Committee for Risk Assessment

RAC

Opinion

proposing harmonised classification and labelling
at EU level of

etridiazole

EC number: 219-991-8
CAS number: 2593-15-9

CLH-O-0000002504-80-02/F

Adopted

4 June 2013

4 June 2013

CLH-O-0000002504-80-02/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 (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: etridiazole

EC number: 219-991-8

CAS number: 2593-15-9

The proposal was submitted by **the Netherlands** and received by the RAC on **28 February 2012.**

In this opinion, all classifications are given firstly in the form of CLP hazard classes and/or categories, the majority of which are consistent with the Globally Harmonised System (GHS) and secondly, according to the notation of 67/548/EEC, the Dangerous Substances Directive (DSD).

PROCESS FOR ADOPTION OF THE OPINION

The Netherlands 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 **28 February 2012**. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by **13 April 2012**.

ADOPTION OF THE OPINION OF THE RAC

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

Co-rapporteur, appointed by RAC: **Boguslaw Baranski**

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

The RAC opinion on the proposed harmonised classification and labelling was reached on **4 June 2013** and the comments received are compiled in Annex 2.

The RAC Opinion was adopted by **consensus**

OPINION OF THE RAC

The RAC adopted the opinion that **etridiazole** should be classified and labelled as follows:

Classification and labelling in accordance with the CLP Regulation

	Index No	International Chemical Identification	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Picto-gram, Signal Word Code(s)	Hazard state-ment Code(s)	Suppl. Hazard statement Code	
Current Annex VI entry	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Carc. 2 Acute Tox. 4 * Acute Tox. 4 * Acute Tox. 3 * Aquatic Acute 1 Aquatic Chronic 1	H351 H302 H312 H331 H400 H410	GHS06 GHS08 GHS09 Dgr	H351 H302 H312 H331 H410		
Dossier submitters proposal	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Keep Carc. 2 Remove (*) from Acute Tox. 4 * Remove Acute Tox. 4 * Remove Acute Tox. 3 * Add Skin Sens. 1B Add STOT SE 3 Keep Aquatic Acute 1 Keep Aquatic Chronic 1	Keep H351 Keep H302 Remove H312 Remove H331 Add H317 Add H335 Keep H400 Keep H410		Keep H351 Keep H302 Remove H312 Remove H331 Add H317 Add H335 Keep H410		Add: Acute M=1 and Chronic M=1
RAC opinion	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Keep Carc. 2 Remove (*) from Acute Tox. 4 * Remove Acute Tox. 4 * Remove Acute Tox. 3 * Add Skin Sens. 1 Keep Aquatic Acute 1 Keep Aquatic Chronic 1	Keep H351 Keep H302 Remove H312 Remove H331 Add H317 Keep H400 Keep H410		Keep H351 Keep H302 Remove H312 Remove H331 Add H317 Keep H410		Add: Acute M=1 and Chronic M=1
Resulting Annex VI	613-133-00-X	etridiazole (ISO);	219-991-8	2593-15-9	Carc. 2 Acute Tox. 4	H351 H302	GHS08 GHS07	H351 H302		

entry if agreed by COM		5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole			Skin Sens. 1 Aquatic Acute 1 Aquatic Chronic 1	H317 H400 H410	GHS09 Wng	H317 H410		 M=1 M=1

Classification and labelling in accordance with the DSD

	Index No	International Chemical Identification	EC No	CAS No	Classification	Labelling	Concentra-tion Limits
Current Annex VI entry	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Carc. Cat. 3; R40 T; R23 Xn; R21-22 N; R50-53	T; N R: 21/22-23-40-50/53 S: (1/2-)36/37-38-45-60-61	
Dossier submitters proposal	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Keep Carc. Cat. 3; R40 Keep Xn; R22 Remove T; R23 Remove Xn; R21 Add Xi; R37 Add R43 Keep N; R50-53	Keep N Remove T Add Xn Keep R: 22-40-50/53 Remove R: 21-23 Add R: 37-43	
RAC opinion	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Keep Carc. Cat. 3; R40 Keep Xn; R22 Remove T; R23 Remove Xn; R21 Add R43 Keep N; R50-53	Keep N Remove T Add Xn Keep R: 22-40-50/53 Remove R: 21-23 Add R: 43	
Resulting Annex VI entry if agreed by COM	613-133-00-X	etridiazole (ISO); 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole	219-991-8	2593-15-9	Carc. Cat. 3; R40 Xn; R22 R43 N; R50-53	Xn; N R: 22-40-43-50/53 S: (2-)-36/37-46-60-61	

SCIENTIFIC GROUNDS FOR THE OPINION

RAC general comment

For the current classification, etridiazole was discussed in the TC-C&L in November 1995, November 1996 and May 1997 (summary records ECBI/94/95, ECBI/45/96 and ECBI/27/97, respectively). Since the basis for the current classification is unknown, it is not possible to indicate in this CLH report which studies were or were not considered for the classification in the 25th ATP to Directive 67/548/EEC.

RAC evaluation of acute toxicity

Summary of the Dossier submitter's proposal
Based on the incongruity between the results of the acute toxicity studies in the DAR and the current classification, it was concluded that the studies in the DAR were not available for the TC-C&L in 1995-1997, even though these studies were conducted already in 1994.

Acute toxicity: oral

In an OECD TG 401 –compliant study on rats, doses of 700, 850, 1000 and 1400 mg/kg bw were tested. The acute oral LD_{50} of etridiazole was determined to be 1141 and 945 mg/kg bw for males and females, respectively. Therefore, the current classification as Acute Tox. 4*; H302 is proposed to be changed to Acute Tox. 4; H302 by removing the *, indicating the removal of the minimum classification from this category.

Acute toxicity: inhalation

It is proposed to remove the current classification Acute Tox. 4*; H331 because in the OECD TG 403 rat study, the acute inhalation LC_{50} values of etridiazole were found to be greater than 5.7 mg/l for both males and females (aerosol exposure with mass median aerodynamic diameter (MMAD) 1.9-3.8 µm; geometric standard deviation (GSD) 1.60-1.81 µm).

Acute toxicity: dermal

It is proposed to remove the current classification Acute Tox. 4*; H312 because in an OECD TG 402 rabbit study no mortality occurred at the tested dose of 5000 mg/kg bw. The observed clinical signs consisted of loss of appetite and decreased defecation and/or no stools. These effects disappeared within a week after treatment. Body weight loss was observed during the first 8 days after treatment in 4/5 males and in 4/5 females. Thus, the acute dermal LD_{50} of etridiazole was found to be >5000 mg/kg bw in male and female rabbits.

Assessment and comparison with the classification criteria
The acute LD_{50} of etridiazole (1141 mg/kg bw in male rats and 945 mg/kg bw in female rats) via the oral route falls within the range of values for classification for acute toxicity 4; H302 (300 <LD_{50}> 2000 mg/kg bw) and Xn; R22 (200 <LD_{50}> 2000 mg/kg bw) in accordance with the CLP and DSD criteria, respectively. For the inhalation and dermal routes, all estimated LD/C_{50} values are above the criteria threshold for classification and labelling (CLP and DSD).

RAC supported the classification as Acute Tox. 4; H302 (Xn; R22) and agreed with the dossier submitter (DS) to remove the classifications as Acute Tox. 3*; H331 (T; R23) and Acute Tox. 4*; H312 (Xn; R21).

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

Summary of the Dossier submitter's proposal

The DS proposed to classify etridiazole for respiratory tract irritation (STOT SE3, H335) based on the results of an OECD TG 403 acute inhalation study in rats and an OECD TG 412 sub-acute repeated dose inhalation toxicity study in rats.

In the acute inhalation study, laboured breathing (M:1/5; F:5/5) was observed in the high-dose group (5.7 mg/l) within 1 day of exposure. After day 1, rapid respiration (M:3/5; F:5/5) and laboured breathing (F:2/5) were observed, but these effects disappeared before the termination of the experiment. At 1.2 mg/l, rapid respiration was observed in all animals on the day of exposure. After day 1, one female still showed rapid respiration but the effect was reversed by the end of the experiment.

In the repeated dose inhalation toxicity study, no NOAEL could be derived for local effects, because squamous metaplasia of the larynx mucosa was observed at all dose levels (LOAEL 15 mg/m^3 = 0.015 mg/l).

According to the DS, the observed findings (breathing difficulties and squamous metaplasia) were signs of reversible respiratory tract irritation. Therefore, for these effects, classification for specific target organ toxicity after a single exposure (STOT SE 3; H335 and Xi; R37) was considered as more appropriate than classification for specific target organ toxicity after repeated exposure.

Comments received during public consultation

The German MSCA did not support the proposed classification as STOT SE 3 because etridiazole had been tested with rather high dust concentrations (about 1000 and 5000 mg/m³). According to the MSCA, it was normal that animals tried to prevent inhalation of such high dust concentrations. Therefore, clinical observations such as laboured breathing and/or rapid respiration would be an expected general reaction in animals exposed to such high concentrations of dust rather than indicating a potential for chemical-specific primary respiratory irritation.

Two MSCAs supported the DS proposal to classify etridiazole as STOT SE 3; H335 and Xi; R37.

Assessment and comparison with the classification criteria

Since human data for respiratory irritation was not available, information obtained from single and repeated inhalation toxicity tests was used in accordance with the CLP Guidance Annex 1:3.8.2.2.1.

The acute inhalation study reported abnormal gait, tremors, decreased activity, increased salivation and labored breathing in exposed rats. While no mortality was seen up to 5.7 mg/l, laboured breathing was seen immediately after exposure and rapid respiration was noted after day 1 in the acute inhalation test. At 1.2 mg/l rapid respiration was observed immediately after exposure in males and females. The effect was reversible on day 1 in all animals apart from one female. At the end of the observation period no treatment-related macroscopic observations were seen. Histopathology data were not available in the CLH report or in the draft assessment report (DAR). Neither nasal discharge nor rhinitis was reported, although it was recognised that the reporting was not very detailed and there was no information on effects during the exposure. Given the dusty nature of the chemical, RAC does not interpret these observations as clear evidence of a chemical-specific, respiratory irritant response.

Rats repeatedly exposed to etridiazole at concentrations of 0.015, 0.075 and 0.2 mg/l showed red nasal discharge with a dose-related trend. However, in the single exposure study, with the higher concentrations of 1.2 and 5.7 mg/l etridiazole, no nasal discharge

was reported. RAC concluded that the effect was likely to occur only after repeated exposure and does not support classification for STOT SE.

As the metaplasia of the larynx reported in rats after repeated exposure was only of minimal severity and as it was unclear whether the effect was a result of single or repeated exposure the effect was not considered as sufficient to support classification for STOT SE.

Overall, RAC concluded that the available evidence did not warrant classification for STOT SE 3; H335 (Xi; R37).

RAC evaluation of skin corrosion/irritation

Summary of the Dossier submitter's proposal
No skin irritation was observed in an OECD TG 404 skin irritation study in rabbits. The dossier submitter did not propose classification for skin corrosion/irritation.

Comments received during public consultation
No specific comments were received on this hazard class.

Assessment and comparison with the classification criteria
As no skin irritation was observed in the rabbit study, RAC agreed with the dossier submitter's conclusion that no classification for skin corrosion/irritation is warranted

RAC evaluation of eye corrosion/irritation

Summary of the Dossier submitter's proposal
No classification for eye corrosion/irritation was proposed by the dossier submitter as only slight eye irritation was observed in an OECD TG 405 eye irritation study in rabbits and no human data were available.

Comments received during public consultation
No specific comments were received for this hazard class.

Assessment and comparison with the classification criteria
Mean scores (24/48/72 h) on eye irritation were reported in the DAR. Due to the low scores for eye irritation RAC agreed with the dossier submitter's conclusion that no classification for eye corrosion/irritation was warranted.

RAC evaluation of respiratory sensitisation

Summary of the Dossier submitter's proposal
No data on respiratory sensitisation was available for the dossier submitter. Accordingly, no classification was proposed for this hazard class.

Comments received during public consultation
No comments were received for this hazard class.

Assessment and comparison with the classification criteria
RAC could not conclude on the classification for respiratory sensitisation due to the lack of data and this should be considered as 'not evaluated'.

RAC evaluation of skin sensitisation

Summary of the Dossier submitter's proposal
In an OECD TG 406 (Magnusson and Kligman) study on 20 control and 20 test female guinea pigs within a group, the following concentrations of etridiazole were tested: 20%

intradermal induction, 100% topical induction, 100 or 50% first challenge and 20, 10, 5 or 1% second challenge. After topical challenge, none to moderate erythema and none to well-defined oedema were observed in the treated animals. Also thickening, dryness and sloughing of the epidermis were noted. In comparison, to the 50% concentration, the effects were more marked after challenge with undiluted etridiazole. All treated animals were sensitised. After topical rechallenge with 20, 10, 5, or 1% etridiazole, the number of sensitised animals was 20, 16, 8, and 1, respectively. Topical challenge and rechallenge in control animals did not induce any dermal reactions. The results of the positive control were not reported. According to the dossier submitter, etridiazole showed skin reactions in 100% of the animals in an adjuvant type guinea pig test method for skin sensitisation.

The dossier submitter proposed to classify etridiazole for Skin Sens. 1B; H317 (CLP) and Xi; R43 (DSD) because over 30% of the animals were considered positive after >1% induction concentration.

Comments received during public consultation

Three MSCAs supported the dossier submitter's proposal to classify etridiazole for Skin Sens. 1B; H317 because skin reactions were observed in ≥30% of the animals treated with >1% etridiazole.

Assessment and comparison with the classification criteria

100% positive responders were observed in a Guinea Pig Maximization Test using 20% etridiazole for the intradermal induction dose.

The CLP criteria for skin sensitisation as amended by the 2nd ATP state that "Skin sensitisers shall be classified in category 1 where data are not sufficient for sub-categorisation" (3.4.2.2.1.1.). In the draft CLP guidance it is stated that:

> *"Classification into sub-categories is only allowed if data are sufficient. Therefore care should be taken when classifying substances into category 1B when category 1A cannot be excluded. In such cases classification into category 1 should be considered. This is particularly important if only data are available from certain tests showing a high response after exposure to a high concentration but where lower concentrations which could show the presence of such effects at lower doses are absent (in line with some test protocols where a maximised dose should be used)".*

RAC agreed with the dossier submitter that etridiazole should be classified as a skin sensitiser because skin reactions were seen in ≥30% of test animals at >1% induction concentrations. RAC did not, however, agree with the dossier submitter's original proposal to classify for skin sensitisation 1B because the data did not allow the exclusion of category 1A. RAC concluded therefore that etridiazole should be classified as Skin Sens. 1; H317 (Xi; R43).

RAC evaluation of repeated dose toxicity (DSD) and specific target organ toxicity (CLP) – repeated exposure (STOT RE)

Summary of the Dossier submitter's proposal

The dossier submitter presented a summary table on the available repeated dose studies. Three rat studies (a 28-day dermal, a 28-day inhalation and a 90-day oral) and a 1-year oral study in dogs were referenced in the CLH report. The detailed study results were given only for the 28-day inhalation study. For other studies only the NOAEL/C was reported. All studies followed OECD TG test guidelines. More information on the studies from the DAR has therefore been added below.

The dossier submitter did not propose to classification for STOT RE.

Comments received during public consultation

According to the one MSCA, the increased platelet counts in male and female rats at ≥64.7 mg/kg and reduced prothrombin time (PTT) and activated thromboplastin time (APTT) at ≥29.5 mg/kg in the oral 90-day study (Richards, 1994) indicated a hypercoagulable status. Thrombus formation in the mouse heart at ≥184.7 mg/kg in the 18-month carcinogenicity study (Goldenthal, 2004) supported their interpretation.. The same MSCA concluded that these effects may be significant (meaning changes which clearly indicate functional disturbance or morphological changes which are toxicologically relevant) but not serious (meaning more profound or serious effects and indicating changes that are of a considerably adverse nature with a significant impact on health) and questioned whether this evidence would be sufficient for STOT RE 2 classification. The DS responded that the increase in platelet counts was small (126% of the control) and was only statistically significant in females. The effects on PTT and APTT were only minor and observed only in males. Furthermore, no increased thrombus formation was observed at these dose levels in the 13-week study.

No other specific comments were received on this endpoint.

Assessment and comparison with the classification criteria

1. Inhalation route:

Effects occurring below the guidance value for STOT RE 2 (600 mg/m³/6 h/d for the 28-day inhalation study as extrapolated from the guidance value for a 90-day inhalation study according to the Haber's rule Table 3.9.2.2, CLP guidance) were considered:

A. Systemic effects

The observed systemic effects included decreased body weight gain (75-78% of control), increased liver weight and liver cell hypertrophy, reduced forelimb grip strengths in males (83% of control) and squamous metaplasia of the larynx. Liver effects were not considered relevant for classification because they were not corroborated by other significant findings indicating significant morphological or functional damage. With regard to the reduced forelimb grip, the DAR did not consider this as a relevant toxic effect, since no effect was observed on hindlimb grip strength in males and no effect on grip strength in females. RAC did not agree with the reasoning in the DAR regarding the irrelevance of the reduced forelimb grip in males because neuronal discords may occur in segments of the spinal cord. However, the significance of the finding is unclear because no indications of associated clinical neuro-functional disorders or histopathological findings were reported. Impaired limb function or ataxia was also observed in other repeated dose studies (mouse carcinogenicity study, developmental toxicity study on rats), but the effects were only seen at maximum tolerated dose (MTD) levels.

B. Local effects on the respiratory tract

Larynx metaplasia was reported as minimal in the CLH report and it was interpreted as a non-adverse lesion. Minimal metaplasia in the ventral area of the larynx is commonly observed in non-treated rats (Kaufmann et al., 2009). As no evidence was seen in control rats, the effect could be attributed to the exposure. However, RAC agrees with the dossier submitter's interpretation of the non-adverse nature of the lesion at this minimal grade and concludes that classification for STOT RE is not warranted for the reported effect.

2. Oral route:

Relevant effects occurring below the guidance value for STOT RE 2 (100 mg/kg bw/d for the 90-day oral study, and 25 mg/kg bw/d for the 1-year oral study as extrapolated from the guidance value for a 90-day oral study according to the Haber's rule Table 3.9.2.2, CLP guidance) were considered.

Increased reticulocyte counts (in both sexes at 1250 mg/kg food) observed in the 90-day oral study in rats as an isolated effect without other relevant findings on red blood cell

parameters indicating anaemic conditions (see the CLP guidance Section 3.9.2.5.2 on haemotoxicity) is not relevant for classification. Decreased PTT and APTT times in males at ≥600 mg/kg food (equal to 29.5 mg/kg bw/d for males and 35.2 mg/kg bw/d for females) were considered as indicative of a hyper-coagulable status by one MSCA. RAC accepted the dossier submitter's argument that the effects are not severe enough to warrant classification. In the case of a biologically significant hypercoagulative status, thrombi should be seen in circulation of major organs and in small peripheral vessels. The 18-month mouse carcinogenicity study reported increased incidences of kidney infarcts and thrombi in the myocardium in mid and high dose (at above 900/1300 mg/kg food, equal to 184.7 and 221.7 mg/kg bw/d in males and females, respectively) after 18 months of administration in diet, which is far above guidance values for classification.

In the 90-day oral study, increased bilirubin in high dose males and increased cholesterol in mid and high dose males indicate disturbances of liver function. However, no corresponding morphological abnormalities were reported in the liver or bile system in the study. It is to be noted that bile duct hyperplasia was observed after chronic administration in rats and mice at doses above guidance values for classification when corrected for duration, and cholangiocarcinomas were increased in rats. In the 90-day oral study, liver weight increases and liver cell hypertrophy were seen in both sexes at ≥600 mg/kg food, being reversible at the end of recovery period. RAC considers these liver effects to be adaptive responses that do not warrant classification.

Increased incidence of hyaline droplets in the kidneys was noted at 600 and 1250 mg/kg food in 9/10 and 10/10 males, respectively. As the incidence was already high in the control males (6/10) and the α2u-globulin-specific staining was positive, these kidney effects are interpreted as a male rat-specific finding and the increase in its incidence is of questionable toxicological significance.

Increased alkaline phosphatase, APTT and cholesterol were noted in dogs in the 1-year dog study. Increased cholesterol levels and increased activity of alkaline phosphatase may indicate disturbed liver/biliary function, but because no corresponding morphological abnormalities (except increase in liver weight) were reported, RAC does not consider these effects as sufficient to warrant classification for STOT RE. RAC concludes that the high mortality rates seen at 45 mg/kg bw in the rabbit teratogenicity study do not either support classification for STOT RE, as it was not known at which time-point during the 13-day treatment the dams died and because no serious adverse effects occurred below the guidance values in the oral 90-day rat study or in the inhalation 28-day rat study.

Overall RAC considered that the observed findings after the exposure via the oral route were not sufficiently serious to justify classification.

3. Dermal route:

Effects occurring below the guidance value for STOT RE 2 (600 mg/kg bw/d for the 28-day dermal study as extrapolated from the guidance value for a 90-day dermal study according to the Haber's rule Table 3.9.2.2, CLP guidance) were liver cell hypertrophy and liver weight increases at ≥400 mg/kg bw/d. As no signs of degenerative lesions or indications of significant functional disorders were observed, RAC considered the liver effects as adaptive and not relevant for classification.

In conclusion RAC supported the dossier submitter's proposal that no classification is warranted for STOT RE.

RAC evaluation of germ cell mutagenicity

Summary of the Dossier submitter's proposal

The dossier submitter did not propose classification for mutagenicity, because in the genotoxicity studies there were no indications for classification.

Comments received during public consultation
No specific comments were received on this hazard class.

Assessment and comparison with the classification criteria
According to the dossier submitter's proposal, many of the in vitro tests were not in accordance with the corresponding guidelines, but based on the three well performed negative in vivo studies (micronucleus test, chromosomal aberration test, UDS test) etridiazole was considered to be non-mutagenic in vivo.

RAC also concluded that the result of the replicative DNA synthesis test did not support the assumption that etridiazole possesses promotor activity with a threshold level.

RAC agreed with the dossier submitter that in vitro and in vivo testing indicates that etridiazole had no genotoxic potential and that classification for germ cell mutagenicity was therefore not warranted.

RAC evaluation of carcinogenicity

Summary of the Dossier submitter's proposal
The dossier submitter proposed classification for Carc. 2; H351 (CLP; DSD: Carc. Cat. 3; R40), in accordance with the current classification, as it was proposed by TC-C&L in November 1995, November 1996 and May 1997 (summary records ECBI/94/95, ECBI/45/96 and ECBI/27/97, respectively).

The proposal was based on the observations in a 2-year rat study (liver tumours, and potentially relevant thyroid and testis tumours), an 18-month mouse study, mechanistic studies and further argumentation in the DAR. Both oral carcinogenicity studies were performed according to the OECD TG guidelines. No studies via inhalation or dermal routes were available.

Comments received during public consultation
During the first PC, comments on this hazard class were received from one MSCA, which agreed with the proposal of the dossier submitter based on a review of the carcinogenicity studies in rat and mouse and of the in vivo mechanistic studies in rat.

During the development of this opinion, carcinogenicity was re-opened for a second (targeted) public consultation in order to strengthen the information base and because this hazard class was not specifically included in the list for which comments had been requested during the first public consultation.

During the targeted public consultation one MSCA supported the conclusion that the mid and high doses in the mouse carcinogenicity study exceeded the MTD. Furthermore, the observations that a number of deaths were related to liver failure and that liver tumours were not the cause of increased mortality were found to be consistent with the interpretation that the maximum tolerated concentration (MTC) was exceeded for the mid and high doses in the mouse study. The classification as Carc. 2 (CLP) was supported by three commenting MSCAs and by industry.

Assessment and comparison with the classification criteria
In total, three rat studies and two mouse studies on carcinogenicity were available. Only the 2-year rat study and the 18-month mouse study, which were predominantly performed in accordance with test guidelines, were considered relevant for the RAC evaluation. The other studies (from 1968 to 1983) were incomplete due to lack of investigated parameters/animals and early termination because of infections or unusually high tumour incidences in controls.

There was an indication of treatment-related increases in tumour rates in the two valid carcinogenicity studies. Only incidences were reported in the study summaries, but the statistics and information about the severities of the effects were lacking.

Rat carcinogenicity study (study 1 in the CLH-report):

In the 109-week carcinogenicity study, rats received etridiazole at 0, 100, 640, or 1280 mg/kg food (equivalent to 0, 5, 30, or 63 mg/kg bw/d for males and 0, 6, 38, or 84 mg/kg bw/d for females). The tumour rates in treated animals as compared to controls were increased in the liver, thyroid, testes, kidneys and mammary glands. Survival rates were not affected by the treatment. A reduction in body weight gain was reported in mid and high dose females. The reduced growth can be assumed to be a consequence of reduced food consumption. The absence of clinical signs and effects on survival support the assumption. However, the food efficiency calculations in the original study report did not reveal marked differences between the control and test groups, although the mean body weight gains of the high dose females relative to their feed intake tended to fall somewhat consistently below control until week 46. With regard to the selected doses, RAC considers that the MTD was not exceeded. The non-neoplastic effects in kidneys and liver are thought to be related to specific toxic/pre-neoplastic effects rather than to non-specific general toxicity.
Compared to controls, the incidences in hepatocellular adenomas were increased in male and female rats at the high dose. In addition, high dose females had increased incidences in hepatocellular carcinomas and cholangiocarcinomas, a tumour type that is rarely found in rats. In general, female rats were more sensitive than male rats. Liver tumours in male rats were mainly benign. The incidences of the liver tumours in concurrent controls were largely in the range of tumour incidences of the conducting laboratory. In conclusion, the liver adenomas/adenocarcinomas and cholangiocarcinomas were considered to be caused by etridiazole treatment.

With respect to non-neoplastic lesions in the liver, there were dose-related increases in incidences of hepatocytomegaly in male and female rats of the mid and high dose groups that was interpreted by RAC to be a precursor lesion in the development of hepatocellular adenomas and carcinomas. Bile duct hyperplasia, bile duct fibrosis, and chronic inflammation of bile ducts may be considered as possible precursor lesions of cholangiocarcinomas and occurred in rats of control and dose groups. Incidences appeared to be higher in high dose females. However, the incidences in controls were already high. Data on severity and distribution of the lesions would have been informative to enable conclusions to be drawn on the dose-response relationship of these lesions conclude on dose-responsiveness. These data were absent in the CLH report and DAR.

In comparison to control incidences, increased incidences of follicular cell adenomas were seen in high dose males and females, but male rats were more sensitive than female rats. Higher incidences of follicular cell adenocarcinomas were only found in males at mid and high dose. In general, the tumour response in females was weaker and predominantly benign. Concurrent control incidences of adenomas in males (12%) were slightly above the laboratory control incidences (range 2-7% for the period 1984-1989, mean 4.7%). There were no adenomas in concurrent female controls, which was consistent with the laboratory historical control values (range 0-1%, mean 0.4% 1984-1989). It was concluded by RAC that thyroid tumours in high dose males and females and mid dose males were caused by the etridiazole treatment.

In testis, interstitial tumour incidences were markedly increased at the high dose (20%). While in the concurrent control this tumour was not found, the historical incidences of the conducting laboratories were 4.7% (range 2.4-13.3 % for the period 1984-1989). A relationship to treatment can be assumed from the increased incidence at the high dose. Tumour findings were in line with increased incidence of the putative precursor lesion, the interstitial cell hyperplasia.

In the kidney, 1 adenoma was observed at 100 and 1280 mg/kg food whereas 4 and 1 adenocarcinomas were observed at 640 and 1280 mg/kg food, respectively, in males. Tubular cell tumours of the kidney are rare tumours in rats. There were no tumours in concurrent controls, which is consistent with the range of laboratory historical control

values (range 0-1.2% for adenomas, 0-2.9% for carcinomas in 1984-1989). Although no clear dose-response relationship was given, since this tumour type is rare, this finding could be of some concern. Moreover, the high incidences of tubular cell karyomegaly in the dose groups only, a finding that can be seen as an early response to renal carcinogens, should be taken into account. Karyomegaly means that the cell nucleus is enlarged and often hyperchromatic, possibly due to polyploidy and abnormal DNA distribution.

The dossier submitter noted that kidney tumours may have resulted from alpha-2-microglobulin accumulation, which is a male rat specific mechanism and not relevant to humans. Neither alpha-2-microglobulin, hyaline droplets, intra-tubular protein casts nor any other nephropathy or related regenerative cellular proliferation were observed in the 109-week study in the rat (neither in the controls of the same age nor in the treated animals). RAC has noted that alpha-2-microglobulin and related effects should not be expected at the end of a chronic study and that RAC should reconsider the conclusion on the human relevance of the kidney tumours. RAC noted that the DAR on the 13-week study did report increased incidences of hyaline droplets at 600 and 1250 mg/kg food at the end of the 90-day treatment period, but no other effects were observed that may have indicated associated cell damage/cell death and secondarily increased (regenerative) cell proliferative activity. After recovery, alpha-2-microglobulin was found in all males of control and dosed groups. No information was available on the severity of hyaline droplets. In the original study results, chronic nephropathy was reported at similar prevalence among the study groups. Evidence of hyaline droplets and positive staining for alpha-2-microglobulin in control and treated rats at the end of the 13-week oral treatment alone does not give sufficient evidence that tumours can be related to this male rat- specific lesion. Furthermore it can not be excluded that the increases in karyomegaly in dosed males and females are precursors to proliferative lesions. Overall, there is some evidence for kidney carcinogenicity. However, there are uncertainties due to these findings being observed at low incidences, only in males and because there was no dose-response relationship. RAC concludes that the mode of action leading to the kidney tumours is unknown but it could also be related to etridiazole treatment.

In mammary glands, increased incidences of fibroadenomas were observed in all treatment groups with highest incidence in high dose females. Control incidence was 18% and the historical control range was 11.6%-69.25% (mean 39%) demonstrating that this tumour type was quite common over 1984-1989 in this rat strain. Due to the high spontaneous incidences RAC considers treatment relationship to be questionable.

Mouse carcinogenicity study (study 2 in the CLH report):

In the 18-month carcinogenicity study, mice received etridiazole in diet at 0, 50, 900 or 1800 mg/kg food. After the first week of dosing, the dietary concentrations were increased to 1300 and 2000 mg/kg food for the mid and high dose groups, respectively, but at week 43, the high dose level was decreased to 1600 mg/kg food due to excessive mortality in this group. Dose levels corresponded to 0, 7.5, 184.7 and 263.3 mg/kg bw/d in males and 0, 9.1, 221.7 and 312.1 mg/kg bw/d in females.

Mortality at high dose groups was 60% and 68% in males and females, respectively, which is still below the percentage (survival 25%, OECD TG Guidance Document 116) when a study should be terminated due to low survival. Mortality at mid-dose was 44% both in males and in females. At high dose, clinical signs such as decreased activity, breathing difficulties, lack of appetite, tremors, hunched posture, impaired limb function and pale and cold skin indicated severely affected general health status. High dose males showed significantly lower body weights than controls from week 3 onwards (91% of the controls) and high dose males and females and mid dose males showed significantly lower food consumption. No numerical values were given either in the CLH report or in the DAR to allow concluding on whether lower body weight was related to the food consumption. Although no clinical signs of disturbed general health status (except lower food intake)

were reported for the mid dose group, RAC agreed that also the mid dose was above the MTD due to high mortality rates.

No statistical information and information on the severity and distribution of non-neoplastic lesions were found in the CLH-report.

Increased liver adenoma incidences were at the same level in mid and high dose males, while increases appeared to be dose-related in mid and high dose females. However, tumour responses may have been influenced by mortality rates, but tumour data were not corrected for life-span. Tumour incidences in controls were low in males and absent in females. Thus tumour responses in dose groups were not confounded by high spontaneous tumour rates. Hepatocellular carcinomas were observed in mid dose males and high dose females. Corresponding findings accompanying the liver tumour development were increased liver weights, enlarged livers, livers with nodules and masses and several degenerative-regenerative hepatocellular lesions were more frequently found in mid and high dose males and females. Like in rats, in comparison to the controls bile duct hyperplasia occurred at higher incidences in mid and high dose males and high dose females, but in contrast to rats, mice did not develop cholangiocarcinomas.

Mode of action consideration
From mutagenicity testing, etridiazole was considered to have no genotoxic potential but other mechanisms may be relevant.

For the liver, the dossier submitter assumed similarities in tumour promoting activities of etridiazole and phenobarbital and analysed liver enzyme activities in mechanistic studies (see Etridiazole additional report, 05, Vol. 3, B6). Etridiazole treatment resulted in liver cell hypertrophy due to increased smooth endoplasmatic reticulum, in a decrease in phase 1 enzymes (CYP450, MCD and ECD), and a slight increase in CYP2B1 and CYP2C13. The conclusion that metabolic enzyme responses were different to phenobarbital was copied from the DAR in the CLH report.

Promoter activity was reported to be separately investigated in initiator and promoter studies using N-nitrosodiethylamine (DEN) as initiator. RAC did not agree with the interpretation of the dossier submitter that increased replicative DNA synthesis supports the assumption that etridiazole possesses promoter activity with a threshold level. It should be noted that promoter activity as such is of minor relevance for classification. Chronic administration of etridiazole alone in the diet induced liver tumours in rats and mice. Finally, the mode of action of etridiazole to induce liver adenomas/carcinomas and cholangiocarcinomas remains unknown.

To conclude on whether one or two species are clearly positive, the tumour data in the mouse are critical. Although no clinical signs of disturbed general health (except lower food intake) were reported, RAC considered the mid and high dose groups in the mouse carcinogenicity study to be above the MTD due to the high mortality rates observed. Therefore, the tumour responses at the mid and high dose were of questionable relevance for the assessment of etridiazole-related carcinogenicity.

The mode of action for the rat thyroid tumours remains unknown. The UDP-gamma transferase activity was increased in the mechanistic study, but there were no effects on rat TSH or changes in T3 or T4. Furthermore, no consistent effects were seen in other studies. RAC concludes that the available data do not support for a rat-specific phenomenon.

RAC did not agree with the dossier submitter's interpretation that the increased tumour rate in rat testes was above the MTD. Also, the observation that Leydig cell tumours in humans are rare does not support the non-relevance of this tumour type for humans. As the mode of action is unknown, the default assumption is that the tumour is of potential relevance for humans.

For the rat kidney tumours, the proposed mode of action (involving alpha-2 microglobulin) does not appear to be plausible. However, the evidence for the kidney tumours resulting from treatment with Etridiazole is weak, because these findings were observed at low incidences, only in males and because there was no dose-response relationship.

Conclusion
In accordance with the criteria in CLP Regulation EC/1272/2008 classification in category 1A for carcinogenicity is not justified (DSD category 1) given that there is no evidence of etridiazole having caused cancer in humans. It is therefore necessary to decide whether to classify etridiazole in CLP category 1B or category 2.
The RAC based its conclusions on carcinogenicity on two available studies, a 104-week study in rats and an 18-month study in mice. The neoplastic lesions observed in the liver at the mid and high dose groups in the mouse study were not considered to be relevant for classification, as both these doses appeared to exceed the maximum tolerable dose (MTD), based on high mortality rates. Regarding the rat study, the RAC concludes that the MTD was not exceeded at any dose. Evident increases in tumour incidences were observed in the liver, thyroid and testes. However, female rats were more prone to developing liver tumours than male rats and liver tumours in male rats were mainly benign. The thyroid tumours were markedly increased in males of the mid and high dose groups, whereas the tumour response in females was weaker and predominantly benign. Because one tumour type was more prevalent in one sex in the rat and because the neoplastic lesions were observed at above the MTD in the mouse study, the RAC concluded that the carcinogenic potential of etridiazole is not sufficient for classification as Carc. 1B. The existing classification of etridiazole as Carc. 2; H351 (DSD: Carc. Cat. 3, R40) is therefore appropriate.

RAC evaluation of reproductive toxicity

Summary of the Dossier submitter's proposal
Currently etridiazole is not classified for reproductive toxicity. The dossier submitter clarified that etridiazole was discussed in the TC-C&L in November 1995, November 1996 and May 1997 (summary records ECBI/94/95, ECBI/45/96 and ECBI/27/97). Since the basis for the current classification is unknown due to a lack of information on the studies used, it is not possible to indicate in the CLH report which studies are new information compared to the studies used for the classification in the 25th ATP to Directive 67/548/EEC.

Sexual Function and Fertility

In a 2-generation oral reproductive toxicity study performed in accordance with OECD TG 416, rats were exposed to etridiazole at 0, 80, 320 and 800 mg/kg food for males and 0, 80, 320 and 640 mg/kg food for females. A decrease in body weight and food consumption was noted among males and females for the F0-generation at 800/640 mg/kg food and for the F1-generation at 320 and 800/640 mg/kg food. No treatment related changes were observed in fertility, oestrus cycle, sperm parameters, mating behaviour, conception or gestation. The dossier submitter concluded that the 2-generation study in rats did not provide any indications for classification for reproductive toxicity.

Developmental toxicity

In a teratogenicity study performed in accordance with OECD TG 414 guideline, rats received etridiazole by gavage 0, 10, 30 or 75 mg/kg bw/d. The NOAEL for maternal effects was set at 30 mg/kg bw/d, based on increased mortality, clinical signs, and decreased body weight. The NOAEL for developmental effects was set at 30 mg/kg bw/d, based on decreased mean foetal weight, anasarca in 2 foetuses, and retarded ossification of various bones. No treatment-related effects were observed in the number of corpora lutea or implantations, the number or percentage of live foetuses, or the sex ratio. There were no morphological changes observed in fetuses that could be attributed to treatment.

In a teratogenicity study in rabbits receiving etridiazole by gavage 0, 1.7, 5, 15 or 45 mg/kg bw/d, the NOAEL for maternal effects was set at 15 mg/kg bw/d based on mortality and decreased body weight. Potential critical effects on e.g. liver, kidney and thyroid were not studied and therefore, the derived maternal NOAEL from this study might not have been accurate. The NOAEL for developmental effects was set at 15 mg/kg bw/d, based on decreased mean foetal weight, a reduction of live foetuses per dam and an increase in dams with resorptions at 45 mg/kg bw/d. No treatment-related effects were observed in the number of corpora lutea or implantation sites, or in the sex ratio. Skeletal examination revealed an increased incidence in missing sternebrae, tail defects and underdeveloped hind limbs. Soft tissue examination showed an increased incidence of crossed hind legs and open eyes. Developmental malformations were observed at doses causing high maternal mortality in rabbits.

The CLP regulation states that maternal mortality over 10% is considered excessive and the data for that dose level shall not normally be considered for further evaluation. In the rat and rabbit developmental studies the maternal mortality was about 20% at the high dose.

The foetal toxicity observed in the rat study was based on retarded ossification. This finding was observed at the dose level causing severe maternal toxicity, including mortality. The dossier submitter concluded that foetal toxicity was secondary to maternal toxicity and therefore not relevant for classification.

For the rabbit developmental study, no historical control data were provided. Missing sternebrae, tail defects, underdeveloped hind limbs, crossed hind legs and open eyes were reported in the high dose group. There were mortalities observed in the dams (3/14) indicating high maternal toxicity. The dams which did not die were not examined for other critical signs of toxicity. Moreover, the mortality at a slightly lower dose level (30 mg/kg bw/d) in the preliminary study also indicated that maternal toxicity at the high dose (45 mg/kg bw/d) was excessive and the data for such dose levels should not normally be considered for further evaluation. Therefore, the dossier submitter concluded that the observed developmental effects were not relevant for classification.

The dossier submitter did not propose classification for reproductive toxicity.

Comments received during public consultation

During the first public consultation, the Two MSCAs agreed with the dossier submitter's proposal for non-classification for this endpoint. A third MSCA suggested that RAC should discuss whether classification for developmental toxicity category 2; H361 (Repr, 2; H361d) would be warranted, noting that the relevant question was whether there was sufficient evidence for classification in category 2 for developmental effects because the toxic dose in the rabbit teratogenicity study also caused serious toxicity in mothers., A fourth MSCA proposed classification as developmental toxicity category 2 and commented that since the historical control data was missing from the rabbit teratogenicity study, the developmental effects could have been a consequence of etridiazole treatment. Moreover, the NOAEL for teratogenic effects was set at 15 mg/kg bw/d based on irreversible structural effects.

During the opinion development process, reproductive toxicity was re-opened for a second (targeted) public consultation in order to strengthen the information base and because this hazard class was not specifically addressed in the list for which comments had been requested during the first public consultation. Industry did not agree with the proposal to classify for developmental toxicity and concluded that the foetal toxicity observed in the developmental studies was based on retarded ossification and that it was observed at dose levels causing severe maternal toxicity, including mortality. The foetal toxicity was considered to be secondary to the maternal toxicity and the retarded ossification of various bones and skeletal malformations observed was not considered relevant for classification for reproductive toxicity. Also, one MSCA supported non-classification for developmental toxicity.

Fertility
RAC agreed with the dossier submitter that in the absence of treatment-related effects on reproductive function in a 2-generation study, a classification could not be justified. (In this study histopathology was conducted on the kidney (no data on other organs) and chronic nephropathy was found in 7/8 F0-males at 800 mg/kg bw/d; this effect was not seen in other male and female groups.)

Developmental toxicity
The developmental study in rats revealed decreased mean foetal weight and anasarca in 2 foetuses and retarded ossification of various bones at 75 mg/kg bw/d. Retarded ossification was frequently observed in sternebrae #5 and #6 and some other bones. At the same dose mortalities (5/25 dams), reduced body weight gain and clinical signs of impaired general health were observed. Body weight loss was seen during the initial phase (GD 6-9 at ≥10 mg/kg bw/d and GD 6-12 at 75 mg/kg bw/d), but there was no data on the quantity of body weight loss in the CLH report. As foetal toxicity occurred only at a dose that induced severe maternal toxicity, the RAC considered these effects to be secondary to severe maternal toxicity.
At 45 mg/kg bw/d the number of live foetuses/dam and pub viability were reduced. Missing sternebrae (3 foetuses in 3 different litters), tail defects (5 foetuses in 2 different litters), underdeveloped hind limbs (4 foetuses in one litter), crossed hind limbs (7 foetuses in 2 different litters) and open eyes (6 foetuses in 2 different litters) were observed at 45 mg/kg bw/d. Mortalities in 3/14 dams at 45 mg/kg bw/d in the main teratogenicity study appeared to be conclusive taking into account that in the range-finding study all rabbits that received 100 or 300 mg/kg bw/d during GD 6-18 died. At the lowest dose of 30 mg/kg bw/d in the range-finding study one rabbit died (the number of 5 dams/group was reported only in the original study report), one of the surviving dams resorbed its entire litter (6 implantation sites and 6 resorption sites), and one of them resorbed 1/8 implantations. The dead dam had also resorbed its entire litter (2 implantation sites and 2 resorption sites). In the negative control group none of the dams died or resorbed their litter.

In the main teratogenicity study, dam body weights were significantly lower than in the negative controls at 45 mg/kg bw/d on day 18 of gestation (absolute weight at GD 18 was 5% lower than on GD 0, while negative controls had 4% absolute weight gain at GD 18). At 45 mg/kg bw/d, a non-statistically significant decrease in body weight compared to controls was noted from day 6 to day 30 of gestation. Other clinical signs were not reported for the dams.

Original study reports indicated that malformed fetuses were not from dams that died prematurely. No viable pups originated from the dams that died (all implanted fetuses were resorbed.

RAC considers missing sternebrae as malformations on the basis of an international harmonisation activity on rat foetal skeletal terminology and classification (Solecki et al., 2001). Also, shortness or any other gross abnormality of the tail was considered to be a malformation by Solecki et al., 2001. Tail defects in combination with underdeveloped hind limbs may be indicative of a caudal dysplasia, which is defined as a severe reduction of caudal structures, including reduction or absence of hindlimbs, tail, and/or sacral area (Makris et al., 2009). However from the available report it cannot be clarified whether the reported effects occurred in combination or not. The RAC did not agree with industry's view that the observed effects can be interpreted as being caused by retarded ossification.

It is to be noted that the abnormalities occurred at a dose at which serious effects indicative of excessive maternal toxicity (21% mortalities, reduced body weight in survivors) had been observed. In survivors, 5% lower absolute body weight on GD 18 compared to GD 0 may be indicative of general toxicity. However no corrected body weight data are available to estimate the extent of the maternal toxic effect. No other signs of maternal toxicity were reported.

RAC concluded that classification for developmental effects at doses causing severe maternal toxicity was not consistent with the CLP criteria (3.7.2.4.4) which state that maternal mortality greater than 10% is considered excessive and that the data for that dose level shall not normally be considered for further evaluation. Accordingly, RAC concluded that the treatment-related effects on pup development at 45 mg/kg bw/d etridiazole co-occurring with maternal mortality greater than 10% do not justify the classification for this hazard class. However, taking into account the dose levels at which mortalities were observed in the range-finding study in rabbits, RAC points out that the highest dose of the main study was incorrectly chosen.

Overall, the observed malformations are considered to be a secondary non-specific consequence of severe maternal toxicity at the same dose. RAC concluded that no classification for reproductive toxicity is warranted.

RAC evaluation of environmental hazards

Summary of the Dossier submitter's proposal
The current Annex VI entry for etridiazole has an environmental classification for both Aquatic Acute 1 and Aquatic Chronic 1. The DS proposed to add both acute and chronic M-factors of 1 to the existing Annex VI entry.

Degradation
The DS provided information on etridiazole's abiotic degradation via hydrolysis and photolysis, and they were not considered relevant routes of degradation for classification of etridiazole.

Information on biotic degradation in a screening test (ready degradation) and in two different water/sediment systems was provided. In a closed bottle test according to OECD TG 301D (EC C.4-E) only 1% biodegradation after 28 days was achieved. In the simulation studies the primary degradation half-lives of etridiazole in the total water/sediment systems were less than two days. Degradation data on etridiazole and its degradation products in soil was also presented in the CLH report. The dossier submitter concluded that etridiazole is not rapidly degradable.

Bioaccumulation
Etridiazole has a measured log K_{ow} of 3.37 (aqueous pH 7 buffer/n-octanol). There is also a study available that measured the bioconcentration factor of etridiazole in bluegill sunfish (*Lepomis macrochirus*) according to the guideline US EPA 165. The BCFs for etridiazole in edible tissue, non-edible tissue and whole fish are 92, 256 and 165 L/kg wet weight, respectively. These values were not corrected for lipid content.

Based on both measured log K_{ow} and BCF values in fish the dossier submitter concluded that etridiazole is not potentially bioaccumulative according to CLP (the BCF is below 500) but fulfils the criteria as a substance having a potential for bioaccumulation according to DSD (the BCF is above 100).

Acute (short-term) aquatic toxicity
Etridiazole's acute toxicity was tested in two fish studies (*Oncorhynchus mykiss*, OECD TG TG 203), three aquatic invertebrate studies (*Daphnia magna*, OECD TG TG 202; *Americamysis bahia* and *Crassostrea virginica*, EPA TG 72-3), and three algal or aquatic plant studies (*Pseudokirchneriella subcapitata* and *Anabaena flos-aquae*, EPA TGs 122-2 and 122-3; and *Lemna gibba*, OECD TG 221). In addition, two acute fish studies, two on invertebrates and two on aquatic algae were reported on etridiazole degradation products (i.e. etridiazole acid and dichloro-etridiazole). According to the DS the lowest reliable L(E)C_{50} value for etridiazole was obtained in the test on *P. subcapitata,* i.e. E_rC_{50} (120 hours)

= 0.49 mg/l (mean measured concentration). In studies with the degradation products, the lowest reliable $L(E)C_{50}$ value of 0.77 mg/l was obtained in fish using dichloro-etridiazole.

Based on the result of the toxicity test with algae, the DS concluded that etridiazole fulfils the CLP criteria for classification with Aquatic Acute 1 with an M-factor of 1.

<u>*Long-term aquatic hazard*</u>
The DS reported that there are adequate chronic toxicity data available for etridiazole on all three trophic levels (one study on fish, EPA TG 72-4; one on invertebrates, EPA TG 72-4 and two on algae, EPA TGs 122-2 and 122-3). No chronic toxicity data were reported for the degradation products, with the exception of the 72-h NOE_rC values for two degradation products in algae which were both higher than 0.1 mg/l. The reported sub-chronic early life stage study in fish resulted in a NOEC value of 0.12 mg/l and the chronic *Daphnia magna* study resulted in a (21-d) NOEC of 0.37 mg/l. The lowest reported NOEC value for etridiazole was in algae (*P. subcapitata*), NOE_rC (72 hours) = 0.027 mg/l. In addition, another NOE_rC (120 hours) of 0.063 mg/l in algae (*Anabaena flos-aquae)* was reported which is also below the threshold value of 0.1 mg/l for classification.

Classification according to CLP. The DS concluded that based on the NOE_rC for algae and information that etridiazole is not rapidly degradable, the substance fulfils the CLP criteria for classification as Aquatic Chronic 1. Since the lowest NOEC value for algae is between 0.01 and ≤ 0.1 mg/l (0.027 mg/l), a chronic M-factor of 1 is assigned to etridiazole.

Classification according to DSD. The DS proposal was based on the lowest acute aquatic toxicity value (0.49 mg/l (120 hours) in aquatic algae) and its non-rapid degradation in the aquatic environment, the DS proposed that etridiazole be classified as N; R50/53 (no specific concentration limits proposed).

Comments received during public consultation
One MSCA raised several issues which were addressed in the RCOM by the DS. Most importantly, they questioned why the 120-h time point was used in defining the E_rC_{50} value rather than 72-h E_rC_{50} in defining acute toxicity to *P. subcapitata*. In the post public consultation response the DS pointed out that the coefficient of variation was too large for controls after 72 and 96-h and therefore it would be best to use the 48-h E_rC_{50} in classification for acute toxicity, since it is most in line with the current OECD guideline.

The DS informed also that the *P. subcapitata* study was revaluated. The coefficient of variation of the section by section specific growth rate of the control and solvent control is > 35%. This is mainly due to a reduced growth rate observed at 72h and 120 h. Therefore the EC_{50} and EC_{10} of the growth rate, based on linear regression, were calculated for the period between 0-48 h, resulting in a value of 0.80 and 0.33 mg/l (based on measured concentrations). These values are comparable to those calculated over the period between 0 -72 h (i.e. 0.80 and 0.22 mg/l, respectively). The DS suggested that the acute toxicity classification should be based on the 48-h EC_{50} in *P. subcapitata* as this value was considered most in line with the current OECD guideline. In addition the DS provided information that Henry's law constant for etridiazole is 3.02 Pa m^3 mol^{-1} at 25 °C. In a study (Dzialo, 1994) on the absorption/desorption of etridiazole, the K_{OC} (L/kg) values for etridiazole were 349 (sandy loam, 2.4 % OC), 195 (clay, 4.2 % OC) and 323 (silt loam, 1.6 % OC). Freundlich adsorption isotherm 1/n values were between 0.84 and 0.92.

Another MSCA agreed with the proposed classification of environmental hazards but had some editorial remarks, including proposing the removal of tables 14a and 14b from the CLH report, since they were withdrawn from the DAR by EFSA.

In response the DS noted that studies of Nag & Yu (1994), Nag & Regis (1998) and Völkel (2000) are considered valid in the DAR (see e.g. DAR addendum May 2010) although these studies have some limitations. The DS considered these results valid and did not support their withdrawal and stating unacceptability of the studies as the reason. However, the

results of the soil studies are not used for classification and labelling since sufficient valid data for the aquatic environment is available. The added value of soil degradation data in the classification decision when adequate aquatic degradation studies are available is unclear and the DS proposed that additional guidance on this be sought from RAC.

Assessment and comparison with the classification criteria

Degradation

According to the Guidance on the Application of CLP Criteria, information on hydrolysis may indicate that a substance is rapidly degradable when the longest half-life (t½) determined within the pH range 4-9 is shorter than 16 days and the degradation products are not classifiable. The available data show that the hydrolysis half-life of etridiazole at 25°C in buffered water solutions with various pH (5.2, 7.1 and 8.9) is 88 to 98 days. Thus all half-lives are well above 16 days, so etridiazole cannot be considered as rapidly degradable based on hydrolysis data.

In two water/sediment systems, treated with [3-^{14}C]-etridiazole at a concentration of 839 µg/l and incubated at 20°C in the dark, etridiazole had primary degradation in the total water/sediment system with half-lives of 1.78 d and 1.92 d. The main degradation products formed were etridiazole acid and dichloro-etridiazole. Etridiazole is primarily degraded with a half-life <16 days but the degradation product dichloro-etridiazole is classifiable (LC_{50} of 0.77 mg/l in fish). Furthermore, the CO_2 production was very low (≤3.1 % after 104 days) indicating that degradation was not ultimate. These data indicate that etridiazole undergoes fast primary degradation but with formation of a classifiable degradation product, and that the ultimate degradation is low over a 28-day period. Since the main degradation product has an acute LC_{50} below 1 mg/l, it is classifiable and therefore the parent substance cannot be considered to be rapidly degradable.

The reported screening study (a closed bottle test according to OECD TG 301D/EC C.4-E) can be considered as an adequate and conclusive study for classification. The degradation rate of etridiazole in the study after 28-days incubation was only 1 % and therefore it did not pass the CLP or DSD criteria for ready biodegradation. The reported water/sediment studies indicated very short half-life for etridiazole but with formation of classifiable metabolites and low ultimate degradation. Therefore, etridiazole should be considered as not rapidly degradable according CLP and not readily degradable according to DSD.

Bioaccumulation

The measured log K_{ow} value and the BCF value from the fish study are both considered as adequately performed and conclusive for classification although the BCF value has not been corrected for lipid content. Both the log K_{ow} of 3.37 and BCF of 165 L/kg are below the threshold values for potentially bioaccumulative substances according to CLP (i.e. log K_{ow} is below 4 and BCF is below 500) but meet the criteria for a potential bioaccumulative substance according to DSD (i.e. log K_{ow} was above 3 and BCF was above 100).

Acute aquatic hazard

Several acute toxicity studies according to different international test guidelines were included in the CLH report. Only one study on etridiazole, i.e. the test on *P. subcapitata* of E_rC_{50} (48 hours) = 0.80 mg/l, is considered as conclusive and meeting the criteria of $L(E)C_{50}$ < 1 mg/l threshold for aquatic acute hazard classification. This value was chosen after re-evaluation of the study by the DS and showed that the coefficient of variation (CV%) of the growth rate in the control cultures was more than 35% due to reduced growth rate at 72-h and 120-h. The 48-h E_rC_{50} in *P. subcapitata* is considered most in line with the current OECD guidelines. Since the EC_{50} value of acute toxicity is ≤ 1 mg/l the CLP classification criteria are met and RAC agrees that etridiazole should be classified as Aquatic Acute 1. Since the value of E_rC_{50} for algae was in the range of 0.1 – 1 mg/l, the value of M-factor should be 1.

Long-term aquatic hazard

CLP classification. Adequate and reliable chronic toxicity studies were reported for fish and crustaceans but the NOEC values in both studies were above the classification threshold value of 0.1 mg/l. RAC evaluated the validity of the reported algal studies and concluded that since both *P. subcapitata* and *A. flos-aquae* studies beyond durations of 48 hours did not meet the validity criteria of CV < 35% for section-by-section specific growth rates in controls, the studies cannot be used for direct conclusions on the classification of chronic toxicity (see details in section Supplemental information - In depth analyses by RAC). Therefore, RAC applies the surrogate approach for long-term hazard classification based on Figure 4.1.1 and Table 4.1.0 (iii) in Annex I to CLP. The lowest acute toxicity value, for the trophic level for which no conclusive chronic toxicity value is available (i.e. algae), is the reported E_rC50 (48h) of 0.80 mg/l (*P. subcapitata*). Since this value was below 1 mg/l and etridiazole was not considered to be rapidly degradable, the RAC concluded that classification as Aquatic Chronic 1 with an M factor 1 was warranted.

DSD classification. Etridiazole has a BCF above 100 and the log K_{ow} above 3 and is not considered rapidly degradable according to DSD. Since the lowest conclusive acute toxicity value (algae E_rC_{50} (48 hours) = 0.80 mg/l) is below the 1 mg/l threshold value for classification, RAC agrees that etridiazole should be classified as N; R50-53 according to DSD with the specific concentration limits N; R50-53: C ≥ 25 % N; R51-53: 2,5 % ≤ C < 25 % R52-53: 0,25 % ≤ C < 2,5 %

Conclusion

The RAC agreed with the dossier submitter's proposal to add an acute M-factor of 1 and a chronic M-factor of 1 to the existing harmonised classification in Annex VI to CLP.

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ANNEXES:

Annex 1 The Background Document (BD) provides the detailed scientific grounds for the opinion. It is based on the CLH report prepared by the dossier submitter; the evaluation performed by the RAC is contained in the RAC boxes.

Annex 2 Comments received on the CLH report, response to comments provided by the dossier submitter and the RAC (excl. confidential information).

Annex 3 Comments and response to comments received during the targeted consultation (21 May – 3 June 2013)