng a.s./L caused effects on *Chaoborus*, which lasted four weeks (Dawo 2005) and five weeks (Huber et al. 2001) after the third application. The time needed for recovery therefore complies with approximately one generation time of *Chaoborus* (six to seven weeks, Sevim 2012). The finding that the time needed for recovery seems to be independent from the number of previous applications with a one week interval, can be explained by the short dissipation time of alpha-cypermethrin in water (Huber et al. 2000b and Stegger and Janz 2014, respectively). Assuming first order kinetics for dissipation and using the DT_{50} of 1 d (Stegger and Janz, 2014, Huber et al. 2000b), the concentration of alpha-cypermethrin after an application of 15 ng a.s./L would be below the NOEC for *Chaoborus* (3 ng a.s./L) after 2.5 d. Since the concentration of alpha-cypermethrin is expected to be negligible one week after the first application of 15 ng a.s./L, there is no build up of alpha-cypermethrin after the second application of 15 ng a.s./L. Therefore, the peaks are toxicologically independent. Assuming a recovery time of six weeks, *Chaoborus* would probably recover within eight weeks after the first application (effect class 3A), if 15 ng a.s./L was applied three times at intervals of one week. Thus, 15 ng a.s./L is likely to be also an appropriate $NOEAEC_{mesocosm}$ for three applications with an interval of approximately one week.

In discussing the effects on *Chaoborus* populations, it is important to consider all factors affecting recovery. The recovery process of *Chaoborus* populations is influenced by the following aspects (based on Sevim 2012, Janz 2014; BASF DocID 2014/1102016) and findings in the review report):

- magnitude of the effect (to what extent is the population impacted)
- dissipation time of the substance in water (e.g. degradation and adsorption) (alpha-cypermethrin: DT_{50} ~ 1 d)
- generation time (depends on temperature, light period and food availability) (usually five to eight weeks during the growing season in Germany)
- number of generations per season (usually three generations in Germany)
- immigration / emigration of flying imagines (availability of unexposed waters)
- oviposition success
- population density (reduced density results in lower mortality due to less cannibalism)
- food availability (presence of prey)

Most of the above listed factors are depending on the season. Therefore the season, in which the test item is applied, should be taken into account. To ensure the possibility of recovery by recolonization, alpha-cypermethrin should be applied early enough in the year. During spring and early summer the conditions are best for recovery. Later in the year the number of flying imagines is usually too low for a complete recovery.

The immigration rate of flying imagines is enhanced by the availability of unexposed waters in the vicinity of the treated area. In each of the reliable evaluated mesocosm studies (except in Sevim 2012) unexposed waters (i.e. the control ponds) were available. However, selection of the water body for oviposition is primarily a function of fish abundance and distance from the site of emergence (Berendonk, 1999; Berendonk and Bonsall, 2002). In the cited papers it has been shown that female imagines of *C. crystallinus* disperse over great distances to find fish-free waterbodies to oviposit and colonize these. Therefore, population recovery in water bodies receiving drift entry of alpha-cypermethrin would also be expected when sources of *C. crystallinus* (such as unexposed waters) are not in the direct vicinity. This is supported by the results of Sevim (2012) which indicated that recovery is also possible in isolated ponds.

The following summary of a modelling study for phantom midge *Chaoborus* sp. population after treatment with alpha-cypermethrin is provided in support of the aquatic risk assessment and has not been evaluated previously on EU level.

Report: CA 8.2.8/4
Strauss T., 2015a
An individual based population model for the phantom midge Chaoborus crystallinus to simulate effects and recovery under Alpha-Cypermethrin (BAS 310 I) exposure
2014/1162681
Guidelines: none
GLP: no

Executive Summary

In this study, the FOCUS R3 stream scenario (late application date) with a twofold application of 10 g alpha-cypermethrin per ha (*i.e.* 200 mL BAS 310 55 I/ha) to leafy vegetables has been selected for modeling. Potential impacts on the population of the phantom midge *Chaoborus crystallinus* under realistic outdoor field conditions were assessed using a combination of different mitigation measures (i.e. buffer zones and drift reducing nozzles).

For this purpose, an individual based population model for *Chaoborus crystallinus* is used. The ecological model has been previously parameterized and intensively tested using large datasets from the laboratory as well as outdoor micro- and mesocosms. The complete life-cycle for *Chaoborus* populations was simulated depending on food conditions, water temperature and photoperiod. The model integrates density-dependent processes as well as immigration and emigration rates of the adults.

Further, a toxicokinetic/toxicodynamic (TKTD) effect model (GUTS approach) was used for the simulation of uptake and elimination rates of alpha-cypermethrin and the prediction of mortality from time-variable exposure, which was successfully coupled with the *Chaoborus* population model. With this modelling approach it is possible to extrapolate effect data from laboratory to field conditions such as outdoor studies (e.g. mesocosm and microcosm studies) without further model calibrations.

The model was tested against control data from several outdoor mesocosm studies first. The biological part of the IBM *Chaoborus* model proved suitable to reproduce the case specific as well as more principle population patterns as found in the mesocosm field studies.
Secondly, the model was tested against three alpha-cypermethrin mesocosm studies. The combination of the *Chaoborus* model and the GUTS model allowed a precise prediction of the impact of alpha-cypermethrin on *Chaoborus crystallinus* populations.
This was demonstrated for a large number of alpha-cypermethrin concentrations and different application patterns (single application in study AL-560-023, two applications in study 2014/1102015, and three applications in study AL-560-056), covering a wide concentration range from 0.06 ng/L to 1.875 µg/L.
The results showed a good match between simulations and measurements for the larval abundances as well as the emergence rate of the adults. This applies also to the timing and magnitude of the toxic effects, and the subsequent degree of recovery.
Finally, simulations with several FOCUS exposure scenarios were conducted to assess the effect size and the duration of the effects under semi-field conditions for *Chaoborus crystallinus*. The ecological scenario was defined as a mesocosm facility with treated and untreated (control) populations, as was used for the model testing previously.
Both, exposure concentrations and water temperatures were derived from the FOCUS R3 scenario.

In summary, the FOCUS R3 step 3 scenario (maximum peak height: 0.042 µg/L) caused stronger effects of longer duration than the FOCUS R3 step 4 scenarios that included mitigation measures (maximum peak heights ranging from 0.0014 to 0.0146 µg/L). In all cases effects lasted shorter than 8 weeks after the exposure started.

The following investigation of phantom midge population development in mesocosm studies is provided in support of the aquatic risk assessment and has not been evaluated previously on EU level.

Report: CA 8.2.8/5
Janz P., 2014a
Phantom midge (Chaoborus spp.) population development in mesocosm studies
2014/1102016
Guidelines: none
GLP: no

Executive Summary

Aquatic mesocosms (model ecosystems) are frequently used to refine aquatic risk assessments when a potential risk is indicated by lower tier studies (e.g. laboratory single species tests). One of the species typically occurring in mesocosms is the phantom midge *Chaoborus* spp.. As top predators in fish-free mesocosm studies chaoborids play a key role in limnetic food webs. Besides they are sensitive to insecticides, especially pyrethroids. For a correct evaluation of mesocosm data a profound knowledge of the biology of the organisms of interest is essential.

Based on a large data pool (data from 19 mesocosm studies, all of which were conducted in the same test system over a period of 15 years) this study provides information on *Chaoborus* population dynamics, their life cycle, the natural variability of *Chaoborus* populations and their influence on the zooplankton community structure. The data of untreated enclosures from 19 mesocosm studies was used to examine the population dynamics of *Chaoborus* and the inter-replicate and temporal variability of *Chaoborus* populations. For the analysis of the influence of *Chaoborus* on the zooplankton community structure, the enclosures in which direct effects exclusively on *Chaoborus* occurred (and consequently potential indirect effects on prey species and competing species) were included additionally.

The composition of the *Chaoborus* populations in the untreated enclosures analyzed in the present study varied significantly between the years. The variation in the *Chaoborus* population structure between the seasons (i.e. within a year) was even higher than between years. It was shown that in the studied test system there usually were three *Chaoborus* generations per year (from the end of April to the beginning of August), each lasting four to six weeks. Calculations of Minimum Detectable Differences (MDDs) showed that the inter-replicate variability was sufficient low in order to reveal population relevant effects of chemicals on *Chaoborus* in mesocosm studies that follow common principles of design and evaluation. Ordination analysis revealed that *Chaoborus* larvae seem to promote high densities of rotifers and reduce the abundance of small crustaceans (nauplii larvae, copepods and small cladocerans).

In the meantime, this report has been published in the peer-reviewed literature as: Janz P., Weltje L., Ebke K.P., Dawo U. 2015. Temporal population dynamics of the phantom midge *Chaoborus crystallinus* and its influence on the zooplankton community. Hydrobiologia 770(1): 273–287.

Analysis of the raw data of 19 mesocosm studies showed that the composition of *Chaoborus* populations varied significantly between the years with the variation in the *Chaoborus* population structure between the seasons (*i.e.* within a year) being higher than between years. The present study confirmed that population relevant effects of chemicals on *Chaoborus* will be picked up in mesocosm studies that follow common principles of design and evaluation. Furthermore, the study revealed that *Chaoborus* larvae seem to promote high densities of rotifers and reduce the abundance of small crustaceans.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Not specified in the study report (data from untreated mesocosm enclosures was used).

B. STUDY DESIGN

Data source: Raw data of 19 mesocosm studies, conducted at TU Munich, Germany in the period between 1999 and 2012.
The data of untreated enclosures was used to examine the population dynamics of *Chaoborus* and the inter-replicate and temporal variability of *Chaoborus* populations. For the analysis of the influence of *Chaoborus* on the zooplankton community structure, the enclosures in which direct effects exclusively on *Chaoborus* occurred (and consequently potential indirect effects on prey species and competing species) were included additionally.

Test system: 11 structurally identical ponds: stainless steel ponds (29 000 L, diameter 5 m, height 1.5 m) with 1 mm black polyethylene foil, the ponds were embedded in the ground at a depth of 1 m; the water surface was positioned approximately at ground level, 20 cm sediment layer (natural sediment obtained from nearby water bodies and soil material obtained during construction of the ponds); 600 - 750 L stainless steel enclosures (diameter: 0.9 - 0.95 m, height: 1.3 - 1.5 m) were introduced into the ponds at least four weeks before the beginning of each mesocosm study.
Macroinvertebrates, zooplankton and phytoplankton were introduced into the ponds with sediment and ditchwater.
Macrophyte species planted: *Myriophyllum spicatum*, *Potamogeton natans*, *Chara intermedia*, *Nitella opaca* and *Elodea Canadensis*; degree of surface cover of macrophytes 20 – 25% at the beginning of each study.
Chaoborus crystallinus was the only *Chaoborus* species present in the test systems. The test systems were described as oligo-mesotrophic. Dilution water: natural water collected from nearby water bodies

Test design: Physico-chemical analysis (dissolved oxygen, conductivity, pH and temperature), *Chaoborus* sampling and sampling of other zooplankton species was usually conducted weekly.

Chaoborus sampling: aquatic stages of *Chaoborus* were collected by two different sampling methods: a) macroinvertebrate artificial substrate sampler and b) net sampling; individuals were identified and counted alive, then released back into the corresponding enclosure. Classification in the different larval stages was done based on body length (Instar I/II < 0.5 cm, Instar III 0.5 - 1 cm and Instar IV > 1 cm).

a) Macroinvertebrate artificial substrate sampler (MASS): artificial substrates consisted of pebble baskets (volume ca. 10 L, pebbles 24/32 mm), plant stems (*Phragmitis communis*) and five horizontal hardboard plates (multiplates, 10 x 10 x 0.4 cm). On each sampling occasion, one artificial substrate sampler was retrieved from each enclosure using a net (mesh size: 250 µm) in order to prevent organisms from escaping.

b) Net sampling: a scoop net (area of the opening: 425 cm^2, mesh size: 250 µm) was used to collect *Chaoborus* specimens.

Other zooplankton species sampling: two depth-integrated water samples (ca. 3 L each) were taken, combined, sieved (mesh size: 63 µm) and preserved with 4% formaldehyde. Specimens were identified and counted using a stereo-microscope at 40 x magnification. Organisms were grouped as Ostracoda, soft-bodied rotifers, rotifers with shell or spines, Cyclopidae, Diaptomidae, Nauplia, small Cladocerans (< 1.4 mm) and large Cladocerans (> 1.4 mm).

Endpoints: Composition of *Chaoborus* populations (presence and dominance of different aquatic life stages); number of *Chaoborus* generations per year; inter-replicate and temporal variability of *Chaoborus* populations; influence of *Chaoborus* on the zooplankton community structure.

Test concentrations: Not relevant, only data from untreated enclosures was used

Test conditions: All studies were conducted at Gruenschwaige Research Station, located approximately 15 km from Freising, Bavaria, Germany. The surrounding area was used by the TU Munich for agricultural research projects, in which no plant protection products or other chemicals were applied.

Analytics: Not relevant.

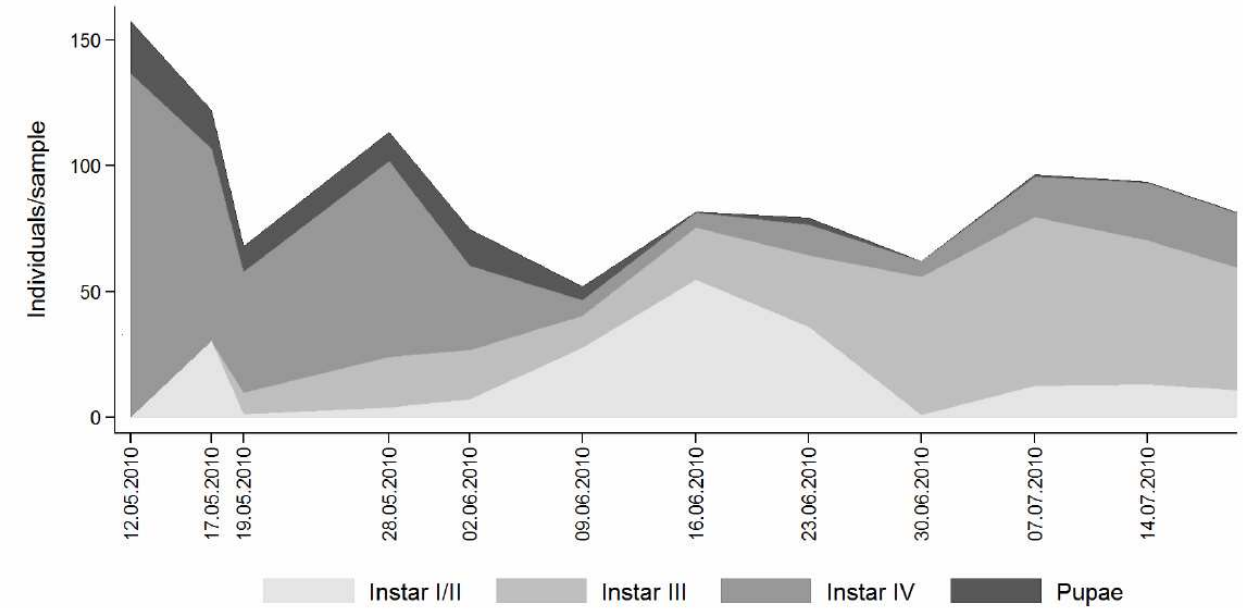
Statistics: Descriptive statistics; calculations of Minimum Detectable Differences (MDDs); multivariate statistical methods (partial Redundancy Analyses (RDAs) and Ordination analysis).

II. RESULTS AND DISCUSSION

Presence and dominance of the aquatic stages

The composition of the *Chaoborus* populations in the untreated enclosures analyzed in the present study varied significantly between the years. Due to its life cycle, the variation in the *Chaoborus* population structure between the seasons (i.e. within a year) was even higher than between years. A typical development of a *Chaoborus* population itemized into the different aquatic life stages is illustrated for the time period from mid-May to mid-July in Figure 8.2.8-1. Instar IV larvae dominated in May, instar III larvae in July and the proportion of instar I/II was highest in June.

Figure 8.2.8-1: Example of a typical development of a *Chaoborus* population in a mesocosm from mid-May to mid-July (data from 2010).



Number of generations per year

The recovery time of populations affected by the test item is an important aspect in using aquatic mesocosm study results for risk assessment. The generation time, reproductive rate and the number of generations per year are among the key factors for the recovery process of populations. It was shown for the studied test system in the south of Germany in Central Europe that there usually were three *Chaoborus* generations per year (from the end of April to the beginning of August), each lasting four to six weeks.

Inter-replicate variability
Inter-replicate variability was predominantly caused by natural variability between parallel enclosures and to a lesser extent by sampling errors. Calculations of Minimum Detectable Differences (MDDs) showed that the inter-replicate variability was sufficient low in order to reveal population relevant effects of chemicals on *Chaoborus* in mesocosm studies that follow common principles of design and evaluation. In addition, it was demonstrated, that effects on *Chaoborus* populations can be measured with high sensitivity using mesocosm test systems. Hence, population relevant effects of chemicals on *Chaoborus* will be picked up in mesocosm studies that follow common principles of design and evaluation.

Temporal variability
The population structure of *Chaoborus* varied significantly between the years and between the seasons (within a year). As indicated by partial Redundancy Analyses (RDAs) the study year could explain 10.8% of the total variance in the *Chaoborus* data set, whereas 23.3% could be explained by the variability within a year.

Influence on the zooplankton community structure
Ordination analysis revealed that *Chaoborus* larvae seem to promote high densities of rotifers and reduce the abundance of small crustaceans (nauplii larvae, copepods and small cladocerans). *Chaoborus* larvae prefer small crustaceans as a food source over rotifers (Kajak and Rybak 1979). Probably, as a consequence of the decrease in crustacean populations caused by *Chaoborus*, the numbers of rotifers increased due to the reduced predator pressure and lower food competition.

In the meantime, this report has been published in the peer-reviewed literature as: Janz P., Weltje L., Ebke K.P., Dawo U. 2015. Temporal population dynamics of the phantom midge *Chaoborus crystallinus* and its influence on the zooplankton community. Hydrobiologia 770(1): 273–287.

III. CONCLUSION

Analysis of the raw data of 19 mesocosm studies showed that the composition of *Chaoborus* populations varied significantly between the years with the variation in the *Chaoborus* population structure between the seasons (*i.e.* within a year) being higher than between years. The present study confirmed that population relevant effects of chemicals on *Chaoborus* will be picked up in mesocosm studies that follow common principles of design and evaluation. Furthermore, the study revealed that *Chaoborus* larvae seem to promote high densities of rotifers and reduce the abundance of small crustaceans.

From the performed literature search the following peer-reviewed scientific study investigating the aquatic toxicity of alpha-cypermethrin to the moorfrog *Rana arvalis* was considered for the aquatic risk assessment of alpha-cypermethrin. Due to missing analytical measurements in this study and the insufficient description of the testing method, exposure duration, etc. it was classified as "not reliable" (RI 3). For details please see the literature search and evaluation files also provided within the submission for Annex I Renewal. Nevertheless, the study is considered to provide some additional information on chronic effects on amphibians (note that a reliable, RI 2, study on acute effects is available – see below). The data have not been used or evaluated during the previous Annex I inclusion process and thus, relevant information and results of the study are described in the following summary.

Report:	CA 8.2.8/6 Greulich K. Pflugmacher S., 2003a Differences in susceptibility of various life stages of amphibians to pesticide exposure 2003/1034399
Guidelines:	none
GLP:	no

Executive Summary

The influence of alpha-cypermethrin on hatching success, mortality, duration of metamorphosis, and growth of moor frog (*Rana arvalis*) tadpoles exposed at various life stages was investigated under worst-case laboratory conditions. Preliminary tests used small clutches of eggs (approximately 10 embryos at stage 10–12) and 10 tadpoles (stage 20) and exposed them for 48 hours to nominal alpha-cypermethrin concentrations of 0.1, 1.0 and 10.0 µg a.s./L in three replicates per treatment. Due to overt toxicity in the 10 µg/L treatment the definitive test was conducted with 0.1 and 1.0 µg/L (the larvae/eggs exposed to 1.0 µg/L also showed some effects). In the study of the life-stage treatment 10 individuals were exposed to nominal alpha-cypermethrin concentrations of 0.1 and 1.0 µg a.s./L. The tadpoles were examined for the mortality and behavioral abnormalities. All experiments were replicated three times. In addition, a water control and a solvent control were set up for all experiments. Body weight and snout-vent length of test organisms were assessed at the end of the experiment, when resorption of the tail was completed. The preliminary test showed overt toxicity in the 10 µg/L treatment and the larvae/eggs exposed to 1.0 µg/L also showed some effects (e.g. impact on hatching rate). However, there was no clear picture concerning growth, where sometimes the treated animals were bigger and sometimes smaller, depending on the exposed life-stage. The definitive test over about 60 days (test duration not clearly indicated in the paper) was conducted with treatments 0.1 and 1.0 µg/L. Growth was assessed by an unconventional method (linear regression of length vs. weight and then comparing slopes with Fisher's exact test). The slope of the 0.1 µg/L treatment was comparable to the control, while the 1.0 µg/L slope showed a significant difference. Further, there was no difference in duration of metamorphosis at 0.1 µg/L. Thus, the 0.1 µg/L treatment may be regarded as a preliminary NOEC.

In the present study, moor frog (*Rana arvalis*) tadpole chronic toxicity of alpha-cypermethrin was investigated. After ca. 60-d exposure to 0.1 µg/L no significant differences compared to the control were found for length, weight and duration of metamorphosis.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193); purity: >99%; purchased from Fluka (Seelze, Germany).

B. STUDY DESIGN

Test species: Moor frog (*Rana arvalis*); eggs (stage 10 - 12) and tadpoles (stage 20 according to Gosner, 1960), two egg clutches were collected from a wetland pond in northeastern Germany in April 2002; one egg clutch used for hatching studies immediately after collection, the other one was reared until tadpoles reached specific developmental stages and then used for studies.

Test design: Preliminary and definitive study: small egg clutches (approx. 10 embryos at stage 10-12) or tadpoles (10 individuals) were exposed to a water control, a solvent control and alpha-cypermethrin treatments. The control and test substance treatments were replicated 3 times respectively.

Preliminary study: Egg clutches and tadpoles were exposed over 48 hours.

Definitive study: Exposure over the whole developmental stage: egg stage, embryo stage and tadpole stage until metamorphosis (resorption of tail completed).

During the experiments animals were observed for mortality and behavioral abnormalities. Body weight and snout-vent length of test organisms were assessed at the end of the experiment, when resorption of the tail was completed.

Endpoints: Hatching success, mortality and sublethal effects (duration of metamorphosis, body weight and length).

Test rates: Water control (artificial, demineralized well water); solvent control (acetonitrile: 100 µL/L); 0.1, 1 and 10 µg alpha-cypermethrin/L in the preliminary studies and 0.1 and 1 µg a.s./L in the definitive study.

Test conditions: Test tanks containing 10 L artificially salted, demineralized water; water temperature: 20°C; light : dark cycle: 14:10 hours; feeding every third day: powdered dry food (Tetramin AZ 40).

Analytics: No analytical measurements conducted.

Statistics: Descriptive statistics; one-way ANOVA followed by Newman-Keuls test ($p < 0.05$) for hatching success data; Fisher`s exact test ($p < 0.05$) for mortality data; regression analysis and Fisher's exact test ($p < 0.05$) for body length and weight data.

II. RESULTS AND DISCUSSION

Analytical results:
Analytical measurements of test item concentrations were not conducted.

Biological results:
The influence of alpha-cypermethrin on hatching success, mortality, duration of metamorphosis, and growth of moor frog (*Rana arvalis*) tadpoles exposed at various life stages was investigated under worst-case laboratory conditions. Preliminary tests used small clutches of eggs (approximately 10 embryos at stage 10–12) and 10 tadpoles (stage 20) and exposed them for 48 hours to nominal alpha-cypermethrin concentrations of 0.1, 1.0 and 10.0 µg a.s./L in three replicates per treatment. Due to overt toxicity in the 10 µg/L treatment the definitive test was conducted with 0.1 and 1.0 µg/L (the larvae/eggs exposed to 1.0 µg/L also showed some effects). In the study of the life-stage treatment 10 individuals were exposed to nominal alpha-cypermethrin concentrations of 0.1 and 1.0 µg a.s./L. The tadpoles were examined for the mortality and behavioral abnormalities. All experiments were replicated three times. In addition, a water control and a solvent control were set up for all experiments. Body weight and snout-vent length of test organisms were assessed at the end of the experiment, when resorption of the tail was completed. The preliminary test showed overt toxicity in the 10 µg/L treatment and the larvae/eggs exposed to 1.0 µg/L also showed some effects (e.g. impact on hatching rate). However, there was no clear picture concerning growth, where sometimes the treated animals were bigger and sometimes smaller, depending on the exposed life-stage. The definitive test over about 60 days (test duration not clearly indicated in the paper) was conducted with treatments 0.1 and 1.0 µg/L. Growth was assessed by an unconventional method (linear regression of length vs. weight and then comparing slopes with Fisher's exact test). The slope of the 0.1 µg/L treatment was comparable to the control, while the 1.0 µg/L slope showed a significant difference. Further, there was no difference in duration of metamorphosis at 0.1 µg/L. Thus, the 0.1 µg/L treatment may be regarded as a preliminary NOEC.

III. CONCLUSION

In the present study, moor frog (*Rana arvalis*) tadpole chronic toxicity of alpha-cypermethrin was investigated. After ca. 60-d exposure to 0.1 µg/L no significant differences compared to the control were found for length, weight and duration of metamorphosis.

From the performed literature search the following peer-reviewed scientific study investigating lethal and sublethal effects of alpha-cypermethrin (and two other insecticides) on two developmental stages of *Xenopus laevis* was considered relevant and reliable (with restrictions, RI 2) for the aquatic risk assessment of alpha-cypermethrin (for details please see the literature search and evaluation files also provided within the submission for Annex I Renewal). The data had not been used or evaluated during the previous Annex I inclusion process and thus, relevant information and the results for alpha-cypermethrin of this study are described in the following summary.

Report:	CA 8.2.8/7 Yu S. et al., 2013a Lethal and sublethal effects of three insectidides on two developmental stages of Xenopus laevis and comparison with other amphibians 2013/1417940
Guidelines:	none
GLP:	no

Executive Summary

Lethal and sublethal effects of alpha-cypermethrin on African clawed frog (*Xenopus laevis*) embryos and larvae were investigated over a period of 96 h under semi-static test conditions. Embryos were exposed to alpha-cypermethrin at nominal concentrations of 10, 20, 40, 80, 160 µg alpha-cypermethrin/L (corresponding to measured concentrations of 4.5, 13.5, 34.4, 77.1 and 160.7 µg a.s./L) in 5 replicates per treatment containing 20 embryos each. Frog larvae were exposed at nominal concentrations of 1.25, 2.5, 5, 10 and 20 µg a.s./L (corresponding to measured concentrations of 1.0, 2.0, 4.5, 8.5 and 22.4 µg a.s./L) in 5 replicates per treatment containing 10 larvae each. In addition, a solvent control was set up for embryo and larvae tests. Embryos and larvae were observed for mortality and sub-lethal effects daily. Body length of larvae was assessed at the end of the experiment. Furthermore, data were used to compare toxicity of *X. laevis* with that of other amphibians.

The biological results are based on measured concentrations. Alpha-cypermethrin caused mortality, malformations, and growth inhibition in both developmental stages at all tested concentrations. Gut abnormalities, axial/tail malformations, and edema were the 3 major malformations observed. Compared with embryos, larvae were more sensitive to alpha-cypermethrin. The results suggest that *X. laevis* larvae may generate more protective toxicity estimates in risk assessments than embryos. Furthermore, it was shown that *Xenopus laevis* may provide useful toxicity thresholds for pyrethroid insecticides.

In a semi-static acute toxicity study with *Xenopus laevis* embryos and larvae the LC_{50} (96 h) values were determined to be 30.6 and 6.9 µg alpha-cypermethrin/L, respectively (based on measured concentrations).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (BAS 310 I, Reg.No. 4 078 193); purity: 99.5%; purchased from ChemService.

B. STUDY DESIGN

Test species: African clawed frog (*Xenopus laevis*); embryos (Nieuwkoop and Faber (NF) stage 8 - 11) and larvae (NF stage 46); derived from laboratory-bred *X. laevis.*

Test design: Experiments were conducted according to the Frog Embryo Teratogenesis Assay-Xenopus (FETAX) protocol (ASTM International, 2004) with modifications. Both experiments were conducted in a semi-static test system with daily renewal of test solutions over an exposure period of 96 hours. Five replicates were set up for each of the five test item concentrations and for the solvent control (acetone) in both experiments.

Embryonic experiment: Embryos were examined with a microscope for viability and developmental stages. Twenty viable embryos from 3 clutches (NF stage 8 -11) were mixed and randomly assigned to each beaker.

Larval experiment: NF stage 46 larvae from 3 clutches were mixed and randomly placed in beakers (10 larvae/beaker).

Embryos and larvae were checked daily for mortality. At the end of the 96 h exposure, surviving larvae were euthanized preserved in 10% formalin and each individual was examined for abnormalities with a microscope. Body length of test organisms was assessed at the end of the experiment. Water quality was measured every other day before water change.

Endpoints: Median lethal concentrations (LC_{50}); median teratogenic concentrations (TC_{50}); teratogenic index (TI); sub-lethal effects (abnormal behavior, body length).

Test rates: Embryonic experiment: Solvent control, 10, 20, 40, 80, 160 µg alpha-cypermethrin/L (nominal) corresponding to measured concentrations of 0, 4.5, 13.5, 34.4, 77.1 and 160.7 µg alpha-cypermethrin/L.

Larval experiment: Solvent control, 1.25, 2.5, 5, 10 and 20 µg alpha-cypermethrin/L (nominal) corresponding to measured concentrations of 0, 1.0, 2.0, 4.5, 8.5 and 22.4 µg alpha-cypermethrin/L.

Test conditions: 50 mL beakers containing 40 mL FETAX solution for the embryonic experiments and 1 L beakers containing 1 L FETAX solution for the larval experiments; temperature: 22 - 24° C; dissolved oxygen: 5.07 - 8.59 mg/L; conductivity: 1610 - 1879 µS/cm; pH: 6.5 - 8.0; average ammonia concentration: 0.36 mg/L (0.04 - 1.32 mg/L); feeding: larvae were given a diet comprised of Nasco frog brittle, Trout Growers Ration, and Nutrafin Max tropical fish food approx. 1 h before water changes.

Analytics: Analytical verification of test item concentrations was conducted using an Agilent 6890 series gas chromatograph equipped with an electron capture detector.

Statistics: Descriptive statistics; LC_{50s} and TC_{50s} were determined using a logistic model in JMP 9.0.0 (SAS Institute); TI was calculated as the ratio LC_{50}/ TC_{50}. Incidence of malformations was compared using the Kruskal–Wallis test. If a statistical significance was indicated, a nonparametric multiple comparison with control was performed. Difference in total length was compared using one-way ANOVA or Welch's test. Pesticide-treated groups were further compared with controls using Dunnett's test. Results were considered significant at $\alpha = 0.05$.

II. RESULTS AND DISCUSSION

Analytical measurements: Analytical verification of test item concentrations was conducted in each test item treatment. Measured concentrations of alpha-cypermethrin in the 10 µg/L and 20 µg/L treatments in the embryonic test were 4.5 µg/L and 13.5 µg/L (corresponding to 45% and 67.5% of nominal concentrations), respectively. This may be due to the absorption of the chemical to glass. However, recovery in the higher concentrations ranged from 86% to 100% of the nominal concentrations. In the larval experiment, the actual concentrations of alpha-cypermethrin ranged from 80% to 112% of the nominal concentrations. The biological results are based on measured concentrations.

Biological results: Mortality rates of 4% and 2% were observed in the solvent controls of the embryonic and larval experiments, respectively. After exposure of embryos over 96 hours, 12%, 38% and 44% mortality was observed at measured concentrations of 4.5, 13.5 and 34.4 µg a.s./L, respectively. In the two highest tested concentrations of 77.1 and 160.7 µg a.s./L all embryos were dead at test end. Larval mortality increased from 16% in the lowest measured concentration of 1.0 µg a.s./L to 88% in the highest concentration of 22.4 µg a.s./L after 96 hours of exposure.
In the embryonic experiments, abnormal behaviors such as twitching and convulsion occurred and were more obvious when embryos were occasionally disturbed during water change. In the larval experiments, alpha-cypermethrin induced abnormal behaviors such as hypersensitivity, twitching, convolution, and swimming in a circle at all tested concentrations. Furthermore, alpha-cypermethrin induced malformations in both embryos and larvae in all concentrations. Gut abnormalities, axial/tail malformations, and edema were the 3 major malformations observed. In both experiments, incidence of total malformations was significantly different among alpha-cypermethrin treatments (Kruskal-Wallis test, $p < 0.05$) and all concentrations caused significantly higher incidence of total malformations compared with controls (non-parametric comparison, $p < 0.05$). The embryonic TC_{50} was determined to be 4.4 µg a.s./L and the TI was calculated to be 6.95 (strong teratogen).
All concentrations of alpha-cypermethrin produced significantly smaller larvae than controls (Dunnett's test, $p < 0.05$).
Comparison of toxicity data for *X. laevis* and data for other amphibians indicate that *Xenopus laevis* larvae had high sensitivity to alpha-cypermethrin/cypermethrin relative to other larval amphibians. The results suggest that *X. laevis* larvae may generate more protective toxicity estimates in risk assessments than embryos. Furthermore, it was shown that *Xenopus laevis* may provide useful toxicity thresholds for pyrethroid insecticides.

The mortality rates and resulting endpoints of both experiments are summarized in Table 8.2.8-3 (embryonic experiment) and Table 8.2.8-4 (larval experiment).

Table 8.2.8-3: Lethal effects of alpha-cypermethrin on African clawed frog (*Xenopus laevis*) embryos after exposure over 96 hours

Concentration [µg a.s./L] (nominal)	**Solvent control**	**10**	**20**	**40**	**80**	**160**
Concentration [µg a.s./L] (measured)	--	**4.5**	**13.5**	**34.4**	**77.1**	**160.7**
Mortality after 96 h [%]	4	12	38	44	100	100
Endpoints [µg a.s./L] (measured)						
Embryonic LC_{50} (96 h)	30.6 (95% confidence limits: 27.1 - 34.7)					

Table 8.2.8-4: Lethal effects of alpha-cypermethrin on African clawed frog (*Xenopus laevis*) larvae after exposure over 96 hours

Concentration [µg a.s./L] (nominal)	**Solvent control**	**1.25**	**2.5**	**5.0**	**10**	**20**
Concentration [µg a.s./L] (measured)	**--**	**1.0**	**2.0**	**4.5**	**8.5**	**22.4**
Mortality after 96 h [%]	2	16	8	48	84	88
Endpoints [µg a.s./L] (measured)						
Larval LC_{50} (96 h)	6.9 (95% confidence limits: 5.7 - 8.5)					

III. CONCLUSION

In a semi-static acute toxicity study with *Xenopus laevis* embryos and larvae the LC_{50} (96 h) values were determined to be 30.6 and 6.9 µg alpha-cypermethrin/L, respectively (based on measured concentrations).

From the performed literature search the following peer-reviewed scientific study investigating the efficacy and environmental fate of alpha-cypermethrin applied to rice fields for the control of chironomid midge larvae was considered relevant and reliable (with restrictions, RI 2) for the aquatic risk assessment of alpha-cypermethrin (for details please see the literature search and evaluation files also provided within the submission for Annex I Renewal). The data had not been used or evaluated during the previous Annex I inclusion process and thus, relevant information and results of this study are described in the following summary.

Report: CA 8.2.8/8
Helliwell S.,Stevens M.M., 2000a
Efficacy and environmental fate of Alpha-Cypermethrin applied to rice fields for the control of chironomid midge larvae (Diptera: Chironomidae)
2000/1024088
Guidelines: none
GLP: no

Executive Summary

The pyrethroid insecticide alpha-cypermethrin was evaluated for the control of chironomid midge larvae in two field trials in New South Wales during 1997 - 1998 (trial 1) and 1998 - ± 1999 (trial 2) rice seasons. In each trial, two bays were designated as untreated controls, two were treated with a chlorpyrifos as a toxicant control and six bays (two at each of three application rates) were treated with alpha-cypermethrin. In trial 1, alpha-cypermethrin was applied at 10, 20, and 30 g a.s./ha, while in trial 2, application rates of 6, 10, and 20 g a.s./ha were evaluated. All treatments were applied 6 days after flooding using a single nozzle hand sprayer. In trial 1, three samples were taken from each bay 4, 9, 14, 19, and 24 days after application (DAA), whilst in trial 2, four samples were taken from each bay 4, 9, 14, 19, and 24 DAA. Concentrations of alpha-cypermethrin in the water column were monitored during each trial.

In trial 1 (1997 – 1998), alpha-cypermethrin applied at 10, 20, and 30 g a.s./ha provided between 54% and 73% control of target Chironominae in the first 19 DAA. Populations of Chironominae were very low during trial 1 with estimated control density at 14 DAA of approximately 485 larvae/m². Alpha-cypermethrin strongly suppressed the non-target group, but also provided significant reductions in target Chironominae at 9 DAA (all rates) and at 14 DAA (10 g a.s./ha rate only). In trial 2 (1998 - 1999), target Chironominae, were far more abundant with an estimated control density 14 DAA of approximately 13100 larvae/m². Alpha-cypermethrin provided > 99% control of Chironominae for 19 DAA at all rates evaluated (6, 10 and 20 g a.s./ha). In trial 2, all treatments significantly reduced populations of target Chironominae at each sampling period up to and including 19 DAA. At test end no statistically significant reductions in larval chironomid populations (including both target Chironominae and 'other Chironomidae') were observed in the 10 and 20 g a.s./ha treatments in trial 1 and the 6 and 10 g a.s./ha treatments in trial 2 when compared to the control populations. Hence, a recovery potential of both target Chironominae and other Chironomidae populations could be observed in the course of both trials after exposure to several alpha-cypermethrin concentrations. NOEAEC values were estimated based on initial (DAT 1) measured water concentrations of alpha-cypermethrin (these endpoints were not reported in the publication).

In two rice field trials, the recovery of chironomid populations was observed following exposure to alpha-cypermethrin at 10 and 20 g a.s./ha over an exposure period of 29 days (trial 1) and at 6 and 10 g a.s./ha over 24 days (trial 2). Based on the effects of alpha-cypermethrin on abundances of chironomid midge larvae and initial (DAT 1) measured water concentrations in the 6 and 10 g a.s./ha treatments, a NOEAEC value of about 0.16 - 0.23 µg a.s./L was derived.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (tested with Dominex® 100 EC, content of a.s.: 100 g alpha-cypermethrin/L); purchased from FMC International AG.

B. STUDY DESIGN

Test species: Naturally-occurring non-biting midges of different taxa; Chironominae (target; predominantly *Chironomus tepperi* Skuse) and 'other chironomids' (predominantly Tanypodinae).

Test site: The test site (size approximately 300 m²) was located at Yanco Agricultural Institute in southwestern New South Wales, Australia. Alternate bays in rows were used as treatment bays, with intervening bays being used as buffer zones. Two rows of nine rectangular bays with earthen banks (each approximately 30 m^2) were used in each trial. The two trials were conducted on adjacent sites on a Birganbigil clay loam soil.

Test design: Field studies conducting during 1997-1998 (trial 1) and 1998-1999 (trial 2). In each trial, two bays were designated as untreated controls, two were treated with a standard chlorpyrifos treatment and six bays (two at each of three application rates) were treated with alpha-cypermethrin. All chemical treatments were applied to the water surface in approximately 5 L of water using a single nozzle hand sprayer. All treatments were applied 6 days after flooding. All treatment and control bays were sown with pregerminated rice (cv. `Namaga', 120 kg dry weight/ha) by hand broadcasting within 2 h of chemical treatments being applied.
Soil core sampling combined with magnesium sulphate flotation was used to quantify larval chironomid populations by using plastic cylinder (96 mm Ø). In trial 1, three samples were taken from each bay 4, 9, 14, 19, 24 and 29 DAA, whilst in trial 2 four samples were taken from each bay 4, 9, 14, 19 and 24 DAA. Extracted larvae were divided into two groups, Chironominae and 'other chironomids' (predominantly Tanypodinae).
Single 1 L composite water samples were taken from the control and alpha-cypermethrin treated bays with five subsamples from random points in each bay. In trial 1, samples were collected at 1, 2, 5, 9, 14, 19 and 25 DAA. In trial 2, samples were collected at 1, 2, 3, 4, 7, 10, 14 and 18 DAA.

Endpoints: NOEAEC, based effects on larval chironomid abundance.

Test concentrations:	Control Treatments (both trials): untreated control, toxicant control (chlorpyrifos, tested as Lorsban® 500 EC (DowElanco, Australia), at 500 g a.s./L applied at 75 g a.s./ha) Alpha-cypermethrin treatment (trial 1): 10, 20 and 30 g a.s./ha Alpha-cypermethrin treatment (trial 2): 6, 10 and 20 g a.s./ha.
Test conditions:	Two rows of nine rectangular bays with earthen banks (each approximately 30 m²; Birganbigil clay loam soil) were used in each trial. Each bay was supplied with water from a central channel. A water depth of approximately 12 cm was maintained in the bays throughout both trials. rice: cv. `Namaga', 120 kg dry weight/ha; mean values of environmental parameters: pH: 9.1; conductivity: 90.2 µS/cm; water temperature: 23.8 °C; rainfall: 0.9 mm (trial 1, first 25 DAA); pH: 7.7; conductivity: 172.3 µS/cm; water temperature: 19.0 °C; rainfall: 1.0 mm (trial 2, first 18 DAA).
Analytics:	Analytical verification of test item concentrations in water was conducted using a Hewlett Packard 5890 Series II gas chromatograph equipped with an electron capture detector.
Statistics:	Descriptive statistics, Data were transformed to $y' = \log_e (y+1)$ and analyzed using ANOVA and Tukey's HSD test ($p < 0.05$). NOEAEC values were estimated based on measured concentrations of alpha-cypermethrin in water (endpoints are not reported in publication).

II. RESULTS AND DISCUSSION

Analytical measurements: Concentrations of alpha-cypermethrin in the water column were monitored during each trial. In both trials, alpha-cypermethrin levels in the water column followed an exponential decay, levels declining to < 0.015 µg/L at 5 DAA for trial 1 and at 7 DAA for trial 2. Concentrations in water for the 20 g a.s./ha rate ranged from 0.23 to 0.41 mg/L 1 DAA down to 0.008 mg/L at 18 DAA, whilst at the 6 g a.s./ha rate alpha-cypermethrin concentrations declined to below the detection limit (0.001 mg/L) at 18 DAA.

Biological results: In trial 1 (1997 – 1998), alpha-cypermethrin applied at 10, 20, and 30 g a.s./ha provided between 54% and 73% control of target Chironominae in the first 19 DAA. Populations of Chironominae were very low during trial 1 with estimated control density 14 DAA of approximately 485 larvae/m². Alpha-cypermethrin strongly suppressed the non-target group, but also provided significant (Tukey's HSD test; $p < 0.05$) reductions in target Chironominae at 9 DAA (all rates) and at 14 DAA (10 g a.s./ha rate only).
In trial 2 (1998 - 1999), target Chironominae, were far more abundant with an estimated control density 14 DAA of approximately 13100 larvae/m². Alpha-cypermethrin provided > 99% control of Chironominae for 19 DAA at all rates evaluated (6, 10 and 20 g a.s./ha). In trial 2, all treatments significantly ($p < 0.05$) reduced populations of target Chironominae at each sampling period up to and including 19 DAA.

At test end no statistically significant reductions in larval chironomid populations (including both target Chironominae and 'other Chironomidae') were observed in the 10 and 20 g a.s./ha treatments in trial 1 and the 6 and 10 g a.s./ha treatments in trial 2 when compared to the control populations. Hence, a recovery potential of both target Chironominae and other Chironomidae populations could be observed in the course of both trials after exposure to several alpha-cypermethrin concentrations. Initial (DAT 1) measured water concentrations were taken from the graphs in the paper. NOEAEC values were based on initial (DAT 1) measured water concentrations of alpha-cypermethrin in the 6 and 10 g a.s./ha treatments (these endpoints were not reported in the publication).

III. CONCLUSION

In two rice field trials, the recovery of chironomid populations was observed following exposure to alpha-cypermethrin at 10 and 20 g a.s./ha over an exposure period of 29 days (trial 1) and at 6 and 10 g a.s./ha over 24 days (trial 2). Based on the effects of alpha-cypermethrin on abundances of chironomid midge larvae and initial (DAT 1) measured water concentrations in the 6 and 10 g a.s./ha treatments, a NOEAEC value of about 0.16 - 0.23 µg a.s./L was derived.

From the performed literature search the following peer-reviewed scientific study investigating the uptake, translocation, and metabolism of the metabolite 3-phenoxybenzoic acid (3-PBA) in the submerged rooted macrophyte water milfoil (*Myriophyllum elatinoides*) was considered relevant and reliable (with restrictions, RI 2) for the aquatic risk assessment of alpha-cypermethrin (for details please see the literature search and evaluation files also provided within the submission for Annex I Renewal). The data had not been used or evaluated during the previous Annex I inclusion process and thus, relevant information and results of this study are described in the following summary.

Report: CA 8.2.8/9
Ando D. et al., 2012a
Uptake, translocation, and metabollism of 3-phenoxybenzioc acid in the submerged rooted macrophyte water milfoil (Myriophyllum elatinoides)
2012/1367684
Guidelines: none
GLP: no

Executive Summary

In a 14-day static toxicity laboratory study, the toxicity and the metabolic pathway of 3-phenoxybenzoic acid (3-PBA; metabolite of alpha-cypermethrin) in water milfoil *Myriophyllum elatinoides* was investigated. Water milfoils were exposed to a water control and to ^{14}C-radiolabeled 3-PBA at an exposure concentration of 3.28 ppm (equivalent to 3280 µg/L; concentration in overlying water or in sediment pore water). Exposure was conducted in a medium-containing shoot/leaves chamber *via* water treatment and in a sediment-containing roots chamber *via* sediment treatment. Plant sampling for ^{14}C-distribution analysis and determination of plant growth (total length and wet weight) was conducted 0.5, 1, 3, 5, 7 and 14 days after test initiation.

No growth inhibition of the plants was confirmed in the sequestered glass chambers after comparing the length (ranging from 1.1 to 1.8 cm) and fresh weight (ranging from 0.7 to 0.11 g) with those grown in the acclimation aquarium during a 14-day incubation period.
The uptake of ^{14}C-radiolabeled 3-PBA dissolved in water by the shoot/leaves amounted to 15.85% of the applied radioactivity (AR) after 0.5 days, and ^{14}C translocation to the roots was minimal during 14 days. Slower uptake of the radioactivity from test item treated sediment by the roots was observed (6.37%AR after 14 days) and 1.71%AR was trans-located to the shoot/leaves. Oxidation, reduction and conjugation of the test item proceeded in the macrophyte.

Water and sediment exposure of the submerged rooted macrophyte *Myriophyllum elatinoides* to 3-phenoxybenzoic acid (3-PBA, metabolite of alpha-cypermethrin) at a concentration of 3.28 ppm caused no growth inhibition after incubation over 14-days. Thus, the NOEC based on growth inhibition can be estimated to be ≥ 3.28 ppm (equivalent to ≥ 3280 µg/L; concentration in overlying water or in sediment pore water).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: 3-phenoxybenzoic acid (3-PBA; metabolite of alpha-cypermethrin); uniformly labeled with ^{14}C at the phenoxyphenyl ring (specific radioactivity: 4.37 GBq/mmol); analytical grade; synthesized in-house.

B. STUDY DESIGN

Test species:	Water milfoil (*Myriophyllum elatinoides* Gaudich), Haloragaceae, a dicotyledonous, submerged rooted macrophyte; purchased from Aqua Rise Co., Osaka, Japan; cultivated in-house for at least 10 days.
Test design:	Static system; 14 day incubation; 3 day rooting phase. 1 test item concentration; each exposure (water and sediment treatments) was conducted in triplicate. Exposure experiments were conducted using a glass vessel partitioned with a welded grass board that included shoot/leaves and roots chambers. The chambers were individually filled with 120 mL of the AAP medium (shoot/leaves chamber) and 35 g of OECD sediment moistened with 20 mL of AAP medium (roots chamber). The test solution was applied to the AAP medium in the shoot/leaves chamber via water treatment or to the sediment in the roots chamber via sediment treatment followed by uniform mixing to establish an exposure concentration of 3.28 ppm (0.167 MBq). The root tip of the sterilized plant (length: 16.5 - 18.3 cm; fresh body weight: 0.34 -0.51 g) was buried into the sediment and the shoot/leaves portion was immersed in the AAP-medium. Sampling was conducted at 0.5, 1, 3, 5, 7 and 14 days after treatment.
Endpoints:	NOEC based on growth inhibition (based on length and fresh weight); uptake rates; metabolite distribution.
Test concentrations:	0 (water control), 3.28 µg ^{14}C-labeled 3-PBA/L (0.167 MBq); equivalent to 3280 µg/L (concentration in overlying water or in sediment pore water).
Test conditions:	Partitioned glass vessel, including a sediment containing root chamber and a medium containing shoot/leaves chamber; root chamber was filled with 35 g artificial sediment (OECD 218) moistened with 20 mL AAP medium; shoot/leaves chamber was filled with 120 mL AAP (American Academy of Pediatrics) water medium; pH of AAP medium: 7.0 ± 0.5; temperature: 20 ± 2 °C; photoperiod: 16 h light : 8 h dark, light intensity: 120 µE*m-^{2}s-^{1}[10].
Analytics:	A reversed-phase HPLC system with scintillation counting was used to analyzed 3-PBA and its degradation products. One-dimesional NMR spectra was measured using a Spectrometer. Liquid chromatography-electrospray ionization-mass spectrometry (LC-ESI-MS) analysis was conducted to measure radioactivity in water, sediment extract, and rinsate/extract of the plants.
Statistics:	No statistical methods were described.

II. RESULTS AND DISCUSSION

Metabolic distribution: Radioactivity in water, sediment and plants was measured 0.5, 1, 3, 5, 7 and 14 days after test initiation. The total recovery of ^{14}C in each treatment was greater than 95% of the applied radioactivity throughout the test period, indicating the insignificant loss of ^{14}C by volatilization or adsorption to the test vessel. In case of the sediment treatment, approximately 80%AR remained as unaltered 3-PBA in the roots chamber, most of which originated from ^{14}C in the interstitial medium water of the sediment. Regarding the water treatment, approximately 80%AR remained in the medium, and no ^{14}C was detected in the sediment phase. The uptake of ^{14}C-radiolabeled 3-PBA dissolved in water by the shoot/leaves amounted to 15.85% of the applied radioactivity (AR) after 0.5 days, and ^{14}C translocation to the roots was minimal during 14 days. Slower uptake of the radioactivity from test item treated sediment by the roots was observed (6.37%AR after 14 days) and 1.71%AR was trans-located to the shoot/leaves. Oxidation, reduction and conjugation of the test item proceeded in the macrophyte.

Plant growth observations: No growth inhibition of the plants was confirmed in the sequestered glass chambers after comparing the length (ranging from 1.1 to 1.8 cm) and fresh weight (ranging from 0.7 to 0.11 g) with those grown in the acclimation aquarium during a 14-day incubation period.

III. CONCLUSION

Water and sediment exposure of the submerged rooted macrophyte *Myriophyllum elatinoides* to 3-phenoxybenzoic acid (3-PBA, metabolite of alpha-cypermethrin) at a concentration of 3.28 ppm caused no growth inhibition after incubation over 14-days. Thus, the NOEC based on growth inhibition can be estimated to be ≥ 3.28 ppm (equivalent to ≥ 3280 µg/L; concentration in overlying water or in sediment pore water).

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CA 8.3 Effects on arthropods

CA 8.3.1 Effects on bees

Table 8.3.1-1: Toxicity of alpha-cypermethrin to arthropods

Substance	Test species	Endpoint	Value	Reference	Study EU agreed?
alpha-cypermethrin	honeybee	24 h acute oral LD_{50}	0.059 µg a.s./bee	Knight, AL-541-002	Yes
		24 h acute contact LD_{50}	0.033 µg a.s./bee		
	honeybee	48 h acute oral LD_{50}	0.246 µg a.s./bee	Barth, 2005/1026137	No, new study
		48 h acute contact LD_{50}	0.030 µg a.s./bee		
	honeybee	10 d chronic LC_{50}	2.29 mg a.s./kg food	Sekine, 2013/1132489	No, new study
		10 d chronic $LD_{50\ (daily)}$	0.11 µg a.s./bee/day		
		10 d chronic $LD_{50\ (overall)}$	1.1 µg a.s./bee		
	honeybee	7 d larvae	acute study (7 day larvae) not valid		No, new study
	honeybee	120 h chronic larvae LD_{50}	> 8.0 ng a.s./larva	Kleebaum, 2014/1162697	No, new study
		120 h chronic larvae LC_{50}	> 0.052 mg a.s./kg food		
		120 h chronic larvae NOED	≥ 8.0 ng a.s./larva		
		120 h chronic larvae NOEC	≥ 0.052 mg a.s./kg food		
	bumble bee	96 h acute oral LD_{50}	0.54 µg a.s./bumblebee	Amsel, 2014/1111111 Amendment, 2014/1315402	No, new study
		96 h acute contact LD_{50}	0.29 µg a.s./bumblebee		

CA 8.3.1.1 Acute toxicity to bees

CA 8.3.1.1.1 Acute oral toxicity

Report: CA 8.3.1.1.1/1
Barth M., 2006a
Acute toxicity of BAS 310 I to the honeybee Apis mellifera L. under laboratory conditions
2005/1026137
Guidelines: OECD 213, OECD 214
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

In a dose response test, young adult worker bees (*Apis mellifera* L.) were exposed to alpha-cypermethrin. The toxicity of the test product was determined in an oral test at concentrations of 0.800, 0.400, 0.200, 0.100 and 0.050 µg a.s./bee (0.650, 0.375, 0.192, 0.100 and 0.050 µg a.s./bee actual intake). Additionally, honeybees were treated with dimethoate as reference item at concentrations ranging from 0.0625 to 0.50 µg a.s./bee (0.499 to 0.0625 µg a.s./bee actual intake). Two control treatments consisting of sucrose solution alone and sucrose solution containing 1% ethyl acetate were carried out**.** The test was conducted with 3 replicates for the treatment groups; each of the test cages contained 10 bees. Assessment of mortality was done after 4, 24 and 48 hours. Additionally behavioral abnormalities were observed within 48 hours after oral exposure.

In the oral toxicity test (48 h) the average mortalities for the test item treatments of 0.800, 0.400, 0.200, 0.100 and 0.050 µg a.s./bee (0.650, 0.375, 0.192, 0.100 and 0.050 µg a.s./bee actual intake) were 100%, 66.75%, 16.7%, 13.3% and 10%, respectively. In the sucrose control and sucrose/ethyl acetate control, average mortalities were 3.3% and 0%, respectively.
At assessments conducted 4 hours after oral exposure of the test item effects on behavior were observed in honeybees consuming doses of 0.05 up to 0.650 µg a.s./bee. At the assessments conducted 24 hours after oral exposure of the test item bees had generally recovered. No different behavior for all surviving bees consuming doses up to 0.375 µg a.s./bee compared to control bees was observed.

The LD_{50} (48 h) was 0.246 µg alpha-cypermethrin (95% CL 0.201 – 0.301 µg alpha-cypermethrin) consumed a.s./bee.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: alpha-cypermethrin (BAS 310 I, Reg. no. 4 078 193); batch COD-000166 (99.3%, analyzed).

B. STUDY DESIGN

Test species: Honeybee (*Apis mellifera carnica* L.), worker bees from a healthy and queen-right colony, age: 4 - 6 weeks, source: Bienenfarm Kern GmbH, Leipzig, Germany, collected in the morning of use and kept under test conditions.

Test design: Oral (LD_{50} test): 48 h; 5 treatment groups; 3 replicates each consisting 10 bees. The mortality was assessed 4, 24 and 48 h after application.

Endpoints: LD_{50}, behavioral abnormalities.

Reference item: Perfekthion EC 400 (dimethoate 408.7 g/L analyzed).

Test concentrations: 0.800, 0.400, 0.200, 0.100 and 0.050 µg a.s./bee. Actual intake: 0.650, 0.375, 0.192, 0.100 and 0.050 µg a.s./bee.

Test conditions: Bees were kept in the dark at 24°C – 25°C (69 - 71% RH) in the oral test. Food: 50 % w/v sucrose solution.

Statistics: Fisher's Exact Binomial Test with Bonferroni Correction for mortality data, $\alpha \leq 0.05$. Probit analysis according to the maximum likelihood method (FINNEY 1971) for calculation of the LD_{50} with 95 % confidence intervals. Statistical program used: ToxRat Professional 2.09 (2004).

II. RESULTS AND DISCUSSION

In the oral toxicity test no statistically significant effects on survival (10.0, 13.3 and 16.7% mortality, respectively) were observed at consumed doses of 0.050, 0.100 and 0.192 µg a.s./bee during 48 hours (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha \leq 0.05$). For the consumed doses of 0.375 and 0.650 µg a.s./bee statistically significant effects of the test item on survival (66.7 and 100.0% mortality) were observed after 48 hours (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha \leq 0.05$). Therefore the LD_{50} (48 h) was calculated as 0.246 µg consumed a.s./bee in the oral toxicity test.

At assessments conducted 4 hours after oral exposure of the test item effects on behavior were observed in honeybees consuming doses of 0.05 up to 0.650 µg a.s./bee. At the assessments conducted 24 hours after oral exposure of the test item bees had generally recovered. No different behavior for all surviving bees consuming doses up to 0.375 µg a.s./bee compared to control bees was observed.

See results are summarized below Table 8.3.1.1.1-1.

Table 8.3.1.1.1-1: Toxicity of alpha-cypermethrin to honeybees (*Apis mellifera* L.) in an oral toxicity test

	Oral test (48 h)
	µg a.s./bee
LD_{50}	0.246
95 %-CL	0.201 - 0.301

III. CONCLUSION

The LD_{50} (48 h) was 0.246 µg alpha-cypermethrin (95 % CL 0.201 – 0.301 µg alpha cypermethrin) consumed a.s./bee.

Report: CA 8.3.1.1.1/2
Amsel K., 2014a
Acute toxicity of BAS 310 I to the bumblebee Bombus terrestris L. under laboratory conditions
2014/1111111
Guidelines: Van der Steen (2001), OECD 213 (1998), OECD 214 (1998), Hanewald et al. (2013), Van der Steen (1996)
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Report: CA 8.3.1.1.1/3
Amsel K., 2014b
Amendment No. 1 to the final report - Acute toxicity of BAS 310 I to the bumblebee Bombus terrestris L. under laboratory conditions
2014/1315402
Guidelines: Van der Steen (2001), OECD 213 (1998), OECD 214 (1998), Hanewald et al. (2013), Van der Steen (1996)
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

In a dose response test, young adult worker bumblebees (*Bombus terrestris* L.) were exposed to BAS 310 I. The acute toxicity of the test item was determined in an oral test at dose rates of 0.13, 0.25, 0.51, 1.01 and 2.02 µg BAS 310 I/bumblebee which is corresponding to 0.13, 0.25, 0.50, 1.00 and 2.00 µg a.s./bumblebee (corrected for purity). The resulting oral uptake was 0.12, 0.24, 0.46, 0.97 and 1.76 µg BAS 310 I/bumblebee. Additionally, bumblebees were treated with Dimethoate EC 400 as reference item at dose rates ranging from 0.25 to 1.50 µg dimethoate/bumblebee (nominal) or with 50% (w/v) sucrose solution and sucrose solution including 1% acetone as controls.

In the oral toxicity test, no mortality occurred in both control groups. In the test item treatment, statistically significant mortality of 53.3, 76.7 and 86.7% occurred after oral consumption of 0.46, 0.97 and 1.76 µg BAS 310 I/bumblebee, after 96 hours. Bumblebees in the dose rate of 0.24 µg consumed BAS 310 I/bumblebee revealed effects on mortality that amounted to 16.7%, whereas no mortality occurred in the lowest dose rate of 0.12 µg consumed BAS 310 I/bumblebee, after 96 hours. Behavioral abnormalities of surviving bumblebees occurred at the higher dose rates of 0.46, 0.97 and 1.76 µg consumed BAS 310 I/bumblebee. Surviving bumblebees showed symptoms of moribundity. No or only slight effects on behavior of surviving bumblebees occurred in the dose rates of 0.12 and 0.24 µg consumed BAS 310 I in the oral toxicity test when compared to the control

The toxicity of BAS 310 I on bumblebees was tested in an acute oral toxicity test. The LD_{50} (96 h) was determined to be 0.54 µg consumed product/bumblebee which is corresponding to 0.54 µg consumed a.s./bumblebee (corrected for purity).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 I, batch no. COD-000595; content: alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193): 99.2% (analyzed).

B. STUDY DESIGN

Test species: Bumblebee (*Bombus terrestris* L.), young adult worker bumblebees derived from a healthy and queen-right micro-hive; source: Biobest Belgium N.V., Westerlo, Belgium; delivered by: Katz Biotech AG, Baruth, Germany; collected from the bumblebee micro-hive in the morning prior to use; starving period: 3 hours.

Test design: In a 96-hour test, adults of *Bombus terrestris* L. were exposed to 5 dose rates of BAS 310 I in treated food (50% (w/v) sucrose solution). In total, 3 treatment groups were set up: 5 dose rates of the test item, two control groups and 4 dose rates of the reference item with 30 replicates per dose and 1 bumblebee per replicate. Assessments of bumblebee mortality and behavioral effects were done after 4, 24, 48, 72 and 96 hours.

Endpoints: Mortality, behavioral impairments.

Reference item: Dimethoate EC 400 (BAS 152 11 I, a.s.: dimethoate, 400 g/L nominal).

Test rates: Controls: 50% (w/v) sucrose solution, 50% (w/v) sucrose solution including 1% acetone; test item: 0.13, 0.25, 0.51, 1.01 and 2.02 µg BAS 310 I/bumblebee which is corresponding to 0.13, 0.25, 0.50, 1.00 and 2.00 µg a.s./bumblebee (corrected for purity); resulting in an actual uptake of 0.12, 0.24, 0.46, 0.97 and 1.76 µg BAS 310 I/bumblebee; reference item: 0.25, 0.46, 0.83 and 1.50 µg dimethoate/bumblebee.

Test conditions: Temperature: 24.9°C – 25.1°C; relative humidity: 57.5% - 60.9%, photoperiod: 24 h darkness, food: 50% (w/v) sucrose solution.

Statistics: Descriptive statistics. Fisher's Exact Binominal test with Bonferroni Correction for mortality data (one-sided greater, $\alpha = 0.05$), Probit analysis using linear weight regression and moving average computation after Thompson for calculation of the LD_{50} values.

II. RESULTS AND DISCUSSION

In the oral toxicity test, no mortality occurred in both control groups. In the test item treatment, statistically significant mortality of 53.3, 76.7 and 86.7% occurred after oral consumption of 0.46, 0.97 and 1.76 µg BAS 310 I/bumblebee, after 96 hours (Fisher's Exact Binominal test with Bonferroni Correction, one-sided greater, $\alpha = 0.05$). Bumblebees in the dose rate of 0.24 µg consumed BAS 310 I/bumblebee revealed effects on mortality that amounted to 16.7%, whereas no mortality occurred in the lowest dose rate of 0.12 µg consumed BAS 310 I/bumblebee, after 96 hours. Behavioral abnormalities of surviving bumblebees occurred at the higher dose rates of 0.46, 0.97 and 1.76 µg consumed BAS 310 I/bumblebee. Surviving bumblebees showed symptoms of moribundity. No or only slight effects on behavior of surviving bumblebees occurred in the dose rates of 0.12 and 0.24 µg consumed BAS 310 I in the oral toxicity test when compared to the control The results are summarized in Table 8.3.1.1.1-2.

Table 8.3.1.1.1-2: Toxicity of BAS 310 I to bumblebee (*Bombus terrestris* L.) in an oral toxicity test

Treatment [µg BAS 310 I/bumblebee]	**Uptake of test item [µg BAS 310 I/bumblebee]**	**Mean mortality [%]**			
		24 h	**48 h**	**72 h**	**96 h**
Control (sucrose)	--	0.0	0.0	0.0	0.0
Control (sucrose + acetone)	--	0.0	0.0	0.0	0.0
0.13	0.12	0.0	0.0	0.0	0.0
0.25	0.24	16.7	16.7	16.7	16.7
0.50	0.46	36.7*	53.3*	53.3*	53.3*
1.00	0.97	56.7*	66.7*	76.7*	76.7*
2.00	1.76	40.0*	73.3*	86.7*	86.7*
Endpoints					
LD_{50} (96 h) [1)]	**[µg consumed a.s./bumblebee]**		**[µg consumed product/bumblebee]**		
	0.54 (95% CL: 0.42 - 0.69)		0.54 (95% CL: 0.42 - 0.69)		

* Statistically significant differences compared to the control (Fisher's Exact Binominal test with Bonferroni Correction, $\alpha = 0.05$).

1) Median lethal dose calculated by Probit analysis (with 95% Confidence Limits).

The LD_{50} value (24 h) for the reference item in the oral toxicity test was determined to be 0.50 µg a.s./bumblebee.

III. CONCLUSION

The toxicity of BAS 310 I on bumblebees was tested in an acute oral toxicity test. The LD_{50} (96 h) was determined to be 0.54 µg consumed product/bumblebee which is corresponding to 0.54 µg consumed a.s./bumblebee (corrected for purity).

CA 8.3.1.1.2 Acute contact toxicity

Report:	CA 8.3.1.1.2/1 Barth M., 2006a Acute toxicity of BAS 310 I to the honeybee Apis mellifera L. under laboratory conditions 2005/1026137
Guidelines:	OECD 213, OECD 214
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

In a dose response test, young adult worker bees (*Apis mellifera* L.) were exposed to alpha-cypermethrin. The toxicity of the test product was determined in an contact test at concentrations of 0.200, 0.096, 0.046, 0.022 and 0.011 µg a.s./bee. Additionally, honeybees were treated with dimethoate as reference standard at concentrations ranging from 0.0625 to 0.50 µg a.s./bee. Three control treatments consisting of deionized water, acetone and 0.1% v/v Tween solution were carried out. The test was conducted with 3 replicates for the treatment groups; each of the test cages contained 10 bees. Assessment of mortality was done after 4, 24 and 48 hours. Additionally behavioral abnormalities were observed within 48 hours after contact exposure.

In the contact toxicity test (48 h) the average mortalities for the test item treatments of 0.200, 0.096, 0.046, 0.022 and 0.011 µg a.s./bee were 100, 93.3, 76.7, 33.3 and 6.7%, respectively. No mortality was observed in the control treatment groups.
Effects on behavior were observed in honeybees after contact exposure to doses of 0.011 up to 0.200 µg a.s./bee at the 4 hour assessment.
Within 48 hours after contact exposure to the test item bees had generally recovered and no different behavior for all surviving bees exposed up to a dose of 0.096 µg a.s./bee compared to control bees was observed.

The LD_{50} (48 h) was 0.030 µg (95% CL 0.025 - 0.036 µg alpha cypermethrin) a.s./bee in the contact toxicity test.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: alpha-cypermethrin (BAS 310 I, Reg. no. 4 078 193); batch COD-000166 (99.3%, analyzed).

B. STUDY DESIGN

Test species: Honeybee (*Apis mellifera carnica* L.), worker bees from a healthy and queen-right colony, age: 4 - 6 weeks, source: Bienenfarm Kern GmbH, Leipzig, Germany, collected in the morning of use and kept under test conditions.

Test design: Contact (LD_{50} test): 48 h; 5 treatment groups, 3 replicates each consisting 10 bees. The mortality was assessed 4, 24 and 48 h after application.

Endpoints: LD_{50}, behavioral abnormalities.

Reference item: Perfekthion EC 400 (dimethoate 408.7 g/L analyzed).

Test concentrations: 0.200, 0.096, 0.046, 0.022 and 0.011 µg a.s./bee.

Test conditions: Bees were kept in the dark at 24°C – 25°C (69% - 71% RH) in the contact test. Food: 50% w/v sucrose solution.

Statistics: Fisher's Exact Binomial Test with Bonferroni Correction for mortality data, $\alpha \leq 0.05$. Probit analysis according to the maximum likelihood method (FINNEY 1971) for calculation of the LD_{50} with 95 % confidence intervals. Statistical program used: ToxRat Professional 2.09 (2004)

II. RESULTS AND DISCUSSION

In the contact toxicity test no statistically significant effects on survival (6.7% mortality) were observed at the tested dose of 0.011 µg a.s./bee after 48 hours (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha \leq 0.05$). For the tested doses of 0.022, 0.046, 0.096 and 0.200 µg a.s./bee statistically significant effects of the test item on survival (33.3, 76.7, 93.3 and 100.0% mortality) were observed after 48 hours (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha \leq 0.05$). Therefore the LD_{50} (48 h) was calculated as 0.030 µg consumed a.s./bee in the contact toxicity test.
In the contact toxicity test effects on behavior were observed in honeybees after contact exposure to doses of 0.011 up to 0.200 µg a.s./bee at the 4 hour assessment.
Within 48 hours after contact exposure to the test item bees had generally recovered and no different behavior for all surviving bees exposed up to a dose of 0.096 µg a.s./bee compared to control bees was observed.

See results are summarized in Table 8.3.1.1.2-1.

Table 8.3.1.1.2-1: Toxicity of alpha-cypermethrin to honeybees (*Apis mellifera* L.) in a contact toxicity test

	Contact test (48 h)
	µg a.s./bee
LD_{50}	0.030
95%-CL	0.025 - 0.036

III. CONCLUSION

The LD_{50} (48 h) was 0.030 µg (95% CL 0.025 - 0.036 µg alpha cypermethrin) a.s./bee in the contact toxicity test.

Report: CA 8.3.1.1.2/2
Amsel K., 2014a
Acute toxicity of BAS 310 I to the bumblebee Bombus terrestris L. under laboratory conditions
2014/1111111
Guidelines: Van der Steen (2001), OECD 213 (1998), OECD 214 (1998), Hanewald et al. (2013), Van der Steen (1996)
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Report: CA 8.3.1.1.2/3
Amsel K., 2014b
Amendment No. 1 to the final report - Acute toxicity of BAS 310 I to the bumblebee Bombus terrestris L. under laboratory conditions
2014/1315402
Guidelines: Van der Steen (2001), OECD 213 (1998), OECD 214 (1998), Hanewald et al. (2013), Van der Steen (1996)
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

In a dose response test, young adult worker bumblebees (*Bombus terrestris* L.) were exposed to BAS 310 I. The acute toxicity of the test item was determined in a contact test at dose rates of 0.06, 0.13, 0.25, 0.51 and 1.01 µg BAS 310 I/bumblebee which is corresponding to 0.06, 0.13, 0.25, 0.50 and 1.00 µg a.s./bumblebee (corrected for purity). Additionally, bumblebees were treated with Dimethoate EC 400 as reference item at dose rates ranging from 2.5 to 10.0 µg dimethoate/bumblebee (nominal) or with deionized water, Tween solution and acetone as controls.

In the contact toxicity test, no mortality occurred in the control group treated with deionized water. After 96 hours of exposure 3.3% mortality occurred in the control groups treated with Tween solution and acetone. In the test item treatment, statistically significant mortality of 43.3, 76.7 and 100.0% occurred after thoracic application of 0.25, 0.51 and 1.01 µg BAS 310 I/bumblebee, after 96 hours. Bumblebees treated with 0.06 and 0.13 µg BAS 310 I/bumblebee revealed effects on mortality that amounted to 6.7 and 20.0%, after 96 hours. Behavioral abnormalities of surviving bumble bees occurred at the higher dose rates of 1.01 and 0.50 µg BAS 310 I/bumblebee. Surviving bumblebees showed symptoms of moribundity. In the other dose rates no effects on behavior of surviving bumblebees occurred when compared to the control.

The toxicity of BAS 310 I on bumblebees was tested in an acute contact toxicity test. The LD_{50} (96 h) was determined to be 0.29 µg product/bumblebee which is corresponding to 0.29 µg a.s./bumblebee (corrected for purity).

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 I, batch no. COD-000595; content: alpha-cypermethrin (BAS 310 I, Reg. No. 4 078 193): 99.2% (analyzed).

B. STUDY DESIGN

Test species: Bumblebee (*Bombus terrestris* L.), young adult worker bumblebees derived from a healthy and queen-right micro-hive; source: Biobest Belgium N.V., Westerlo, Belgium; delivered by: Katz Biotech AG, Baruth, Germany; collected from the bumblebee micro-hive in the morning prior to use.

Test design: In a 96-hour test, adults of *Bombus terrestris* were exposed to 5 doses of BAS 310 I in an appropriate carrier (Tween as wetting agent) placed on the dorsal bumblebee thorax. In total, 3 treatment groups were set up: 5 dose rates of the test item, 3 control groups and 4 dose rates of the reference item with 3 replicates per dose and 10 bumblebees per replicate, respectively.
Assessments of bumblebee mortality and behavioral effects were done after 4, 24, 48, 72 and 96 hours.

Endpoints: Mortality, behavioral impairments.

Reference item: Dimethoate EC 400 (BAS 152 11 I, a.s.: dimethoate, 400 g/L nominal).

Test rates: Controls: deionized water, 1.0 % (v/v) Tween solution, pure acetone; test item: 0.06, 0.13, 0.25, 0.51 and 1.01 µg BAS 310 I/bumblebee which is corresponding to 0.06, 0.13, 0.25, 0.50 and 1.00 µg a.s./bumblebee (corrected for purity); reference item: 2.5, 4.0, 6.3 and 10.0 µg dimethoate/bumblebee.

Test conditions: Temperature: 24.9°C – 25.1°C; relative humidity: 57.5% - 60.9%, photoperiod: 24 h darkness, food: 50% (w/v) sucrose solution.

Statistics: Descriptive statistics. Fisher's Exact Binominal test with Bonferroni Correction for mortality data (one-sided greater, $\alpha = 0.05$), Probit analysis using linear weight regression and linear maximum likelihood regression for calculation of the LD_{50} values.

II. RESULTS AND DISCUSSION

In the contact toxicity test, no mortality occurred in the control group treated with deionized water. After 96 hours of exposure 3.3% mortality occurred in the control groups treated with Tween solution and acetone. In the test item treatment, statistically significant mortality of 43.3, 76.7 and 100.0% occurred after thoracic application of 0.25, 0.51 and 1.01 µg BAS 310 I/bumblebee, after 96 hours (Fisher's Exact Binominal test with Bonferroni Correction, one-sided greater, $\alpha = 0.05$). Bumblebees treated with 0.06 and 0.13 µg BAS 310 I/bumblebee revealed effects on mortality that amounted to 6.7 and 20.0%, after 96 hours. Behavioral abnormalities of surviving bumblebees occurred at the higher dose rates of 1.01 and 0.50 µg BAS 310 I/bumblebee. Surviving bumblebees showed symptoms of moribundity. In the other dose rates no effects on behavior of surviving bumblebees occurred when compared to the control. The results are summarized in Table 8.3.1.1.2-2.

Table 8.3.1.1.2-2: Toxicity of BAS 310 I to bumblebee (*Bombus terrestris* L.) in a contact toxicity test

Treatment [µg BAS 310 I/bumblebee]	Mean mortality [%]				
	24 h	48 h	72 h	96 h	
Water	0.0	0.0	0.0	0.0	--
Tween	0.0	0.0	3.3	3.3	--
Acetone	0.0	0.0	0.0	3.3	0.0
0.06	0.0	0.0	3.3	6.7	3.5 [2)]
0.13	0.0	0.0	20.0*	20.0	17.2 [2)]
0.25	0.0	33.3*	40.0*	43.3*	41.4 *[2)]
0.51	0.0	50.0*	70.0*	76.7*	75.9 *[2)]
1.01	53.3*	86.7*	96.7*	100.0*	--
Endpoints					
LD_{50} (96 h) [1)]	[µg a.s./bumblebee]		[µg product/bumblebee]		
	0.29 (95% CL: 0.23 - 0.37)		0.29 (95% CL: 0.23 - 0.37)		

* Statistically significant differences compared to the control (Fisher's Exact Binominal test with Bonferroni Correction, $\alpha = 0.05$).

1) Median lethal dose calculated by Probit analysis (with 95% Confidence Limits).

2) Corrected mortality according to Schneider-Orelli, 1947.

The LD_{50} value (24 h) for the reference item in the contact toxicity test was determined to be 5.9 µg a.s./bumblebee.

III. CONCLUSION

The toxicity of BAS 310 I on bumblebees was tested in an acute contact toxicity test. The LD_{50} (96 h) was determined to be 0.29 µg product/bumblebee which is corresponding to 0.29 µg a.s./bumblebee (corrected for purity).

CA 8.3.1.2 Chronic toxicity to bees

Report:	CA 8.3.1.2/1 Sekine T., 2014a Chronic oral toxicity test of BAS 310 I on the honey bee (Apis mellifera L.) in the laboratory 2013/1132489
Guidelines:	OECD 213 (1998), CEB No. 230 (2003)
GLP:	yes (certified by Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden)

Executive Summary

In a 10-day chronic oral toxicity test, one day old worker honeybees (*Apis mellifera* L.) were exposed to a daily application of BAS 310 I diluted in the bee food (50 % (*w/w*) aqueous syrup solution). The test item BAS 310 I was daily administered to the bees in sugar solution at the following concentrations: 0.4, 1.0, 2.5, 6.25, and 15.625 mg a.s./kg food. The actual daily mean doses were 0.014, 0.033, 0.098, 0.35, and 1.13 µg a.s./bee/day after 10 days, corresponding to overall doses of 0.14, 0.33, 0.98, 3.2, and 6.8 µg a.s./bee. Untreated diet as well as diet treated with the solvent served as water and solvent controls, respectively.

After 10 days of exposure, a mean mortality of 3.3 % and 6.7 % were observed in the water and solvent control, respectively. In the test item group mean mortalities ranged between 20.0% at an actual dose of 0.04 µg a.s./bee/day and 100 % at the highest test dose. At a dose of 1.13 µg a.s./be/day bees displaying moving coordination problems and/or apathy were observed from day 1 to 3. A single surviving bee showed cramps on day 4 and 5 before dying. A dose of 0.35 µg a.s./bee/day led to moving coordination problems and/or apathy from day 2 to 9. 0.098 µg a.s./bee/day resulted in moving coordination problems and/or apathy from day 2 to 10. At a dose of 0.033 µg a.s./bee/day moving coordination problems and/or apathy were observed from day 2 to 10, except on day 8. At 0.014 µg a.s./bee/day only single bees showed uncoordinated movements on the days 2, 3, 5 and 6. Intensity and frequency of the behavioral abnormalities were dose dependent.

In a 10-day chronic toxicity feeding study with BAS 310 I, the $LD_{50\ (daily)}$ was determined to be 0.11 µg a.s./bee/day. Consequently, the LD_{50} for the entire testing period ($LD_{50\ (overall)}$) was 1.1 µg a.s./bee. This corresponds to a LC_{50} of 2.29 mg a.s./kg food.
The $NOED_{daily}$ was determined to be < 0.014 µg a.s./bee/day and the $NOED_{overall}$ < 0.14 µg a.s./bee. The NOEC was < 0.4 mg a.s./kg food.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: alpha-cypermethrin (BAS 310 I, Reg. no. 4 078 193); batch No.: COD-000595: 99.2 % (analyzed; tolerance ± 1.0 %).

B. STUDY DESIGN

Test species: Honey bee (*Apis mellifera* L.); young female worker bees (one day old); obtained from a healthy and queen-right colony, bred by IBACON, collected in the morning of use.

Test design: 10-day chronic oral feeding test in the laboratory (dose response test). Freshly emerged honey bees were daily provided with 5 freshly prepared doses of test item treated sugar solutions (50 % *w/w* Apiinvert) *ad libitum* over a period of 10 days. An untreated water control, a solvent control and a reference item were included in this study. 3 replicates per treatment, each consisting of 10 bees per test cage. Assessments of bee mortality and behavioral effects were done daily over the 10 days test period.

Endpoint: Mortality, behavioral impairments.

Reference item: BAS 152 11 I (Perfekthion EC), 400.0 g/L dimethoate (nominal).

Test concentrations: Control: untreated diet (50 % (*w*/*w*) aqueous syrup solution), solvent control: untreated diet with solvent (50 % (*w*/*w*) aqueous syrup solution plus 5 % acetone).
Test item: 15.625, 6.25, 2.5, 1.0 and 0.4 mg a.s./kg food. Target Dose Level: 0.625, 0.25, 0.1, 0.04 and 0.016 µg a.s/bee/day. Actual Mean Dose Level: 1.13, 0.35, 0.098, 0.033 and 0.014 µg a.s./bee/day (based on daily actual intake).
Reference item: 1 mg a.s./kg food. Target Dose Level: 0.04 µg a.s/bee/day. Actual mean dose level: 0.034 µg a.s/ bee/day (based on daily actual intake).

Test conditions: Temperature: 33°C – 34°C, mean relative humidity: 34 % – 78 %, photoperiod: darkness (except during assessments), food: 50 % aqueous syrup solution (Apiinvert, 30 %sucrose, 31 % glucose, 39 % fructose).

Statistics: Descriptive statistics; the LC_{50} as well as $LD_{50\ (daily)}$ and $LD_{50\ (overall)}$ values of the test item were estimated according to moving average computations (Thompson and Weil, 1952). The NOEC as well as the $NOED_{daily}$ and $NOED_{overall}$ of the test item were determined using Fisher's Exact Test (pairwise comparison, one-sided greater, $\alpha = 0.05$).

II. RESULTS AND DISCUSSION

In a chronic oral toxicity test, the test item BAS 310 I was daily administered to the honeybees via food at the following concentrations: 0.4, 1.0, 2.5, 6.25, and 15.625 mg a.s./kg food. The actual daily mean doses were 0.014, 0.033, 0.098, 0.35, and 1.13 µg a.s./bee/day after 10 days, corresponding to overall doses of 0.14, 0.33, 0.98, 3.2, and 6.8 µg a.s./bee.

Mortality occurred in all test item treated dose levels ranging from 20.0 % at a dose 0.033 µg a.s./bee/day to 100 % at the highest test dose at the test end. There was 3.3 % mortality in the water control and 6.7 % in the solvent control.
At a dose of 1.13 µg a.s./be/day, honeybees displaying moving coordination problems and/or apathy were observed from day 1 to 3. A single surviving bee showed cramps on day 4 and 5 before dying. A dose of 0.35 µg a.s./bee/day led to moving coordination problems and/or apathy from day 2 to 9. The dose of 0.098 µg a.s./bee/day resulted in moving coordination problems and/or apathy from day 2 to 10. At a dose of 0.033 µg a.s./bee/day, moving coordination problems and/or apathy were observed from day 2 to 10, except on day 8. At 0.014 µg a.s./bee/day only single bees showed uncoordinated movements on day 2, 3, 5 and 6. Intensity and frequency of the behavioral abnormalities were dose dependent. Because control mortality was ≤ 15 % and the mortality of the reference item was ≥ 50 % the test can be regarded as valid.

The results are summarized in Table 8.3.1.2-1 and Table 8.3.1.2-2.

Table 8.3.1.2-1: Mortality and behavioural abnormalities of honeybees (*Apis millifera* L.) exposed to BAS 310 F in a 10-day chronic oral toxicity test

		Treatment							
Day	**Effect mean [%] 1)**	**Control**		**BAS 310 I [mg a.s./kg food]**					
		water	**solvent**	**0.40**	**1.0**	**2.5**	**6.25**	**15.625**	**reference item**
1	mortality	3.3	0.0	0.0	0.0	6.7	0.0	0.0	0.0
	behav. abnorm.	0.0	0.0	0.0	0.0	0.0	0.0	23.3	0.0
2	mortality	3.3	0.0	6.7	6.7	23.3	20.0	30.0	0.0
	behav. abnorm.	0.0	0.0	3.3	3.3	13.3	40.0	40.0	0.0
3	mortality	3.3	0.0	13.3	13.3	30.0	63.3	93.3	3.3
	behav. abnorm.	0.0	0.0	3.3	30.0	20.0	20.0	6.7	3.3
4	mortality	3.3	0.0	20.0	13.3	33.3	66.7	96.7	13.3
	behav. abnorm.	0.0	0.0	0.0	26.7	33.3	20.0	3.3	0.0
5	mean mortality	3.3	0.0	20.0	16.7	36.7	70.0	96.7	23.3
	behav. abnorm.	0.0	0.0	13.3	13.3	16.7	16.7	3.3	0.0
6	mortality	3.3	0.0	20.0	16.7	36.7	76.7	100.0	63.3
	behav. abnorm.	0.0	0.0	10.0	13.3	23.3	20.0	0.0	10.0
7	mortality	3.3	0.0	20.0	16.7	36.7	83.3	100.0	90.0
	behav. abnorm.	0.0	3.3	0.0	13.3	23.3	13.3	0.0	10.0
8	mean mortality	3.3	6.7	20.0	16.7	40.0	86.7	100.0	96.7
	behav. abnorm.	0.0	0.0	0.0	0.0	13.3	6.7	0.0	3.3
9	mortality	3.3	6.7	20.0	16.7	46.7	86.7	100.0	100.0
	behav. abnorm.	0.0	0.0	0.0	6.7	6.7	10.0	0.0	0.0

10	mortality	3.3	6.7	30.0	20.0	46.7	100.0	100.0	100.0
	behav. abnorm.	0.0	0.0	0.0	10.0	30.0	0.0	0.0	0.0

[1)] Results are averages from three replicates (10 honeybees, each) per concentration or control, respectively.
behav. abnorm. = behavioral abnormalities.

Table 8.3.1.2-2: Cumulative mortality and toxicity endpoints of honeybees (*Apis mellifera* L.) exposed to BAS 310 I in a chronic oral toxicity test

Treatment [BAS 310 I]			Mortality after 10 days	
Actual daily mean doses [µg a.s./bee/day]	**Overall doses [µg a.s./bee]**	**Concentration [mg a.s./kg food]**	**Cumulative mortality [%]**	**Corrected cumulative mortality [%]**
Solvent control	Solvent control	Solvent control	6.7	--
0.014	0.14	0.4	30.0	25.0
0.033	0.33	1.0	20.0	14.3
0.098	0.98	2.5	46.7	42.9
0.35	3.2	6.25	100.0	100.0
1.13	6.8	15.625	100.0	100.0

Endpoints *		**10 days**
Test item dose	**$LD_{50\ (daily)}$**	0.11 µg a.s./bee/day (95 % CL: 0.071 – 0.18)
	$LD_{50\ (overall)}$	1.10 µg a.s./bee (95 % CL: 0.71 – 1.8)
	NOED (daily) 1)	< 0.014 µg a.s./bee/day
	NOED (overall) 1)	< 0.14 µg a.s./bee
Test item concentration [mg a.s./kg food]	**LC_{50}**	2.29 mg a.s./kg food (95 % CL: 1.56 – 3.36)
	NOEC 1)	< 0.4 mg a.s./kg food

* The LC_{50}/LD_{50} was estimated according to moving average computations, corrected by solvent control mortality using Abbott's formula; CL. = lower and upper confidence limits.

1) The NOEC/NOED was estimated using Fisher's Exact Test (pairwise comparison, one-sided greater, $\alpha = 0.05$).

The reference item dimethoate caused 100 % mortality at day 10 at a concentration 1 mg dimethoate/kg food (1 ppm), corresponding to an actual dose of 0.034 µg a.s./bee/day.

III. CONCLUSION

In a 10-day chronic toxicity feeding study with BAS 310 I, the $LD_{50\ (daily)}$ was determined to be 0.11 µg a.s./bee/day. Consequently, the LD_{50} for the entire testing period ($LD_{50\ (overall)}$) was 1.1 µg a.s./bee. This corresponds to a LC_{50} of 2.29 mg a.s./kg food.
The $NOED_{daily}$ was determined to be < 0.014 µg a.s./bee/day and the $NOED_{overall}$ < 0.14 µg a.s./bee. The NOEC was < 0.4 mg a.s./kg food.

CA 8.3.1.3 Effects on honeybee development and other honeybee life stages

The acute study on honeybee larvae (DocID 2014/1090816) was conducted, but was not valid and would need to be repeated in the bee season of 2015 in order to obtain a reliable endpoint. A new acute study could potentially be submitted at the end of September 2015. However, the submission already comprises a study addressing the repeated exposure to honeybee larvae (BASF DocID 2014/1162697). As just one of the bee larvae studies (acute or chronic) is required to address honeybee larvae in tier 1, the missing study is actually obsolete. Furthermore, the honeybee brood will be addressed in several higher tier studies (see chapter 10.3).

Report:	CA 8.3.1.3/1 Kleebaum K., 2014a Chronic toxicity of BAS 310 I honeybee larvae Apis mellifera L. under laboratory conditions (in vitro) 2014/1162697
Guidelines:	OECD 237 (2013) Honey bee (Apis mellifera) larval toxicity test single exposure, OECD Draft Test Guideline on Honey bee (Apis mellifera) Larval Toxicity Test Repeated Exposure (February 2014)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

In a chronic feeding toxicity test, honeybee larvae (*Apis mellifera* L.) were exposed to repeated applications of BAS 310 I (alpha-cypermethrin) diluted in the larvae food. The toxicity of the test item was determined at doses of 1.0, 2.0, 4.0, 6.0 and 8.0 ng a.s./larva (corresponding to 1.0, 2.0, 4.0, 6.0 and 8.1 ng product/larva). The concentrations of test item in the diet were 0.006, 0.013, 0.026, 0.039 and 0.052 mg a.s./kg food. Additionally, honeybee larvae were treated with dimethoate as reference item and with an untreated diet as well as with a diet treated with the solvent as controls, respectively.

In the larval toxicity test, the control group (untreated diet) showed a mortality of 0.0% after 96 hours (D7) and 5.6% after 120 hours (D8). The solvent control showed a mortality of 0.0% after 96 hours (D7) and 2.8% after 120 hours (D8). In the test item group, none of the larvae fed with 1.0, 2.0, 4.0, 6.0 and 8.0 ng a.s./larva revealed a mortality, which was statistically significantly different in comparison to the control groups after 96 hours (D7) and 120 hours (D8), respectively.

In a chronic larval toxicity test with BAS 310 I, the NOED (96 h and 120 h) was ≥ 8.0 ng a.s./larva and the corresponding NOEC (96 h and 120 h) ≥ 0.052 mg a.s./kg food. Accordingly, the LD_{50} (96 h) was determined to be > 8.0 ng a.s./larva, which is equivalent to a LC_{50} (96 h) of > 0.052 mg a.s./kg food. The LD_{50} (120 h) was calculated to be > 8.0 ng a.s./larva, which is equivalent to a LC_{50} (120 h) of > 0.052 mg a.s./kg food.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: alpha-cypermethrin (BAS 310 I, Reg. no. 4 078 193); batch no.: COD-000595: 99.2 % (analyzed; tolerance ± 1.0 %).

B. STUDY DESIGN

Test species: *Apis mellifera iberica* P. (honeybee), synchronized first instar larvae (L1, one day old); derived from three healthy and queen-right colonies; source: Beekeeper Joaquin Cordero, Cazalla (Sevilla), Spain.

Test design: One day old honeybee larvae (D1) of *Apis mellifera* L. were transferred from brood combs to polystyrene grafting cells in 48-well cell culture plates 3 days before start of the treatment. On 4 successive days (D3 to D6) the larvae were chronically exposed to BAS 310 I diluted in the larvae food (aqueous sugar solution mixed with royal jelly). After these applications no additional feeding of the larvae has been carried out. In total, 3 treatment groups were set up: 5 doses of the test item, 2 untreated control groups and 1 dose of the reference item with 3 replicates per dose and 12 larvae per replicate. Assessments of larval mortality were done after 24, 48, 72, 96 and 120 hours (respectively D4, D5, D6, D7 and D8). Additionally other observations as small body size or large quantities of remaining food after 96 and 120 hours (on D7 and D8) were noted. In an analytical phase of the study the concentration of the active substance in the test item base stock solution was determined.

Endpoints: Mortality, quantitative observations: body size, remaining food.

Reference item: Dimethoate tech. (analysed purity: 99.8 % w/w).

Test concentrations: Control: untreated diet (50% aqueous sugar solution + 50% royal jelly), solvent control: diet containing 2% v/v acetone; BAS 310 I: 1.0, 2.0, 4.0, 6.0 and 8.0 ng a.s./larva (corresponding to 1.0, 2.0, 4.0, 6.0 and 8.1 ng product/larva), the concentrations of test item in the diet were 0.006, 0.013, 0.026, 0.039 and 0.052 mg a.s./kg food; reference item: 6.2 µg dimethoate/larva.

Test conditions: Temperature: 34.0°C – 34.7°C, relative humidity: 95.2% - 95.8%, photoperiod: darkness (except during assessments), food: 50% aqueous sugar solution and 50% royal jelly.

Statistics: Descriptive statistics; Fisher's Exact Binominal test with Bonferroni Correction for mortality results and for determination of NOEC/NOED (one-sided greater, $\alpha = 0.05$).

II. RESULTS AND DISCUSSION

In the larval toxicity test, the control group (untreated diet) showed a mortality of 0.0% after 96 hours (D7) and 5.6% after 120 hours (D8). The solvent control showed a mortality of 0.0% after 96 hours (D7) and 2.8% after 120 hours (D8). In the test item group, none of the larvae fed with 1.0, 2.0, 4.0, 6.0 and 8.0 ng a.s./larva revealed a mortality, which was statistically significantly different in comparison to the control groups after 96 hours (D7) and 120 hours (D8), respectively. The results are summarized in Table 8.3.1.3-1.

Table 8.3.1.3-1: Toxicity of BAS 310 I to *Apis mellifera* L. in an acute larval toxicity test

Treatment		Mortality after 96 hours (D7)			Mortality after 120 hours (D8)		
Dosage [ng a.s./larva]	Concentration [mg a.s./kg food]	mean mortality [%] absolute	mean mortality [%] corrected [1]	mean OO [%] [2]	mean mortality [%] absolute	mean mortality [%] corrected [1]	mean OO [%] [2]
Control	Control	0.0	--	0.0	5.6	--	0.0
Solvent control	Solvent control	0.0	--	0.0	2.8	--	0.0
1.0	0.006	5.6	--	0.0	5.6	2.9	9.1
2.0	0.013	5.6	--	26.0	5.6	2.9	6.1
4.0	0.026	2.8	--	22.5	5.6	2.9	6.1
6.0	0.039	8.3	--	39.1	13.9	11.4	15.9
8.0	0.052	8.3	--	30.3	8.3	5.7	21.2

Endpoints		96 hours (D7)	120 hours (D8)
Test item dose [ng a.s./larva]	LD_{50}	> 8.0	> 8.0
	NOED [3]	≥ 8.0	≥ 8.0
Test item concentrations [mg a.s./kg food]	LC_{50}	> 0.052	> 0.052
	NOEC [3]	≥ 0.052	≥ 0.052

[1] Corrected mortality (according to Schneider-Orelli 1947).

[2] OO: Other observations (large quantities of remaining food, smaller body size of larva); Calculation are performed with non-rounded values.

[3] Fisher's Exact Binominal test with Bonferroni Correction, $\alpha = 0.05$, one sided greater.

In the reference item treatment group of 6.2 µg dimethoate/larva a mortality of 61.1% on D7 and 61.1% (corrected by control mortality: 58.8%) on D8 was determined.

III. CONCLUSION

In a chronic larval toxicity test with BAS 310 I, the NOED (96 h and 120 h) was ≥ 8.0 ng a.s./larva and the corresponding NOEC (96 h and 120 h) ≥ 0.052 mg a.s./kg food. Accordingly, the LD_{50} (96 h) was determined to be > 8.0 ng a.s./larva, which is equivalent to a LC_{50} (96 h) of > 0.052 mg a.s./kg food. The LD_{50} (120 h) was calculated to be > 8.0 ng a.s./larva, which is equivalent to a LC_{50} (120 h) of > 0.052 mg a.s./kg food.

CA 8.3.1.4 Sub-lethal effects

No new studies are available.

CA 8.3.2 Effects on non-target arthropods other than bees

No new studies are available.

CA 8.3.2.1 Effects on Aphidius rhopalosiphi

No new studies are available.

CA 8.3.2.2 Effects on Typhlodromus pyri

No new studies are available.

CA 8.4 Effects on non-target soil meso- and macrofauna

Table 8.4-1 Toxicity to non-target soil meso- and macrofauna of alpha-cypermethrin and relevant metabolites

Substance	Species	Endpoint	Value [mg/kg dry soil]	Reference	Study EU agreed?
alpha-cypermethrin	*Eisenia fetida*	LC_{50}	> 100 > 50 $_{CORR}$ [1)]	Inglesfield, Sherwood, CY-531-002	Yes
alpha-cypermethrin	*Eisenia fetida*	NOEC	≥ 4.0	2014/1135439	No, new study
3-PBA (ME 310 I 011, CL 206128)	*Eisenia fetida*	LC_{50}	214.67 107 $_{CORR}$ [2)]	Staab, 2001/1014597	Yes
DCVA (ME 310 I 001, CL 912554)	*Eisenia fetida*	LC_{50}	198.08 99 $_{CORR}$ [2)]	Staab, 2001/1014603	Yes
3-PBA (ME 310 I 011, CL 206128)	*Eisenia fetida*	NOEC	4.8	2014/1135451	No, new study
DCVA (ME 310 I 001, CL 912554)	*Eisenia fetida*	NOEC	6.25	2014/1135452	No, new study
3-PBA (ME 310 I 011, CL 206128)	*Folsomia candida*	NOEC	200	2014/1135435	No, new study
DCVA (ME 310 I 001, CL 912554)	*Folsomia candida*	NOEC	≥ 8.0	2014/1135436	No, new study
3-PBA (ME 310 I 011, CL 206128)	*Hypoaspis aculeifer*	NOEC	≥ 1000	2014/1135437	No, new study
DCVA (ME 310 I 001, CL 912554)	*Hypoaspis aculeifer*	NOEC	125	2014/1135438	No, new study
alpha-cypermethrin [3)]	*Eisenia fetida*	LC_{50}	762	Hartnik et al. 2007/1070505	No, new study
		NOEC	< 4.65		
	Folsomia candida	LC_{50}	> 258		
		NOEC	8.43		
alpha-cypermethrin [3)]	*Hypoaspis aculeifer*	EC_{10}	0.5	Sechi et al. 2013/1417920	No, new study
	Eisenia fetida	EC_{10}	4.8		

[1)] Toxicity endpoint is re-adjusted using a soil factor of 2 to address the organic content of the soil (peat 10%), and the log Pow of the substance is > 2.

[2)] Studies were carried out with 10% peat and no log P_{ow} values are available for the metabolites CL 206128 and CL 912554. Therefore, the endpoints were corrected by a safety factor of 2.

[3)] Peer-reviewed scientific studies presented as additional information (for details see chapter CA 8.4.2 below).

CA 8.4.1 Earthworms – sub-lethal effects

Report: CA 8.4.1/1
Friedrich S., 2014b
Sublethal toxicity of BAS 310 I (Alpha-Cypermethrin) to the earthworm Eisenia fetida in artificial soil
2014/1135439
Guidelines: OECD 222 (2004)
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

The effects of BAS 310 I (alpha-cypermethrin) on mortality, biomass development and reproduction were investigated in an extended laboratory study over 56 days. Five test concentrations (0.25, 0.5, 1, 2, 4 mg a.s./kg dry soil) were incorporated into the soil (10% peat) with four replicates per treatment (each containing 10 worms). An untreated control with 8 replicates was included. The reference item was tested in a separate study. Assessment of worm mortality, body weight and feeding activity was carried out after 28 days, assessment of reproduction (number of juveniles) was carried out after 56 days.

BAS 310 I (alpha-cypermethrin) did not show any statistically significant effects compared to the control on mortality and body weight. The mortality of adult worms was 0 – 2.5% in the treated variants and 1.3% in the control group. The weight change of adult worms was about 25.9 – 29.4% in the treated variants and 28.6% in the control group. The reproduction rates were not statistically significantly different compared the control up to and including a concentration of 4 mg a.s./kg dry soil. No behavioral abnormalities were observed in any of the treatment groups. The feeding activity in all the treated groups was comparable to the control.

In a 56-day earthworm reproduction study with BAS 310 I (alpha-cypermethrin), no adverse effects on survival, biomass development and reproduction could be determined at concentrations up to and including 4 mg a.s./kg dry soil. The NOEC for mortality, biomass and reproduction was determined to be ≥ 4 mg a.s./kg dry soil, the highest concentration tested.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 I (alpha-cypermethrin); batch no.: COD-000595; purity: 99.2% (tolerance ± 1.0%) (analyzed).

B. STUDY DESIGN

Test species:	Earthworm (*Eisenia fetida*), adult worms (with clitellum and weight of 254 – 443 mg/worm), approximately 3 months old; source: W. Neudorff GmbH KG, followed by in-house culture.
Test design:	56-day test in treated artificial soil according to OECD 222 (10% peat), different concentrations of the test item were incorporated into the soil; 6 treatment groups (5 test item concentrations, control); 4 replicates for the test item treatments, 8 replicates for the control, 10 worms each. Assessment of adult worm mortality, behavioral effects and biomass development after 28 days was done. Reproduction rate after additional 28 days (assessed 56 days after application) was determined.
Endpoints:	Mortality, weight change, feeding activity and reproduction rate.
Reference item:	Nutdazim 50 Flow (carbendazim, SC 500). The effects of the reference item were investigated in a separate study.
Test concentrations:	0.25, 0.5, 1, 2, 4 mg a.s./kg dry soil (based on analyzed purity).
Test conditions:	Artificial soil according to OECD 222 (with 10% peat); pH 6.00 – 6.05 at test initiation, 5.68 – 5.74 at test termination; water content 56.0 – 56.2% of maximum water holding capacity (WHC) at test initiation and 55.1 – 56.2% of WHC at test termination; temperature: 18.1 – 19.6°C; photoperiod: 16 h light : 8 h dark, light intensity: 540 lux.
Statistics:	Fisher's Exact Binomial Test for mortality ($\alpha = 0.05$, one-sided greater), Williams-t-test for weight change and reproduction ($\alpha = 0.05$, one-sided smaller).

II. RESULTS AND DISCUSSION

BAS 310 I (alpha-cypermethrin) did not show any statistically significant effects compared to the control on mortality and body weight (Fisher's Exact Binomial Test ($\alpha = 0.05$, one-sided greater) for mortality and Williams-t-test for weight change ($\alpha = 0.05$, one-sided smaller)). The mortality of adult worms was 0 – 2.5% in the treated variants and 1.3% in the control group. The weight change of adult worms was about 25.9 – 29.4% in the treated variants and 28.6% in the control group.

The reproduction rates were not statistically significantly different compared the control up to and including a concentration of 4 mg a.s./kg dry soil (Williams-t-test for reproduction ($\alpha = 0.05$, one-sided smaller)). No behavioral abnormalities were observed in any of the treatment groups. The feeding activity in all the treated groups was comparable to the control. The results are summarized in Table 8.4.1-1.

Table 8.4.1-1: Effects of BAS 310 I (alpha-cypermethrin) on earthworms (*Eisenia fetida)* in a 56-day reproduction study

BAS 310 I [mg a.s./kg dry soil]	**Control**	**0.25**	**0.5**	**1**	**2**	**4**
Mortality (day 28) [%]	1.3	0.0	2.5	0.0	0.0	2.5
Weight change (day 28) [%]	28.6	29.4	25.9	29.0	26.9	26.9
No. of juveniles (day 56)	135.8	143.3	130.3	120.8	129.0	111.5
Reproduction (day 56) [% of control]	100	105.5	95.9	89.0	95.0	82.1
Endpoints [mg a.s./kg dry soil]						
NOEC (day 28)	≥ 4					
NOEC (day 56)	≥ 4					

In a separate study the reference item Nutdazim 50 Flow had a significant effect on biomass increase and reproduction of *Eisenia fetida*. The reproduction rate was clearly inhibited by 39% and 100% compared to the control at the tested concentrations of 5 and 10 mg product/kg dry soil.

III. CONCLUSION

In a 56-day earthworm reproduction study with BAS 310 I (alpha-cypermethrin), no adverse effects on survival, biomass development and reproduction could be determined at concentrations up to and including 4 mg a.s./kg dry soil. The NOEC for mortality, biomass and reproduction was determined to be ≥ 4 mg a.s./kg dry soil, the highest concentration tested.

Report:	CA 8.4.1/2 Witte B., 2014a Effects of Reg.No. 130213 (metabolite of BAS 310 I, Alpha-cypermethrin, CL 206128) on reproduction and growth of earthworms Eisenia fetida in artificial soil 2014/1135451
Guidelines:	OECD 222 - Earthworm reproduction Test (2004), ISO 11268-2 (2012)
GLP:	yes (certified by Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden)

Executive Summary

The effects of Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin; CL 206128) on mortality, biomass development and reproduction of the earthworm *Eisenia fetida* (Annelida: Oligochaeta) were investigated in a laboratory study over 56 days. Seven concentrations (0.12, 0.31, 0.77, 1.92, 4.8, 12 and 30 mg CL 206128/kg dry soil) were incorporated into the soil with four replicates per treatment (each containing 10 worms). An untreated control with 8 replicates was included. The reference item was tested in a separate study. Assessment of worm mortality, body weight and feeding activity was carried out after 28 days, assessment of reproduction (number of juveniles) was carried out after 56 days.

No mortality was observed in any treatment group. The body weight changes of the earthworms after 4 weeks exposure to CL 206128 were not statistically significantly different compared to the control up to and including the highest test concentration of 30 mg/kg dry soil. The reproduction rates were not statistically significantly different compared to the control up to and including the test concentration of 4.8 mg/kg dry soil. At the test concentration of 12 mg/kg dry soil and above the reproduction was statistically significantly reduced compared to the control. No behavioral abnormalities were observed in any of the treatment groups. The feeding activity in all the treated groups was comparable to the control.

In an earthworm reproduction and growth study with Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) the No Observed Effect Concentration (NOEC) for mortality, growth and feeding activity of the earthworm *Eisenia fetida* was equal to or greater than 30 mg/kg dry soil, *i.e.* the highest concentration tested. The No Observed Effect Concentration (NOEC) for reproduction was determined to be 4.8 mg/kg dry soil. The EC_{10} was determined to be 8.3 mg/kg dry soil and the EC_{20} was determined to be 35.0 mg/kg dry soil.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin; CL 206128); batch no.: AC12251-34; purity: 100.0% (tolerance ± 1.0%).

B. STUDY DESIGN

Test species: Earthworm (*Eisenia fetida*), adult worms (with clitellum and weight range 300 to 600 mg), approximately 10 months old, source: in-house culture.

Test design: 56-day test in treated artificial soil prepared according to OECD 222; different concentrations of the test item were incorporated into the soil; 8 treatment groups (7 test item concentrations, control); 4 replicates for the test item treatments and 8 replicates for the control with 10 worms each. Assessment of adult worm mortality, behavioral effects and biomass development was carried out after 28 days exposure of adult worms in treated artificial soil. Reproduction rate (number of offspring) was assessed after additional 28 days (assessed 56 days after application).

Endpoints: Mortality, weight change, feeding activity and reproduction rate.

Reference item: Carbendazim (499 g/kg nominal). The effects of the reference item were investigated in a separate study.

Test concentrations: Control, 0.12, 0.31, 0.77, 1.92, 4.8, 12 and 30 mg Reg. No. 130 213/kg dry soil.

Test conditions: Artificial soil according to OECD 222; pH 6.0 to 6.2 at test initiation, pH 6.0 to 6.1 at test termination; water content 51.7% - 57.4% of maximum water holding capacity (WHC) at test initiation and 54.6% - 59.6% of the maximum WHC at test termination; temperature: 18°C - 22°C; photoperiod: 16 h light : 8 h dark, light intensity: within the range of 400 - 800 lux.

Statistics: Descriptive statistics, Williams t-test (α = 0.05, two-sided for body weight change and one-sided smaller for reproduction). Probit analysis for EC_x values.

II. RESULTS AND DISCUSSION

No mortality was observed in any treatment group. The body weight changes of the earthworms after 4 weeks exposure to CL 206128 were not statistically significantly different compared to the control up to and including the highest test concentration of 30 mg/kg dry soil (Williams t-test, α = 0.05, two-sided).
The reproduction rates were not statistically significantly different compared to the control up to and including the test concentration of 4.8 mg/kg dry soil (Williams t-test, α = 0.05, one-sided smaller). At the test concentration of 12 mg/kg dry soil and above the reproduction was statistically significantly reduced compared to the control. No behavioral abnormalities were observed in any of the treatment groups. The feeding activity in all the treated groups was comparable to the control. The results are summarized in Table 8.4.1-2.

Table 8.4.1-2: Effects of CL 206128 on earthworms (*Eisenia fetida)* in a 56-day reproduction study

CL 206128 [mg/kg dry soil]	**Control**	**0.12**	**0.31**	**0.77**	**1.92**	**4.8**	**12**	**30**
Mortality (day 28) [%]	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Weight change (day 28) [%]	43.9	39.7	42.1	41.3	41.8	43.5	41.4	42.5
No. of juveniles (day 56)	221	226	233	195	224	211	194 *	178 *
Reproduction (day 56) [% of control]	--	102.0	105.1	87.9	101.2	95.5	87.7	80.3
Food consumption [g]	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0
Endpoints [mg/kg dry soil]								
NOEC (day 28) (mortality and weight)	≥ 30							
NOEC (day 56) (reproduction)	4.8							
EC_{10} [1)]	8.3							
EC_{20} [1)]	35.0							

* Statistically significantly different compared to the control (Williams-t-test, α = 0.05, one-sided smaller).
1) Probit analysis.

In the most recent test with the reference item, there were statistically significant effects on reproduction at a concentration of 1.95 mg carbendazim/kg soil and higher, which is in line with the guideline OECD. The EC_{50} for reproduction was calculated to be 1.87 mg carbendazim/kg dry soil.

III. CONCLUSION

In an earthworm reproduction and growth study with Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) the No Observed Effect Concentration (NOEC) for mortality, growth and feeding activity of the earthworm *Eisenia fetida* was equal to or greater than 30 mg/kg dry soil, *i.e.* the highest concentration tested. The No Observed Effect Concentration (NOEC) for reproduction was determined to be 4.8 mg/kg dry soil. The EC_{10} was determined to be 8.3 mg/kg dry soil and the EC_{20} was determined to be 35.0 mg/kg dry soil.

Report:	CA 8.4.1/3 Witte B., 2014b Effects of Reg.No. 4080830 (metabolite of BAS 310 I, Alpha-cypermethrin; CL 912554) on reproduction and growth of earthworms Eisenia fetida in artificial soil 2014/1135452
Guidelines:	OECD 222 (2004), ISO 11268-2 (2012)
GLP:	yes (certified by Hessisches Ministerium fuer Umwelt, Energie, Landwirtschaft und Verbraucherschutz, Wiesbaden)

Executive Summary

The effects of Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin; CL 912554) on mortality, biomass development and reproduction of the earthworm *Eisenia fetida* (Annelida: Oligochaeta) were investigated in a laboratory study over 56 days. Five concentrations (6.25, 12.5, 25, 50 and 100 mg CL 912554/kg dry soil) were incorporated into the soil with four replicates per treatment each containing 10 worms. An untreated control with 8 replicates was included. The reference item was tested in a separate study. Assessment of worm mortality, body weight and feeding activity was carried out after 28 days, assessment of reproduction (number of juveniles) was carried out after 56 days.

No mortality was observed in any treatment group. The body weight changes were not statistically significantly different compared to the control up to and including the concentration of 12.5 mg/kg dry soil. At the concentration of 25 mg/kg dry soil and above the body weight change was statistically significantly reduced compared to the control. The reproduction rates were not statistically significantly different compared to those in the control at the concentration of 6.25 mg/kg dry soil. At the concentration of 12.5 mg/kg dry soil and above the reproduction was statistically significantly reduced compared to the control. No behavioral abnormalities were observed in any of the treatment groups, except at the highest test item treated group of 100 mg/kg dry soil, where the adult worms after 28 days were tensed and coiled. The feeding activity in all the treated groups was comparable to the control, except at the highest test concentration of 100 mg/kg dry soil the food intake was slightly reduced.

In a 56-day earthworm reproduction study with Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin; CL 912554) the NOEC for mortality was equal to or greater than 100 mg/kg dry soil, the NOEC for biomass was 12.5 mg/kg dry soil and the NOEC for reproduction was 6.25 mg/kg dry soil. The EC_{10} was determined to be 14.8 mg/kg dry soil, the EC_{20} was determined to be 21.0 mg/kg dry soil and the EC_{50} was determined to be 40.8 mg/kg dry soil.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin; CL 912554); batch no.: AC12717-65; purity: 99.5% (tolerance ± 1.0%) (analyzed).

B. STUDY DESIGN

Test species: Earthworm (*Eisenia fetida*), adult worms (with clitellum and weight 300 - 576 mg), approximately 7 months old, source: in-house culture.

Test design: 56-day test in treated artificial soil prepared according to OECD 222; different concentrations of the test item were incorporated into the soil; 6 treatment groups (5 test item concentrations, control); 4 replicates for the test item treatments and 8 replicates for the control with 10 worms each. Assessment of adult worm mortality, behavioral effects and biomass development was carried out after 28 days exposure of adult worms in treated artificial soil. Reproduction rate (number of offspring) was assessed after additional 28 days (assessed 56 days after application).

Endpoints: Mortality, weight change, feeding activity and reproduction rate.

Reference item: Carbendazim (499 g/kg nominal). The effects of the reference item were investigated in a separate study.

Test concentrations: Control, 6.25, 12.5, 25, 50 and 100 mg CL 912554/kg dry soil.

Test conditions: Artificial soil according to OECD 222; pH 5.6 - 5.7 at test initiation, pH 5.8 - 6.1 at test termination; water content 53.4% - 53.8% of maximum water holding capacity (WHC) at test initiation and 54.7% to 60.9% of the maximum WHC at test termination; temperature: 18°C - 22°C; photoperiod: 16 h light : 8 h dark, light intensity: 400 - 800 lux.

Statistics: Descriptive statistics. Fisher's Exact Test ($\alpha = 0.05$, one-sided smaller) for mortality, Williams t-test ($\alpha = 0.05$, two-sided) for body weight change, Bonferroni-Welch t-test ($\alpha = 0.05$, one-sided smaller) for reproduction, Probit analysis (EC_x).

II. RESULTS AND DISCUSSION

No mortality was observed in any treatment group.

The body weight changes were not statistically significantly different compared to the control up to and including the concentration of 12.5 mg/kg dry soil. At the concentration of 25 mg/kg dry soil and above the body weight change was statistically significantly reduced compared to the control (Williams t-test, $\alpha = 0.05$, two-sided).

The reproduction rates were not statistically significantly different compared to those in the control at the concentration of 6.25 mg/kg dry soil. At the concentration of 12.5 mg/kg dry soil and above the reproduction was statistically significantly reduced compared to the control (Bonferroni-Welch t-test, $\alpha = 0.05$, one-sided smaller).

No behavioral abnormalities were observed in any of the treatment groups, except at the highest test item treated group of 100 mg/kg dry soil, where the adult worms after 28 days were tensed and coiled. The feeding activity in all the treated groups was comparable to the control, except at the highest test concentration of 100 mg/kg dry soil the food intake was slightly reduced. Results are summarized in Table 8.4.1-3.

Table 8.4.1-3: Effects of CL 912554 on earthworms (*Eisenia fetida)* in a 56-day reproduction study

CL 912554 [mg/kg dry soil]	**Control**	**6.25**	**12.5**	**25**	**50**	**100**
Mortality (day 28) [%]	0.0	0.0	0.0	0.0	0.0	0.0
Weight change (day 28) [%]	55.6	50.0	53.9	40.1 *	37.0 *	23.4 *
No. of juveniles (day 56)	223	264	177 **	170 **	110 **	1 **
Reproduction (day 56) [% of control]	--	118.0	79.2	76.1	49.0	0.2
Food consumption [g]	25.0	25.0	25.0	25.0	25.0	22.0
Endpoints [mg/kg dry soil]						
NOEC (day 28) (mortality)	≥ 100					
NOEC (day 28) (weight)	12.5					
NOEC (day 56) (reproduction)	6.25					
EC_{10}[1)] (day 56)	14.8					
EC_{20}[1)] (day 56)	21.0					
EC_{50}[1)] (day 56)	40.8					

* Statistically significantly different compared to the control (Williams-t-test, $\alpha = 0.05$, two-sided).
** Statistically significantly different compared to the control (Bonferroni-Welch t-test, $\alpha = 0.05$, one-sided smaller).
1) Probit analysis.

In the most recent test with the reference item carbendazim, there were statistically significant effects on reproduction at a concentration of 1.95 mg carbendazim/kg dry soil and higher, which is in line with the guideline OECD 222. The EC_{50} for reproduction was calculated as 1.87 mg carbendazim/kg dry soil.

III. CONCLUSION

In a 56-day earthworm reproduction study with Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) the NOEC for mortality was equal to or greater than 100 mg/kg dry soil, the NOEC for biomass was 12.5 mg/kg dry soil and the NOEC for reproduction was 6.25 mg/kg dry soil. The EC_{10} was determined to be 14.8 mg/kg dry soil, the EC_{20} was determined to be 21.0 mg/kg dry soil and the EC_{50} was determined to be 40.8 mg/kg dry soil.

From the performed literature search, the following peer-reviewed scientific study on non-target soil invertebrates was considered relevant for the terrestrial risk assessment of alpha-cypermethrin. Due to the unrealistic high concentration of alpha-cypermethrin tested in the study it was classified as being "not reliable" (Reliability Index (RI) 3). For details please see the literature search and evaluation files also provided in the submission package for Annex I Renewal. Nevertheless, the study is quite reasonably performed and in general to a certain extent comparable with current guidelines (e.g. ISO 11267:1999). Thus, the endpoints for the relevant species *Eisenia fetida* and *Folsomia candida* are presented as additional information for the terrestrial risk assessment and have not been evaluated previously.

Report:	CA 8.4.1/4 Hartnik T. et al., 2007b Toxicity of the pesticide Alpha-Cypermethrin to four soil nontarget invertebrates and implications for risk assessment 2007/1070505
Guidelines:	none
GLP:	no

Executive Summary

The aim of this toxicity study was to assess acute and chronic effects of alpha-cypermethrin on different soil invertebrate species. In this abstract the focus is on the effects of alpha-cypermethrin on the relevant test species, i.e. the earthworm *Eisenia fetida* and the soil non-target arthropod *Folsomia candida.* Other issues discussed in the paper (*i.e.* the comparison to aquatic toxicity and the effects of alpha-cypermethrin on other non-target soil invertebrate species) are not summarized here, because there are no comparable endpoints, no relevant species and not relevant for the risk assessment.

Tests were conducted in test soil spiked with alpha-cypermethrin. All tests were performed according to standardized OECD or International Organization for Standardization (ISO) guidelines except that natural soil was used instead of artificial OECD soil. For the assay with *E. fetida*, soil concentrations of 7.5, 30, 100, 300, and 1000 mg a.s./kg dry soil and for the assay with *F. candida*, nominal soil concentrations of 0.9, 3, 9, 30, 90, and 300 mg a.s./kg dry soil were used. For all tests, the test item concentrations were mixed thoroughly into the batch of soil.

All tests passed the validity criteria of the test protocols. For adult *E. fetida*, a LC_{50} value of 762 mg a.s./kg dry soil, was estimated. For springtails, the LC_{50} value was considerably higher than 258 mg a.s./kg dry soil, the highest concentration used in the test (for springtails, because lethality was less than 15% in this treatment.

Bioassays with the two invertebrate species *Eisenia fetida* and *Folsomia candida* in soil showed that despite a low acute toxicity, alpha-cypermethrin has a profound chronic toxicity toward soil-living organisms.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (technical grade; purity: 97.6%) provided by Inter-Trade Denmark (Bindslev, Denmark).

B. STUDY DESIGN

Test species: Earthworms *Eisenia fetida* (adults aged 24 - 30 weeks with a well-developed clitellum), springtails *Folsomia candida* (aged 10-12 days).

Test design: Different concentrations of the test item were incorporated into the test soil. For each bioassay, five (for *E. fetida*) and six (for *F. candida*) concentration levels, ranging from 7.5 to 1000 and from 0.9 to 300 mg alpha-cypermethrin/kg dry soil for *E. fetida* and *F. candida*, were mixed homogenously into the soil, respectively.
Reproduction test with *E. fetida:* A modified version of OECD 222 was used. Three replicates of 600 g of spiked soil per concentration level were used; for the controls, six replicates were prepared. Each replicate contained 10 worms weighing 372 ± 28 mg at test start. Cocoon production and mortality were assessed after 28 d.
Reproduction test with *F. candida:* A modified version of ISO 11267 was used in natural test soil. Four replicates for each test item concentration, six replicates for the control, each replicate containing 10 collembolans. Mortality (number of surviving adults) and number of living juveniles were assessed after 21 d.

Endpoints: Mortality and effects on reproduction were determined.

Test concentrations: *E. fetida*: 7.5, 30, 100, 300, and 1000 mg a.s./kg dry soil;
F. candida: 0.9, 3.0, 9.0, 30, 90, and 300 mg a.s./kg dry soil;

Test conditions: All tests were performed in natural test soil.
E. fetida: temperature: 20 ±2°C; photoperiod: 16:8-h light:dark; culture medium: mixture of sphagnum peat, potting soil, and chopped vegetables.

F. candida: 20 ±2°C; photoperiod: 16:8-h light:dark; cultured on artificial substrate; food: baker's yeast.

Test soil:

Parameter	**Soil type: sandy loam**
Organic carbon (g/100 g dry soil)	2.2
pH	6.5
Sand /silt / clay (%)	72.4 / 17.5 / 10.1
Water holding capacity (%)	34
Cation exchange capacity ($mmol_c$ (+)/kg)	132
Base saturation (%)	100
Exchangeable acidity ($mmol_c$ (+)/kg)	< 0.1

Statistics: Descriptive statistics; ANOVA followed by Dunnett's test ($\alpha = 0.05$) for determination of no-observed-effect concentration (NOEC) for sublethal endpoints (JMP, Ver 5.0; SAS Institute, Cary, NC, USA). Non-linear regression analysis for dose-response functions (GraphPad Prism, Ver 4.0; GraphPad Software, San Diego, CA, USA). Median effect concentrations (EC_{50}) was estimated by iteration using the three-parametric sigmoidal function.

II. RESULTS AND DISCUSSION

All tests passed the validity criteria of the test protocols. For adult *E. fetida*, a LC_{50} value of 762 mg a.s./kg dry soil, was estimated. For springtails, the LC_{50} value was considerably higher than 258 mg a.s./kg dry soil, the highest concentration used in the test (for springtails, because lethality was less than 15% in this treatment. A summary of the results is presented in the table below.

Table 8.4.1-4: Calculated toxicity parameters (median lethal concentration LC_{50}, median effective concentration EC_{50}, 10% effective concentration EC_{10}, and no-observed-effect concentrations, NOEC) of alpha-cypermethrin for *Eisenia fetida* and*Folsomia candida* [#]

Test species	LC_{50} [mg a.s./kg dry soil]	EC_{50} [mg a.s./kg dry soil]	EC_{10} [mg a.s./kg dry soil]	NOEC [mg a.s./kg dry soil]
Eisenia fetida	762 (510 - 1139)	31.0 (14.8 - 68.0)	1.57 (-1.01 - 4.15)	< 4.65
Folsomia candida	> 258	60.3 (32.0 - 113)	3.69 (-0.87 - 8.24)	8.43

[#] Ranges in parentheses represent the 95% confidence intervals.

III. CONCLUSION

Bioassays with the two invertebrate species (*Eisenia fetida* and *Folsomia candida*) in soil showed that despite a low acute toxicity, alpha-cypermethrin has a profound chronic toxicity toward soil-living organisms.

From the performed literature search, the following peer-reviewed scientific study on non-target soil invertebrates was considered as relevant for the terrestrial risk assessment of alpha-cypermethrin. Due to the unrealistic high concentration of alpha-cypermethrin tested in the study and the extrapolated endpoints it was classified as being “not reliable” (Reliability Index (RI) 3), as these endpoints are not measured but had been calculated far below the tested concentrations. For details please see the literature search and evaluation files also provided in the submission package for Annex I Renewal. Nevertheless, the study, but the study may be used as supplemental information as it was in general quite reasonably performed and to a certain extent comparable with current guidelines (e.g. OECD guideline 222). Thus, the endpoints for the relevant species *Eisenia fetida* and *Hypoaspis aculeifer* are presented as additional information for the terrestrial risk assessment and have not been evaluated previously.

Report: CA 8.4.1/5
Sechi V. et al., 2013b
Species composition of a soil invertebrate multi-species test system determines the level of ecotoxicity
2013/1417920
Guidelines: none
GLP: no
(certified by none)

Executive Summary

The system was capable of detecting population dynamics and species interactions not present in single species tests, and included interactions between faunal and microbial communities. More relevant here was the aim of the study to test the performance of two different non-target species in soil spiked with a range of alpha-cypermethrin concentrations. In this abstract the focus is on the effects of alpha-cypermethrin on the relevant test species, i.e. the earthworm *Eisenia fetida* and the soil non-target arthropod *Hypoaspis aculeifer*. The effects on other species which are not relevant for the risk assessment are not summarized here as there are no comparable endpoints as well as extrapolated (no measured) endpoints. Therefore these species as well as their endpoints are not relevant for the risk assessment.

The two different communities responded differently to the insecticide treatments. Alpha-cypermethrin had a general negative effect on the soil fauna, i.e. the soil non-target arthropod *Hypoaspis aculeifer and* the fresh body weight of *E. fetida*. The two different communities responded in a different manner to the insecticide. A positive effect due to the presence of the earthworms in the COM-F was observed, where in contrast the predator mite was more abundant in the COM-C.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Alpha-cypermethrin (technical grade; purity, 97.6%), provided by Inter-Trade Denmark (Bindslev, Denmark)

B. STUDY DESIGN

Test species relevant:

Functional group	Species	No. individuals	No. individuals m^{-2}
Predator mite	*Hypoaspis aculeifer*	10 females	1270
Lumbricid detrivore	*Eisenia fetida*	5 juveniles	635

Test design:	Two different faunal communities (COM-F, COM-C) were constructed, including three trophic levels and different functional groups. Both communities included the predator mite *Hypoaspis aculeifer,* but differed concerning the earthworms as only into one juveniles of the earthworm *Eisenia fetida* were added (COM-F). Besides a non-spiked control, three different alpha-cypermethrin concentrations were used. Due to technical constraints, four replicates were set up for the 1 and 25 mg a.s./kg dry soil treatment groups and eight replicates were set up for the control and the 5 mg a.s./dry soil treatments. Microarthropods were extracted from four representative soil subsamples (120 g of fresh soil each) from each cylinder using a Macfadyen high gradient extractor. Individuals of *E. fetida* were carefully collected from the soil by hand sorting. After extraction, all species were identified and counted under a binocular microscope.
Endpoints:	EC_{10} (extrapolated not measured), EC_{50}, population development (abundance).
Test concentrations:	Untreated control, three concentrations of alpha-cypermethrin: 1, 5, and 25 mg/kg dry soil.
Test conditions:	Each mesocosm was covered by a transparent plastic lid with holes for ventilation and stored at 15°C with a 12:12 h light-dark periodicity. Mesocosms were weighted and moisture was adjusted weekly by spraying water onto the soil surface in order to maintain water content of approximately 20%. Every week lost food was replenished, and moistened dried cattle manure (1:3 (*w:w*) manure:water) was added weekly on the surface of the soil in order to avoid food limitation.
Test soil:	Natural agricultural soil; sandy loam (38.0% coarse sand, 40.6% fine sand, 10.0% silt, 12.3% clay); 30% water holding capacity; pH 6.2.
Analytics:	PLFA (phospholipid fatty acid) analysis to determine main taxa for the soil microbial community. The total amount of PFLA was used as an estimator of the microbial biomass. GC-MS analyses to determine the concentration of alpha-cypermethrin in the soil after 8 weeks (20 subsamples of 5 g soil each) in both communities.
Statistics:	Descriptive statistics; mixed effect models; F-test; linear regression of log-transformed abundances of a species was used to estimate EC_{10} and EC values using the estimate function of SAS/PROC NLMIXED (SAS Institute Inc., 2011); quantitative PLFA data was analyzed using a GLM procedure to test for main effects and interactions, and Bonferroni post-test to compare effect of individual insecticide concentrations. Principal component analysis was performed on the data sets from the PLFA analysis using the Unscrambler software v. 7.6 (CAMO ASA, NO).

II. RESULTS AND DISCUSSION

The two different communities responded differently to the insecticide treatments. Alpha-cypermethrin had a general negative effect on the soil fauna, i.e. the soil non-target arthropod *Hypoaspis aculeifer and* the fresh body weight of *E. fetida*. The two different communities responded in a different manner to the insecticide. A positive effect due to the presence of the earthworms in the COM-F was observed, where in contrast the predator mite was more abundant in the COM-C.

Soil mites
The predatory mite *H. aculeifer* was significantly affected by the insecticide ($p < 0.001$), too.

Earthworms
No mortality of earthworms was observed in any of the test containers. The biomass of *E. fetida* increased with time at all alpha-cypermethrin concentrations. A negative effect at 5 and 25 mg a.s./kg dry soil was observed ($p < 0.05$). After eight weeks, the biomass was reduced by 16% at the highest tested concentration.

Soil microbial community
At week 4, the total PLFA content in soil was lower (varying from 10 to 100%) in the COM-C compared to COM-F, tending to decrease (not significantly) with increasing insecticide concentration. After 8 weeks, it was similar in the two communities and unaffected by the insecticide concentration. There were no systematic trends in relation to insecticide treatment (no significant main effect of insecticide concentration).

A summary of the results is presented in the table below.

Table 8.4.1-5: EC_{10} and the EC_{50} values of the tested species after week 4 and 8 *

Species	Time [week]	Community	EC_{10} [mg a.s./kg dry soil]	95% C.L	EC_{50} [mg a.s./kg dry soil]	95% C.L
Hypoaspis aculeifer	4	COM-C	2.7	1.2 – 4.2	13.4	5.8 – 21.1
	8	COM-C	0.5	0.3 – 0.7	3.2	1.7 – 4.7
	8	COM-F	2.8	0.1 – 5.5	14.1	0.6 – 27.6
Eisenia fetida	4	COM-F	4.8	1.6 – 7.9	23.8	8.2 – 39.5
	8	COM-F	11.5	2.0 – 21.0	> 25	--

* EC_{50} and EC_{10} values have been estimated only for significant declines of species abundances with alpha-cypermethrin concentration and similarly for the fresh body weight of *E. fetida*.
C.L. = Confidence limits.
COM-C = Community with *E. crypticus.*
COM-F = Community with *E. fetida.*

Table 8.4.1-6: Effect on individual species abundances in the mesocosms by the factors (time, community and alpha-cypermethrin) and interactions of those

Species	Time	COM	alpha-cypermethrin	Time*Conc.	COM*Conc.	Time* COM
Hypoaspis aculeifer		* ↓	* ↓		*	
Eisenia fetida	* ↑		* ↓	*		

COM: indicates community effect, COM-F vs. COM-C effect; Time: indicates different effects after 4 and 8 weeks; alpha-cypermethrin: Pesticide effect (Conc. 0, 1, 5, 25 mg/kg dry soil). Significance is indicated by:*: $P < 0.05$; **: $P < 0.01$. ↑ and ↓ indicate positive (↑) or negative effects (↓) driven by the factors on the population abundance.

III. CONCLUSION

The two different communities responded differently to the insecticide treatments. Alpha-cypermethrin had a negative effect on the soil non-target arthropod *Hypoaspis aculeifer and* the fresh body weight of *E. fetida*. A positive effect due to the presence of the earthworms in the COM-F was observed, where in contrast the predator mite was more abundant in the COM-C.

CA 8.4.2 Effects on non-target soil meso- and macrofauna (other than earthworms)

Report: CA 8.4.2/1
Friedrich S., 2014a
Effects of Reg.No. 130213 (metabolite of BAS 310 I, alpha-Cypermethrin, CL 206128) on the reproduction of the collembolan Folsomia candida
2014/1135435
Guidelines: OECD 232 (2009), ISO 11267 (1999)
GLP: yes
(certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

The effects of Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) on mortality and reproduction of the collembolan, *Folsomia candida*, were investigated in a chronic laboratory experiment over a time period of 28 days. The test item was mixed into artificial soil at rates of 25, 50, 100, 200 and 400 mg CL 206128/kg dry soil. For the solvent control, the soil was left untreated. 4 replicates were prepared for the treatment groups and 8 replicates were prepared for the solvent control, each containing 10 springtails. Assessment of mortality and reproduction was made 28 days after treatment.

Statistically significant differences on mortality compared to the solvent control were observed at a concentration of 400 mg CL 206128/kg dry soil. Mortality rates of 0% - 50.0% were recorded in the test item treatment groups. 3.8% parental mortality was observed in the solvent control. Statistically significant differences compared to the solvent control on number of juveniles were recorded at a concentration of 400 mg CL 206128/kg dry soil. The mean reproduction in the solvent control reached 659 juveniles. Reproduction rates in 25, 50, 100, 200 and 400 mg CL 206128/kg dry soil were 642, 664, 669, 583 and 473 juveniles, respectively.

In a 28-day collembolan reproduction study with Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) the NOEC based on mortality and reproduction was determined to be 200 mg/kg dry soil.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128), batch No.: AC12251-34, analyzed purity: 100% (tolerance ± 1.0%)

B. STUDY DESIGN

Test species: Collembola (*Folsomia candida*), age: 9 - 12 days; source: in-house culture.

Test design: 28-day test in treated artificial soil (with 5% peat); different concentrations of the test item were mixed homogeneously into the soil which was filled in glass vessels before collembolans were introduced on top of the soil. 6 treatment groups (5 test item concentrations, solvent control) were set up with 4 replicates for the test item treatments and 8 replicates for the solvent control and each containing 10 juvenile collembolans. Feeding of collembola with about 2 mg dry yeast at the beginning of the test for each test vessel and additional feeding on day 14. Assessment of adult collembolans mortality, reproduction rate (number of juveniles) and behavioral effects was carried out after 28 days.

Endpoints: Mortality and reproduction rate after 28 days.

Reference item: Boric acid (100% analyzed). The effects of the reference item were investigated in a separate study.

Test concentrations: Control, 25, 50, 100, 200 and 400 mg CL 206128/kg dry soil.

Test conditions: Artificial soil according to OECD 232 with a peat content of 5%; water content: 58.5% of the maximum water holding capacity (WHC) at test initiation and 57.4% - 57.8% of the maximum WHC at test termination; pH 6.00 - 6.12 at test initiation, pH 5.65 - 5.74 at test termination; temperature 18.1°C - 19.3°C; photoperiod: 16 h light : 8 h dark; light intensity 520 lux.

Statistics: Descriptive statistics. Fisher's Exact Binominal test with Bonferroni Correction for mortality ($\alpha = 0.05$, one-sided greater), Williams-t-test for reproduction ($\alpha = 0.05$, one-sided smaller).

II. RESULTS AND DISCUSSION

Statistically significant differences on mortality compared to the solvent control were observed at a concentration of 400 mg CL 206128/kg dry soil (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha = 0.05$, one-sided greater). Mortality rates of 0% - 50.0% were recorded in the test item treatment groups. 3.8% parental mortality was observed in the solvent control. Statistically significant differences compared to the solvent control on number of juveniles were recorded at a concentration of 400 mg CL 206128/kg dry soil (Williams-t-test, $\alpha = 0.05$, one-sided smaller). The mean reproduction in the solvent control reached 659 juveniles. Reproduction rates in 25, 50, 100, 200 and 400 mg CL 206128/kg dry soil were 642, 664, 669, 583 and 473 juveniles, respectively. The results are summarized in Table 8.4.2-1.

Table 8.4.2-1: Effects of Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) on collembola (*Folsomia candida*) in a 28-day reproduction study

CL 206128 [mg/kg dry soil]	Solvent control	25	50	100	200	400
Mortality (day 28) [%]	3.8	5.0	5.0	0.0	5.0	50.0 *
No. of juveniles (day 28)	659	642	664	669	583	473
Reproduction in [%] of control (day 28)	100	97	101	102	89	72 *
Endpoints [mg/kg dry soil]						
NOEC (mortality/reproduction)	200					
LOEC (mortality/reproduction)	400					
LC_{50} (95% confidence limits) [1)]	400 (284 - 563)					
EC_{50} [2)]	> 400					

1) Based on Moving average computation.

2) Based on estimation of the data.

* statistically significantly different compared to the control [Fisher's Exact Binominal test with Bonferroni Correction for mortality ($\alpha = 0.05$, one-sided greater), Williams-t-test for reproduction ($\alpha = 0.05$, one-sided smaller)].

III. CONCLUSION

In a 28-day collembolan reproduction study with Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) the NOEC based on mortality and reproduction was determined to be 200 mg/kg dry soil.

Report:	CA 8.4.2/2 Friedrich S., 2015a Effects of Reg. No. 4080830 (metabolite of BAS 310 I, Alpha-Cypermethrin, CL 912554) on the reproduction of the collembolan Folsomia candida 2014/1135436
Guidelines:	OECD 232 (2009), ISO 11267 (1999)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

The effects of Reg. No. 4080830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) on mortality and reproduction of the collembola, *Folsomia candida*, were investigated in a chronic laboratory experiment over a time period of 28 days. The test item was mixed into artificial soil at rates of 0.25, 0.5, 1.0, 2.0, 4.0 and 8.0 mg CL 912554/kg dry soil. For the solvent control, the soil was left untreated. 4 replicates were prepared for the treatment groups and 8 replicates were prepared for the solvent control, each containing 10 springtails. Assessment of mortality and reproduction was made 28 days after treatment.

No statistically significant effect on parental mortality was found for any concentration tested. Mortality rates of 2.5% - 5.0% were recorded in the test item treatment groups. In the solvent control the mortality rate was 2.5%. No statistically significant effects on the number of juveniles compared to the solvent control were recorded at any concentration tested. The mean reproduction in the solvent control reached 951 juveniles. Reproduction rates in 0.25, 0.5, 1.0, 2.0, 4.0 and 8.0 mg CL 912554/kg dry soil were 943, 920, 946, 907, 950 and 880 juveniles, respectively.

In a 28-day collembolan reproduction study with Reg. No. 4080830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) the NOEC based on mortality and reproduction was determined to be ≥ 8.0 mg/kg dry soil.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Reg. No. 4080830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554), batch No.: AC12717-65, analyzed purity: 99.5% (tolerance ± 1.0%)

B. STUDY DESIGN

Test species: Collembola (*Folsomia candida*), age: 9 - 12 days; source: in-house culture.

Test design: 28-day test in treated artificial soil (with 5% peat); different concentrations of the test item were mixed homogeneously into the soil which was filled in glass vessels before collembolans were introduced on top of the soil. 7 treatment groups (6 test item concentrations, solvent control) were set up with 4 replicates for the test item treatments and 8 replicates for the solvent control and each containing 10 juvenile collembolans. Feeding of collembola with about 2 mg dry yeast at the beginning of the test for each test vessel and on day 14. Assessment of adult mortality, reproduction rate (number of juveniles) and behavioral effects was carried out after 28 days.

Endpoints: Mortality and reproduction rate after 28 days.

Reference item: Boric acid (100% analyzed). The effects of the reference item were investigated in a separate study.

Test concentrations: Control, 0.25, 0.5, 1.0, 2.0, 4.0 and 8.0 mg CL 912554/kg dry soil.

Test conditions: Artificial soil according to OECD 232 with a peat content of 5%; water content: 58.3% - 58.8% of the maximum water holding capacity (WHC) at test initiation and 57.1% - 58.1% of the maximum WHC at test termination; pH 6.05 - 6.11 at test initiation, pH 5.72 - 5.75 at test termination; temperature 18.5°C - 21.8°C; photoperiod: 16 h light : 8 h dark; light intensity 460 lux.

Statistics: Descriptive statistics. Fisher's Exact Binominal test with Bonferroni Correction for mortality ($\alpha = 0.05$, one-sided greater), Williams-t-test for reproduction ($\alpha = 0.05$, one-sided smaller).

II. RESULTS AND DISCUSSION

No statistically significant effect (Fisher's Exact Binomial Test with Bonferroni Correction, α = 0.05, one-sided greater) on parental mortality was found for any concentration tested. Mortality rates of 2.5% - 5.0% were recorded in the test item treatment groups. In the solvent control the mortality rate was 2.5%. No statistically significant effects (Williams-t-test, α = 0.05, one-sided smaller) on the number of juveniles compared to the solvent control were recorded at any concentration tested. The mean reproduction in the solvent control reached 951 juveniles. Reproduction rates in 0.25, 0.5, 1.0, 2.0, 4.0 and 8.0 mg CL 912554/kg dry soil were 943, 920, 946, 907, 950 and 880 juveniles, respectively. The results are summarized in Table 8.4.2-1.

Table 8.4.2-2: Effects of Reg. No. 4080830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) on collembola (*Folsomia candida*) in a 28-day reproduction study

CL 912554 [mg/kg dry soil]	Solvent control	0.25	0.5	1.0	2.0	4.0	8.0
Mortality (day 28) [%]	2.5	5.0	2.5	2.5	2.5	2.5	2.5
No. of juveniles (day 28)	951	943	920	946	907	950	880
Reproduction in [%] of control (day 28)	100	99	97	99	95	100	92
Endpoints [mg/kg dry soil]							
NOEC (mortality/reproduction)	≥ 8.0						
LOEC (mortality/reproduction)	> 8.0						
LC_{50} [1)]	> 8.0						
EC_{50} [1)]	> 8.0						

1) Based on estimation of the data.

III. CONCLUSION

In a 28-day collembolan reproduction study with Reg. No. 4080830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) the NOEC based on mortality and reproduction was determined to be ≥ 8.0 mg/kg dry soil.

Report:	CA 8.4.2/3 Schulz L., 2014a Effects of Reg.No. 130213 (metabolite of BAS 310 I, alpha-Cypermethrin, CL 206128) on the reproduction of the predatory mite Hypoaspis aculeifer 2014/1135437
Guidelines:	OECD 226 (2008)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

The effects of Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) on mortality and reproduction of the predatory mite *Hypoaspis aculeifer* were investigated in a chronic laboratory study over a time period of 14 days. The test item was mixed into artificial soil at concentrations of 62.5, 125, 250, 500 and 1000 mg/kg dry soil. For the control treatment, the soil was prepared with acetone (solvent control). 8 replicates and 4 replicates were prepared for the solvent control and test item treatment groups, respectively, each containing 10 adult predatory mites (females). Assessment of mortality and reproduction was carried out after the 14-day exposure of the predatory mites.

Mortality rates of 0.0 - 7.5% were recorded in the test item treatment groups. In the solvent control the mortality rate was 0.0%. The observed mortality rates for adults mortality in the test item treatment groups compared to the control were not statistically significantly different. Differences between the behavior and the morphology of the mites in the solvent control and the test item treatment groups were not observed. Reproduction rates in the 62.5, 125, 250, 500 and 1000 mg/kg dry soil were 240.3, 252.3, 239.8, 229.3 and 248.0 juveniles, respectively. The mean reproduction in the solvent control reached 243.0 juveniles. The test item showed no statistically significantly adverse effects on reproduction at all tested concentrations compared to the control.

In a 14-day *Hypoaspis aculeifer* reproduction study with Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128), the LC_{50} and the EC_{50} were estimated to be higher than 1000 mg/kg dry soil. The NOEC for mortality and for reproduction was determined to be ≥ 1000 mg/kg dry soil, the highest concentration tested.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128), batch No.: AC12251-34, analyzed purity: 100.0% (tolerance ± 1.0%).

B. STUDY DESIGN

Test species: *Hypoaspis aculeifer* (Canestrini), adult mites with an age difference of 2 days; source: in-house culture.

Test design: The effects of Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128) on mortality and reproduction of the predatory mite *Hypoaspis aculeifer* were investigated in a chronic laboratory study over a time period of 14 days according to OECD 226 (2008). The different concentrations of the test item were homogeneously mixed into the artificial soil which was then used to fill glass vessels after which the predatory mites were introduced on top of the soil; 6 treatment groups (5 test item concentrations, solvent control); 8 replicates for the solvent control group and 4 replicates for the test item treatment groups each with 10 predatory mites. Feeding of mites with *Tyrophagus putrescentiae* at the beginning and *ad libitum* during the test. Assessment of adult mortality and reproduction effects was carried out after 14 days.

Endpoints: Mortality and reproduction rate after 14 days.

Reference item: Dimethoate (analyzed purity: 99.8%, tolerance ± 1.0%). The effects of the reference item were investigated in a separate study.

Test concentrations: Control, test item: 62.5, 125, 250, 500 and 1000 mg/kg dry soil.

Test conditions: Artificial soil according to OECD 226, pH 5.5 - 6.0 at test initiation, pH 5.5 - 5.8 at test termination; water content at test initiation 48.04 - 50.83% of maximum water holding capacity (WHC) and 45.45 - 48.42% of maximum WHC at test termination; temperature 19.7 - 21.1°C; photoperiod: 16 h light : 8 h dark; light intensity: 504 lux.

Statistics: Fisher's Exact Binomial Test with Bonferroni Correction for mortality ($\alpha = 0.05$, one-sided greater), Dunnett-t-test for reproduction ($\alpha = 0.05$, one-sided smaller).

II. RESULTS AND DISCUSSION

Mortality rates of 0.0 - 7.5% were recorded in the test item treatment groups. In the solvent control the mortality rate was 0.0%. The observed mortality rates for adults mortality in the test item treatment groups compared to the control were not statistically significantly different (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha = 0.05$, one-sided greater). Differences between the behavior and the morphology of the mites in the solvent control and the test item treatment groups were not observed.

Reproduction rates in the 62.5, 125, 250, 500 and 1000 mg/kg dry soil were 240.3, 252.3, 239.8, 229.3 and 248.0 juveniles, respectively. The mean reproduction in the solvent control reached 243.0 juveniles. The test item showed no statistically significantly adverse effects on reproduction at all tested concentrations compared to the control (Dunnett-t-test, $\alpha = 0.05$, one-sided smaller). The results are summarized in Table 8.4.2-3.

Table 8.4.2-3: Effects of CL 206128 on *Hypoaspis aculeifer* mortality and reproduction (14 days)

CL 206128 [mg/kg dry soil]	**Control**	**62.5**	**125**	**250**	**500**	**1000**
Mortality [%]	0.0	0.0	2.5	2.5	7.5	0.0
No. of juveniles (day 14)	243.0	240.3	252.3	239.8	229.3	248.0
Reproduction (day 14) [% of control]	100	99	104	99	94	102
Endpoints [mg/kg dry soil]						
NOEC (mortality)	≥ 1000					
NOEC (reproduction)	≥ 1000					
LC_{50}[1)]	> 1000					
EC_{50}[1)]	> 1000					

1) Based on estimation of the data.

In a separate study the EC_{50} (reproduction) of the reference item dimethoate (analyzed purity: 99.8%, tolerance ± 1.0%) was calculated to be 6.2 mg/kg dry soil.

III. CONCLUSION

In a 14-day *Hypoaspis aculeifer* reproduction study with Reg. No. 130 213 (metabolite of BAS 310 I, alpha-cypermethrin, CL 206128), the LC_{50} and the EC_{50} were estimated to be higher than 1000 mg/kg dry soil. The NOEC for mortality and for reproduction was determined to be ≥ 1000 mg/kg dry soil, the highest concentration tested.

Report:	CA 8.4.2/4 Schulz L., 2014b Effects of Reg.No. 4080830 (metabolite of BAS 310 I, alpha-Cypermethrin, CL 912554) on the reproduction of the predatory mite Hypoaspis aculeifer 2014/1135438
Guidelines:	OECD 226 (2008)
GLP:	yes (certified by Saechsisches Staatsministerium fuer Umwelt und Landwirtschaft, Dresden, Germany)

Executive Summary

The effects of Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) on mortality and reproduction of the predatory mite *Hypoaspis aculeifer* were investigated in a chronic laboratory study over a time period of 14 days. The test item was mixed into artificial soil at concentrations of 31.25, 62.5, 125, 250 and 500 mg/kg dry soil. For the control treatment, the soil was prepared with acetone (solvent control). 8 replicates and 4 replicates were prepared for the solvent control and test item treatment groups, respectively, each containing 10 adult predatory mites (females). Assessment of mortality and reproduction was carried out after the 14-day exposure of the predatory mites.

Mortality rates of 2.5 - 12.5% were recorded in the test item treatment groups. In the solvent control the mortality rate was 2.5%. The observed mortality rates for adults mortality in the test item treatment groups compared to the control were not statistically significantly different. Differences between the behavior and the morphology of the mites in the solvent control and the test item treatment groups were not observed. Reproduction rates in the concentrations of 31.25, 62.5, 125, 250 and 500 mg/kg dry soil were 183.8, 207.8, 176.0, 156.0 and 107.5 juveniles, respectively. The mean reproduction in the solvent control reached 199.5 juveniles. The test item showed no statistically significantly adverse effects on reproduction up to and including 125 mg/kg dry soil. However, CL 912554 caused statistically significant effects on reproduction at 250 and 500 mg/kg dry soil.

In a 14-day *Hypoaspis aculeifer* reproduction study with Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554), the LC_{50} and the EC_{50} were estimated to be higher than 500 mg/kg dry soil, the highest concentration tested. The NOEC for mortality and for reproduction were determined to be ≥ 500 mg/kg dry soil and 125 mg/kg dry soil, respectively.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554), batch No.: AC12717-65, analyzed purity: 99.5% (tolerance ± 1.0%).

B. STUDY DESIGN

Test species: *Hypoaspis aculeifer* (Canestrini), adult mites with an age difference of 2 days; source: in-house culture.

Test design: The effects of Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554) on mortality and reproduction of the predatory mite species *Hypoaspis aculeifer* were investigated in a chronic laboratory study over a time period of 14 days according to OECD 226 (2008).
The different concentrations of the test item were homogeneously mixed into the artificial soil which was then used to fill glass vessels after which the predatory mites were introduced on top of the soil; 6 treatment groups (5 test item concentrations, solvent control); 8 replicates for the solvent control group and 4 replicates for the test item treatment group each with 10 predatory mites. Feeding of mites with *Tyrophagus putrescentiae* at the beginning and *ad libitum* during the test. Assessment of adult mortality and reproduction effects was carried out after 14 days.

Endpoints: Mortality and reproduction rate after 14 days.

Reference item: Dimethoate (analyzed purity: 99.8%, tolerance ± 1.0%). The effects of the reference item were investigated in a separate study.

Test concentrations: Control, 31.25, 62.5, 125, 250 and 500 mg/kg dry soil.

Test conditions: Artificial soil according to OECD 226, pH 5.5 - 5.7 at test initiation, pH 5.5 - 5.6 at test termination; water content at test initiation 48.91 - 52.42% of maximum water holding capacity (WHC) and 48.11 - 49.83% of maximum WHC at test termination; temperature 19.7 - 20.7°C; photoperiod: 16 h light : 8 h dark; light intensity: 520 lux.

Statistics: Fisher's Exact Binomial Test with Bonferroni Correction for mortality ($\alpha = 0.05$, one-sided greater), Williams-t-test for reproduction ($\alpha = 0.05$, one-sided smaller), Probit analysis for EC_x estimation.

II. RESULTS AND DISCUSSION

Mortality rates of 2.5 - 12.5% were recorded in the test item treatment groups. In the solvent control the mortality rate was 2.5%. The observed mortality rates for adults mortality in the test item treatment groups compared to the control were not statistically significantly different (Fisher's Exact Binomial Test with Bonferroni Correction, $\alpha = 0.05$, one-sided greater). Differences between the behavior and the morphology of the mites in the solvent control and the test item treatment groups were not observed.

Reproduction rates in the concentrations of 31.25, 62.5, 125, 250 and 500 mg/kg dry soil were 183.8, 207.8, 176.0, 156.0 and 107.5 juveniles, respectively. The mean reproduction in the solvent control reached 199.5 juveniles. The test item showed no statistically significantly adverse effects on reproduction up to and including 125 mg/kg dry soil. However, CL 912554 caused statistically significant effects on reproduction at 250 and 500 mg/kg dry soil (Williams-t-test, $\alpha = 0.05$, one-sided smaller). The results are summarized in Table 8.4.2-4.

Table 8.4.2-4: Effects of CL 912554 on *Hypoaspis aculeifer* mortality and reproduction (14 days)

CL 912554 [mg/kg dry soil]	**Control**	**31.25**	**62.5**	**125**	**250**	**500**
Mortality [%]	2.5	2.5	5.0	5.0	12.5	5.0
No. of juveniles (day 14)	199.5	183.8	207.8	176.0	156.0 *	107.5 *
Reproduction (day 14) [% of control]	100	92	104	88	78	54
Endpoints [mg/kg dry soil]						
NOEC (mortality)	≥ 500					
NOEC (reproduction)	125					
LC_{50} [1)]	> 500					
EC_{50} [1)]	> 500					

* Statistically significantly different compared to the control (Williams-t-test, $\alpha = 0.05$, one-sided smaller).
1) Based on estimation of the data.

In a separate study the EC_{50} (reproduction) of the reference item dimethoate (analyzed purity: 99.8%, tolerance ± 1.0%) was calculated to be 6.2 mg/kg dry soil.

III. CONCLUSION

In a 14-day *Hypoaspis aculeifer* reproduction study with Reg. No. 4 080 830 (metabolite of BAS 310 I, alpha-cypermethrin, CL 912554), the LC_{50} and the EC_{50} were estimated to be higher than 500 mg/kg dry soil, the highest concentration tested. The NOEC for mortality and for reproduction were determined to be ≥ 500 mg/kg dry soil and 125 mg/kg dry soil, respectively.

CA 8.4.2.1 Species level testing

Not triggered; no new studies are available.

From the performed literature search, two peer-reviewed scientific studies on non-target soil invertebrates were considered relevant and reliable (with restrictions; RI 2) for the terrestrial risk assessment of alpha-cypermethrin. Please refer to chapter CA 8.4.1 for details.

CA 8.5 Effects on nitrogen transformation

Table 8.5-1 Toxicity to nitrogen transformation of alpha-cypermethrin and metabolites

Test substance	Endpoint	NOEC [mg/kg dry soil]	Reference	Study EU agreed?
alpha-cypermethrin	Effects on nitrogen transformation	0.4	Review report SANCO/4335/2000-final, Feb. 2004; Monograph Vol. 1, Sep. 1999	Yes
alpha-cypermethrin	Effects on nitrogen transformation	100	Koelzer, 2006/1008040	No, new study
CL 206128 (mPBA, ME 310 I 011)	Effects on nitrogen transformation	0.02	Review report SANCO/4335/2000-final, Feb. 2004	Yes
CL 912554 (DCVA, ME 310 I 001)	Effects on nitrogen transformation	0.0144	Review report SANCO/4335/2000-final, Feb. 2004	Yes

Report:	CA 8.5/1 Koelzer U., 2006a Effects of BAS 310 I (Reg.No. 4078193) on the activity of the soil microflora - Nitrogen transformation test (ECx) 2006/1008040
Guidelines:	OECD 216 (2000)
GLP:	yes (certified by Ministerium fuer Umwelt und Verkehr Baden-Wuerttemberg, Stuttgart)

Executive Summary

The effect of BAS°310 I on nitrogen transformation was tested in a lucerne-enriched silty sand soil. BAS°310 I was applied to samples of the soil in a laboratory at test concentrations of 1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil. The treated soils and untreated control were incubated at 20 ± 2 °C in the dark for 28 days. Triplicate samples of each treatment were removed for analysis of NH_4-nitrogen and NO_3-nitrogen 0, 7, 14 and 28 days after application.

No adverse effects of BAS°310°I on nitrogen transformation could be observed at all test item concentrations (1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil) after 28 days. Only slight deviations from the control between + 0.728% and + 4.95% were measured after 28 days.

Based on the results of this study, BAS°310°I caused no adverse effects (< 25% deviation from control according to OECD 216) on the soil nitrogen transformation (measured as NO_3-N production) in silty sand soil tested at concentrations up to 100 mg a.s./kg dry soil after 28 days of incubation.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 I, batch no. COD-000166, content: alpha-cypermethrin (BAS°310°I, Reg. No. 4 078 193), purity: 99.3% (w/w).

B. STUDY DESIGN

Test soil: Biologically active agricultural soil: silty sand soil, pH 6.75, 1.17% C_{org}, WHC: 27.3%.

Test design: Determination of the N-transformation (NO_3-nitrogen production) in soil enriched with lucerne meal (concentration in soil 0.5%). Comparison of test item treated soil with a non-treated soil. NH_4-nitrogen formed from organically bound nitrogen and NO_3-nitrogen formed from the nitrification process were determined using an Ionanalyser (Orion Research Inc). Sampling scheme: 0, 7, 14 and 28 days after treatment. Sub-samples (3 replicates) were withdrawn from the bulk batches and subjected to the measurement.

Endpoints: Effects on the NO_3-nitrogen production 0, 7, 14 and 28 days after application.

Test concentrations: Control, 1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil; test concentrations related to a soil depth of 5 cm and a soil density of 1.5 g/cm^3.

Reference item: Dinoterb (purity: 97.5%). The reference item was tested at 13.3 mg/kg dry soil in a separate study.

Test conditions: Soil moisture: approx. 45 % of maximum water holding capacity, water content: 11.1 %, pH 6.75. Soil samples were incubated at 20 ± 2 °C while stored in glass bottles in the dark.

Statistics: Descriptive statistics. Dunnett's t-Test ($\alpha = 0.05$).

II. RESULTS AND DISCUSSION

No adverse effects of BAS°310°I on nitrogen transformation could be observed at all test item concentrations (1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil) after 28 days. Only slight deviations from the control between + 0.728% and + 4.95% were measured after 28 days. Effects in all test concentrations were not statistically significantly different compared to the control (Dunnett's t-Test; $\alpha = 0.05$). The results are summarized below in Table 8.5-2.

Table 8.5-2: Effects of BAS°310°I on soil micro-organisms (nitrogen transformation) on days 7, 14 and 28 of incubation

Soil (days)	Control	1.0 mg BAS°310°I/kg dry soil		3.3 mg BAS°310°I/kg dry soil		10 mg BAS°310°I/kg dry soil		33 mg BAS°310°I/kg dry soil		100 mg BAS°310°I/kg dry soil	
	NO_3-N [mg/kg dry soil	NO_3-N [mg/kg dry soil]	% Deviation from control [1)]	NO_3-N [mg/kg dry soil]	% Deviation from control [1)]	NO_3-N [mg/kg dry soil]	% Deviation from control [1)]	NO_3-N [mg/kg dry soil]	% Deviation from control [1)]	NO_3-N [mg/kg dry soil]	% Deviation from control [1)]
Silty sand soil (7 d)	32.5	32.1	-1.07	33.2	+2.06	31.5	-2.93	31.4	-3.32	33.0	+1.67
Silty sand soil (14 d)	41.4	43.7	+5.66	43.8	+5.82	42.5	+2.58	42.9	+3.70	40.5	-2.09
Silty sand soil (28 d)	43.5	45.4	+4.22	43.8	+0.728	45.7	+4.95	45.1	+3.55	45.5	+4.47

[1)] Based on NO_3-nitrogen production; - = inhibition, + = stimulation.

The reference item Dinoterb produced a stimulation of nitrogen transformation of +34.7% at 13.3 mg/kg dry soil after 28 days incubation.

III. CONCLUSION

Based on the results of this study, BAS°310°I caused no adverse effects (< 25% deviation from control according to OECD 216) on the soil nitrogen transformation (measured as NO_3-N production) in silty sand soil tested at concentrations up to 100 mg a.s./kg dry soil after 28 days of incubation.

CA 8.6 Effects on terrestrial non-target higher plants

No new studies are available.

CA 8.6.1 Summary of screening data

No new studies are available.

CA 8.6.2 Testing on non-target plants

No new studies are available.

CA 8.7 Effects on other terrestrial organisms (flora and fauna)

The following study is no data requirement for the risk assessment and is presented as additional information to the dossier.

Report:	CA 8.7/1 Koelzer U., 2006b Effects of BAS 310 I (Reg.No. 4078193) on the activity of the soil microflora - Carbon transformation test (ECx) 2006/1008041
Guidelines:	OECD 217 (2000)
GLP:	yes (certified by Ministerium fuer Umwelt und Verkehr Baden-Wuerttemberg, Stuttgart)

Executive Summary

The effect of BAS°310 I on carbon transformation was tested in a silty sand soil. BAS°310 I was applied to samples of the soil in a laboratory at test concentrations of 1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil. The treated soils and untreated control were incubated at 20 ± 2 °C in the dark for 28 days. Triplicate samples of each treatment were removed for analysis of carbon transformation (oxygen consumption) 0, 7, 14, 28 and 56 days after application.

No adverse effects of BAS°310°I on carbon transformation could be observed at all test item concentrations (1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil) after 28 days. An additional measurement after 56 days confirmed that result. Only slight deviations from the control between -1.37% and -10.1% were measured after 56 days.

Based on the results of this study, BAS°310°I caused no adverse effects (< 25% deviation from control according to OECD 217) on the soil carbon transformation (measured as oxygen consumption) in silty sand soil tested at concentrations up to 100 mg a.s./kg dry soil after 28 days of incubation. An additional measurement after 56 days confirmed that result.

I. MATERIAL AND METHODS

A. MATERIALS

Test item: BAS 310 I, batch no. COD-000166, content: alpha-cypermethrin (BAS°310°I, Reg. No. 4 078 193), purity: 99.3% (w/w).

B. STUDY DESIGN

Test soil:	Biologically active agricultural soil: silty sand soil, pH 6.75, 1.17% C_{org}, WHC: 27.3%.
Test design:	Determination of carbon transformation in soil after addition of glucose (concentration in soil 0.3%). Comparison of test item treated soil with a non-treated soil. A "OxiTop Control®" system was used to measure the oxygen consumption over a period of maximum 24 hours at different sampling intervals. Sampling scheme: 0, 7, 14, 28 and 56 days after treatment, sub-samples were withdrawn from the bulk batches and subjected to the measurement.
Endpoints:	Effects on O_2 consumption 0, 7, 14, 28 and 56 days after application.
Test concentrations:	Control, 1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil; test concentrations related to a soil depth of 5 cm and a soil density of 1.5 g/cm^3.
Reference item:	Dinoterb (purity: 97.5%). The reference item was tested at 13.3 mg/kg dry soil in a separate study.
Test conditions:	Soil moisture: approx. 45% of maximum water holding capacity, water content: 11.4%; pH 6.75. Soil samples were incubated at 20 ± 2 °C while stored in glass bottles in the dark.
Statistics:	Descriptive statistics. Multiple-t-test, Dunnett's t-Test ($\alpha = 0.05$).

II. RESULTS AND DISCUSSION

No adverse effects of BAS°310°I on carbon transformation could be observed at all test item concentrations (1.0, 3.3, 10, 33 and 100 mg BAS°310°I/kg dry soil) after 28 days. An additional measurement after 56 days confirmed that result. Only slight deviations from the control between -1.37% and -10.1% were measured after 56 days. Effects of all test concentrations were not statistically significantly different compared to the control (Multiple-t-test, Dunnett's t-Test; $\alpha = 0.05$). The results are summarized below in Table 8.7-1.

Table 8.7-1: Effects of BAS°310°I on soil micro-organisms (carbon transformation) on days 7, 14 and 28 and 56 of incubation

Soil (days)	Control	1.0 mg BAS°310°I/kg dry soil		3.3 mg BAS°310°I/kg dry soil		10 mg BAS°310°I/kg dry soil		33 mg BAS°310°I/kg dry soil		100 mg BAS°310°I/kg dry soil	
	O_2 consumption [mg/h/kg dry soil]	O_2 consumption [mg/h/kg dry soil]	% Deviation from control [1)]	O_2 consumption [mg/h/kg dry soil]	% Deviation from control [1)]	O_2 consumption [mg/h/kg dry soil]	% Deviation from control [1)]	O_2 consumption [mg/h/kg dry soil]	% Deviation from control [1)]	O_2 consumption [mg/h/kg dry soil]	% Deviation from control [1)]
Silty sand soil (7 d)	4.84	4.19	-13.4	3.70	-23.6	4.44	-8.35	4.24	-12.5	3.38	-30.2
Silty sand soil (14 d)	4.28	4.56	+6.40	3.85	-10.1	4.72	+10.3	4.08	-4.82	4.18	-2.30
Silty sand soil (28 d)	3.77	4.38	+16.1	3.94	+4.38	4.31	+14.1	3.30	-12.6	3.89	+3.15
Silty sand soil (56 d)	4.68	4.25	-9.13	4.43	-5.27	4.62	-1.37	4.21	-10.1	4.61	-1.50

[1)] Based on O_2 consumtion; - = inhibition, + = stimulation.

The reference item Dinoterb produced an inhibition of carbon transformation of 35.1% at 13.3 mg/kg dry soil after 28 days incubation.

III. CONCLUSION

Based on the results of this study, BAS°310°I caused no adverse effects (< 25% deviation from control according to OECD 217) on the soil carbon transformation (measured as oxygen consumption) in silty sand soil tested at concentrations up to 100 mg a.s./kg dry soil after 28 days of incubation. An additional measurement after 56 days confirmed that result.

CA 8.8 Effects on biological methods for sewage treatment

The results of the already peer-reviewed and accepted study are still valid and they are summarized in Table 8.8-1. No new study has been performed.

Table 8.8-1: Effects on biological methods for sewage treatment

Test item	Study type	Endpoint [mg a.s./L]	Reference (BASF DocID)	EU agreed
BAS 310 I (alpha-cypermethrin)	Respiration inhibition test (inhibition of oxygen consumption activated sludge from wastewater plant)	No inhibitory effect on microbial respiration up to 1000 mg a.s./L	AL-123-092 amended by AL-690-005	yes

CA 8.9 Monitoring data

According to the knowledge of the applicant, there are currently no published ecotoxicological monitoring data available for alpha-cypermethrin or its metabolites, which would provide additional knowledge on the ecotoxicological assessment not covered by this dossier.

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Alpha-Cypermethrin

Document M-CA, Section 9

LITERATURE DATA

Compiled by:

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Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 9 LITERATURE DATA

A literature search on alpha-cypermethrin and the common product trade names was performed by the BASF Group Information Center. The Literature Search Report on alpha-cypermethrin describes the general search and evaluation process as well as details on search profiles, search histories and summary tables.

The complete search report is provided in K-CA 9 (BASF DocID 2014/1320877).

The first step of the search result processing based on summary records was done by the Information Center and involved the separation into "hits" and "ballast" (obviously irrelevant records). The “ballast” was not further processed.

The “hits” were further evaluated by the scientific experts and categorized into “not relevant”, “not reliable”, and “used for dossier”. This is documented in EXCEL files which are attached to the search report in K-CA 9 with the file names as listed below (alphabetical order):

Analytics and Product Chemistry:	
	Alpha-Cypermethrin Literature Analytics and Product Chemistry
Ecotoxicology:	Alpha-cypermethrin Literature Ecotox aquatic
	Alpha-cypermethrin Literature Ecotox general
	Alpha-cypermethrin Literature Ecotox terrestric
	Alpha-cypermethrin Literature Ecotox wildlife
E-fate:	Alpha-cypermethrin Literature E-fate
Consumer Safety:	Alpha-cypermethrin Literature Metabolism and Residues in Animals
	Alpha-cypermethrin Literature Metabolism and Residues in Plants
Toxicology:	Alpha-cypermethrin Literature Toxicology

The hits in “Alpha-cypermethrin Literature Analytics and Product Chemistry”, did not contribute to the risk assessment as no literature was identified as either relevant or reliable for further evaluation; no new endpoints or new relevant analytical residue methods have been identified. Hence, none were therefore further discussed in the dossier.

The hits in “E-fate" did not contribute to the risk assessment, but three publications were further discussed in the dossier as supportive information.

The hits in “Metabolism and Residues in Animals”, “Metabolism and Residues in Plants”, “Ecotox general “, “Ecotox wildlife “, did not contribute to the risk assessment and were therefore not further discussed in the dossier.

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Alpha-Cypermethrin

Document M-CA, Section 10

CLASSIFICATION AND LABELLING OF THE ACTIVE SUBSTANCE

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[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

Table of Contents

CA 10 CLASSIFICATION AND LABELLING OF THE ACTIVE SUBSTANCE

The following harmonized classification and labelling was adopted for alpha-cypermethrin:

Legislation	Classification	Labelling	Concentration limits
Regulation (EC) No 1272/2008	Hazard class and category code: Acute Tox. 3 (oral) STOT SE 3 STOT RE 2 Aquatic Acute 1 Aquatic Chronic 1 Hazard statement code: H301, H335, H373, H400, H410	Pictogram signal word code: GHS06 GHS08 GHS09 Danger Hazard statement code: H301, H335, H373, H410	M-factor = 1000

New studies were performed leading to a change of classification for acute inhalation toxicity. Therefore, BASF proposed the following classification and labelling for alpha-cypermethrin:

Legislation	Classification	Labelling	Concentration limits
Regulation (EC) No 1272/2008	Hazard class and category code: Acute Tox. 3 (oral) **Acute Tox. 4 (inhalation)** STOT SE 3 STOT RE 2 Aquatic Acute 1 Aquatic Chronic 1 Hazard statement code: H301, **H332,** H335, H373, H400, H410	Pictogram signal word code: GHS06 GHS08 GHS09 Danger Hazard statement code: H301, **H332,** H335, H373, H410	M-factor = 1000