Committee for Risk Assessment

RAC

Opinion

proposing harmonised classification and labelling
at EU level of

metaflumizone (ISO); (*EZ*)-2'-[2-(4-cyanophenyl)-1-(α,α,α -trifluoro-m-tolyl)ethylidene]-[4-(trifluoromethoxy)phenyl]carbanilohydrazide [*E*-isomer ≥ 90%, *Z*-isomer ≤10% relative content] [1]

(*E*)-2'-[2-(4-cyanophenyl)-1-(α,α,α -trifluoro-m-tolyl)ethylidene]-[4-(trifluoromethoxy)phenyl]carbanilohydrazide [2]

EC Number: -
CAS Number: [1] 139968-49-3; [2] 852403-68-0

CLH-O-0000001412-86-179/F

Adopted

5 December 2017

5 December 2017

CLH-O-0000001412-86-179/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:	**metaflumizone (ISO); (EZ)-2'-[2-(4-cyanophenyl)-1-(α,α,α -trifluoro-m-tolyl)ethylidene]-[4-(trifluoromethoxy)phenyl]carbanilohydrazide [E-isomer > 90%, Z-isomer <10% relative content] [1], (E)-2'-[2-(4-cyanophenyl)-1-(α,α,α -trifluoro-m-tolyl)ethylidene]-[4-(trifluoromethoxy)phenyl]carbanilohydrazide [2]**
EC Number:	**-**
CAS Number:	**[1] 139968-49-3; [2] 852403-68-0**

The proposal was submitted bythe **United Kingdom** and received by RAC on **7 November 2016.**

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 **20 December 2016**. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by **13 February 2017**.

ADOPTION OF THE OPINION OF RAC

Rapporteur, appointed by RAC: **Agnes Schulte**

Co-Rapporteur, appointed by RAC: **Katalin Gruiz**

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 **5 December 2017** 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 statement Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	No current Annex VI entry										
Dossier submitter's proposal	616-RST-VW-Y	metaflumizone (ISO); (*EZ*)-2'-[2-(4-cyanophenyl)-1-(α,α,α-trifluoro-m-tolyl)ethylidene]-[4-(trifluoromethoxy)phenyl]carbanilohydrazide [*E*-isomer ≥ 90%, *Z*-isomer ≤10% relative content] [1], (*E*)-2'-[2-(4-cyanophenyl)-1-(α,α,α-trifluoro-m-tolyl)ethylidene]-[4-(trifluoromethoxy)phenyl]carbanilohydrazide [2],]		[1] 139968-49-3 [2] 852403-68-0	Repr. 2 Lact. STOT RE 2	H361d H362 H373 (oral, inhalation)	GHS08 Wng	H361d H362 H373 (oral, inhalation)			
RAC opinion	616-RST-VW-Y			[1] 139968-49-3 [2] 852403-68-0	Repr. 2 Lact. STOT RE 2	H361fd H362 H373	GHS08 Wng	H361fd H362 H373			
Resulting Annex VI entry if agreed by COM	616-RST-VW-Y			[1] 139968-49-3; [2] 852403-68-0	Repr. 2 Lact. STOT RE 2	H361fd H362 H373	GHS08 Wng	H361fd H362 H373			

GROUNDS FOR ADOPTION OF THE OPINION

RAC general comment

There is no existing entry in Annex VI of CLP for metaflumizone. Therefore, all human health and environmental endpoints have been evaluated.

Metaflumizone consists of two stereoisomers: in the substance evaluated the E and Z isomer content was 91% and 6.35%, on average respectively.

With regard to impurities, there is no concern for classification at the proposed levels. Individual impurities are discussed in the DAR in detail.

RAC evaluation of physical hazards

Summary of the Dossier Submitter's proposal

Information provided in the CLH report summarised that metaflumizone is not flammable and not self-ignitable and does not evolve flammable gases in contact with water. Metaflumizone has no explosive properties.

The dossier submitter (DS) proposed no classification for physical hazards.

Comments received during public consultation

No comments on physical hazards were received during the public consultation.

Assessment and comparison with the classification criteria

Metaflumizone does not meet the relevant CLP criteria and therefore RAC supports the proposal of the DS not to classify metaflumizone for physical hazards.

HUMAN HEALTH HAZARD EVALUATION

RAC evaluation of acute toxicity

Summary of the Dossier Submitter's proposal

The CLH dossier presented acute toxicity OECD Test Guideline studies on all routes of exposure in rats. In addition, a guideline-compliant acute oral toxicity study in mice is available. The oral LD_{50} values in a mouse and rat study and the dermal LD_{50} value in a rat study were found to be > 5000 mg/kg bw for males and females combined. The inhalation LC_{50} value in a 4-hour rat study (nose-only exposure) was found to be above the tested concentration of 5.3 mg/L (mass median aerodynamic diameters 3.6 ± 3.2 µm) for males and females combined. These values are all above the guidance values for classification for acute toxicity. Therefore, the DS proposed no classification for acute toxicity for all routes.

Comments received during public consultation

One MSCA asked for the exact concentration tested in the acute inhalation study. No other comments were received.

Assessment and comparison with the classification criteria

The oral LD_{50} values in a mouse and rat study of above 5000 mg/kg bw does not justify acute toxicity category 4 (oral; 300 < LD_{50} ≤ 2000 mg/kg bw). The LC_{50} value in a 4-h rat study (nose-only exposure) of 5.3 mg/L does not justify acute toxicity category 4 (inhalation; 1.0 < LC_{50} ≤ 5.0 mg/L for dusts and mists). The dermal LD_{50} value in a rat study of above > 5000 mg/kg bw does not justify acute toxicity 4 (dermal; 1000 < LD_{50} ≤ 2000 mg/kg bw). Apart from one animal dying 15 min after dermal application, no other mortalities were seen in the available acute toxicity studies in rats and mice.

RAC agrees with the conclusions of the DS that all LD_{50} and LC_{50} estimates were above the guidance values for classification and **no classification is warranted for acute toxicity for the oral, inhalation and dermal routes.**

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

Summary of the Dossier Submitter's proposal

The DS concluded that classification as STOT SE 1 or 2 is not warranted since no signs of specific target organ toxicity were identified in the acute oral, inhalation and dermal toxicity studies.

Category 3 for specific target organ toxicity following a single dose is reserved for substances or mixtures causing transient target organ effects such as narcotic effects and upper respiratory irritation. The DS concluded that STOT SE 3 classification is not warranted based on the observations in the acute inhalation study where rats showed accelerated respiration, squatting posture and red discolouration of the lobes of the lungs. In a 28-d repeated dose inhalation study in rats, minimal hypertrophy of the respiratory epithelium was noted in all animals at a dose of 0.7 mg/L and in 2/5 males and females at 0.1 mg/L. Minimal to slight multifocal hyperplasia of type II cells and higher incidence of alveolar macrophages was observed in animals from a dose of 0.1 mg/L.

Comments received during public consultation

No comments on STOT SE were received.

Assessment and comparison with the classification criteria

RAC agrees with the DS that STOT SE 1 and 2 are not warranted. No specific target organ toxicity after single exposure was identified.

Regarding STOT SE 3, no evidence of narcotic effects and no unequivocal evidence of transient irritation of the upper or lower respiratory tract were observed. Accelerated respiration was observed during exposure and could either be considered as signs of toxicity or as signs of irritation. Red discoloration of the lung lobes was noted at the end of the 14-d observation period and is not considered to indicate a transient (acute) hyperaemia. Hypertrophy of the respiratory epithelium and hyperplasia of type II alveolar cells are effects that need repeated exposure and

thus are to be considered under STOT RE. As no specific evidence on the transient nature and the sensory/cytotoxic irritation is given and animals were tested at relatively high test concentrations, RAC agrees with the Dossier Submitter on **no classification for STOT SE**

RAC evaluation of skin corrosion/irritation

Summary of the Dossier Submitter's proposal

An OECD TG study with three male New Zealand white rabbits did not reveal observations of erythema or oedema at any time-point during the study after exposure to metaflumizone (500 mg) in distilled water (0.5 mL) under semi-occlusive conditions for four hours. The DS concluded that classification of metaflumizone as a skin irritant is not warranted.

Comments received during public consultation

No comments on skin corrosion/irritation were received.

Assessment and comparison with the classification criteria

RAC agrees with the DS's proposal that **classification for skin irritation/corrosion is not warranted** based on the available study (using water as a vehicle).

RAC evaluation of serious eye damage/irritation

Summary of the Dossier Submitter's proposal

Metaflumizone tested in a OECD TG study was shown to cause minimal irritation in the form of conjunctival erythema in three rabbits (grade 1) at 24 hours, which was cleared by 48 hours post-instillation. There was no evidence of corneal opacity or iritis in any of the rabbits. The DS considered that metaflumizone does not meet the criteria for classification as an eye irritant.

Comments received during public consultation

No comments on serious eye damage/irritation were received.

Assessment and comparison with the classification criteria

The average score for conjunctival erythema for each rabbit was 0.33 and there was no corneal opacity or iritis. Regarding classification for eye irritation, this is below the average score for conjunctival erythema specified in the CLP criteria of ≥ 2 (24-72 h) in at least two out of three animals.

RAC agrees that **no classification for serious eye damage/irritation is warranted**.

RAC evaluation of skin sensitisation

Summary of the Dossier Submitter's proposal

Metaflumizone did not cause skin sensitisation in a OECD TG maximisation test in Guinea pigs. According to the DS's proposal, no classification is justified for this hazard class.

The CLH dossier informs about intradermal and topical induction doses that caused moderate to intense erythema. The test guideline recommends that selected concentrations should cause mild to moderate skin irritation.

Comments received during public consultation

No comments on skin sensitisation were received.

Assessment and comparison with the classification criteria

Metaflumizone did not cause a positive response in a guideline-compliant maximisation test in Guinea pigs. RAC agrees with the DS's proposal that **classification for skin sensitisation is not warranted**.

RAC evaluation of germ cell mutagenicity

Summary of the Dossier Submitter's proposal

Based on the available data, the DS did not propose to classify metaflumizone for germ cell mutagenicity.

***In vitro* tests**

A bacterial gene mutation test (Ames test, OECD TG 471, GLP) and a mammalian cell gene mutation test (HPRT test, Chinese hamster V79 cells, OECD TG 476, GLP) were negative with and without metabolic activation. A mammalian chromosomal aberration test with V79 cells showed negative results when metaflumizone was tested in the presence of metabolic activation. When tested in the absence of metabolic activation there was a statistically significant increase of chromosomal aberrations (OECD TG 473, GLP).

***In vivo* tests**

A micronucleus test (OECD TG 474, GLP) in mice was negative up to the highest tested dose of 2000 mg/kg bw after i.p. injection. Clinical signs were observed at all tested doses. No information was given on cytotoxic effects (PCE/NCE).

Furthermore, an unscheduled DNA synthesis test (indicator test, OECD TG 486, GLP) in rats was negative after oral administration (gavage) of doses up to 2000 mg/kg bw. No signs of clinical toxicity were observed.

In summary, the induction of clastogenic effects in a positive *in vitro* chromosomal aberration test was not confirmed in a negative *in vivo* mutagenicity test (micronucleus test) in mice. Overall, the DS concluded that metaflumizone does not have mutagenic potential *in vivo*.

Comments received during public consultation

No comments on germ cell mutagenicity were submitted.

Assessment and comparison with the classification criteria

RAC concluded in agreement with the DS's proposal that no classification for germ cell mutagenicity is warranted.

Metaflumizone did not induce gene mutations in bacteria or in a mammalian cell culture (V79 cells) *in vitro* but showed clastogenic activity in a mammalian cell culture (V79 cells) *in vitro* only in the absence of metabolic activation. Based on the negative *in vivo* micronucleus test no mutagenicity was induced in somatic cells (criterion for classification as Category 2). Information on the induction of germ cell mutagenicity (criterion for classification as Category 1B) is not available.

RAC considers that metaflumizone does not meet the classification criteria as defined in CLP, and **no classification as a germ cell mutagen** is therefore proposed.

RAC evaluation of carcinogenicity

Summary of the Dossier Submitter's proposal

The DS concluded, based on two carcinogenicity studies by gavage that were included in the CLH report, that no metaflumizone-related increase in neoplastic findings were observed in rats at 30, 60, and 300/200 mg/kg bw/d (24 months, OECD TG 453, GLP), or in mice at 100, 250, and 1000 mg/kg bw/d (18 months, OECD TG 451, GLP) and thus no classification was proposed.

A slightly increased mortality in females at 300/200 mg/kg bw/d was noted during the first 12 months; however, the pattern of mortality (2 animals at 4 and 5 months and 1 animal at months 6, 7, 9 and 11, respectively) was considered by the DS, and in the DAR, as not indicating a relationship to treatment.

Non-neoplastic findings in rats were reduced bw and bw gain in females at 300 mg/kg bw/d that resulted in lowering of the dose to 200 mg/kg bw/d during week 3 of the study. Increased incidences of hepatocellular hypertrophy in males at ≥ 60 mg/kg bw/d and females at 200 mg/kg bw/d as well as basophilic hepatocellular alteration in males at 60 mg/kg bw/d were seen.

In mice, lower final bw was seen in males at 1000 mg/kg bw/d. In addition, increased erythrocyte turnover through haemolysis (increased red blood cell; RBC), decreased mean corpuscular hemoglobin concentration (MCHC, in males, also at 250 mg/kg bw/d), decreased mean cell haematocrit (MCH, females at 12 and 18 months), increased reticulocytes (at 18 months) increased splenic haemosiderosis in males at 12 and 18 months and females at 18 months) were seen.

Comments received during public consultation

No specific comments regarding carcinogenicity were received.

Assessment and comparison with the classification criteria

RAC does not agree with the interpretation that the slight increase in the incidence of basophilic hepatocellular alterations in male rats at doses ≥ 60 mg/kg bw/d was of an adaptive nature. Basophilic alterations could be linked to liver tumorigenesis. However, these lesions were also observed spontaneously and might or might not progress to liver tumours or also regress spontaneously. In the rat study on metaflumizone, the incidences of basophilic hepatocellular

alterations in males were 25% (15/60), 26.7% (16/60) at 60 and 300 mg/kg bw/d, respectively, versus 12% (7/60) in the controls.

No increase in liver tumours was seen in treated male and female rats and no increase of basophilic foci was seen in treated female rats. A summary table on the tumour incidences was not included in either the CLH report or in the DAR study summary; thus any further details of these studies could not be evaluated by RAC.

As no treatment-related increase in tumour responses were reported and no concern was identified for somatic cell mutagenicity, RAC supports the DS's proposal that **no classification for carcinogenicity is warranted**.

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

Summary of the Dossier Submitter's proposal

In the repeated dose studies included in the CLH report, an increase in mortality was noted in animals exposed to metaflumizone. This was not observed in every study, but was observed in studies conducted via both the oral (mouse and dog) and inhalation (rat) routes of exposure at doses relevant for classification (see table 16 of the Background Document (BD)). A clear basis for the increase in mortality has not been established, but animals were described as having reduced general condition and often marked weight loss. Significant effects on bodyweight and bodyweight gain were observed consistently in the available studies at doses relevant for classification (table 16 of the BD). Effects on organs such as the spleen, thymus, uterus and ovaries were prevalent; however, these were considered secondary to the marked weight loss and weight gain effects. Whilst the underlying cause of the increased mortality and the bodyweight effects is unclear, the severity of the findings indicates that classification is warranted. In general, the effects occurred at doses relevant for classification as STOT RE 2; however, in some cases, bodyweight was markedly reduced at doses much lower than this and relevant for STOT RE 1. In these cases, the severity of the effect was lower and varied between sexes and therefore, classification as STOT RE 2 was considered more appropriate by the DS. As the cause of mortality has not been established, no tissue/organ was been proposed as the target organ. These effects were observed via the oral and inhalation routes of exposure, no effects of relevance to classification were observed via the dermal route. Therefore, the DS considered it appropriate to specify the oral and inhalation routes of exposure in the hazard statement.

Comments received during public consultation

Three MSCAs supported the proposed classification for STOT RE, two in agreement with Category 2, another tending towards Category 1.

One MSCA questioned whether palatability could have contributed to the effects on body weight since the toxicity was observed at lower doses in the diet than by gavage administration. Another MSCA proposed to consider the 90-d study with 15 rats/sex/group (study no. 20, table 16 BD) as the most reliable study considering that the available 28-d studies included only 5 animals/sex/group.

Assessment and comparison with the classification criteria

Please note that table 16 of the BD summarises the effects on mortality and body weights that were seen below the guidance values for STOT RE 2 and STOT RE 1. The order of the studies reflects the relative importance of arguments supporting the classification proposal.

Regarding the effects at doses with relevance for STOT RE 1:

In all rat studies (oral and inhalation) that showed lower body weight gain (BWG) and lower (absolute) body weight at doses relevant for STOT RE 1 (studies no. 18, 20, 14 and 25, in the right-hand column of table 16 in the BD), a lower food consumption (FC) was also indicated. For the oral studies (no. 18 and 20) the level of reduction in FC was at a similar range as the body weight effects.

In the two inhalation studies (no. 14 and 25), rats showed either lower BWG or, in the case of the female rats in study 25, even body weight loss. Food consumption was lower (-15 to – 33%) in female rats in both studies, while lower FC (up to -8%) was observed in the male rats of study 25 from week 1 to 3 (except the last week) (see table 16 of the BD).

In the mouse study (no. 23) lower BWG was observed at 8.2 mg/kg bw/d (-21%) which was in the same range as the reduction of the food consumption at this dose (-16%) (this information is in addition to table 16 of the BD). However, the reduction in FC in these groups of male mice did not show a clear dose response; the effect on BWG and FC were even stronger in males at 4.3 mg/kg bw/d (FC – 20%, BWG -47% in comparison to control values). In female mice, the food consumption was lower in all dose groups than in the controls, gaining significance in the low and high dose groups without a dose-response relationship. These effects did not match the BWG which was highest in the low dose group.

In conclusion, the effects on the BWG and absolute body weight were reflected by the FC or in the case of the mouse study, both BWG and FC were reduced, but did not show a clear dose-response relationship. Overall, the effects observed at doses relevant for STOT RE 1 are not considered to be serious effects that warrant classification per se. However, some uncertainties remain as the lower food consumption in the rat inhalation study appears unrelated to palatability.

Regarding the effects at doses relevant for STOT RE 2:

- *12-month (gavage) dog study (no. 24)*

Beagle dogs (5/sex/group) were administered capsules containing metaflumizone at doses of 0, 6, 12, 30 or 60/40/30 mg/kg bw/d.

Premature sacrifices of 2/5 female dogs (receiving 30 mg/kg bw/d by gavage administration) due to their reduced general state, vomiting, ataxia and lateral position warrant classification with STOT RE 2.

This dose, 30 mg/kg bw/d, is below the guidance values for STOT RE 2 (≤ 100 mg/kg bw/d for 90 days corresponds to ≤ 43 mg/kg bw/d for 215 days, the date at which one of the two females were sacrificed (one at day 215 and the second at day 237)).

While the sacrifice of two females and one male of the top dose on day 57 should be attributed to the dose of 60 mg/kg bw/d that was administered until day 49, sacrifice of one additional male and female on day 250 and 226 of this dose group could be attributed to the dose level of 40 mg/kg bw/d (which was administered after day 49) which is still at a dose that warrants STOT RE 2. The high dose was further reduced to 30 mg/kg bw/d on day 245.

One male given 12 mg/kg bw/d was found dead on day 317. The study report indicated that this death was not treatment-related, but did not explain the reasons. These additional sacrifices/deaths support classification as STOT RE 2.

According to the report, lower body weights, reduced BWG and/or body weight loss and reduced FC in treated dogs were noted at ≥ 30 mg/kg bw/d. Lower BWG in female dogs (-33%, -21% and -39%, respectively) were observed at 12, 30 and 60/40/30 mg/kg bw/d (low, mid and high doses). No information on the percentage of reduced FC was, however, given. Thus reduced BGW after gavage administration of metaflumizone, with no information on the contribution of reduced FC on the impaired growth, is considered as supporting evidence of non-specific toxicity of metaflumizone ≥ 12 mg/kg bw/d.

A higher incidence rate of haemosiderosis in Kupffer cells of the liver at ≥ 12 mg/kg bw/d and lower MCHC at ≥ 30 mg/kg bw/d may indicate an (intravascular) haemolytic, hypochromic anaemia. Treatment-related effects on other blood parameters were not seen (not fully documented in detail, the DAR indicated a tendency to lower Hb and higher total bilirubin from day 173 at ≥ 30 mg/kg bw/d). No data on the grading of haemosiderosis was reported. Spleen weights increased up to 57% in both sexes at all doses without a clear dose-response relationship. In male animals, due to the magnitude of the increase, it was considered as toxicologically relevant in the study report (Table B.6.24 in the DAR). The statement in the study report that there was no histopathological correlate to this increase in spleen weight seems not plausible, or the study was limited in the methods to identify the composition in the spleen compartments. No cause of death or obvious mode of action was identified.

Although no clear picture of the severity of the anaemic effects was obvious, haemosiderosis and hypochromasia at dose levels of ≥ 12 mg/kg bw/d are adverse effects that are not sufficient in themselves to support classification, but could be interpreted as supporting the proposed classification. It should be noted that table 16 of the BD indicates mortality in 2/5 male dogs, which deviates from the original summary in the DAR. It can also be noted that an increased spleen weight may be due to different effects that may occur simultaneously such as increased (haemolytic) erythrocyte degradation and macrophage activation in the red pulpa, haemosiderin deposition and/or hyperaemia.

- *28-d (inhalation, nose-only) rat study (no. 14)*

Wistar rats (5/sex/group) were exposed to metaflumizone at 0, 0.03, 0.1 and 0.7 mg/L.

One male rat died on day 17 at 0.7 mg/L, three female rats were sacrificed on days 9-10. Severe ataxia, apathy, lateral/abdominal position, tremor, splayed limbs and reduced general condition were reported for these animals and comparable signs were seen for surviving animals at this dose. These adverse effects at 0.7 mg/L occurred at the upper range of the guidance value for classification ($0.06 < C \leq 0.6$ mg/L for a dust aerosol). Due to their delayed occurrence during the study, the mortalities/moribundities could not be attributed to acute inhalation toxicity and are considered to support classification as STOT RE 2.

Lower BWG at 0.1 and 0.7 mg/L in male and female rats and even body weight loss at 0.7 mg/L (final body weight was 20/30% lower compared to day 0 in the male/female rats) was linked to lower FC (-40% and –64% in males and females, respectively). Unlike in the discussion of the diet studies in rats, reduced food consumption throughout the inhalation study cannot be explained by palatability problems and should therefore be considered as an effect secondary to the poor general health status and/or erosions/ulcer observed in the glandular stomach at 0.7 mg/L. However, no data on the incidences/severity of erosions/ulcer were given in the study summary in the DAR or the CLH report.

Besides the observed systemic toxic effects, minimum to marked alveolar inflammation and histiocytosis, minimum to slight multifocal hyperplasia of type II pneumocytes at 0.1 and 0.7 mg/L and histiocytic granulomas in BALT/mediastinal lymph nodes at 0.7 mg/L indicated that metaflumizone is accumulated in the lungs, induces granulomatous alveolar and lymph node inflammation and alveolar hyperplasia, and is transported to local lymph nodes.

In the nose, hypertrophy of respiratory epithelium was seen in all animals at 0.7 mg/L and in two males and females at 0.1 mg/L. Although most of the effects on the respiratory tract were not reported to be of marked severity, they are considered as adverse and their occurrence started at 0.1 mg/L, a concentration which is close to STOT RE 1, these effects are considered to support the proposal for STOT RE 2.

Lesions in the thymus, lymph nodes and spleen, indicating an immunosuppressive/cytotoxic effect on T-lymphocytes at 0.7 mg/L and with lower severity/incidences at 0.1 mg/L, support the need for classification. The DS's view that the effect on the immune system is secondary to the reduced BWG and stress may justified. However, an independent immunotoxic effect cannot be ruled out.

Please note that the guidance value range of $0.2 < C \leq 1$ mg/L referred to in table 16 of the BD refers to vapour.

- *28-d (inhalation, whole body) rat study (no. 25)*

In the second 28-d inhalation study, Wistar rats (10/sex/group) were exposed to 0 or 0.03 mg/L only (i.e. control and the low concentration of the study mentioned above). Even at this low concentration, lower body weight, BWG and FC were observed. A slight increase in the severity of adrenal cortical cytoplasmic vacuolation in female rats and an increase in the grade of haemosiderin deposition in male and female rats were also seen.

- *28-d (diet) mouse study (no. 22)*

Mice (5/sex/group) received 0, 50, 200 or 800 ppm metaflumizone in their diet (equivalent to 0, 10, 42 and 101 mg/kg bw/d for both males and females).

Mortalities at 800 ppm during week 1 and 2 in 5/5 male and 4/5 female mice and clinical signs of toxicity (ataxia, convulsions) support the proposal for classification as STOT RE 2 based on the guidance values of $30 < C \leq 300$ mg/kg bw/d for this type and duration of study. Body weight loss, lower BWG and reduced FC, splenic atrophy in males and females at 800 ppm and lower mean BWG in females at 200 ppm (42 mg/kg bw/d) were additional effects supporting classification as STOT RE 2.

- *28-d/90-d (gavage) range-finding rat study (no. 21)*

Rats (5/sex/group) were administered metaflumizone in 0.5% aqueous carboxymethylcellulose (CMC) by gavage at dose levels of 0, 100, 500, 1000 mg/kg bw/d for 28 days or 0, 100 mg/kg bw/d for 90 days. In the 90-d part after one week, the initial dose of 1000 mg/kg bw/d was reduced to 100 mg/kg bw/d because of the marked weight loss observed.

At doses with relevance for classification with STOT RE; lower final bw (-7% in males, -9% in females) and lower (mean) BWG (-12% in males, -23% in females) were noted at 100 mg/kg bw/d after 28 days. Mean food consumption was lower than in controls (- 7% in males, - 17% in females) after 28 days. Body weight gain was reduced in female only (-13%) at 1000 (initial)/100 mg/kg bw/d after 90 days. Effects were much stronger at 500 and 1000 mg/kg bw/d (outside the guidance values of relevance for STOT RE). This gavage study showed that the effects on growth were severe, but much less than after diet administration at comparable or lower doses.

- *28-d, 90-d (dietary) and two-generation (gavage) rat studies (no. 18, 20, 28)*

Lower BWG and final body weight at doses with relevance for classification with STOT RE 2 were seen in rats of the 28-d study (no. 18), 90-d study (no. 20) and two-generation study (no. 28) (see table 16 of the BD). Reduction in growth at doses relevant for classification could be considered as consistent with the effects seen after gavage administration and in the inhalation studies where the palatability could be ruled out as the cause. In comparison to the gavage studies, the effects on body weight seem to occur at lower doses in the diet studies. As the FC was reduced at similar ranges as the reduction in BWG in the oral diet studies in the rat, these

effects may at least in part be due to lower FC. Mortalities/moribundities were not seen in the diet studies in rats.

Thus, these studies and the observed findings at doses relevant for classification are less crucial for the classification with STOT RE 2.

Carcinogenicity study

Sprague-Dawley rats (80/sex/group), animals were administered metaflumizone in carboxymethylcellulose (0, 30, 60 and 200/300 mg/kg bw/d) by gavage.

The effect on the body weight was most prominent in the diet studies and were discussed as being due to the palatability. A lower body weight (-11% until week 48) was noted in the carcinogenicity study in female rats after oral administration of 300 mg/kg bw/d which lead to a reduction of the high dose to 200 mg/kg bw/d during week three.

In conclusion, RAC agrees with the DS's proposal that classification with STOT RE 2 is warranted based on the observation of mortality/moribund condition in dogs, mice and rats from gavage, diet and inhalation studies and severe clinical signs of toxicity and reduced bw/BWG at doses with relevance for STOT RE 2 observed in these studies. Additional supporting evidence consisting of haemolytic effects (dogs), chronic granulomatous lung inflammatory/granulomatous responses and indications of immunotoxic effects on T-lymphocytes (in rats, following inhalation) were given.

RAC agrees with the DS's proposal not to specify a given organ systems as non-specific toxicity is predominant. The specification of the routes (oral and inhalation) seems acceptable based on the lack of effects in the dermal rat study at doses within the guidance value range for classification. However, 'starry sky [1] ' cells in the thymus, increased lymphocyte necrosis/apoptosis in the mesenteric lymph nodes at ≥ 300 mg/kg bw/d and reduced (T-cell) cellularity in the perioarteriolar lymphoid sheath of the spleen, and diffuse atrophy of the mandibular lymph nodes at 1000 mg/kg bw/d were observed and thus indicated that – similar to the immunotoxic effects in the 28-d inhalation study – immunotoxic effects were seen after repeated dermal exposure.

Moreover, as the gavage/capsule administration studies indicated that dogs could be more sensitive than rats and no information on dermal route in dogs is given, RAC prefers not to indicate specific routes of exposure.

Overall, RAC agrees to classify metaflumizone as **STOT RE 2; H373 (May cause damage to organs through prolonged or repeated exposure)**, with no indication of specific organs or routes of exposure.

RAC evaluation of reproductive toxicity

Summary of the Dossier Submitter's proposal

Effects on fertility and offspring

The DS summarised the effects on fertility and offspring seen in a two-generation reproductive toxicity study in rats (no. 28, OECD TG 416, GLP) as a decrease in the male and female fertility

[1] The 'starry sky' appearance of the thymus, observed in the rat dermal study (no. 26), indicates an increased (apoptotic) necrosis of thymus cells which are phagocytised by numerous macrophages containing cellular/nuclear debris of the apoptotic lymphocytes in their cytoplasm. Apoptosis of thymic lymphocytes may occur at a low extent as a physiological process in healthy individuals (without the 'starry sky' appearance).

indices in animals in the F0 generation at the top dose (75 mg/kg bw/d, mating A). This was observed in the presence of maternal toxicity, characterised by reduced body weight and BWG and general poor state. When the same animals were administered a reduced dose of 50 mg/kg bw/d and re-mated, signs of toxicity were observed (poor general condition) but body weight was only reduced by 7%. Fertility index was again decreased in comparison to controls, but to a lesser extent and remained within the historical control data (HCD). As animals were able to mate successfully during mating B and there were no effects in the F1 generation, it was not proposed by the DS to classify for effects on fertility.

Some evidence of reproductive toxicity occurring during gestation was indicated by a reduction in total numbers of live foetuses at birth. This was observed in mid- and high-dose groups of both generations in a dose-dependent manner. Statistical significance was only observed in the top-dose group of the F1B generation, although it was observed at a higher magnitude in the F1A generation at 75 mg/kg bw/d.

Complete litter loss occurred in the F1A generation dosed with 75 mg/kg bw/d (the only group having significant body weight effects) and to a lesser extent in the top-dose F1B generation. Complete litter loss was also noted in one dam of the F1A mid-dose group. All dams that lost litters also showed inadequate nursing behaviour and/or increased cannibalisation; this latter was considered to be the cause of the litter loss. Increased litter losses, increased cannibalisation and inadequate nursing are responses that are very commonly observed when dams are stressed due to ill-health. The DS regarded these findings as not relevant to humans and therefore, did not considered it appropriate to classify for these effects.

The tables below contain a summary (from the DAR) of the results from this two-generation study in rats by gavage:

Table: Pre-mating bodyweight and food consumption in F0 animals

Observations/study week	**Dose level (mating A/mating B)** (mg/kg bw/d)			
	0/0	**12/12**	**30/20**	**75/50**
	Parental Generation (Mating A)			
	Males			
Body weight (g): Week 10	336.9	335.3	348.5	338.2
Weight gain (g): Weeks 0-10	221.0	221.0	234.5	224.6
Food consumption (g/d): Weeks 0-10	21.1	21.1	21.5	20.7
Females				
Body weight (g): Week 10	204.8	205.3	200.6	176.1**
Weight gain (g): Weeks 0-10	105.9	105.4	101.3	76.9**
Food consumption (g/d): Weeks 0-10	15.3	15.5	15.8	13.3
Parental Generation (Mating B)				
Males				
Body weight (g): Week 10	413.2	412.4	429.3	423.5
Weight gain (g): Weeks 0-10	31.5	31.5	33.25	32.7
Food consumption (g/d): Weeks 0-10	20.2	20.6	20.5	19.8
Females				
Body weight (g): Week 10	239.2	241.5	237.0	221.9**
Weight gain (g): Weeks 0-10	10.3	13.4	10.5	9.9
Food consumption (g/d): Weeks 0-10	15.0	15.7	16.1	15.1

* significantly different to controls, $p < 0.05$; ** $p < 0.01$

Table: Reproductive performance of F0 and F1 parents

Observation	**Dose level** (mg/kg bw/d)			
F0 (Mating A)	**0**	**12**	**30**	**75**
Pre-coital interval (d)	2.3 ± 1.1	2.7 ± 1.1	2.6 ± 1.0	3.0 ± 1.2
		Males		
Placed with females (#)	24	25	25	25
Successfully mated (#)	24	25	25	24
Mating index (%)	100	100	100	96
Fertility index (%)	96	100	84	72*
Intercurrent deaths (#)	1	-	-	-
		Females		
Placed with males (#)	25	25	25	25
Successfully mated (#)	25	25	25	24
Mating index (%)	100	100	100	96
Fertility index (%)	96	100	84	75*
Intercurrent deaths (#)	-	-	-	-
Gestation interval (d)	22.1 ± 0.3	22.1 ± 0.3	22.2 ± 0.5	22.2 ± 0.4
Litters (#)	24	25	21	18
F0 (Mating B)				
Dose level (mg/kg bw/d)	**0**	**12**	**20**	**50**
Pre-coital interval (d)	2.4 ± 1.1	2.8 ± 1.2	2.8 ± 0.9	2.4 ± 1.0
		Males		
Placed with females (#)	24	25	24	24
Successfully mated (#)	24	25	24	24
Mating index (%)	100	100	100	100
Fertility index (%)	100	100	88	88
Intercurrent deaths (#)	-	-	-	-
		Females		
Placed with males (#)	25	25	24	24
Successfully mated (#)	25	25	24	24
Mating index (%)	100	100	100	100
Fertility index (%)	100	100	88	88
Intercurrent deaths (#)	0	0	0	0
Gestation interval (d)	22.3 ± 0.6	22.4 ± 0.6	22.4 ± 0.5	22.4 ± 0.5
Litters (#)	25	25	21	20
F1				
Dose level (mg/kg bw/d)	**0**	**12**	**20**	**50**
		Males		
Placed with females (#)	24	24	25	25
Successfully mated (#)	23	24	25	25
Mating index (%)	96	100	100	100
Fertility index (%)	96	96	92	96
		Females		
Placed with males (#)	24	25	25	25
Successfully mated (#)	23	25	25	25
Mating index (%)	96	100	100	100
Fertility index (%)	100	96	92	96
Gestation interval (d)	22.1 ± 0.29	22.0 ± 0.46	22.1 ± 0.34	22.2 ± 0.41

* significantly different to controls, $p < 0.05$; ** $p < 0.01$

Table: Litter parameters for F1A, F1B and F2 generations

Parameter	**Dose level** (F1A/F1B and F2) (mg/kg bw/d)			
Dose level (mg/kg bw/d)	**-**	**12/12**	**30/20**	**75/50**
		F1A Generation		
Live foetuses (#)	257	279*	197	163
Dead foetuses	0	5*	2	2
Sex ratio Day 0 (% males)	44.4	46.6	57.9	44.2
Deaths: Days 0-4 (%)	1 (0.4)	3 (1.1)	3 (1.5)	12 (7.4)
Deaths: Days 4-21 (%)	0 (0)	0 (0)	1 (0.5)	38 (23.6)
Litter size: PND 0	10.7 ± 1.9	11.2 ± 2.4	9.4 ± 3.0	9.1 ± 2.1
PND 4 (pre-cull)	10.6 ± 2.0	11.0 ± 2.4	9.2 ± 3.1	8.4 ± 2.7
PND 4 (post-cull)	7.8 ± 0.6	7.8 ± 0.8	7.2 ± 1.9	7.1 ± 1.9
PND 7	7.8 ± 0.6	7.8 ± 0.8	7.2 ± 1.9	5.8 ± 3.0
PND 14	7.8 ± 0.6	7.8 ± 0.8	7.2 ± 1.9	5.0 ± 3.5
PND 21	7.8 ± 0.6	7.8 ± 0.8	7.2 ± 1.9	5.0 ± 3.5
Gestation index (%)	100	100	100	100
Live birth index (%)	100	98	99	99
Viability index (%)	99	99	98	93**
Lactation index (%)	100	100	99	70**
		F1B generation		
Dose level (mg/kg bw/d)	Control	12	20	50
Live foetuses (#)	227	232	194	179*
Dead foetuses	5	4	1	12*
Sex ratio Day 0 (% males)	48.0	51.3	44.3	46.9
Deaths: Days 0-4 (%)	1 (0.4)	1 (0.4)	3 (1.5)	15 (8.4)
Deaths: Days 4-21 (%)	0 (0)	2 (0.8)	0 (0)	11 (6.2)
Litter size: PND 0	9.1 ± 2.8	9.3 ± 3.6	9.2 ± 2.7	8.9 ± 2.5
PND 4 (pre-cull)	9.0 ± 2.8	9.2 ± 3.6	9.1 ± 2.6	8.2 ± 3.4
PND 4 (post-cull)	7.3 ± 1.8	7.0 ± 2.2	7.4 ± 1.4	6.9 ± 2.4
PND 7	7.3 ± 1.8	6.9 ± 2.2	7.4 ± 1.4	6.5 ± 2.9
PND 14	7.3 ± 1.8	6.9 ± 2.2	7.4 ± 1.4	6.3 ± 2.9
PND 21	7.3 ± 1.8	6.9 ± 2.2	7.4 ± 1.4	6.3 ± 2.9
Gestation index (%)	96	96	100	95
Live birth index (%)	98	98	99	94*
Viability index (%)	100	100	98	92*
Lactation index (%)	100	99	100	92*
		F2 generation		
Implantation sites (#)	269 ± 11.7	279 ± 11.6	261 ± 11.3	262 ± 10.9
Live foetuses (#)		262	244	234
Dead foetuses	249	1	3	5
Sex ratio Day 0 (% males)	46.6	47.3	47.5	50.0
Deaths: Days 0-4 (%)	14 (5.6)	5 (1.9)	6 (2.4)	8 (3.4)
Deaths: Days 4-21 (%)	1 (0.4)	0 (0)	1 (0.4)	3 (1.3)
Litter size: PND 0	10.8 ± 1.8	10.9 ± 2.2	10.6 ± 2.1	9.8 ± 2.9
PND 4 (pre-cull)	10.2 ± 2.9	10.7 ± 2.5	10.3 ± 2.1	9.4 ± 3.2
PND 4 (post-cull)	7.7 ± 1.7	7.8 ± 1.0	7.9 ± 0.5	7.3 ± 1.9
PND 7	7.6 ± 1.7	7.8 ± 1.0	7.8 ± 0.5	7.2 ± 1.9
PND 14	7.6 ± 1.7	7.8 ± 1.0	7.8 ± 0.5	7.2 ± 2.0
PND 21	7.6 ± 1.7	7.8 ± 1.0	7.8 ± 0.5	7.2 ± 2.0
Gestation index (%)	100	100	100	100
Live birth index (%)	98	100	99	98
Viability index (%)	94	98	98	97
Lactation index (%)	99	100	99	98

* significantly different to controls, $p < 0.05$; ** $p < 0.01$

Key elements from the DAR on the developmental toxicity study in rabbits:

Table: Foetal findings

Observation	Dose level (mg/kg bw/d)			
	0	**30**	**100**	**300**
Mated (#)	25	25	25	25
Pregnant (#)	21	23	19	23
Non-pregnant (#)	4	2	6	2
Died/sacrificed (#)	1	-	-	3
Died/sacrificed pregnant (#)	-	-	-	1
Died/sacrificed non-pregnant (#)	1	-	-	2
Aborted (#)	1	-	-	2
Premature delivery (#)	-	-	-	-
Litters (#)	20	23	19	20
Corpora lutea (#)	152	171	165	155
(#/dam)	(7.6 ± 1.5)	(7.4 ± 1.4)	(8.7 ± 1.3)	(7.8 ± 1.6)
Implantations (#)	129	146	148	138
(#/dam)	(6.4 ± 2.2)	(6.3 ± 2.0)	(7.8 ± 1.4)	(6.9 ± 2.1)
Live foetuses (#)	121	129	131	130
(#/dam)	(6.1 ± 2.4)	(5.6 ± 1.8)	(6.9 ± 1.8)	(6.5 ± 2.1)
Dead foetuses	0	0	0	1
(#/dam)	0	0	0	0.1 ± 0.2
Resorptions (#)	8	17	17	7
Early	8	8	14	5
Late	0	9	3	2
Resorptions/dam (#)	0.4 ± 0.6	0.7 ± 1.0	0.9 ± 0.9	0.3 ± 0.5
Early	0.4 ± 0.6	0.3 ± 0.7	0.7 ± 0.9	0.3 ± 0.4
Late	0.0 ± 0.0	0.4 ± 0.7 *	0.2 ± 0.4	0.1 ± 0.3
Total resorptions (#)	7	10	11	7
Foetal weight (g)	36.3 ± 3.7	38.1 ± 3.7	34.3 ± 3.9	33.6 ± 5.5
Males	36.2 ± 4.3	37.4 ± 4.6	34.0 ± 3.6	33.6 ± 5.8
Females	36.5 ± 3.9	38.3 ± 4.3	34.3 ± 4.0	33.5 ± 5.9
Litter weight (g)	214.2 ± 75.3	209.5 ± 55.4	232.1 ± 48.2	212.4 ± 66.7
Runts (#)[a]	4 (3.3)	2 (1.6)	7 (5.3)	18 (13.8)
Sex ratio (% male)	47.1	47.3	48.9	42.3
Pre-implantation loss (%)	15.6 ± 21.9	15.0 ± 20.4	10.3 ± 11.7	11.4 ± 19.6
Post-implantation loss (%)	8.4 ± 14.4	10.2 ± 12.7	11.9 ± 13.1	6.1 ± 9.5
Gravid uterine weight (g)	297.8 ± 98.2	297.3 ± 73.3	321.0 ± 63.4	302.2 ± 86.8

[a]foetuses with a body weight ≤ 75% of control mean, * significantly different to controls, $p < 0.05$; ** $p < 0.01$

Table: Developmental toxicity

Parameter			Dose level (mg/kg bw/d)				
			Historical control	**0**	**30**	**100**	**300**
Absent subclavian artery	**Foetal incidence**	(#)	3/805	0/121	1/129	1/131	3/131
		(%)	0.4 (0-2.1)	0	0.8	0.8	2.3
	Litter incidence	(#)	3/118	0/20	1/23	1/19	3/20
		%	2.5 (0-13.6)	0	4.3	5.3	15
	Affected litters	(%)	0.4 (0-1.9)	0 0	0.6 ±3.0	0.7 ±2.9	3.1* ±8.4
Incomplete ossification of sternebra	**Foetal incidence**	(#)	197/805	42/121	45/129	66/131	58/131
		(%)	24.5 (17.1-29.5)	35	35	50	46
	Litter incidence	(#)	92/118	15/20	18/23	18/19	18/20
		(%)	78.0 (70.8-87.5)	75	78	95	90
	Affected litters	(%)	24.2 (16.6-31.6)	29.7 ±24.4	32.3 ±27.3	47.5* ±28.2	46.5** ±23.4

* significantly different to controls, $p < 0.05$; ** $p < 0.01$

Developmental effects

In a developmental study in rats (OECD TG 414, GLP), there were no treatment-related external variations, skeletal variations or visceral effects observed. In this study (no. 35) time-mated female Wistar rats (25/dose) were treated with metaflumizone suspended in 0.5% carboxymethylcellulose (0, 15, 40 or 120 mg/kg bw/d) via oral gavage on days 6-19 of gestation. There was no mortality or clinical signs in dams. Bodyweight gain was significantly reduced between gestational days 6 and 8 (54%) and days 6 and 19 (12%) resulting in an overall bodyweight loss of 22%. According to the DAR, a 6% reduction on food consumption was noted at 120 mg/kg bw/d on GD 6-19. There were no treatment-related external or skeletal variations or visceral effects observed in foetuses.

In a developmental study in rabbits (OECD 414, GLP), serious maternal toxicity occurred leading to premature sacrifice of a number of the dams. However, there was an increased incidence of absent subclavian artery in foetuses of the high-dose group (3/131 (2.3%) versus 0 in controls; 3/20 litters). Whilst this finding has been observed in historical controls, it is rare, occurring in only one other study of the 33 conducted in the same period in the same laboratory, with an incidence of 3/145 foetuses (2.1%) in 3/22 litter. Whilst it is plausible that the malformation arose spontaneously, in the absence of any incidences in the concurrent control and given the rare nature of this finding, it is difficult to dismiss it as not being treatment-related. The DS considered that the absence of subclavian arteria seen constituted some evidence and hence does not justify Category 1B, but proposed classification with Category 2. The incidence of absent subclavian artery occurred in one foetus from each of the low- and mid-dose groups and three foetuses in the top-dose group. This led to an incidence that was marginally above the HCD.

The DS interpreted the clinical relevance of this malformation as unclear and because of this and the fact that it occurred in the presence of maternal toxicity, and in one species only, classification in Category 2 for developmental toxicity (Repr. 2, H361d) was deemed appropriate.

Effects on lactation

The DS proposed to classify for effects on lactation based on the increased incidence of pup deaths during PND 0-4 and PND 4-21 in F1A pups and to a lesser extent in the F1B generation. Lower pup body weight during PND 1-7 and empty stomach at necropsy was considered as supporting evidence.

Metaflumizone is a lipophilic substance, as evidenced by its physical and chemical properties, and as such might be expected to be transferred to milk. An additional study was carried out to assess this (no. 37). In this study, residues of metaflumizone were found in milk at levels approximately 10 times less than the dose administered (peaking at 14.6 mg/kg bw/d). The amount transferred to pups was 3-28 times less than that found in breast milk (peaking at 4.0 mg/kg bw/d when dosing of the dam continued throughout the post-natal period). These findings suggested that metaflumizone can enter the breast milk and in levels that have been shown to cause body weight effects in repeated dose dietary studies in rats (no. 20). It is difficult to ascertain whether metaflumizone caused an effect on pups directly through the milk or if it was a secondary effect due to impaired maternal care from dams suffering from ill-health. However, these effects on pups occurred during the time period when the mother provided the sole means of nutrition; therefore the DS proposed to classify for effects on lactation.

Comments received during public consultation

Three MSCAs agreed with the DS's view on no classification for fertility effects and two MSCAs supported classification for lactation. One MSCA expressed its general agreement with the proposed classification proposals for human health.

Two MSCAs supported the classification for effects on development.

In total four MSCAs agreed with the classification proposal for Repr. 2 H361d; another MSCA proposed Category 1B. One MSCA supported the Category 2 classification for developmental effects based on the incidence of absent clavicola which is a rare finding but has been observed in control animals previously. Another MSCA agreed based on the incidence of absent subclavian artery outside the historical control incidence. One MSCA addressed the weaknesses of the available HCD on absent clavian artery which lack information on median and mean values, and considered also the incidences in the low- and mid-dose groups as above the median/mean historical control incidences.

The manufacturer disagreed with the proposed classification as Repr. 2 H361d. As their main evidence, they proposed to disregard one case of absent clavian artery at 300 mg/kg bw/d as it was observed in a dead pup and they required a correction of the final incidence at the top dose to 1.5% instead of 2.3%. It was highlighted that the arterial branching patterns vary considerably in humans and rabbits and that there was no evidence of malformation or atrophy in the upper limbs of the subjected rabbit foetuses. Thus the effect should be considered as a variation. Based on insufficient severity of the effect and that the incidences were considered within the HCD, the manufacturer disagreed with the proposal. It was suggested to allocate the absent clavian artery in one foetus at 100 mg/kg bw/d to a syndrome as also a right sided aortic arch and a ventricular septal defect were seen.

Assessment and comparison with the classification criteria

Effects on fertility and offspring

Systemic toxicity in the high-dose F0 parental generation (75 or 50 mg/kg bw/d) was reported as an increased occurrence of a poor general state, statistically significant decreased food consumption, statistically significant lower body weights and impaired body weight gain. There were no indications of treatment-related organ weight changes, gross lesions, or microscopic findings. At 75 mg/kg bw/d, lower bw (-14%) and BWG (-27%) during weeks 0 to 10 (premating), associated with lower food consumption (-13%) in comparison with controls, were seen. However, no evidence on other clinical signs of toxicity is given in the DAR or the CLH report. RAC considers it unlikely that the slight difference in the final bw is responsible for increased oestrus cycle length, lower fertility index or complete litter losses shortly after birth in 4/18 dams (the latter may be more relevant for developmental toxicity), perinatal pup deaths and lower viability of pups at PND 0-4. Due to missing information, it is questioned whether the parental animals were severely affected by general toxicity as they were able to mate at similar rates as controls. It cannot be ruled out that the cause of prolonged cycle length in two dams was a metaflumizone-related effect rather than a sign of non-specific toxicity.

Moreover, it is uncertain whether the reduced pup viability can only be attributed to improper nursing behaviour, as a non-specific maternal effect, as one litter loss occurred in a dam with normal nursing behaviour.

When comparing the effects in these parental animals at 75 mg/kg bw/d with the effects observed in the 28/90-d gavage study in rats (no. 21) it can be concluded that marked weight loss was observed only at 1000 mg/kg bw/d and led to a dose reduction during week 1. No mortalities and no clinical signs were observed at 500 mg/kg bw/d. Body weights, BWG and FC were significantly lower at 500 mg/kg bw/d and non-significantly reduced at 100 mg/kg bw/d after 28 days. A slight decrease in FC and BWG (-13%) was observed in females at 1000/100 mg/kg bw/d after 90 days of treatment. The observation that gavage doses of 500 mg/kg bw/d were tolerated for 28 days without any clinical signs of toxicity, and only slight effects on BWG and FC observed at 100 mg/kg bw/d after 28 days and 90 days of treatment questions whether the slight effects

on body weight at 75 and 50 mg/kg bw/d – in the absence of any clinical sign of toxicity, and otherwise consistent with observations in repeated dose gavage administration of 100 mg/kg bw/d – should be considered as severe maternal effects.

Comparing the effects in rats seen in gavage studies with dose-responses seen in diet studies, indicated that effects of comparable magnitude (lower BWG of about 12-23%, lower FC of about 7-17% seen in the 28/90-d study, no. 21) at 100 mg/kg bw/d correspond to effects seen in diet studies at 4-7 mg/kg bw/d in the 90-d study (no. 20) or at 23.8/27.2 mg/kg bw/d in the 28-d study (no. 18). It is assumed that absorption after gavage administration (possible due to lipophilicity of metaflumizone) is markedly lower than after diet administration (factor of more than 10 for the 90-d study part, factor of more than 3 for the 28-d study design).

The outcome of the second mating (mating B) at 50 mg/kg bw/d may also be relevant for classification. In this part of the study, animals from study day 126 received metaflumizone at reduced doses of 0, 12, 20 or 50 mg/kg bw/d by oral gavage, and parental rats were treated for a 10-week period (pre-mating) and were mated with the same partner as for their first mating to produce a second litter (F1B). Treatment resulted in a significantly lower body weight (–7%) in F0-females at week 10 in comparison with the controls. Food consumption was unchanged, and the BWG was slightly (non-significantly) lower (-4%). No effects on body weight and FC were seen in F0 males. The DAR summary stated that F0 high-dose females were in poor general state, but neither the DAR nor the CLH report reported details on the nature of the signs and their incidences.

Compared with 75 mg/kg bw/d, the dose of 50 mg/kg bw/d resulted in a slightly lower body weight (-7%) and minor effects on the BWG (-4%) that could not explain the increased number of dead pups at 50 mg/kg bw/d. In contrast to 75 mg/kg, 50 mg/kg bw/d did not result in a prolongation of the oestrus cycle. The mating index was unaffected at both 75 and 50 mg/kg bw/d in F0 and F1 females indicating that the general health status at 50 mg/kg bw/d was sufficient for mating performance and conception. The fertility index was slightly (non-significantly) lower at 20 and 50 mg/kg bw/d in both parental generations.

The number of stillborn foetuses was increased at 50 mg/kg bw/d in the F0 generation (12 versus 5 in controls) but not in the pups from F1 dams. While the viability during PND 0-4 in the F1B pups was significantly lower (92% versus 100% in controls), no such effect was seen in the F2 pups (5 at 50 mg/kg bw/d versus 4 in controls). As the $LogK_{OW}$ values for metaflumizone's isomers are rather high, a potential for accumulation could not be ruled out, in particular as high levels of radioactivity were seen in female sex organs in the accumulation study. This may indicate that the internal doses at mating B might have been higher than just 50 mg/kg bw/d. Regarding the impaired nursing behaviours as a sign of poor general state, it is noted that the DAR reported that two females at 50 mg/kg bw/d showed improper nursing behaviour, losing all of their pups shortly after birth, as did a third female which showed normal nursing behaviour.

RAC questioned whether the dosing was high enough for the chosen gavage administration, whether the increased number of stillborn pups and reduced pup viability should be regarded as a fertility effect, and indicated that ovary effects were also seen in repeated dose studies in rats. The Industry representative emphasised the palatability problems in the diet studies as rats selectively had chosen food without the test substance instead of food with the test substance. The prolonged oestrus cycle was in their view only attributable to two dams, one that died later on with severe chronic nephropathy and the other that showed normalised oestrus cycle after receiving 50 mg/kg bw/d in mating B.

According to the CLP criteria, adverse effects on fertility and reproductive performance are normally not relevant for classification purposes if the adverse effects were seen at dose levels causing marked systemic toxicity (e.g. lethality, dramatic reduction in absolute body weight, coma). In the absence of marked toxicity at 50 and at 75 mg/kg bw/d the effects on fertility and

offspring could not considered to be secondary effects to the slightly reduced body weight/BWG. Therefore RAC agreed (in contrast to the original proposal for no classification by the DS) **to classify metaflumizone as Repr. 2; H361f for fertility**.

Lactation

Increased incidences/rates of pup death at PND 4 and PND 21 and significantly lower lactation indices were observed in the F1A generation (in dams receiving 75 mg/kg bw/d) and F1B generation (in dams receiving 50 mg/kg bw/d). While a stronger effect was seen at 75 mg/kg bw/d during PND 4-21, pups from dams following gavage exposure to 50 mg/kg bw/d showed a higher death rate during PND 0-4.

The number of dams showing improved nursing behaviour, after the second mating with lower doses, was not available to RAC. The DAR indicated only 2 dams at 50 mg/kg bw/d, thus the contribution of the nursing behaviour remains unclear. As the bw was (non-significantly) lower (-21%) in comparison with controls in F1A pups and less severe in F1B and F2 pup generations (-9% and -8%), this effect points either to a direct effect that was still ongoing during the first PND or to effect via lactation.

No treatment-related effect on the lactation index was observed in the F2 generation.

According to the CLP criteria, clear evidence on the offspring due to transfer to the milk or adverse effect on the quality of the milk or indications that the substance is present at potentially toxic levels in breast milk justifies classification for lactation effects. For RAC, the lower lactation index and the reduced survival at PND 4-21 where milk is the only nutrition source justifies classification for lactation effects. As pointed out in the CLH report, additional studies have found residues of metaflumizone in the milk after birth and during the lactation period. As BWG effects were seen at lower doses in diet studies compared to gavage studies, it seems plausible that even low doses in milk can have effects on the growth.

In conclusion, RAC agrees with the DS to classify metaflumizone for **effects on or via lactation, Lact.; H362**.

Developmental effects

RAC agrees with the DS's view that no treatment-related external variations, skeletal variations or visceral effects were observed (see summary tables B.6.48 and B.49 in the DAR).

The crucial effect seen in the developmental toxicity study in rabbits was an increased incidence of absent subclavian artery in foetuses at the high dose compared to controls. In addition, a significant increase of incomplete ossification of sternebra (a minor lesion) was observed at mid and high dose.

There were clinical signs of toxicity and sacrifices due to moribundity that demonstrated maternal toxicity in 4/25 rabbits at 300 mg/kg bw/d. Two non-pregnant and one pregnant animal at the top dose were sacrificed. Two litters were aborted at the top dose versus one in controls. The number of live foetuses was not affected in any of the dose groups, while the foetal weight was slightly (non-significantly) lower at the top dose in comparison with controls.

Neither the CLP dossier nor the DAR summary allow a conclusion on whether the malformation in 3/20 litters was observed in those dams that were sacrificed (one pregnant dam) or demonstrated clinical signs of toxicity. Although it is likely to assume, it remains unclear from the summary table and reporting whether the clinical signs reported for 4/25 animals at 300 mg/kg bw/d (lateral position, ataxia, poor general state, no defecation or blood in the bedding) were seen in the two non-pregnant animals and the pregnant animal that were sacrificed. Some more information is found in the RCOM (DS response to comment 7): prior to abortion one of two dams that aborted exhibited clinical signs from day 26 of gestation. Another dam was sacrificed in moribund status on day 28 after clinical signs seen from day 24 of gestation (RAC

assumed that the information given in the RCOM that all foetuses found to be stunted relates to this animal). Obviously, visceral effects were not assessed in the two aborted litters and hence may be underestimated. Unequivocally, maternal toxicity is observed at the gavage dose of 300 mg/kg bw/d, the number of sacrificed rabbits (2 non-pregnant, 1 pregnant) is in the 10% range above which data should not be considered for classification. Based on the assumption that the malformations observed related to those 20 dams that were not in poor general condition, these effects may be of relevance for classification. This is supported by the lack of effects on the bw/BWG, FC or corrected uterus weight.

The CLP criteria states in such cases that developmental effects that occur in the presence of maternal toxicity are considered to be evidence of developmental toxicity unless it can be unequivocally demonstrated that the developmental effect are secondary to maternal toxicity. Classification (in this case in category 2) should be considered where there is a significant toxic effect in the offspring, e.g. irreversible effects such as structural malformations, embryo/foetal lethality, significant postnatal functional deficiencies.

For metaflumizone the observed malformations were not unequivocally demonstrated to be (only) secondary to maternal toxicity. Absent subclavian artery is a rare malformation in control animals, was absent in the study control group and its incidence increased with dose (0/20, 1/23, 1/19, 3/20 litters at 0, 30, 100, 300 mg/kg bw/d, respectively). Comparison with the provided HCD indicated that the incidence (2.3%) at the high dose was slightly above the upper limit of historical control incidences (HCD range of 0-2.1%, from 1995-2008). The study on metaflumizone was finalised in 2004.

The DS indicated that the clinical relevance of this malformation is unclear and pointed out in their response to the comment from the MSCA that the effect of the absent subclavian artery on the rabbit development is unknown as it is not known whether the arterial branching pattern would compensate in some way. The study outcome remains unclear as information on whether other branches were compensating or alterations in other branches have been seen is not available. The manufacturer in his comment (no. 8) stated that no evidence of malformation or atrophy of the upper limbs were seen in the affected rabbit foetuses.

Although no skeletal deficiencies were identified in the treated rabbits and compensation by other vascular structures must be assumed, uncertainties remain about the clinical relevance in rabbits as no follow-up information on living foetuses with absent subclavian artery is available. With regard to the situation in humans, RAC was not aware of data/publications on consequences of absent subclavian arteries, however an aberrant subclavian artery in humans can lead to dysphagia (through disturbance in swallowing by compression of the oesophagus) or aneurysma of the adjacent descending thoracic aorta that can cause serious complications in patients and needs surgical intervention (Kouchoukos *et al.*, 2007). Aberrant subclavian arteries are reported as rarely occurring in humans (0.5-1.0% in the population).

RAC considers the effect 'absent subclavian artery' as a malformation and does not agree with the Industry comment interpreting the effect as a variation. The classification as a malformation has also been agreed by experts contributing to the IPCS Harmonisation Project on Terminology in Developmental Toxicology (Solecki *et al.*, 2003).

RAC follows the argumentation of the DS that the incidence was outside the HCD and that the finding occurred at a dose that produced maternal toxicity in rabbits. However, whether the incidence should be considered as marginally increased compared to the HCD where one study out of 34 (see Table 21 in the CLH dossier) showed an incidence of 2.05% and two other studies showed incidences of 0.58% and 0.64%, leading to a mean incidence of 0.09%, seems debatable.

RAC considered that for a lipophilic substance, the choice of a lipid vehicle would have been more appropriate than the use of aqueous CMC solutions and questioned whether the actual doses were possibly lower than the nominal ones administered. RAC members emphasised that also

the single cases of absent subclavian arteries in the low and mid dose groups contribute to the evidence. Other members considered that the increased number of stillborn pups and reduced pup viability during PND 0-4 seen in the two-generation study in rats are rather developmental effects than fertility effects, and suggested to take this evidence as supporting for the classification for developmental toxicity. RAC noted the remaining uncertainties on the long-term consequences from the absent artery.

RAC agrees with the DS's proposal that classification with **category 2 for developmental toxicity, Repr. 2; H361d,** is warranted.

Overall, RAC agrees to classify metaflumizone as **Repr. 2; H361fd and Lact., H362**.

ENVIRONMENTAL HAZARD EVALUATION

RAC evaluation of aquatic hazards (acute and chronic)

Summary of the Dossier Submitter's proposal

Water solubility of the mixture of E/Z (92.2:7.8) according to EEC method A6 1.4.1 (column elution method) (DAR B.2.1.11)

- pH 5: 1.35 µg/L
- pH 7: 1.81 µg/L
- pH 9: 1.73 µg/L
- In deionized water: 1.79 µg/L
- Isomers: E: 1.43 µg/L; Z: 2.03 µg/L

Partition coefficient – EEC method A8 (HPLC) (DAR B.2.1.13)

- LogPow at pH 5 and 30 °C: Z isomer: 4.4 E isomer: 5.1
- LogPow at pH 7 and 20 °C: Z isomer: 4.2 E isomer: 4.9
- LogPow at pH 3 and 20 °C: Z isomer: 3.8 E isomer: 4.4

Dissociation: no dissociation in water.

Degradation

- ***Aquatic hydrolysis*** (Fang, 2004a) GLP, US EPA N 161-1. Incubation in dark for 30 days with two different radiolabelled metaflumizones: benzonitrile-U-^{14}C (B) and trifluoromethoxyphenyl-U-^{14}C (T). The degradation pathway has been identified.
  - DT_{50} at pH 4 25 °C – B: 5.4 and T: 6.5 days
  - DT_{50} at pH 4 12 °C – B: 16.1 and T: 18.4 days
  - DT_{50} at pH 5 25 °C – B: 31.4 and T: 27.3 days
  - DT_{50} at pH 5 12 °C – B: 88.8 and T: 77.2 days

  The DS has, thus, concluded that metaflumizone is hydrolytically stable

- ***Aquatic photolysis*** (Ta, 2004d; Ta, 2004e) GLP, US EPA N 161-2, later re-assessed by Gericke, 2011c using FOCUS kinetics guidance (2006). Incubation at pH 9 for up to 15 days at 22 °C under constant irradiation. B and T radiolabelled metaflumizone was used. Aqueous photolysis pathway proposed. Mineralization 1.7%.
  - Results of Gericke study between 2.4–4.2 days.

  Susceptible for photodegradation when irradiated, but under aquatic environmental conditions photolyticpotential is limited.

- ***Biodegradation***
  - ***Ready biodegradation*** (Heim *et al*., 2002) GLP, OECD 301 (CO_2 evolution). 1.1–1.8% degradation was observed over 29 days.

    The DS has, thus, concluded that metaflumizone is not readily biodegradable.

  - ***Simulation testing***
    Water–sediment simulation tests (Rosenwald, 2003; Schriever, 2010)
    US EPA N 162-4, at 20 °C, in dark, under aerobic conditions, for 100 days.
    Dissipation $DT_{50\ water}$: 0.01 days for both study (two different lakes).
    Re-evaluated $DT_{50\ water}$ (Schriever, 2010): 0.556 and 0.733 days.
    Dissipation $DT_{50\ total\ system}$: 394 (lake 1) and 715 days (lake 2).
    Re-evaluated $DT_{50\ total\ system}$: 322 and 581 days
    Mineralization: 3.7 and 4.5%.
  - Simulation test using 12 hour light–dark cycle (Ta, 2004f; Schriever, 2010c)
    Dissipation $DT_{50\ water}$: 0.5–1.5 d,
    Re-evaluated $DT_{50\ water}$ (Schriever, 2010c): 0.298 d (dark); 0.218 d (irradiated);
    Dissipation $DT_{50\ total\ system}$: 3.6, 8.4 and 23.2 days (different radiolabels).
    Re-evaluated (Schriever, 2010c):376 days in dark (only sediment, dark: 674 d); 6.32 days irradiated (only sediment:3–38 days)
    Mineralization: 16.3, 35.5 and 23.9% (different radiolabels).
    Due to irradiation conditions of low realism metaflumizone should be considered not ultimately degraded or transformed to non-classifiable products within 28 days.

The overall conclusion of the DS was that the substance is **not rapidly degradable** for the purpose of classification and labelling.

Aquatic Bioaccumulation

- ***Kow*** (Holman & Petry, 2001)
  E isomer: Log K_{OW} 5.1 at pH 5, 30 °C
  Z isomer: Log K_{OW} 4.4 at pH 5, 30 °C
  Class, 2006a,b
  E isomer: Log K_{OW} 4.9 at pH 7, 20 °C and 4.4 at pH 3, 20 °C
  Z isomer: Log K_{OW} 4.2 at pH 7, 20 °C and 3.8 at pH 3, 20 °C
- ***Experimental aquatic BCF*** – Unpublished study performed according to GLP, OECD 305, radioactive metaflumizone
  42 days exposure period, flow through, *Lepomis macrochirus, non-steady state*
  $BCF_{whole\ fish}$: 7,800 to 8,100 L/kg
  Lipid normalized $BCF_{whole\ fish}$: 5,769 to 4,099 L/kg
  2008 – 28 days flow through, *Cyprinus carpio, steady state*
  BCF: 1,986 to 2,117 l/kg
  Lipid normalized BCF: 2,736 to 2,916 l/kg
  Depuration half-life $DT_{50\ whole\ fish}$: 14–17 days and 14.7–16 days.

Metaflumizone has the potential to bioaccumulate.

Aquatic toxicity

- ***Fish***
  - ***Acute aquatic toxicity:*** 3 fish studies are considered relevant by the DS: 96 h, flow-through test, endpoint: mortality. The highest (dissolved) conc. (mm) did not reached 50%, but only 40.5% and 25% mortality.

US EPA 72-1, OECD 203, GLP, purity: 96.3%, E/Z 92:8	Rainbow Trout (*Oncorhynchus mykiss*)	LC_{50} >0.0378 mg/L (mm)	2001e
US EPA 72-1, OECD 203, GLP, purity: 96.3%, E/Z 92:8	Bluegill Sunfish (*Lepomis macrochirus*)	LC_{50} >0.349 mg/L (mm)	2001c
US EPA 72-3, OECD 203, GLP, purity: 96.3%, E/Z 92:8	Fathead Minnow (*Pimephales promelas*)	LC_{50} >0.257 mg/L (mm)	2001d

Two more studies were introduced in the CLH Report, which are not considered by the DS as appropriate for classification purposes. The DS's argumentation was: "*Given the decline in ^{14}C in the aqueous phase over the study duration, the eMS (evaluating member state) does not feel the study 96-hour LC_{50} based on initial measured concentrations of metaflumizone is appropriate for classification. In addition, given the spiked sediment, and that catfish are bottom feeders, it is unclear what concentration the test organisms were exposed to over the study period. On this basis, the eMS considers the study does not provide a valid endpoint for hazard classification*".

The DS considered that toxicity studies using spiked water-sediment systems were not valid for the purpose of hazard classification due to the exposure route (largely due to issues related to sediment ingestion and/or adsorption).

- ***Chronic toxicity:*** early life-stage toxicity, measured in flow-through test system, the highest tested nominal concentration was 0.002 mg/L. No toxic effect was measured even in the highest concentration, so the NOEC is higher than or equal to the highest tested concentration.

OECD 210, GLP, purity: 96.3%, E:Z=92:8	Rainbow trout (*Oncorhynchus mykiss*)	Hatching success, survival, growth	93 days	NOEC ≥0.00147 (mm) based on no effects at the highest exposure concentration	2002a
OECD 210, GLP, purity: 96.3%, E:Z=92:8	Fathead minnow (*Pimephales promelas*)	Hatching success, survival, growth	41 days	NOEC ≥0.001157 (mm) based on no effects at the highest exposure concentration	2002

The DS stated that no statistical differences were observed between the controls and any treatments for any endpoint in both studies. Therefore, the NOEC was higher than the highest tested concentration (≥0.00147 mg/L) in the 93 days and ≥0.00116 mg/L in the 41 days study based on mean measured concentrations).

- ***Invertebrates***
  - ***Aquatic acute:*** 2 relevant 48 h Daphnia tests: one static, one flow-through. Endpoint: immobilization. In the Aufderheide *et al*. (2001f) study, the highest (dissolved) concentration did not reach 50% immobilization, only a maximum of 35%. The EC_{50} for mean measured concentrations of the Weltje and Glaser (2006b) study was 2.86, much above water solubility of metaflumizone.

Daphnia sp US EPA 72-2, GLP, purity: 96.3%, E:Z=92:8	*Daphnia magna*	Flow-through	EC50 >0.331 mg/L (mm)	Aufderheide *et al.*, 2001f
Daphnia sp OECD 202, GLP, purity: 99.7%, Z isomer	*Daphnia magna*	Static	EC50 = 2.86 mg/L (mm)	Weltje & Glaeser, 2006b

- ***Aquatic chronic:*** one 21 day, semi-static study is cited by the DS (Olivieri
- *et al.*,2001) . The highest initial nominal concentration was 0.002 mg/L. The NOEC result – similar to fish – reflected that no toxicity was measured in the highest exposure concentrations.

OECD 211, GLP, purity: 96.3%, E:Z=92:8	*Daphnia magna*	Survival; reproduction, growth	NOEC ≥0.00147 (mm) based on no effects at the highest exposure concentration	Olivieri *et al*, 2001

- ***Aquatic plants/algae:*** freshwater and marine algae 72 h and *Lemna* 7 days static tests and their results are summarized in the table below.
  - ***Aquatic acute and chronic:*** 5 growth inhibition studies did not reveal any effects below the limit of water solubility.

OECD 201, GLP, purity: 96.3%, E:Z 92:8	*Pseudokirchneriella subcapitata*, freshwater algae	Cell multiplication inhibition	ErC_{50} >0.0256 (mm) ErC_{10} >0.0256 (mm)	Aufderheide *et al*, 2001a
OECD 201, GLP, purity: 96.9%, E:Z 94:6	*Anabaena flos-aquae* freshwater algae	Cell multiplication inhibition	ErC_{50} >0.225 (mm) ErC_{10} >0.225 (mm)	Hicks & Holmes, 2004b
OECD 201, GLP, purity: 96.9%, E:Z 94:6	*Navicula pelliculosa* freshwater algae	Cell multiplication inhibition	ErC_{50} >0.0264 (mm) ErC_{10} >0.0264 (mm)	Hicks & Holmes, 2004c
OECD 201, GLP, purity: 96.3%, E:Z 92:8	*Skeletonema costatum,* marine algae	Cell multiplication inhibition	ErC_{50} >0.011 (mm) ErC_{10} >0.011 (mm)	Hicks & Holmes, 2004d
OECD 221, GLP, purity: 96.9%, E:Z 94	*Lemna gibba duckweed*	Growth	ErC50 >0.314 (mm) NOErC ≥ 0.314 (mm)	Hicks & Holmes, 2004a

- ***Toxicity of metaflumizone to other aquatic and sediment dwelling organisms***

Two amphipod crustaceans and four *Chironomus* midge studies were introduced by the DS, all applying spiked sediments as the route of exposure.

Two Chironomus studies were available using aqueous exposure in a water-sediment system. However, it is not possible to use the quoted study endpoints for hazard classification for two reasons: (i) analytical data are not available for the full exposure series to validate aqueous exposure concentrations; (ii) the sediment compartment is present in the test vessels, so it is unclear if a contribution of the toxicity was due to sediment contact/ingestion.

The final conclusion of the DS is that no aquatic classification is proposed.

Comments received during public consultation

Two member states commented on the CLH report:

One agreed with the proposal of no classification for environmental hazards and added an editorial correction for the EC_{50} value of the Aufderheide et al. (2001f) *Daphnia magna* study. The DS stated that the latter was a typographical error and RAC subsequently used the correct value in the opinion.

The second MS disagreed with the DS proposal of no classification for environmental hazards and instead proposed a classification of Aquatic acute 1 (H400) and Aquatic chronic 1 (H410) (acute M-factor of 1 and chronic M-factor of 10). They also disagreed with the experimental value provided for water solubility. The DS responded that each study was considered individually and the presented results were based on analytical measurement and visual observations to reflect dissolved concentrations. Furthermore, in all acute and chronic studies considered, no effects up to the highest tested concentrations were observed, for the standard test species (fish, daphnia, algae and aquatic plants).

Additionally, this MS considered that Study 4 (2002) on short-term toxicity to fish as well performed and valid acute study with *Ictalarus punctatus,* of which the mean measured concentration (geometric mean) in the spiked water from the start (0 hours) to the end of the test (96 hours) is 0.154 mg/L. The DS did not accept this non-standard study because of the uncertainty in the exposure conditions, i.e. the concentration of metaflumizone in water and the rapid dissipation of metaflumizone from water, the spiked water and sediment and the potential ingestion of significant amounts of sediment by the fish.

Finally, the MS argued for the consideration of the chironomid study results to derive a Chronic Category 1 (M-factor=10) classification. The DS stated that these results reflected the highest tested exposure concentration and that at these exposure levels no significant effects were observed. Consequently, the substance should not be classified for environmental hazard based on these results.

The DS acknowledged that *C. riparius* data can be used in principle to derive a classification, but the study included in the CLH report (Weltje, 2005) has significant limitations, such as:

- The test system was static with no test item renewal
- Analytical data are not available to validate the aqueous exposure concentrations over the full study period
- The data support the rapid distribution of metaflumizone to the sediment phase, below detection limit in water by day 28

RAC agrees with the argumentation provided by the DS in all these points.

Assessment and comparison with the classification criteria

Degradation

Metaflumizone only undergoes hydrolysis at low pH, it is hydrolytically stable at pH 7 and 9. Hydrolysis DT_{50} values of the isomers at pH 5, 25°C are between 27.3–31.4 days, at 12°C between 77.2–88.8 days >16 days. Data on hydrolysis might be considered for classification purposes only when the longest half-life determined within the pH range 4–9 is shorter than 16 days. Accordingly, metaflumizone is hydrolytically stable.

Metaflumizone is susceptible to photodegradation, but in the aquatic environmental compartments the potential for aquatic photolysis is usually limited by local conditions such as turbidity. The applicability of metaflumizone photolysis results for classification purposes is therefore limited.

In a ready biodegradation study (OECD 301, 29 days), 1.1–1.8% degradation was observed.

Metaflumizone is therefore not considered to be readily biodegradable.

In an aerobic water-sediment study performed in the dark, DT_{50} values for metaflumizone were between 394 and 715 days >28 days. Metaflumizone is not ultimately degraded within 28 days or transformed to non-classifiable products.

RAC's overall conclusion on degradation is that metaflumizone is not rapidly degradable for the purpose of classification and labelling.

Aquatic bioaccumulation

The measured Log K_{OW} of metaflumizone at aquatic environmental pH is above the CLP trigger value of 4 and the experimental fish BCFs are above the trigger of 500 L/kg (up to 8,000 L/kg).

RAC's conclusion on bioaccumulation is that metaflumizone is a substance with a potential to bioaccumulate.

Aquatic toxicity

Aquatic acute toxicity data on metaflumizone are available for fish, invertebrates, algae and aquatic plants. No acute L(E)C_{50} effects were observed up to the quoted limit of water solubility. **Metaflumizone does not warrant classification for Aquatic Acute toxicity**.

Aquatic chronic toxicity data on metaflumizone are available for standard species of fish, invertebrates, algae and aquatic plants. Exposure concentrations in the chronic studies range up to or above 0.002 mg/L (nominal) to reflect the quoted water solubility. In each case, the NOEC was equal to or greater than the highest tested concentration. Metaflumizone showed no chronic effects up to the limit of water solubility for the purpose of classification, so it does not warrant classification for Aquatic Chronic toxicity.

As Metaflumizone has no acute toxicity up to the measured water solubility, is not rapidly degradable and has a potential to bioaccumulate, classification as Aquatic Chronic 4 requires consideration. However, as there are reliable NOECs from long-term toxicity tests that show no toxicity up to the water solubility limit, **Metaflumizone does not warrant classification for Aquatic Chronic toxicity**.

Additional references

Kouchoukos, NT. and Masetti P (2007) Aberrant Subclavian artery and Kommerll aneurysm: Surgical treatment with a standard approach. The Journal of Thoracic and Cardiovascular Surgery, 133:888-892.

Solecki, R. et al. (2003) Harmonization of rat fetal external and visceral terminology and classification. Report of the Fourth Workshop on the Terminology in Developmental Toxicology. Berlin, 18-20 April 2002. Reprod Toxicol 17:625-637.

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).