Committee for Risk Assessment

RAC

Opinion

proposing harmonised classification and labelling
at EU level of

imiprothrin (ISO); reaction mass of: [2,4-dioxo-(2-propyn-1-yl)imidazolidin-3-yl]methyl(1R)-cis-chrysanthemate; [2,4-dioxo-(2-propyn-1-yl)imidazolidin-3-yl]methyl(1R)-trans-chrysanthemate

EC Number: 428-790-6
CAS Number: 72963-72-5

CLH-O-0000001412-86-197/F

Adopted

9 March 2018

9 March 2018

CLH-O-0000001412-86-197/F

OPINION OF THE COMMITTEE FOR RISK ASSESSMENT ON A DOSSIER PROPOSING HARMONISED CLASSIFICATION AND LABELLING AT EU LEVEL

In accordance with Article 37 (4) of Regulation (EC) No 1272/2008, the Classification, Labelling and Packaging (CLP) Regulation, the Committee for Risk Assessment (RAC) has adopted an opinion on the proposal for harmonised classification and labelling (CLH) of:

Chemical name: **imiprothrin (ISO); reaction mass of: [2,4-dioxo-(2-propyn-1-yl)imidazolidin-3-yl]methyl(1R)-cis-chrysanthemate; [2,4-dioxo-(2-propyn-1-yl)imidazolidin-3-yl]methyl(1R)-trans-chrysanthemate**

EC Number: **428-790-6**

CAS Number: **72963-72-5**

The proposal was submitted by **United Kingdom** and received by RAC on **13 February 2017.**

In this opinion, all classification and labelling elements are given in accordance with the CLP Regulation.

PROCESS FOR ADOPTION OF THE OPINION

United Kingdom has submitted a CLH dossier containing a proposal together with the justification and background information documented in a CLH report. The CLH report was made publicly available in accordance with the requirements of the CLP Regulation at ***http://echa.europa.eu/harmonised-classification-and-labelling-consultation/*** on **14 March 2017**. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by **28 April 2017**.

ADOPTION OF THE OPINION OF RAC

Rapporteur, appointed by RAC: **Marja Pronk**

The opinion takes into account the comments provided by MSCAs and concerned parties in accordance with Article 37(4) of the CLP Regulation and the comments received are compiled in Annex 2.

The RAC opinion on the proposed harmonised classification and labelling was adopted on **9 March 2018** by **consensus**.

Classification and labelling in accordance with the CLP Regulation (Regulation (EC) 1272/2008)

	Index No	International Chemical Identification	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors and ATE	Notes
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Pictogram, Signal Word Code(s)	Hazard state-ment Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	613-259-00-5	imiprothrin (ISO); reaction mass of: [2,4-dioxo-(2-propyn-1-yl)imidazolidin-3-yl]methyl(1R)-cis-chrysanthemate; [2,4-dioxo-(2-propyn-1-yl)imidazolidin-3-yl]methyl(1R)-trans-chrysanthemate	428-790-6	72963-72-5	Acute Tox. 4* Aquatic Acute 1 Aquatic Chronic 1	H302 H400 H410	GHS07 GHS09 Wng	H302 H410			
Dossier submitters proposal	613-259-00-5		428-790-6	72963-72-5	**Modify** Acute Tox. 4 **Add** Acute Tox. 4 Repr. 2 **Retain** Aquatic Acute 1 Aquatic Chronic 1	**Retain** H302 H400 H410 **Add** H332 H361d	**Retain** GHS07 GHS09 Wng **Add** GHS08	**Retain** H302 H410 **Add** H332 H361d		**Add** Oral: ATE = 550 mg/kg bw Inh.: ATE = 1.4 mg/L M=10 M=10	
RAC opinion	613-259-00-5		428-790-6	72963-72-5	**Modify** Acute Tox. 4 **Add** Acute Tox. 4 Carc. 2 STOT SE 2 **Retain** Aquatic Acute 1 Aquatic Chronic 1	**Retain** H302 **Add** H332 H351 H371 (nervous system; oral, inhalation) **Retain** H400 H410	**Retain** GHS07 GHS09 Wng **Add** GHS08	**Retain** H302 H410 **Add** H332 H351 H371 (nervous system; oral, inhalation)		**Add** Oral: ATE = 550 mg/kg bw Inh.: ATE = 1.4 mg/L M=10 M=10	
Resulting Annex VI entry if agreed by COM	613-259-00-5		428-790-6	72963-72-5	Carc. 2 Acute Tox. 4 Acute Tox. 4 STOT SE 2 Aquatic Acute 1 Aquatic Chronic 1	H351 H332 H302 H371 (nervous system; oral, inhalation) H400 H410	GHS07 GHS08 GHS09 Wng	H302 H332 H351 H371 (nervous system; oral, inhalation) H410		Oral: ATE = 550 mg/kg bw Inh.: ATE = 1.4 mg/L M=10 M=10	

GROUNDS FOR ADOPTION OF THE OPINION

RAC general comment

The Dossier Submitter (DS) proposed the classification of imiprothrin, a synthetic pyrethroid insecticide, for acute toxicity (oral, inhalation), reproductive toxicity (developmental toxicity) and hazard to the aquatic environment. Studies on acute toxicity (dermal), mutagenicity, carcinogenicity, reproductive toxicity (fertility) and STOT (single and repeated exposure) were also made available and assessed. However, the DS considered that the results for these latter hazard classes were conclusive, but not sufficient for classification.

HUMAN HEALTH HAZARD EVALUATION

RAC evaluation of acute toxicity

Summary of the Dossier Submitter's proposal

Acute toxicity studies via the dermal, oral and inhalation routes of exposure were conducted in rats and mice using imiprothrin and the Manufacturing Use Product. The Manufacturing Use Product is a mixture (50% imiprothrin, 50% isopropyl myristate). The DS considered that the studies using the Manufacturing Use Product could also be used to support the classification proposal given the low acute toxicity of isopropyl myristate. Similar LD_{50} values were obtained in the oral and dermal acute toxicity studies with the Manufacturing Use Product and the pure substance imiprothrin.

In addition to the acute toxicity studies an acute neurotoxicity study with imiprothrin in rats was also available.

Acute oral toxicity

Acute Oral Studies with Imiprothrin

Rat study (Study SGT-20-0026, 1992)

In a GLP OECD TG 401 comparable study, SD rats (5/sex/group) were treated by gavage with imiprothrin in corn oil at dose levels of 500, 1000, 1400, 2000, 2800 or 4000 mg/kg bw (males) and 500, 700, 1000, 1400, 2000 or 2800 mg/kg bw (females). At a dose of 500 mg/kg bw, no abnormal signs were observed. Deaths in females were reported at ≥700 mg/kg bw. In males, death was reported at ≥1000 mg/kg bw. Observations of abnormal clinical signs included tremor, a decrease in spontaneous activity, excretion of an oily substance, urinary incontinence, stained fur, ataxic gait, prone position, lateral position and irregular respiration. The animals that died during the observation period showed retention of an oily substance in the stomach and autolysis of the intestine. In surviving animals, no treatment-related gross pathological changes were observed. The LD_{50} value was 1800 mg/kg bw in males and 900 mg/kg bw in females.

Mouse study (Study SGT-20-0028, 1992)

In a GLP OECD TG 401 comparable study, CD-1 mice (5/sex/group) were treated by gavage with imiprothrin in corn oil at dose levels of 300, 380, 480, 600, 760 or 950 mg/kg bw (both sexes). Clinical signs began 30 minutes after dosing and included decrease in spontaneous activity (at ≥380 mg/kg bw, males/females); tremors (females at ≥380 mg/kg bw, males at ≥480 mg/kg bw); excretion of oily substance (males); clonic convulsion (females at ≥480 mg/kg bw, excluding the 600 mg/kg bw dose group). All signs in surviving animals disappeared within 4 hours. Deaths were reported at ≥480 mg/kg bw in females and at ≥760 mg/kg bw in males. All

deaths were reported to be preceded by tremors and clonic convulsions. The animals that died during the observation period showed retention of an oily substance in the stomach and autolysis of the intestine. The LD_{50} value was 724 mg/kg bw in males and 550 mg/kg bw in females.

Acute Oral Studies with Manufacturing Use Product

Rat study (Study SGT-20-0030, 1992)

In a GLP OECD TG 401 comparable study, SD rats (5/sex/group) were treated by gavage with Manufacturing Use Product at dose levels of 1000, 2000, 3200, 4000 or 5000 mg/kg bw (males) and 1000, 2000, 2600, 3200 or 4000 mg/kg bw (females). In the cases where mortality was observed it occurred 24 h post dosing. With all other doses except 3200 mg/kg bw the clinical signs in surviving animal disappeared within 3 days. Irregular respiration (females), blotted fur and urinary incontinence were observed. At doses ≥2000 mg/kg bw there was decrease in spontaneous activity, tremor, prone position (females). Ataxic gait was observed in females at 2000 and 3200 mg/kg bw, and in males at 2000, 3200, 4000 and 5000 mg/kg bw. When dosing at 4000 mg/kg bw (females) and 5000 mg/kg bw (males), clonic convulsion was observed. The LD_{50} value for Manufacturing Use Product was 4500 mg/kg bw in males and 2400 mg/kg bw in females, equivalent to 2250 and 1200 mg/kg bw of imiprothrin, respectively.

Mouse study (Study SGT-20-0032, 1992)

In a GLP OECD TG 401 comparable study, CD-1 mice (5/sex/group) were treated by gavage with Manufacturing Use Product at dose levels of 500, 680, 910, 1230, 1660 or 2240 mg/kg bw (both sexes). Clinical signs began 30 minutes after dosing and included decrease in spontaneous activity (at ≥680 mg/kg bw, males/females); tremors (females at ≥680 mg/kg bw, males at ≥910 mg/kg bw); prone position (males at 910 mg/kg bw); clonic convulsion (males at ≥910 mg/kg bw; females at ≥1230 mg/kg bw); irregular respiration; excretion of oily substance (males); ataxic gait (males at ≥1660 mg/kg bw, females at 1660 mg/kg bw). All signs in surviving animals disappeared within 1 day. The LD_{50} value for Manufacturing Use Product was 1350 mg/kg bw in males and 1300 mg/kg bw in females, equivalent to 675 and 650 mg/kg bw of imiprothrin, respectively.

Acute Neurotoxicity Study with Imiprothrin

In a GLP compliant US-EPA 81-8 acute neurotoxicity study (Study SGT-51-0073, 1995), SD rats (4/sex/dose, replicated over 3 days) were treated by oral gavage with imiprothrin in corn oil at doses of 0, 200, 600 or 1000 mg/kg bw (males) and 0, 100, 300 or 1000 mg/kg bw (females). A few females in all treated group displayed flicking of the forelimbs. No structural changes to nervous system tissues were detected (neuropathology was conducted for the control and high dose animals). At 300 mg/kg bw (females only) fur staining along the ventral thoracic, abdominal and urogenital regions was observed. Tremors were noted in one female. At 600 mg/kg bw (males only) it was reported slight ataxic gait in one male. At 1000 mg/kg bw two females were reported dead. There was increase in tremors, wet muzzle and overall gait incapacity (females). Decrease in motor activity, arousal, body tone and extensor thrust (females) was reported. Slight tremors were noted in one male and slight ataxic gait was exhibited by one male. The LD_{50} was >1000 mg/kg bw for both sexes.

Conclusion

Taking all oral studies together, the DS concluded that imiprothrin should be classified as Acute Tox. 4; H302 (Harmful if swallowed) with an ATE value of 550 mg/kg on the basis of the lowest obtained LD_{50} value of 550 mg/kg bw in female mice.

Acute dermal toxicity

Acute Dermal Study with Imiprothrin

In a GLP OECD TG 402 comparable study (Study SGT-20-0027, 1992), SD rats (5/sex) were exposed to a single application of imiprothrin in corn oil at 2000 mg/kg bw. The area of exposure was covered with a semi-occlusive dressing for 24 hours and the animals were subsequently observed for 14 days. No deaths or clinical signs of toxicity were reported. The LD_{50} was >2000 mg/kg bw for both sexes.

Acute Dermal Study with Manufacturing Use Product

In a GLP OECD TG 402 comparable study (Study SGT-20-0031, 1992), SD rats (5/sex) were exposed to a single application of Manufacturing Use Product at 2000 mg/kg bw. The area of exposure was covered with a semi-occlusive dressing for 24 hours and the animals were subsequently observed for 14 days. No deaths or clinical signs of toxicity were reported. The LD_{50} was >2000 mg/kg bw for both sexes (equivalent to >1000 mg/kg bw imiprothrin).

Conclusion

The DS concluded that imiprothrin does not fulfil the criteria and should therefore not be classified for acute toxicity following the dermal route of exposure.

Acute inhalation toxicity

Acute Inhalation Study with Imiprothrin

In a GLP OECD TG 403 comparable study (Study SGT-10-0003, 1991), SD rats (5/sex/group) were exposed (whole body) to aerosolised imiprothrin in corn oil at a concentration of 418 or 1200 mg/m^3 (equivalent to 0.418 and 1.2 mg/L, respectively) for four hours. The mass median aerodynamic diameter (MMAD) was 0.74-0.85 µm. No animals died during the study. No detailed observation of the animals was possible during the extent of the test due to the density of the mist caused by the aerosol. Nevertheless, signs of irregular respiration, dark red staining around the nose and wet fur were noted. These signs disappeared between 1 and 8 days post dose. At 1200 mg/m^3, the animals exhibited an exaggerated startle response, tip toe gait and loss of abdominal and sub-mandibular hair in females. At study termination the only reported clinical observation was the hair loss. No significant macroscopic or microscopic findings were noted at necropsy. The LC_{50} was established as being >1.2 mg/L for both males and females.

Acute Inhalation Study with Manufacturing Use Product

In a GLP OECD TG 403 comparable study (Study SGT-30-0064, 1993), SD rats (5/sex/group) were exposed (whole body) to aerosolised Manufacturing Use Product undiluted at a concentration of 2810, 3620 or 4430 mg/m^3 (equivalent to 2.81, 3.62 and 4.43 mg/L) for four hours. The MMAD was 3.19-3.75 µm. Deaths were reported in both males and females at doses ≥2.81 mg/L. At 4.43 mg/L, all animals died, but it was not possible to make precise clinical observations during the exposure period due to the dense aerosol mist generated. At lower doses, the following abnormal signs were noted: muscular fibrillation, irregular respiration, lacrimation, nasal discharge, a red substance around the snout, salivation, urinary incontinence, ataxic gait, tip toe gait, wet fur, ocular discharge, tremor and hypersensitivity in males. There were no significant treatment-related findings at necropsy. The LC_{50} was established as being 3.6-4.4 mg/L for males and 2.8-3.6 mg/L for females, equivalent to 1.8-2.2 and 1.4-1.8 mg/L imiprothrin for males and females, respectively.

Conclusion

Given that the study with Manufacturing Use Product used higher concentrations of imiprothrin, and that the toxicity in that study seems to be driven by imiprothrin rather than isopropyl myristate (which is reported to have a very high LC_{50} of >33-41 mg/L), the DS concluded that the Manufacturing Use Product LC_{50} values place imiprothrin in Category 4 (Acute Tox. 4; H332; LC_{50} values between 1 and 5 mg/L). On this basis, an Acute Toxicity Estimate (ATE) value of 1.4mg/L for the inhalation route (dusts and mists) was proposed.

Comments received during public consultation

One MSCA asked for clarification on the number of deaths in the oral mouse study with imiprothrin at the dose level of 760 mg/kg bw. This was clarified by the DS (3/5). Another MSCA expressed support for the proposed classification for acute oral and inhalation toxicity, but found the presentation of the inhalation data confusing.

Assessment and comparison with the classification criteria

As described above, eight guideline studies investigating the effects of a single dose of imiprothrin via oral, dermal and inhalation routes are available (four studies with imiprothrin as test substance, four studies with the Manufacturing Use Product containing 50% imiprothrin and 50% isopropyl myristate). In addition there is a guideline acute oral neurotoxicity test available with imiprothrin in rats. RAC agrees with the DS that also the studies using the Manufacturing Use Product are relevant for classification, given the low acute toxicity of isopropyl myristate, oral and dermal LD_{50} values in the range of those found for neat imiprothrin, and the higher imiprothrin concentration tested for inhalation. In the table below the LD_{50}/LC_{50} values as observed in the nine available studies are presented and compared with the classification criteria. From this, it follows that imiprothrin fulfils the criteria for classification in category 4 for the oral and inhalation route, but not for the dermal route.

Table: Overview of LD_{50}/LC_{50} values (expressed as imiprothrin dose in mg/kg bw (oral/dermal) or in mg/L (inhalation)) in acute toxicity studies with imiprothrin (neat and in Manufacturing Use Product)

	Acute oral		**Acute dermal**		**Acute inhalation**	
	Study with imiprothrin	Study with Manufacturing Use Product	Study with imiprothrin	Study with Manufacturing Use Product	Study with imiprothrin	Study with Manufacturing Use Product
Rat						
male	1800	2250	>2000	>1000	>1.2	1.8-2.2
female	900	1200	>2000	>1000	>1.2	1.4-1.8
Rat (neurotox) **male/female**	>1000					
Mouse						
male	724	675				
female	550	650				
Criteria Category 4	300-2000		1000-2000		1-5 (dusts and mists; 4h)	
	Fulfilled		Not fulfilled		Fulfilled	

Overall, RAC agrees with the proposal of the DS and considers that imiprothrin should be classified as **Acute Tox. 4; H302** and **Acute Tox 4; H332**, with **ATE values** respectively of **550 mg/kg bw (oral)** and **1.4 mg/L (inhalation)**.

RAC evaluation of specific target organ toxicity – single exposure (STOT SE)

Summary of the Dossier Submitter's proposal

There are eight acute toxicity studies and one acute oral neurotoxicity study available investigating the effects of a single dose of imiprothrin. The results of these studies have been described in detail in the section on 'Acute toxicity' above. As to clinical signs of toxicity, these were observed following oral and inhalation exposure, but not following dermal administration. The clinical signs observed in the oral and inhalation studies included ataxic gait, tremor, decreases in spontaneous activity, urinary incontinence and clonic convulsions. These effects are most likely related to the neurotoxic mode of action of imiprothrin, a synthetic pyrethroid insecticide that acts on the sodium channel in the nerve membranes of the invertebrate nervous system. Sodium channel modulators cause pronounced repetitive activity and a prolongation of the transient increase in sodium permeability of the nerve membranes, resulting in continual nerve impulse transmission leading to tremors and death. In the acute oral and inhalation toxicity studies, some neurotoxic effects occurred at high doses that were also lethal, but some effects were also observed at doses below the LD_{50}/LC_{50} values and in the range of classification with STOT SE 2 (oral: between 300 and 2000 mg/kg bw; inhalation: between 1 and 5 mg/L/4h). Given however the mortalities in these studies, and the fact that the neurotoxic effects in the acute oral neurotoxicity study were limited to the high dose of 1000 mg/kg bw at which two animals died, the DS proposed to classify for acute toxicity rather than for STOT SE 2.

Comments received during public consultation

One MSCA commented that an additional classification in STOT SE 1 should be considered in view of clinical signs of (acute) neurotoxicity following inhalation at clearly sublethal doses and below the guidance value of 1 mg/L/4h in a 28-day study in rats. The DS in response indicated that the findings in the 28-day study do not describe clearly whether the effects were observed after a single exposure. Considering all of the data, and the nature of the general acute hazard, the DS felt that the proposed classification for acute toxicity was already adequate.

Assessment and comparison with the classification criteria

Following single administration, clinical signs indicative of neurotoxicity were observed for the oral and inhalation route, but not for the dermal route.

In the acute oral toxicity studies with imiprothrin and Manufacturing Use Product in rats and mice, clinical signs indicative of neurotoxicity appeared 30-60 minutes post dosing. In surviving animals these signs disappeared within 3 days (rats) or 1 day (mice). Whereas the more serious effects (like e.g. clonic convulsions) were mostly observed at the higher, lethal doses, some effects were also seen at doses not resulting in mortality.

In the imiprothrin study in rats, neurotoxic effects only occurred at doses that also caused mortality. The same was true for female rats in the Manufacturing Use Product study, but in male rats tremor, a decrease in spontaneous activity and ataxic gait were observed at non-lethal doses

corresponding to 1000 and 1600 mg imiprothrin/kg bw. Irregular respiration was additionally noted in males at 1600 mg/kg bw.

In the imiprothrin study in mice, tremor and a decrease in spontaneous activity were seen at non-lethal doses of 380 (no mortality in males and females) and 480 mg/kg bw (no mortality in males). A decrease in spontaneous activity and tremor were also observed at non-lethal doses corresponding to 340 (no mortality in males and females) and 460 (no mortality in males) mg imiprothrin/kg bw in the Manufacturing Use Product study in mice. Males at the latter dose additionally showed prone position, clonic convulsion and irregular respiration.

In the acute oral neurotoxicity study in rats at imiprothrin doses of 0, 200, 600 or 1000 mg/kg bw for males and 0, 100, 300 or 1000 mg/kg bw for females, no histopathological lesions in nervous system tissues were found. Treatment-related mortalities only occurred at 1000 mg/kg bw (2/12 females, dying on the day of treatment). At 300 mg/kg bw, one or two females showed ungroomed fur and fur staining, and one female had tremor. At 600 mg/kg bw, one male showed slight ataxic gait. At 1000 mg/kg bw, there was one male with slight ataxic gait and slight tremors. Females at 1000 mg/kg bw showed several effects, including severe tremors in the head, body and/or limbs after dosing, wet muzzle, overall gait incapacity, decreases in locomotor activity, arousal, extensor thrust and body tone, delays for the positional passivity test and altered olfactory response and visual placing test response. In addition, a few females in this group showed no/reduced response for toe/tail pinch testing, corneal/pinna reflexes and an increase for the auricular startle test. Grip strengths were also slightly reduced for this group, and motor activity levels were markedly reduced.

In the acute inhalation toxicity studies with imiprothrin and Manufacturing Use Product in rats, clinical signs indicative of neurotoxicity appeared from 30-60 minutes post dosing. They disappeared within a couple of hours to 7 days at the latest.

In the acute toxicity study with imiprothrin, male and female rats showed irregular respiration, tip toe gait and ataxic gait at 0.418 and 1.2 mg/L and an exaggerated startle response at 1.2 mg/L, but no mortality. In the acute study with Manufacturing Use Product in rats, signs of neurotoxicity were observed at all tested concentrations (corresponding to 1.4-2.2 mg imiprothrin/L) and included muscular fibrillation, ataxic and tip toe gait, decrease in spontaneous activity in both sexes and irregular respiration and hypersensitivity in males. Death was however also seen at these concentrations.

Clinical signs of toxicity characteristic of neurotoxicity have also been observed at the highest tested concentration of 186 mg/m^3 (0.186 mg/L) in a 28-day inhalation study in rats, in the absence of mortalities. These signs included decreased spontaneous activity (in 1 to 9 males and females), tip toe gait (in 1 to 6 animals), and jumping, hypersensitivity and tremor (in 1 female). Irregular respiration (in 1 to 10 animals), nasal discharge (in 1 to 4 animals), salivation (in 1 to 3 animals) and urinary incontinence (in 1 to 3 animals) were also seen. RAC notes that the degree of severity and the exact onset of these signs is not reported (although they are likely to be acute in nature, as supported by the absence of histopathological or functional long term findings investigated through detailed examination and FOB in a 90-day oral neurotoxicity study in rats).

Conclusion

Neurotoxicity was consistently observed across all acute oral and inhalation studies, at both lethal and non-lethal doses. Whereas RAC notes that for lethality the substance is already proposed to be classified, the fact that effects are also seen at non-lethal doses makes it necessary to consider if additional classification for STOT SE is warranted. As the overall profile of toxic signs is not typical of narcosis, classification with STOT SE 3 is not appropriate. This leaves STOT SE 1 or 2. The lowest non-lethal doses at which the neurotoxic effects are observed fall within the guidance values for STOT SE 2 (300 < C ≤ 2000 mg/kg bw) for the oral route and for STOT SE 1 (≤ 1

mg/L) for the inhalation route. RAC notes though, that details on the severity and incidence of each finding in the acute toxicity studies is missing, that most findings were transient in nature, and that their relevance to fulfil the severity criteria for STOT SE 1/2 is not totally clear. It is further noted that the sublethal dose levels with neurotoxic findings were mostly within a factor 2 lower than the lethal dose levels, with the exception of two rat studies with imiprothrin (the acute inhalation study and the acute oral neurotoxicity study in males) where no lethality was seen. Nevertheless, given the consistent picture, and further supported by the fact that imiprothrin belongs to the group of pyrethroids, which is known to induce neurotoxic effects, RAC considers it important for classification to note the neurotoxic properties of imiprothrin in this case.

RAC therefore concluded that **classification as STOT SE 2; H371 for its effects on the nervous system by the oral and inhalation route** is warranted.

RAC evaluation of specific target organ toxicity– repeated exposure (STOT RE)

Summary of the Dossier Submitter's proposal

Six repeated dose oral toxicity studies (all under GLP and guideline complaint) were available; two in rats (for 90 days and 2 years), two in mice (for 90 days and 18 months) and two in dogs (for 90 days and 1 year). One GLP guideline 28 day inhalation study and one GLP guideline 21 day dermal study (both in rats) were also available. In addition, there was a GLP guideline 90 day oral repeated dose neurotoxicity study in rats. In the table below the effects in these studies at relevant doses for classification are presented.

Table: Summary of repeated dose toxicity studies with imiprothrin

Study	**Dose levels**	**Target organ(s)** **NOAEL/C**	**Effects at relevant doses for classification**
ORAL			
90 day (diet) SD rat (12/sex/group) OECD TG 408 GLP (Study SGT-20-0040, 1992)	0, **100**, 3000, 6000, 10000 ppm equivalent to m: 0, 5.9, 179, 350, 611 mg/kg bw/day f: 0, 7, 197, 399, 657 mg/kg bw/day Guidance value for classification **≤ 100** mg/kg bw/day	Salivary gland, liver, red blood cells NOAEL 100 ppm	No
90 day (diet) Neurotoxicity study SD rat (12/sex/group) US EPA 82-7 GLP (Study SGT-51-0074, 1995)	0, **1000**, 3000 and 10000 ppm equivalent to m: 0, 62, 191 and 648 mg/kg bw/day f: 0, 74, 219, 722 mg/kg bw/day Guidance value for classification **≤ 100** mg/kg bw/day	None (only body weight affected) $NOAEL_{general\ tox}$ 1000 ppm $NOAEL_{neurotox}$ >10000 ppm	No

Study	Dose levels	Target organ(s) NOAEL/C	Effects at relevant doses for classification
2 year (diet) SD rat (64/sex/group; interim sacrifice of 14/sex/group after 52 weeks) OECD TG 453 GLP (Study SGT-50-0069, 1995)	0, **50, 250**, 2500, 5000 ppm equivalent to m: 0, 2, 9, 90 or 180 mg/kg bw/day f: 0, 2, 11, 109, 219 mg/kg bw/day Guidance value for classification **≤ 12.5** mg/ kg bw/day	Salivary gland, liver, red blood cells NOAEL 250 ppm	No
90 day (diet) CD-1 Mouse (12/sex/group) OECD TG 408 GLP (Study SGT-20-0021, 1992)	0, 1000, 3000, 5000, 7000 ppm equivalent to m: 0, 130, 371, 643, 883 mg/kg bw/day f: 0, 150, 435, 803, 1239 mg/kg bw/day Guidance value for classification **≤ 100** mg/ kg bw/day)	Liver, red blood cells NOAEL 1000 ppm	No
18 month (diet) CD-1 Mouse (66/sex/group; interim sacrifice of 15/sex/group after 52 weeks) US EPA 83-2 GLP (Study SGT-50-0070, 1994)	0, **100**, 3500, 7000 ppm equivalent to m: 0, 10, 354, 702 mg/kg bw/day f: 0, 12, 409, 814 mg/kg bw/day Guidance value for classification **≤ 16.7** mg/ kg bw/day	Liver, red blood cells NOAEL 100 ppm	No
90 day (capsule) Beagle dog (4/sex/group; additional 2/sex in control and high dose groups for 6-week recovery period) OECD TG 409 GLP (Study SGT-20-0051, 1992)	0, **10, 100**, 1000 mg/kg bw/day Guidance value for classification **≤ 100** mg/ kg bw/day	Salivary gland, liver, red blood cells; no longer seen in recovery animals NOAEL 10 mg/kg bw/day	100 mg/kg bw/day: <u>Observations</u> ↑salivation, loose & watery faeces <u>Organ weights</u> ↑salivary gland: 15% f; 28% m (relative) ↑ liver: 11% f; 14% m (relative)
1 year (capsule) Beagle dog (4/sex/group) OECD TG 452 GLP (Study SGT-41-0065, 1994)	0, **5**, 50, 500 mg/kg bw/day Guidance value for classification **≤ 25** mg/kg bw/day	Salivary gland, liver, red blood cells NOAEL 5 mg/kg bw/day	No
INHALATION			
28 day	0 (vehicle and air controls), **2.4, 22, 186** mg/m^3	Salivary gland, liver,	186 mg/m^3 (0.186 mg/L): <u>Observations</u>

Study	Dose levels	Target organ(s) NOAEL/C	Effects at relevant doses for classification
SD rat (10/sex/group) US EPA 82-4 GLP (Study SGT-20-0056, 1992)	(0.0024, 0.022, 0.186 mg/L) Whole body, 4h/day Guidance values for classification (dusts and mists) Cat. 1 **≤ 0.06** mg/L/6h/day Cat. 2 **0.06 < C ≤ 0.6** mg/L/6h/day	red blood cells NOAEC 22 mg/m^3	↓ body weight (bw): 7% f; 14% m ↓ bw gain: 20% f; 27% m Clinical signs of toxicity characteristic of neurotoxicity including decreased spontaneous activity (1 to 9 m and f), tip toe gait (1 to 6 m and f), hypersensitivity (1 f) and tremor (1 f) <u>Organ weights</u> ↑salivary gland: 52% f; 58% m (relative) ↓ thymus: 17% m; 23% f (absolute) ↑ liver: 11% m; 21% f (relative) ↑ kidneys: 11% m; 16% f (relative) ↑ brain: 9% f; 13% m (relative) ↑ ovaries: 11% (relative) ↑ testes: 19% (relative) ↑ thyroid: 31% m (relative) ↑ adrenals: 19% f (relative) <u>Haematology</u> ↓(<10%) in Hb, Hc and erythrocyte numbers: m and f ↑ reticulocyte count: 27% m; 87% f ↓ prolongation of activated partial thromboplastin: 43% f <u>Clinical Chemistry</u> ↑ total cholesterol: 20% f; 22% m ↓ triglyceride: 56% m <u>Histopathology</u> ↑basophilic staining (slight) of acinar cells in salivary gland: 9 m and 10 f
DERMAL			
21 day SD rat (5/sex/group) US EPA 82-2 GLP (Study SGT-51-0072, 1995)	0, **100, 300**, 1000 mg/kg bw/day Semi-occluded, 6h/day Guidance value for classification **≤ 800** mg/kg bw/day	Skin, salivary gland $NOAEL_{systemic}$ 300 mg/kg bw/day $NOAEL_{local}$ 300 mg/kg bw/day	No

The liver, salivary sub-mandibular gland and red blood cells were identified by the DS as the target organs for toxicity in the repeated dose studies. However, it was concluded that there were no consistent significant adverse effects nor supporting histopathology at doses at or below the guidance values for classification. Therefore the DS concluded that a STOT RE classification is not warranted for imiprothrin.

Comments received during public consultation

No specific comments were received on this hazard class.

Assessment and comparison with the classification criteria

Following repeated oral administration, hepatotoxicity and haematotoxicity were reported across all three species, the mouse being less sensitive than the rat or the dog. In addition, the salivary gland was also a target organ for toxicity in the rat and dog. Upon repeated inhalation some clinical signs of neurotoxicity were observed in rats, as well as effects on liver, red blood cells

and salivary gland. The latter organ was also affected in rats following repeated dermal administration.

In the available studies, the effects on the target organs were most consistently observed at doses that are above the guidance values for classification for STOT RE. Only in the 90 day oral study in dogs and the 28 day inhalation study in rats effects were observed at or below the guidance values for classification. In dogs, it concerned increases in weights of the liver and salivary sub-mandibular gland at 100 mg/kg bw/day, but as there were no supporting histopathological findings in these organs, these effects in the 90 day oral dog study do not warrant classification.

In rats, several effects were observed at 0.186 mg/L. The effects on the liver (increases in weight and total cholesterol, decrease in triglycerides) and red blood cells (indicative of regenerative anaemia) were however not supported by histopathological findings in the relevant organs. The increase in salivary gland weight at 0.186 mg/L was associated with an increased incidence of basophilic staining of the acinar cells, but the staining was described as only slight. Weights of some other organs were also affected, but without accompanying histopathological findings. Finally, some clinical signs of neurotoxicity were observed, but RAC notes that the degree of severity and the exact onset of these signs is not reported. They are likely to be acute in nature, as supported by the absence of histopathological or functional long term findings investigated through detailed examination and FOB in the 90-day oral neurotoxicity study in rats. Overall, RAC considers the effects observed at a dose below the guidance value in the 28 day inhalation rat study not to warrant classification.

In conclusion, RAC agrees with the Dossier Submitter proposal that **no classification for STOT RE is warranted**.

RAC evaluation of germ cell mutagenicity

Summary of the Dossier Submitter's proposal

Three guideline/GLP *in vitro* studies assessing the mutagenic potential of imiprothrin are available. Clear negative results were observed *in vitro* in a bacterial reverse mutation test (Study SGT-20-0023, 1992) and in a mammalian cell gene mutation (hprt) assay conducted in Chinese hamster lung fibroblasts (V79) (Study SGT-20-0046, 1992). In a chromosome aberration test in Chinese hamster lung cells (Study SGT-20-0024, 1992), a dose-related increase in structural aberrations was noted with exogenous metabolic activation at the two highest concentrations tested (75 and 100 µg/mL). An increased incidence of polyploidy cells was observed at all dose levels. These effects were not observed in the absence of metabolic activation.

In two guideline/GLP studies assessing the mutagenic potential of imiprothrin *in vivo*, no evidence of genotoxicity was found in a mouse bone marrow micronucleus test following intraperitoneal administration (Study SGT-20-0041, 1992) and in an Unscheduled DNA Synthesis (UDS) assay in rat hepatocytes following oral administration (Study SGT-20-0045, 1992). Although in the micronucleus test only a slight reduction in PCE/NCE ratio was observed, general systemic toxicity including mortality (at the highest dose) was seen. Further, in view of toxicokinetic data suggesting wide distribution of imiprothrin, the DS presumed that imiprothrin would have reached the bone marrow, and therefore considered the result of the micronucleus test clearly negative.

The DS considered that the positive results obtained in the *in vitro* mammalian chromosome aberration test in Chinese hamster lung cells could be disregarded in face of the negative results

obtained in both *in vivo* studies. Overall, the DS concluded that there is no evidence to suggest that imiprothrin is genotoxic *in vivo* and that classification is therefore not warranted.

Comments received during public consultation

2 MSCAs commented that a genotoxic effect in somatic cells cannot be ruled out in view of a positive *in vitro* chromosome aberration assay in lung cells following metabolic activation, and no *in vivo* clastogenicity study available in a metabolically active organ. One of these MSCAs argued that neither the *in vivo* micronucleus test in bone marrow nor the *in vivo* UDS test in liver could adequately negate the positive *in vitro* finding. This MSCA further pointed to a structural alert for *in vivo* clastogenicity within the imiprothrin structure, but concluded that, overall, the criteria for classification as Muta. 2 are not met.

The DS in response stated that isolated positive results are not unusual and that it could be a false positive. With evidence from radiolabelled imiprothrin studies that the substance is widely distributed to a wide range of organs and tissue, the DS considered it reasonable to presume that also the bone marrow had been reached and therefore considered the available *in vivo* test data to be adequately reassuring.

Assessment and comparison with the classification criteria

Imiprothrin tested negative in a bacterial reverse mutation assay and a mammalian cell gene mutation assay, but showed *in vitro* clastogenic activity in a mammalian chromosome aberration test in Chinese hamster lung cells following metabolic activation. Although this effect was not expressed *in vivo* in a conventional bone marrow micronucleus test, and imiprothrin also tested negative in a rat liver UDS assay, RAC notes that these tests do not inform on the clastogenic potential in metabolically active organs like the liver or the lung. Whereas genotoxicity in somatic cells can therefore not be totally ruled out, RAC considers **no classification** to be justified given that the available data do not meet the criteria for classification.

RAC evaluation of carcinogenicity

Summary of the Dossier Submitter's proposal

Two carcinogenicity studies were available; one in rats and one in mice.

2 year rat study

In a 2 year carcinogenicity study (Study SGT-50-0069, 1995) conducted under GLP and conform to OECD TG 453 guideline, imiprothrin was administered to SD rats (50/sex/dose) at 0, 50, 250, 2500 or 5000 ppm (males: 0, 2, 9, 90, 180 mg/kg bw/day; females: 0, 2, 11, 109, 219 mg/kg bw/day) in the diet for 2 years. A satellite group of animals (14/sex/dose) was maintained on the same regimen and was sacrificed after 52 weeks. Neoplastic lesions were observed in the liver and the lungs, as shown in the table below.

Table: Summary of neoplastic findings (incidence) in the 2 year rat study

	Males (50 animals in all dose groups)					**Females** (50 animals in all dose groups)				
Doses (mg/kg bw/day)	**0**	**2**	**9**	**90**	**180**	**0**	**2**	**11**	**109**	**219**
LIVER										
Adenoma	1	1	1	0	4	0	1	0	3	1
Haemangioma	0	0	0	0	1	0	0	0	0	0
Carcinoma	1	0	0	0	0	0	0	0	0	1
LUNG										
Adenoma	0	0	0	0	2	0	0	0	0	0
Adenocarcinoma	0	0	0	0	0	0	1	0	0	0

No historical control data available

As to the findings in the liver, the DS considered that, given the low numbers of tumours observed in the treated animals, the presence of both benign and malignant tumours in control males, and the absence of any clear dose-response, it is unlikely that the tumour findings were treatment-related. The DS further considered that the limited number of tumour findings in the lung (lung adenocarcinoma in females at the low dose and lung adenoma in males at the top dose) do not indicate that imiprothrin is carcinogenic.

18 month mice study

In an 18 month carcinogenicity study (Study SGT-50-0070, 1994) conducted under GLP and conform to OECD TG 451 guideline, imiprothrin was administered to CD-1 mice (51/sex/dose) at 0, 100, 3500 or 7000 ppm (males: 0, 10, 354, 702 mg/kg bw/day; females: 0, 12, 409 or 814 mg/kg bw/day) in the diet for 18 months. A satellite group of animals (15/sex/dose) was maintained on the same regimen and was sacrificed after 52 weeks. Similar to rats, neoplastic lesions were observed in the liver and the lungs, as shown in the table below.

Table: Summary of neoplastic findings in the 18 month mouse study

	Males					**Females**				
Doses (mg/kg bw/day)	**0**	**10**	**354**	**702**	**HCD**	**0**	**12**	**409**	**814**	**HCD**
LIVER (incidence)					n/a					n/a
No. of mice	51	51	51	50[a]		51	51	51	51	
Foci of cellular alteration	6	4	10	12		1	0	4	6	
Adenoma	14	13	13	21		0	0	2	1	
Carcinoma	5	7	1	6		0	0	0	0	
LUNG (%)										
No. of mice	51	50[b]	51	50[a]		50[b]	51	51	49[c]	
Adenoma	5.9	14.0	7.8	6.0	12.2 (3.9-21.6)	6.0	3.9	7.8	16.3	5.4 (0-13.7)
Adenocarcinoma	9.8	8.0	11.8	26.0*	5.9 (2.8-9.8)	6.0	3.9	9.8	8.2	4.4 (1.9-6)

[a] One animal which died accidentally was excluded from the analysis

[b] The autolysed lung specimen from one animal was excluded from the analysis

[c] The lung tissues from 2 animals were lost due to cannibalism

* Significantly different from control group at $P < 0.05$

HCD = laboratory historical control data; mean (range) from 6 studies (1988-1999), one of which was the imiprothrin study

n/a = no historical control data available

Liver adenoma and carcinoma were observed in the male, but not the female, control group. The malignant liver lesions in males were not dose-related and averaged the same frequency as seen in the control group. In the top dose males an increase in adenoma was seen, but with a relatively high frequency of these benign tumours in concurrent controls the DS considered the significance of this apparent dose-related effect unclear. In the female mice there were very few liver tumours observed and no dose-response was evident. According to the study report, no statistical

significance was present for males or females. On the basis of these data, the DS considered that imiprothrin has not been found to produce a clear hepatocarcinogenic effect in mice.

In female mice, an increase (not statistically significant) in lung adenoma was seen at the top dose. The incidence at this dose (16.3%) was outside the laboratory historical control range (0-13.7%) but within the broader historical control range from the animal supplier (1.67-26.67%; not considered relevant by RAC). No increase in lung adenocarcinoma was seen, possibly due to the increased mortality rate in females at the higher doses (survival rates were 86.3, 80.4, 72.5 and 54.9% at 0, 12, 409 and 814 mg/kg bw/day, respectively).

In male mice, a statistically significant increase in lung adenocarcinoma was observed at the top dose. The incidence at this dose (26%) (as well as the 11.8% incidence at the mid dose) exceeded the laboratory historical control range (2.8-9.8%), and was at the upper limit of the animal supplier historical control range (1.43-26%; not considered relevant by RAC). Benign lung tumours were reported in all treatment groups, but the incidence of adenoma did not show a dose-response relationship.

As the lung tumour profile indicated an uncertain picture of the carcinogenic effects of imiprothrin in mice, an additional examination was performed to evaluate alveolar proliferating lesions in all lobes (the original examination concerned sections from the left lobe of the lung and other lobes bearing macroscopic lesions). When looking at the incidences of lung adenoma and adenocarcinoma in the combination of original and additional lung sections (see table below), further incidences of both benign and malignant lung tumours were observed in the additional sections in all dose groups (except for adenocarcinoma in males at the top dose), but there was no longer statistical significance seen.

Table: Lung tumour incidences in the original and additional lung sections combined

	Males					**Females**				
Doses (mg/kg bw/day)	**0**	**10**	**354**	**702**	**HCD**	**0**	**12**	**409**	**814**	**HCD**
LUNG (%)					n/a					n/a
No. of mice	*51*	*51*	*51*	*50*[a]		51	51	51	49[b]	
Adenoma	19.6	21.6	19.6	18.0		17.6	13.7	21.6	30.6	
Adenocarcinoma	11.8	9.8	13.7	26.0		9.8	5.9	11.8	12.2	

[a] One animal which died accidentally was excluded from the analysis

[b] The lung tissues from 2 animals were lost due to cannibalism

HCD = laboratory historical control data; mean (range)

n/a = no historical control data available

According to the DS, the interpretation of the lung findings in mice is not straightforward. Although an increase with dose was found for lung adenocarcinoma in male mice, a similar increase was not seen in females. It is unclear whether the reduced survival of females at the top dose was a factor in this apparent difference in sensitivity between the sexes. Similarly, in the absence of any other information suggesting a sex-specific response of the mouse lung to imiprothrin, the malignant tumours may not have been treatment-related. Further doubts about the significance of the tumour findings is cast by the observation of benign lung tumours in control and all dose groups in both sexes. Adenocarcinoma was also observed in control animals.

Overall, the DS considered that no clear treatment-related findings were observed in rats or mice, although an increased incidence of malignant tumours was evident in lungs of top dose male mice (compared to concurrent and historical control rates). In deciding on whether classification is warranted or not, the DS took into account that there was no evidence of mutagenicity, and that a prominent effect was only seen in the lungs of male mice at the top dose. Furthermore, the DS considered that the concern for a carcinogenic potential of imiprothrin is lowered by the relatively high background incidence of tumours and the lack of a mechanistic basis for the

findings. On the basis of both the strength and weight of evidence, the DS concluded that imiprothrin does not warrant classification for carcinogenicity.

Comments received during public consultation

Two MSCA's commented that classification for carcinogenicity category 2 should be considered, one MSCA in view of the lung tumours in male mice, the other MSCA in light of significant treatment-related increases in lung adenocarcinoma in male mice (positive trend), supported by related findings in rats and indications for neoplastic change in livers of rats and mice (positive trends). Both MSCA's highlighted the relevance of comparing the incidence of lung tumours to the laboratory historical control data (HCD) rather than to the supplier HCD (as done in the CLH report). The MSCA's also highlighted that a mutagenic effect cannot be excluded (in view of the positive *in vitro* chromosomal aberration with metabolic activation and the structural alert identified). One of the MSCA's finally pointed to structural similarities with another pyrethroid insecticide acting on the sodium channel and classified as a carcinogen as a potential basis for read-across.

The DS in response indicated that, to their opinion, a prominent effect was only seen in lungs of male mice at the top dose, but that the data are not sufficiently convincing to classify imiprothrin for carcinogenicity, given the relatively high background incidence of the tumours and the lack of a mechanistic basis for the findings. Given that the available data on imiprothrin were sufficient to conclude on 'no classification', read-across to data on other substances was not considered necessary by the DS.

Assessment and comparison with the classification criteria

In the two oral bioassays available (one in rats, one in mice), imiprothrin slightly increased the incidences of lung and liver tumours at the highest dose levels in rats and mice, in male animals in particular.

Rat study

Slightly increased incidences (not statistically significant; HCD not available) of lung adenoma (2/50 vs 0/50 in controls) and liver adenoma (4/50 vs 1/50 in controls) were observed in male rats at the highest dietary dose of 5000 ppm, at which there was in addition one isolated case of haemangioma. No increase in lung or liver carcinoma was observed in males, and aside from an increase in pitted foci of the liver no treatment-related histopathological findings were noted in the liver or lungs. Lung and liver tumour incidences were not increased in female rats, and these organs did not show abnormal histopathological findings, aside from an increase in pitted foci of the liver at the top dose. Based on these results, there is very limited evidence for carcinogenicity of imiprothrin in rats, with only a slight, not statistically significant increase in benign tumours in one sex only. These findings do not warrant classification.

Mouse study

Male mice at the highest dietary dose of 7000 ppm showed increased incidences of liver adenoma and lung adenocarcinoma. As to the liver adenoma, the increase (21/50 vs 14/51 in controls) was not statistically significant (HCD not available). Males at this dose did not show an increased incidence of liver carcinoma (6/50 vs 5/51 in controls). In contrast to males, very few liver tumours were observed in females (no carcinoma in any dose group or in controls, 0/51, 0/51, 2/51 and 1/51 adenoma at 0, 100, 3500 and 7000 ppm, respectively). Females did show an increase (not statistically significant) in foci of cellular alteration though, as did males. Non-neoplastic liver changes upon imiprothrin treatment included increases in liver weight and hepatocellular hypertrophy in males and females of the mid and high dose groups.

As to the lung adenocarcinoma, the increase in male mice at the high dose (26% vs 9.8% in controls) was statistically significant and outside the laboratory HCD (mean 5.9%, range 2.8-9.8%). No increase in lung adenoma was seen in male mice (6% vs 5.9% in controls). Female mice at the high dose did not show an increased incidence of lung adenocarcinoma (8.2% vs 6% in controls), but the incidence of lung adenoma was increased (16.3% vs 6% in controls). The difference with the control females was not statistically significant, but the incidence was slightly above the HCD (mean 5.4%, range 0-13.7%).

RAC considers the slight, not statistically significant increase in benign liver tumours in one sex in one species (male mice only) does not warrant classification.

The interpretation of the malignant lung tumour findings in one sex at the top dose only is more difficult. RAC notes that at the mid dose the incidence of lung adenocarcinoma in male mice also exceeded the laboratory historical control range and that the increase was dose-related (positive trend). RAC further notes that survival of male mice was not affected by treatment and was well above the test guideline value of 25%. Males at the high (and mid) dose did show a marked decrease in body weight gain though over the whole treatment period (approximately 40% (and 20%) of control body weight gain), associated with a significant reduction in food consumption. This could point to these dose levels being above the maximum tolerated dose (MTD). A similar effect on body weight gain and food consumption was seen in females at the high dose, where, in contrast to males, treatment did result in reduced survival (54.9% vs 86.3% in controls; yet, well above 25%) but not in increased incidences of lung adenocarcinoma. However, according to information from Industry, for mice, changes in body weight are more suitable for the evaluation of systemic effects of a tested chemical than changes in body weight gain, given that in mice variation in body weight gain is normally more noticeable than variation in body weight. When looking at the final body weights, the reductions, as compared to controls, were indeed much less marked (for males at the mid and high dose the decrease was approximately 9 and 15%, respectively; for females this was approximately 10 and 22%, respectively), indicating that the mid and high dose levels were not clearly above the MTD. Whether the increased mortality in females may have been a factor in the apparent sex difference is difficult to say; it may not have been too much of a confounding factor, given that the higher mortality was mainly observed towards the end of the study (after 69 weeks of treatment at the high dose). Besides, females did show increases in lung adenoma and in lung adenoma and adenocarcinoma combined (positive trends). RAC finally notes that, aside from genotoxicity data, other mechanistic data as to the possible mode of action of lung tumour formation by imiprothrin is lacking.

All in all, the increase in lung adenocarcinoma constitutes limited evidence for carcinogenicity of imiprothrin in mice. Since:

- the increase is marked and dose-related;
- it has not been convincingly shown that the elevated lung tumour incidences at the highest dose level are linked to a bad health status of the exposed males, in view of the relatively moderate reductions in body weight;
- a contribution of genotoxicity cannot totally be excluded, given that *in vitro,* imiprothrin was shown to be clastogenic in lung cells, with no *in vivo* studies in metabolically active tissues available to counteract this;
- the available data do not convincingly indicate that the lung tumours are not relevant for humans,

RAC considers the lung tumours to warrant classification in category 2. This category is considered appropriate in view of the experimental data indicating a weak carcinogenic potential of imiprothrin, expressed in one species and one sex only.

RAC therefore concludes that **classification as Carc. 2; H351 is warranted**.

RAC evaluation of reproductive toxicity

Summary of the Dossier Submitter's proposal

For the endpoint reproductive toxicity, one two-generation study in rats is available, as well as two developmental toxicity studies (one in rats, one in rabbits).

Fertility

In a two-generation study (Study SGT-41-0067, 1994) conducted under GLP and conform to OECD TG 416, imiprothrin was administered to SD rats (30/sex/group) at 0, 200, 2000 or 6000 ppm in the diet for two generations. The achieved test material intakes for P1 parental animals were 9-20, 95-179 and 288-543 mg/kg bw/day (males) and 12-31, 110-325 and 346-909 mg/kg bw/day (females) at 200, 2000 and 6000 ppm, respectively. F1 animals received 11-30, 115-300 and 350-972 mg/kg bw/day (males) and 12-29, 117-297 and 366-883 mg/kg bw/day (females) at 200, 2000 and 6000 ppm, respectively. The main effects observed in the parental generations included decreased food consumption, body weight and body weight gain (at 6000 ppm in males and females of P1 and F1, and at 2000 ppm in F1 females), increased incidences of splenic haemosiderosis (at 6000 ppm in males and females of P1 and F1, and at 2000 ppm in P1 females), and increased liver weights without histopathology findings (at 6000 ppm in P1 males and females and F1 females). In pups at the top dose, a significant decrease in pup weight was seen from day 1 (9%) to day 21 (21%) of lactation. Pups showed no gross abnormalities, but a few minor skeletal abnormalities (increased number of unilateral/bilateral 14th rib, thoracic vertebrae and rib pairs, decreased number of lumbar vertebrae), indicative of disturbed ossification, were observed in F2 litters at 2000 and (more prominently) 6000 ppm. These were considered most likely to have been due to the poor nutritional state of the dams. There were no adverse effects on fertility parameters including mating, gestational indices and length, pup viability and lactation performance.

Given the lack of adverse effects on fertility, the DS concluded that imiprothrin does not warrant classification for fertility.

Developmental toxicity

<u>Rats</u>

In an extended developmental toxicity study (Study SGT-21-0048, 1992) conducted under GLP and with a method similar to OECD TG 414, imiprothrin was administered to female SD rats (36/dose) at 0, 50, 200 and 600 mg/kg bw/day in corn oil, by gavage, on days 6-17 of gestation. Two thirds of dams were sacrificed on day 20 and the foetuses were examined for developmental toxicity, with the remaining one third of dams allowed to deliver offspring to produce the F1 generation.

Maternal toxicity was evident at the top dose, with clear signs of toxicity (such as tremor, clonic convulsions and staggering gate), death (3/36, with 1/3 due to a dosing error) and reductions in food consumption, body weight and body weight gain on several gestation days (but no longer at the end of gestation). The latter reductions were, to a more limited degree, also seen at the mid dose, as well as minor signs of toxicity immediately after dosing.

Foetal weights at 600 mg/kg bw/day were slightly (not statistically significantly) decreased (6%). There was a significant increase in number of foetuses with visceral anomalies, mainly thymic remnants in the neck seen at 600 mg/kg bw/day (22% compared to 3% in control). An increased incidence of unilateral dilatation of the renal pelvis was observed at 50 mg/kg bw/day (6/125 vs 0/119 in control), but this was considered an incidental finding since the effect was not seen at higher dose levels. No treatment-related skeletal anomalies were observed. However, increased

incidences of skeletal variations were seen: higher incidences of lumbar rib were found at 200 and 600 mg/kg bw/day (16%, 20%, 48% and 68% at 0, 50, 200 and 600 mg/kg bw/day, respectively). Foetuses at 600 mg/kg bw/day additionally showed pre-sacral vertebrae (12% vs 1% in control), splitting of the vertebral body (14% compared to 1%), and reduced ossification of 5th and 6th vertebrae.

In this extended study no abnormal effects were observed on mating, fertility and gestation of F1 parental animals, neither were there differences in viability index after birth in the treated groups, or abnormalities following necropsy of F1 parental animals and foetuses from F1 dams. There were also no differences in any F1 groups in the motor coordination, learning ability and emotional behaviour tests.

Since considerable overt signs of toxicity including mortality were evident in dams at the high dose, the DS considered it possible that the foetal effects could be considered as secondary consequences of maternal toxicity. At the mid dose, however, the maternal toxicity was limited to transient reductions in body weight gain.

Rabbits

In a developmental toxicity study (Study SGT-20-0025, 1992) conducted under GLP and conform OECD TG 414, imiprothrin was administered to female JW-NIBS rabbits (15/dose) at 0, 30, 100 and 300 mg/kg bw/day in corn oil, by gavage, on days 6-18 of gestation. In a follow-up study, additional animals (20/group) were dosed at 0, 3, 10 or 30 mg/kg bw/day on days 6-18 of gestation, to further investigate some of the findings in the main study.

The top dose of 300 mg/kg bw/day was clearly maternally toxic with clinical signs, death (2 animals, 1 of which may have been due to a gavage error, plus 1 moribund animal), abortions or premature labour (5 animals, 1 of which was a dead animal), and statistically significantly reduced body weight gain (by 68-200%, throughout gestation), food consumption (by 29-78%, throughout treatment period) and body weight (by 7-8% on days 15-18 of gestation). Abortion or premature labour was also seen in 1 animal at 100 mg/kg bw/day, but as this also occurred in 1 control animal, the finding was considered spontaneous. Body weight gain and food consumption were also reduced at 100 mg/kg bw/day throughout gestation and treatment period, respectively, but the differences with controls were not statistically significant. No maternal toxicity was seen at 30 mg/kg bw/day.

A statistically significant reduction in foetal body weight was observed at 300 mg/kg bw/day (15-16%). Also at 100 mg/kg bw/day the foetal bodyweight tended to be reduced (4.3-4.9%; not statistically significant). No foetus showed any abnormal external characteristics. Visceral examination showed minor abnormalities and variations in each treated group, which were not different from the control group. One type of skeletal malformation was observed (fusion of the nasal bone) in addition to two types of skeletal variation (hypoplasia of the frontal bone and 27 pre-sacral vertebrae).

The incidences are shown in the table below.

Table: Incidences of skeletal malformations and variations observed in the rabbit developmental toxicity study

	Dose (mg/kg bw/day)				
	0	30	100	300	HCD
Fusion of the nasal bone					
Number of pups with effect	**1/74 (1.4%)**	**0/75 (0%)**	**1/70 (1.4%)**	**9/64* (14.1%)**	**Mean: 0.1% Range: 0.0-1.4%**
Litter incidence	1/12 (8.3%)	0/11 (0%)	1/10 (10%)	4/8 (50%)	
Foetal incidences within affected litters	1/7	N/A	1/9	3/8 1/8 3/8 2/10	
Hypoplasia of the frontal bone					
Number of pups with effect	**0/74 (0%)**	**0/75 (0%)**	**2/70 (2.9%)**	**10/64 (15.6%)**	**Mean: 0.1% Range: 0.0-1.4%**
Litter incidence	0/12 (0%)	0/11 (0%)	1/10 (10%)	2/8 (25%)	
Foetal incidences within affected litters	N/A	N/A	2/8	4/8 6/10	
27 Pre-sacral vertebrae					
Number of pups with effect	**1/74 (1.4%)**	**6/75 (8.0%)**	**7/70 (10.0%)**	**11/64 (17.2%)**	**Mean: 3.4% Range: 0.0-8.6%**
Litter incidence	1/12 (8.3%)	4/11 (36.4%)	3/10 (30%)	3/8 (37.5%)	
Foetal incidences within affected litters	1/7	1/5 3/8 1/8 1/4	3/6 1/5 3/7	6/10 1/8 4/6	

HCD: historical control data from 11 studies (1989-1992); one of which was the main oral study in rabbits and one of which was the additional study in rabbits.

* Significantly different from control group at $P < 0.05$

Fusion of the nasal bone, which the DS considered to be a malformation, was observed at the top dose in 50% of the litters with an incidence (14.1%) greatly exceeding the historical control range (0-1.4%). Hypoplasia of the frontal bone was observed at the top dose (in 2/8 litters, one litter being the same litter in which 1/8 pups showed fusion in the nasal bone, indicating a possible cause for concern for craniofacial development) and the mid dose (in 1/10 litters; however, it was also noted in 3/7 pups of a dam that was excluded from analysis due to gavage error). The incidences at the mid (2.9%) and high dose (15.6%) exceeded the historical control range (0-1.4%). It was postulated by the study author that the hypoplasia (which is considered a variation) may have been a retarded ossification related to lower foetal body weights (and thus suppression of food consumption in the dams).

A dose-related (but not statistically significant) increase in 27 pre-sacral vertebrae (regarded as a skeletal variation) was observed in all groups, with statistical significant foetal incidences at the mid dose (10.0%) and high dose (17.2%) exceeding the historical control range (0-8.6%). This effect was additionally observed in one pup of a mid-dose dam that was excluded from the analysis due to gavage error. Given that no definite conclusion could be reached regarding treatment-relationship, a follow-up study was conducted in rabbits to further investigate the 27 pre-sacral vertebrae findings.

In the follow-up study, 27 pre-sacral vertebrae was observed in 2, 6, 5 and 6 foetuses (1.8, 5.2, 3.9 and 4.8%) in the control, 3, 10 and 30 mg/kg bw/day groups respectively. The study authors concluded that there was no tendency for 27 pre-sacral vertebrae to increase in a dose dependant manner, as it had done in the main study. Various minor anomalies and skeletal variations in addition to 27 pre-sacral vertebrae were also observed, but the incidences were not different between the treated and control groups.

The DS considered the results of the rabbit study to indicate a potential for imiprothrin to induce adverse developmental effects in foetuses, including fusion of the nasal bone, hypoplasia of the frontal bone and 27 pre-sacral vertebrae. However, the DS also noted that the effects were mainly seen at maternally toxic doses.

Conclusion

Some of the developmental findings at the top dose levels in rats and rabbits (600 and 300 mg/kg bw/day, respectively) might be related to the high level of maternal toxicity. However, the DS considered that the secondary nature of all the developmental effects has not been unequivocally demonstrated, as supported by some developmental effects being seen at dose levels where maternal toxicity was limited (200 and 100 mg/kg bw/day in rats and rabbits, respectively) or absent. According to the DS, effects that cannot be dismissed completely and may form a possible cause for concern for developmental toxicity are the malformation (fusion of the nasal bone) in rabbits at 300 mg/kg bw/day, the dose-related increase in hypoplasia of the frontal bone and 27 pre-sacral vertebrae at 100 and 300 mg/kg bw/day in rabbits, and the dose-related increase in lumbar rib in the rat at 200 and 600 mg/kg bw/day. With reference to the ECETOC Guidance on Evaluation of Reproductive Toxicity Data, the DS considered that the increase in supernumery ribs and small (hypoplastic) skull bones as observed in the rat and rabbit studies could be insufficient to support classification, since these findings are designated a low-moderate level of concern. However, with reference to the same ECETOC Guidance, the DS considered the malformation in rabbits to support classification as this finding has a high level of concern. Adding to the weight of evidence are the dose-response observed for most findings, and the effects occurring in more than one litter, at least at the top dose. Had the hypoplasia and fusion of the nasal bone occurred in the absence of maternal toxicity, the DS considered that a classification for developmental toxicity in category 1B could have been warranted. Given however that the maternal toxicity reduces somewhat the level of concern, the DS considered category 2 more appropriate.

Lactation

The DS did not propose classification for effects on or via lactation because imiprothirid does not meet the criteria, for two out of three criteria due to lack of data (i.e., there is no human evidence available indicating a hazard to babies, and the ability of imiprothrin to partition into the breast milk has not been investigated). For the third criterion, the two-generation study in rats showed no evidence for an effect of imiprothrin on lactation performance.

Comments received during public consultation

One MSCA agreed to the proposal for classification in reproductive toxicity category 2 (H361d), based on the skeletal malformations in the rabbit but not the rat study.

Industry provided five expert statements (four confidential and one public), all in support of no classification for developmental toxicity. The main arguments given for no classification are:

- Fusion of the nasal bone in rabbits was only observed at the severely maternally toxic dose of 300 mg/kg bw/day (above MTD).
- Fusion of the nasal bone as observed in the rabbit study was observed only in the proximal part of the nasal suture. Because the bridge of the nasal region does not experience the radial expansion that occurs in the dome of the skull, partial joining of nasal bones across the midline will not result in anatomical malformations similar to craniosynostosis. Such nasal bone fusions would likely remodel over time, and should therefore be considered a skeletal variation rather than a malformation.

- In the rabbit study there was indeed no evidence of fusion between the nasal bones and the adjacent bones comprising the facial skeleton nor any indication of dysmorphology of the anterior part of the cranium or palate.
- Hypoplasia of the frontal bone as observed in rabbits at doses of 100 and 300 mg/kg bw/day was due to delayed ossification as a consequence of a lower foetal body weight derived from dams that were strongly affected by the treatment (reduced food consumption and body weight loss).
- Delayed ossification is a more appropriate descriptor than hypoplasia, since the overall shape of the frontal bone, as well as of the dome of the skull, was reported to be normal.
- 27 pre-sacral vertebrae is a common skeletal variation in rabbits, often in conjunction with 13th rib; they are not mechanistic precursors to malformations.
- Pre-sacral vertebrae and lumbar (or supernumery) ribs are also a common finding in rodents. The sensitive period for the induction of additional ribs/vertebra is early in the gestation period (GD 8-12), when axial specification is being established. Maternal toxicity during this period has been shown to be associated with the induction of supernumery ribs.
- The rib and vertebrae findings in rats showed substantial post-natal resolution, indicating they signify delayed ossification rather than a structural alteration in development.
- The increases in 27 pre-sacral vertebrae in the rabbit study at 100 and 300 mg/kg bw/day and in lumbar ribs/pre-sacral vertebrae in rats at 200 and 600 mg/kg bw/day can be explained as a consequence of maternal toxicity, that was already evident in both species in the early stages of embryonic development.

Two individuals did not support the proposed classification based on the findings at the top dose in the rabbit study because at this dose there was extensive maternal toxicity. One individual additionally noted that the study authors described the fusion of nasal bones only as a minor finding that was not accompanied by further evidence of fusion or abnormality of any other cranial bones.

The DS in response indicated that the proposed classification is primarily based on the findings in rabbits. According to the DS, it is the finding of fusion of the nasal bone (which with reference to the ECETOC Guidance on Evaluation of Reproductive Toxicity Data is to be considered a malformation) in this species, in combination with hypoplasia of the frontal bone, that gives rise to a cause for concern for craniofacial development. The DS acknowledged that the maternal toxicity may have contributed to the findings in rabbit foetuses (otherwise category 1B could have been justified), but still did not consider this to be unequivocal evidence that the observed effects were a secondary non-specific consequence of lower foetal bodyweight. Consequently, the DS stood by their proposal for category 2.

Assessment and comparison with the classification criteria

Fertility

In view of the absence of findings on fertility parameters in the two-generation study in rats, RAC supports the DS conclusion that imiprothrin does not need to be classified for effects on fertility and sexual development.

Developmental toxicity

In the rat developmental toxicity study, increases in a number of skeletal variations and a visceral finding (thymic remnants in the neck) were observed. These effects are indicative of delayed ossification or a manifestation of developmental delay, and showed partial or complete post-natal resolution. With the exception of lumbar ribs, all other effects were only observed at the highest dose of 600 mg/kg bw/day, a dose that was clearly maternally toxic. An increase in lumbar ribs

was additionally observed at the mid dose of 200 mg/kg bw/day, but also at this dose there was maternal toxicity, with decreased food consumption and body weight gain (in particular during gestation days 9-18). Delayed ossification was also seen in the rat two-generation study at maternally toxic dose levels. RAC considers the effects observed in rats not to constitute a high level of concern; they are considered insufficient to warrant classification.

In the rabbit developmental toxicity study, effects observed included increases in 27 pre-sacral vertebrae and in hypoplasia of frontal bone (both skeletal variations), and in fusion of nasal bone. Similar to the rats, RAC does not consider the skeletal variations, which occurred at maternally toxic dose levels of 100 and 300 mg/kg bw/day, to constitute a high level of concern; they are considered insufficient to warrant classification. As to the fusion of the nasal bone, the DS considered this to be a malformation. RAC however notes that it was a partial fusion, not a full-length fusion/hypoplasia. According to the DS it is the finding of the malformation in combination with the hypoplasia of the frontal bone that gives cause for concern for craniofacial development. RAC however notes that there was no indication of fusion of other cranial bones, and that the overall shape of the frontal bone, as well as of the dome of the skull, was apparently normal. It is moreover noted that an increase in fusion of the nasal bone was only observed at a dose that was clearly above the MTD (with clinical signs of toxicity, 2/15 dams dying, 5/15 dams having abortions, and consistently lower food consumption and body weight gain during organogenesis). Normally effects observed at dose levels above the MTD should be carefully taken into consideration as they could be secondary non-specific consequences of maternal toxicity.

Given the total weight of evidence, RAC considers that **classification for developmental toxicity is not warranted**.

Lactation

RAC supports the conclusion of the DS that imiprothrin does not meet the criteria in the CLP Regulation and therefore does not need to be classified for effects on or via lactation.

ENVIRONMENTAL HAZARD EVALUATION

RAC evaluation of aquatic hazards (acute and chronic)

Summary of the Dossier Submitter's proposal

Imiprothrin is a biocidal active substance in the scope of the Biocidal Products Regulation (Regulation (EU) No 528/2012) and is currently classified as Aquatic Acute 1 (H400) and Chronic 1 (H410) in Annex VI to the CLP Regulation (added at the 1st ATP).

Environmental testing was conducted using imiprothrin active substance as either *trans* or *cis* isomer (both in R-configuration) which together comprise approximately 90% purity in an approximate ratio of *trans*:*cis* of 80:20.

The DS proposed to revise the existing harmonised entry by adding M-factors of 10 for both acute and chronic hazards. Aquatic acute toxicity data are available for fish, invertebrates and algae. The lowest acute value is a 96-hrs LC_{50} of 0.038 mg/L for Rainbow trout (*Oncorhynchus mykiss*) resulting in a classification as Aquatic Acute 1 (H400) with an acute M-factor of 10. Chronic aquatic toxicity data on imiprothrin for fish and invertebrates are not available and a chronic 72-hrs NOErC of 1.3 mg/L for algae would not result in an Aquatic Chronic classification. However, algae are not the most acutely sensitive trophic level. Due to the lack of a full chronic dataset, the surrogate approach was applied by the DS using available acute fish and invertebrate

toxicity data. Imiprothrin is not considered rapidly degradable for classification purposes, consequently the chronic hazard classification would result in Aquatic Chronic 1 (H410) with a chronic M-factor of 10.

Degradation

Hydrolysis of imiprothrin was tested according to US EPA Subdivision N guideline 161-1 (similar to OECD TG 111) following GLP principles. The substance was hydrolytically stable at pH 5, while hydrolysis was observed at pHs 7 and 9, increasing with alkalinity (half-lives of 58.6 days at pH 7 and 0.746 days at pH 9 at a study temperature of 25°C, respectively). Converting these values to 12°C results in half-lives of 166 days at pH 7 and 2.11 days at pH 9. One major hydrolysis product was formed, CPG (N-carbamoyl-N-propargylglycine; formed as a result of hydrolysis of the intermediate product PGH (1-propargylimidazolidine-2,4-dione)) reaching a maximum of 26.5 and 89.8% at pH 7 and 9, respectively. Based on structural similarity to other pyrethroids, KCA (chrysanthemic acid) was also considered as a relevant hydrolysis product.

Photodegradation of imiprothrin was not tested. However, consideration of structurally similar pyrethroids indicates that photodegradation may occur under experimental conditions with the formation of degradants such as KCA and imidazolidone.

Ready biodegradation was tested following the Japanese MITI-I method (similar to OECD TG 301 C). Despite the study not being conducted according to GLP principles, it was considered valid under Regulation (EU) No 528/2012. The study was run using 100 mg/L imiprothrin and a closed system at 25°C and pH 7. While primary degradation did occur with a residual amount of imiprothrin of 58% at day 28, the biodegradation rate based on oxygen consumption was 2% indicating minimal mineralisation. The main primary degradation products were PGH and KCA, amounting to 50 and 45%, respectively.

In an aerobic water/sediment simulation study conducted according to OECD TG 308 and following GLP principles, radiolabelled 1R-cis and 1R-trans isomers of imiprothrin were used. The study was run at 20°C±2°C in the dark for 101 days. The test item was observed to dissipate from the water column to sediment. Primary degradation was rapid with imiprothrin DT_{50} total system values between 1.37 and 5.4 days at 20°C (2.6 and 10.2 days at 12°C; recalculated by the DS). Ultimate degradation was slower with mineralisation at 7.5 to 11.2% applied radioactivity (% AR) on day 31 and 39.6 to 52.3% AR by study termination on day 101, indicating that imiprothrin does not have an ultimate degradation half-life of <16 days. The DT_{50} total system values for the three major degradants (PGH, CPG and PG (propargylglycine)) showed that they have longer half-lives than imiprothrin.

Overall, the DS concluded that the available information on degradation of imiprothrin was not sufficient to show that the substance is ultimately degraded within 28 days (equivalent to a half-life of <16 days) or transformed to non-classifiable degradants. As a result imiprothrin was considered non-rapidly degradable for classification purposes.

Bioaccumulation

A Log Kow of 2.9 (at 25°C and pH 6.2–6.6) was measured for imiprothrin following the EC method A.8. (Shake flask method). An experimental aquatic BCF study in Bluegill Sunfish (*L. macrochirus*) following OECD TG 305 and GLP principles showed a lipid normalised whole fish BCF of 124 to 144 L/kg (based on total ^{14}C-residues) and of 4.6-5.8 L/kg (based on ^{14}C-imiprothrin). The DS concluded that imiprothrin does not meet the CLP criteria for bioaccumulation, given both the Log Kow and the experimentally derived BCF were below the CLP trigger values of ≥ 4 (for Log Kow) and ≥500 (for BCF), respectively.

Aquatic toxicity

Valid aquatic acute toxicity data are available for fish, invertebrates and algae with fish being the most sensitive trophic level. Valid aquatic chronic toxicity data are available for algae only, while data for fish and aquatic invertebrates are lacking. All values were based on mean measured concentrations. A summary of the relevant information on aquatic toxicity is presented in Table 7.

Table 7. Summary of relevant information from aquatic toxicity studies on imiprothrin

Guideline / GLP status	Species	Endpoint	Exposure		Results		Reference
			Design	Duration	Endpoint	Toxicity (mg/L)	
Acute toxicity to fish US EPA FIFRA 72-1, GLP, purity: 92.9%	Bluegill Sunfish (*Lepomis macrochirus*)	Mortality	Flow-through	96 hours	LC_{50}	0.07 (mm)	ABC Laboratories, Inc, USA (1993a)
Acute toxicity to fish US EPA FIFRA 72-1, GLP, purity: 92.9%	Rainbow Trout (*Oncorhynchus mykiss*)	Mortality	Flow-through	96 hours	LC_{50}	**0.038** (mm)	ABC Laboratories, Inc, USA (1993b)
Daphnia sp Acute Immobilisation US EPA FIFRA 72-2, GLP, purity: 92.9%	*Daphnia magna*	Acute immobili-sation	Flow-through	48 hours	EC_{50}	0.051 (mm)	Bowman and Stuerman, 1993c
Freshwater Algal Growth Inhibition OECD TG 201, GLP, purity: 91.6%	*Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*)	Cell multipli-cation inhibition	Static	72 hours	ErC_{50} NOErC	>7.8 (mm) **1.3** (mm)	Bell, 1996a

Two acute fish studies (following US EPA FIFRA 72-1 guideline and according to GLP principles) were conducted using Bluegill Sunfish (*L. macrochirus*) and Rainbow trout (*O. mykiss*) under flow-through conditions over a period of 96 hours. In both studies exposure concentrations were prepared using the solvent dimethylformamide (DMF; 0.1 mL/L)) and a solvent control was included. The reported 96-hrs LC_{50} values were 0.07 mg/L (mm) in the first test and 0.038 mg/L (mm) in the second test. The measured concentrations of imiprothrin remained within 80–120% for all measured samples in the first test and increased up to 133% at 96 hours in the second test.

An acute study with *D. magna* was conducted under flow-through conditions following US EPA FIFRA 72-2 guideline and according to GLP principles, resulting in a 48-hrs EC_{50} of 0.051 mg/L (mm).

A static algal growth inhibition study was conducted following OECD TG 201 and according to GLP principles. The 72-hrs ErC_{50} was reported to be >7.8 mg/L (mm) and the 72-hrs NOErC 1.3 mg/L (mm). It was noted that greater inhibition was observed at 48 hours, indicating that the 48-hrs ErC_{50} would be between 3.2 and 7.8 mg/L based on mean measured concentrations. The DS concluded that for the purpose of classification, endpoints based on an exposure of 72 or 96 hours are preferred. Therefore, the reported endpoints at 72 hours are used for classification purposes.

Based on the available information for aquatic acute toxicity, the DS concluded that imiprothrin meets the classification criteria as Aquatic Acute 1 with an M-factor of 10 based on the lowest 96-hrs LC_{50} of 0.038 mg/L for rainbow trout. Due to the lack of reliable chronic toxicity data for the acutely most sensitive trophic level, the DS applied the surrogate approach. Considering that imiprothrin is non-rapidly degradable, this resulted in a classification as Aquatic Chronic 1 with an M-factor of 10.

Comments received during public consultation

Two MSCAs supported the environmental classification as proposed by the DS.

Assessment and comparison with the classification criteria

The measured water solubility of imiprothrin is 93.5 mg/L at 25°C and pH 6.5. Imiprothrin is not anticipated to dissociate. Experimental data indicate the vapour pressure is low at 1.86 x 10^{-6} Pa at 25°C. The Henry's Law Constant of 6.33 x 10-6 Pa m^3 mol^{-1} indicates that imiprothrin is unlikely to partition significantly from the water phase to air. Imiprothrin is also surface active (surface tension 46.6 mN/m at 21°C). Measured data indicate that imiprothrin is likely to be moderately mobile in soil, Log K_{oc} of 2.43.

Degradation

Imiprothrin is hydrolytically stable at pH 5 and it undergoes hydrolysis with increasing alkalinity. Hydrolysis DT_{50} values at 12°C are 166 days at pH 7 and 2.11 days at pH 9, at 25°C 58.6 days at pH 7 and 0.746 days at pH 9. Two hydrolysis products were considered relevant, CPG (N-carbamoyl-N-propargylglycine) and KCA (chrysanthemic acid). Data on hydrolysis might be considered for classification purposes only when the longest half-life determined within the pH range 4-9 is less than 16 days (corresponding to a degradation of >70% within 28 days). Accordingly, imiprothrin is hydrolytically stable.

In a 28-day ready biodegradability study 2% mineralisation was observed, while primary degradation amounted to 42%. Imiprothrin is considered not readily biodegradable.

In an aerobic water/sediment simulation study, whole system DT_{50}s for imiprothrin were between 2.6 and 10.2 days at 12°C. Mineralisation was observed with 7.5 to 11.2 % AR on day 31 and 39.6 to 52.3% AR by day 101. Total system DT_{50}s for three principal degradants were between 7.4 and 97.7 days. The degradation data does not support that imiprothrin would fulfil the criteria for ultimate degradation in the aquatic environment with a half-life of <16 days (corresponding to a degradation of >70% within 28 days) or that it is transformed to non-classifiable products in that period.

Overall conclusion on degradation: RAC agrees with the DS's proposal to consider imiprothrin as not rapidly degradable for the purpose of classification and labelling.

Aquatic Bioaccumulation

Lipid normalised whole fish BCFs based on total ^{14}C-residues were 124-144 L/kg, and 4.6-5.8 L/kg based on ^{14}C-imiprothrin. The measured BCFs values are below the CLP criterion of BCF ≥500. The low bioaccumulation potential of imiprothrin is also supported by the experimental (shake-method flask) Log K_{ow} of 2.9, which is below the CLP trigger value of Log K_{ow} ≥4. It is noted that for surface-active substances, the shake-flask method is not the most suitable experimental method to determine the Log K_{ow} due to micelle/emulsion formation. According to the REACH Guidance (Chapter R.7a section 7.1.8.5), in many cases a calculated K_{ow} value based on the octanol and water solubilities will be the first choice for surfactants. In this regard, a Log

K_{ow} value of 2.98 was calculated by RAC using KOWIN (v1.68 estimate). This value is in the same range as the experimental Log K_{ow} value of 2.90 for imiprothrin. It is considered that at best the experimental Log K_{ow} value could be used as supportive data since experimental BCF values are available. Therefore, RAC agrees with the DS proposal to consider imiprothrin as a substance with a low potential to bioaccumulate.

Aquatic toxicity

Aquatic acute toxicity data on imiprothrin are available for fish, invertebrates and algae (table above). Acute endpoints for fish and invertebrates lie in the range of 0.01 to 0.1 mg/L. The lowest acute aquatic toxicity value is a 96-h LC_{50} of 0.038 mg/L for the fish rainbow trout. According to Tables 4.1.0(a) and 4.1.3 of the CLP guidance, imiprothrin should be classified as Aquatic Acute 1 with an acute M-factor of 10.

Aquatic chronic toxicity data on imiprothrin is available for one trophic level, algae. In the absence of adequate long-term toxicity data for fish and aquatic invertebrates, the surrogate method is applied as recommended in CLP guidance section 4.1.3.3 and Table 4.1.0. The substance is considered not rapidly degradable and does not fulfil the criteria for bioaccumulation.

- Classification based on adequate chronic toxicity data. Algae long-term testing provide a 72-h NOErC of 1.3 mg/L. The NOErC is above 1 mg/L and the substance is not rapidly degradable. Imiprothrin does not fulfil the criteria for chronic classification, based on Table 4.1.0 (b)(i).
- Classification based on surrogate data for fish and aquatic invertebrates. The lowest acute toxicity value is a 96-h LC_{50} of 0.038 mg/L for rainbow trout. The 96-h LC_{50} is ≤1 mg/L and the substance is not rapidly degradable. Imiprothrin fulfils the criteria of category Chronic 1, based on Table 4.1.0(b)(iii).
- Overall conclusion: category Chronic 1 applies following the most stringent outcome.
- The M-factor is based on the acute aquatic toxicity between 0.01 and 0.1 mg/L.

Conclusion on Classification

RAC concludes that imiprothrin fulfils the CLP criteria for classification as **Aquatic Acute 1** with an **M-factor of 10** and **Aquatic Chronic 1** with an **M-factor of 10**.

ANNEXES:

Annex 1 The Background Document (BD) gives the detailed scientific grounds for the opinion. The BD is based on the CLH report prepared by the Dossier Submitter; the evaluation performed by RAC is contained in 'RAC boxes'.

Annex 2 Comments received on the CLH report, response to comments provided by the Dossier Submitter and RAC (excluding confidential information).