Committee for Risk Assessment

RAC

Opinion

proposing harmonised classification and labelling
at EU level of

propiconazole (ISO); (2*RS*,4*RS*;2*RS*,4*SR*)-1-{[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl}-1*H*-1,2,4-triazole

EC Number: 262-104-4
CAS Number: 60207-90-1

CLH-O-0000001412-86-139/F

Adopted

9 December 2016

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9 December 2016

CLH-O-0000001412-86-139/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: **propiconazole (ISO); (2*RS*,4*RS*;2*RS*,4*SR*)-1-{[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl}-1*H*-1,2,4-triazole**

EC Number: **262-104-4**

CAS Number: **60207-90-1**

The proposal was submitted by **Finland** and received by RAC on **27 November 2015.**

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

PROCESS FOR ADOPTION OF THE OPINION

Finland 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 **1 April 2016**. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by **16 May 2016**.

ADOPTION OF THE OPINION OF RAC

Rapporteur, appointed by RAC: **Miguel A. Sogorb**

Co-Rapporteur, appointed by RAC: **João Carvalho**

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

The RAC opinion on the proposed harmonised classification and labelling was adopted on **9 December 2016** 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	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	613-205-00-0	propiconazole (ISO); (2*RS*,4*RS*;2*RS*,4*SR*)-1-{[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl}-1*H*-1,2,4-triazole	262-104-4	60207-90-1	Acute Tox. 4 * Skin Sens. 1 Aquatic Acute 1 Aquatic Chronic 1	H302 H317 H400 H410	GHS07 GHS09 Wng	H302 H317 H410			
Dossier submitters proposal	613-205-00-0	propiconazole (ISO); (2*RS*,4*RS*;2*RS*,4*SR*)-1-{[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl}-1*H*-1,2,4-triazole	262-104-4	60207-90-1	**Retain** Skin Sens. 1 Aquatic Acute 1 Aquatic Chronic 1 **Add** Repr. 2 **Modify** Acute Tox. 4	**Retain** H317 H400 H410 H302 **Add** H361d	**Retain** GHS07 GHS09 Wng **Add** GHS08	**Retain** H302 H317 H410 **Add** H361d		**Add** M=1 M=1	
RAC opinion	613-205-00-0	propiconazole (ISO); (2*RS*,4*RS*;2*RS*,4*SR*)-1-{[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl}-1*H*-1,2,4-triazole	262-104-4	60207-90-1	**Retain** Skin Sens. 1 Aquatic Acute 1 Aquatic Chronic 1 **Add** Repr. 1B **Modify** Acute Tox. 4	**Retain** H317 H400 H410 H302 **Add** H360D	**Retain** GHS07 GHS09 **Add** GHS08 **Modify** Dgr	**Retain** H302 H317 H410 **Add** H360D		**Add** M=1 M=1	
Resulting Annex VI entry if agreed by COM	613-205-00-0	propiconazole (ISO); (2*RS*,4*RS*;2*RS*,4*SR*)-1-{[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl}-1*H*-1,2,4-triazole	262-104-4	60207-90-1	Repr. 1B Acute Tox. 4 Skin Sens. 1 Aquatic Acute 1 Aquatic Chronic 1	H360D H302 H317 H400 H410	GHS08 GHS07 GHS09 Dgr	H360D H302 H317 H410		M=1 M=1	

GROUNDS FOR ADOPTION OF THE OPINION

RAC general comment

Propiconazole is an active substance in the meaning of EU Regulation 1107/2009 concerning the placing of plant protection products on the market and in the meaning of EU regulation 528/2012 concerning the making available on the market and use of biocidal products. Propiconazole has the following current entry in Annex VI of CLP regulation:
Acute Toxicity 4* (H302); Skin Sensitisation 1 (H317); Aquatic Acute 1 (H400) and Aquatic Chronic 1 (H410).

The Dossier Submitter (DS) proposed the following harmonised classification, based on previous European assessments[1] and on open, peer-reviewed scientific literature:
Acute Toxicity 4 (H302); Skin Sensitisation 1 (H317); Reproductive Toxicity 2 (H361d); Aquatic Acute 1 (H400, M-factor of 1) and Aquatic Chronic 1 (H410, M-factor of 1).

The DS reviewed only the hazards acute toxicity, skin sensitisation, STOT RE, carcinogenicity and reprotoxicity in the CLH dossier but also included information about germ cell mutagenicity as supporting information for the carcinogenicity endpoint. Nonetheless, during the Public Consultation (PC) some comments addressed to germ cell mutagenicity, skin and eye corrosion/irritation were also received.

HUMAN HEALTH HAZARD EVALUATION

RAC evaluation of acute toxicity

Summary of the Dossier Submitter's proposal

The DS proposed classification of propiconazole for acute oral toxicity as Category 4 on the basis of an acute oral toxicity up-and-down test (OECD TG 425) in rat showing an LD50 of 550 mg/kg bw.

The DS proposed no classification of propiconazole for acute dermal toxicity on the basis of two independent studies in rat showing LD50 higher than 4000 and 5000 mg/kg bw and a third study in rabbit showing an LD50 higher than 6000 mg/kg bw. In the three cases the studies were performed following OECD TG 402.

A single study of acute inhalation toxicity (OECD TG 403) showing an LC50 higher than 5800 mg/m3 made the DS conclude on no classification of propiconazole for acute toxicity by inhalation route.

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[1] a) Draft Assessment Report (DAR; Finland 1998);
b) DAR Addenda (Finland, 2002);
c) Draft Renewal Assessment Report (dRAR, Finland 2015), and
d) Competent Authority Report on the Document IIA (CAR; Finland 2015).

Comments received during public consultation

Four Member State Competent Authorities (MSCAs) supported the classification proposed by the DS.

Assessment and comparison with the classification criteria

The three tables below summarise the acute oral, dermal and inhalation animal toxicity studies, respectively, that were assessed by the DS in the CLH report. No cases of poisoning of humans with propiconazole have been reported.

Table: Summary of acute oral toxicity studies with propiconazole. In all cases propiconazole was administered by gavage.

Method	Species Sex Nº group	Dose level	Results	Reference
OECD TG 425 (Up-and-Down Procedure) EPA OPPTS 870.1100 GLP	Rat RjHan:WI Females 1-3/group	175, 550, 2000 mg/kg bw	LD_{50} = 550 mg/kg bw Mortalities at 2000 (2/2) and 550 (1/3) mg/kg bw No mortality (0/1) at 175 mg/kg bw Clinical signs: decreased activity, prone position, incoordination, lateral position and hunched back were observed in both animals treated at 2000 mg/kg bw	dRAR B.6.2.1.3
Similar to OECD TG 401	Mouse Tif:MAG (SPF) 5 males and 5 females	800, 1500, 2500 or 3000 mg/kg	LD_{50} = 1490 mg/kg bw 1/10 deaths at 800 mg/kg, 4/10 deaths at 1500 mg/kg, 9/10 deaths at 2500 mg/kg, 10/10 deaths at 3000 mg/kg Clinical signs: sedation, dyspnoea, ruffled fur and lateral, ventral and/or curved body position	DAR IIA 5.2.1/02

Table: Summary of acute dermal toxicity studies with propiconazole.

Method	Species Sex Nº group	Dose level	Results	Reference
OECD TG 402 EPA OPPTS 870.1200 EC 440/2008 GLP	Rat RjHan:WI 5 males and 5 females	5000 mg/kg bw Coverage: semi-occlusive 24 hours	LD_{50} > 5000 mg/kg bw No mortalities	dRAR B.6.2.2.3
OECD TG 402	Rat Tif:RAIf (SPF)	3000, 4000 mg/kg Coverage: occlusive 24 hours	LD_{50} > 4000 mg/kg bw No mortalities Clinical signs from 2 days after dosing included dyspnoea, ruffled fur and	DAR II A 5.2.2 / 01

	5 males and 5 females		curved body position in both groups, with full recovery within 9 days.	
Similar to OECD TG 402	Rabbit New Zealand White 3 males and 3 females	0, 2000, 6000 mg/kg bw Coverage: occlusive 24 hours	LD_{50} > 6000 mg/kg bw No mortalities No clinical signs or effect on body weight. No abnormal findings were observed at necropsy.	DAR II A 5.2.2/02

Table: Summary of acute inhalation toxicity studies with propiconazole. The assay was performed with propiconazole of 91.1% purity.

Method	Species Sex Nº group	Dose level	Results	Reference
OECD TG 403 GLP	Rat Tif:RAIf (SPF) 5 males and 5 females	0, 5836 ± 186 mg/m3 Inhalation: Aerosol (nose only) 4 hours	LC50 (4 h): > 5800 mg/m³ No mortalities Signs of systemic toxicity (ruffled fur, dyspnoea, abnormal body positions and reduced activity) were seen in control and, with a greater severity, in test animals.	DAR IIA 5.2.3/01

Comparison with the criteria

The acute oral test in mouse yielded an LD_{50} of 1490 mg/kg bw. However, rat was noted to be the most sensitive species with a LD_{50} of 550 mg/kg bw. The classification should be based on the most appropriate sensitive species tested and in this case the LD_{50} for rat is within the limits $300 < LD_{50} \leq 2000$ mg/kg bw. Therefore, RAC is in agreement with the DS, and concludes on classification of propiconazole as **Acute Oral Toxicity Category 4 (H302: Harmful if swallowed).**

The limit concentration for triggering classification for the dermal route is 2000 mg/kg. The available information shows that doses of up to 4000-6000 mg/kg bw of propiconazole did not cause fatalities. Thus, RAC agrees with the DS that propiconazole **does not fulfil the criteria for classification for dermal acute toxicity.**

The limit concentration for triggering classification for the inhalation route is 5000 mg/m^3. The available information shows that a dose of 5800 mg/m^3 of propiconazole did not cause fatalities. Thus, RAC agrees with the DS that propiconazole **does not fulfil the criteria for classification for inhalation acute toxicity.**

RAC evaluation of skin corrosion/irritation

Summary of the Dossier Submitter's proposal

This hazard was not reviewed in the CLH report.

Comments received during public consultation

One MSCA submitted a comment indicating that this hazard should be considered on the basis of a relevant FAO/WHO assessment [1]. The DS replied that this hazard had not been evaluated in the CLH dossier.

Assessment and comparison with the classification criteria

RAC can only form opinions on hazard classes that have been proposed for review in the CLH dossier and which proposal has been subject to public consultation. Therefore, RAC did not evaluate this hazard class.

RAC evaluation of serious eye damage/irritation

Summary of the Dossier Submitter's proposal

This hazard was not reviewed in the CLH report.

Comments received during public consultation

One MSCA submitted a comment indicating that this hazard should be considered on the basis of a relevant FAO/WHO assessment [2]. The DS replied that this hazard had not been evaluated in the CLH dossier.

Assessment and comparison with the classification criteria

RAC can only form opinions on hazard classes that have been proposed for review in the CLH dossier and which proposal has been subject to public consultation. Therefore, RAC did not evaluate this hazard class.

RAC evaluation of skin sensitisation

Summary of the Dossier Submitter's proposal

The DS proposed to retain the current classification in Annex VI of propiconazole as skin sensitizer Category 1 (H317) because the available data (an OECD TG 406 study showing 30% and 50% of sensitisation 24 hours and 48 hours after challenge with 30% propiconazole and a few reports of humans where exposure to propiconazole caused skin reactions) does not warrant revision of the assigned classification.

[1] FAO Plant Production and Protection Paper, 178, 2004 - Pesticide residues in food – 2004 (Report of the Joint Meeting of the FAO Panel of Experts on Pesticide Residues in Food and the Environment and WHO the Core Assessment Group)

[2] FAO Plant Production and Protection Paper, 178, 2004 - Pesticide residues in food – 2004 (Report of the Joint Meeting of the FAO Panel of Experts on Pesticide Residues in Food and the Environment and WHO the Core Assessment Group)

Comments received during public consultation

Three MSCAs supported the classification proposed by the DS.

A manufacturer commented agreeing with the proposal to retain Category 1. However, the company submitted a new study where an intradermal injection induction dose of 1% propiconazole caused 0% of sensitisation and therefore would allow discarding category 1A and instead specify the subcategory leading to Category 1B. The DS assessed the new study and noted that in the induction phase skin reactions observed in control animals were similar to the test animals, while 24 hours after challenge one control animal (that received only vehicle) showed significant dermal response. The DS considered the new study not acceptable for classification purposes because of the non-specific positive reactions in vehicle control animals. RAC also noted that no positive controls were included in this new study and therefore negative results might be interpreted as an intrinsic resistance of the animals to sensitisation.

Assessment and comparison with the classification criteria

The table below summarises the available animal studies with propiconazole.

Table: Summary of animal studies on skin sensitisation with propiconazole.

Method	Species Strain Nº/group	Dose levels	Results Conclusion	Reference Acceptability
Guinea pig maximisation test OECD TG 406 GLP	Guinea pig Himalayan Spotted (GOHI Ibm:GOHI (SPF)) 10 animals/sex/ test group 5 animals/sex/ vehicle control group	Day 0: Induction: intradermal 5% propiconazole, in peanut oil Day 8: Induction: 100% propiconazole (or vaseline) occlusive for 48 hours Day 21: Challenge: 30% propiconazole (or vaseline) occlusive for 24 hours	Test group: 6/20 (24 h after challenge) 10/20 (48 h after challenge) Vehicle control: 0/10 (24 h after challenge) 0/10 (48 h after challenge) **SENSITISING**	DAR IIA 5.2.6 **Acceptable**
Optimization test Similar to the method recommended in the "Appraisal of the Safety of Chemicals in Foods, Drugs and Cosmetics" (1959), the US Association of Food and Drug Officials (AFDO). non-GLP	Guinea pig Pirbright White 10 animals/sex/ group	Day 0: Induction: intradermal injections 0.1% propiconazole in propylene glycol Day 10: Challenge: 10% propiconazole occlusive dressing in vaseline (24 hours)	Test group: 2/20 (after intradermal challenge) 3/19 (24 h after epidermal challenge) Vehicle control group: 4/19 (after intradermal challenge) 0/18 (48 h after challenge) **INCONCLUSIVE**	DAR IIA 5.2.6/01 **Not acceptable**

Comparison with the criteria

A preliminary test showed that an intradermal dose of 5.0% propiconazole caused mild to moderate irritation at the injection site with no clinical signs. The same preliminary study also showed that dermal application of 100% and 30% propiconazole caused mild to moderate irritation and no irritation, respectively. The main test was performed using intradermal induction with 5.0% propiconazole and a first epidermal occlusive challenge with 100% propiconazole 8 days after induction and a second epidermal occlusive challenge with 30% propiconazole. These conditions caused positive skin reactions in 2 males and 4 females of the test group animals 24 h after completion of the application, and in 5 males and 5 females at the 48 h examination. Therefore the rates of sensitization 24 and 48 hours after induction were 30 and 50%, respectively. No reactions were observed in animals treated with vehicle control. There were no mortalities during the test, and no remarkable clinical observations were reported. Body weights of the test animals were not affected by treatment.

A second skin sensitisation study by using an optimization test was available. This optimization test was considered to be acceptable according to an earlier version of OECD TG 406, but not according to the present OECD test guideline or according to directive 92/69/EEC B.6. Regardless of the employed method, two additional reasons for disregarding the results of this study were that the sensitivity of the strain of Guinea pig employed in the test has not been checked and that two vehicle control animals and one animal in the test group died during the study. In this optimization test the induction was performed with 10 intracutaneous injections of a 0.1% dilution of propiconazole in propylene glycol. During the second and third weeks of the induction period the test material was incorporated in a mixture of vehicle with complete Bacto adjuvant (complete Freund). After a two week long treatment free period one intracutaneous injection of the test dilution (0.1% propiconazole in propylene glycol) was given as first challenge. Ten days after the intracutaneous challenge injection a sub-irritant dose of the test compound (10% in vaseline) was applied epicutaneously under an occlusive dressing which was left in place for 24 hours. The incidence of positive animals immediately after and 24 hours after the intradermal challenge injection was 16 and 16%, respectively; while an incidence of 21% in the vehicle control group was found immediately after challenge. Thus, this study was considered inconclusive and non-acceptable for classification purposes.

There is information on a few cases in humans where exposure to propiconazole has caused skin reactions: i) Medical surveillance of employees in production, formulation and packaging plants revealed 4 cases of local skin reactions among 139 exposed individuals during the period 1982-2000; ii) A few cases where chest pain and local skin reactions have been experienced when exposed via inhalation or skin contact to a product containing propiconazole have been reported; iii) One case of allergic contact dermatitis diagnosed through patch testing with 3.07 mg propiconazole/mL among 60 individuals. In contrast to the information suggesting certain capability of propiconazole to induce sensitisation in humans an epicutaneous test with 1% technical grade propiconazole conducted with 20 human volunteers gave no evidence of sensitisation or skin irritation.

Overall, the human data does not give clear evidence of a potential skin sensitising effect of propiconazole, while the valid Guinea pig maximisation test showed that an intradermal induction of 5% propiconazole sensitised 50% of animals 48 hours after challenge. This response is within the range required in the ECHA "Guidance on the Application of the CLP Criteria" for classifying propiconazole as Category 1B (because the response was higher than ≥ 30% with an intradermal induction dose higher than 1%). However, RAC notes that induction was not tested at concentrations of 1% and lower and therefore it is unknown if the response at 1% would have been higher than 60% (criteria requested for classification as Category 1A). Therefore, category

1A cannot be excluded with the available information and RAC supports the DS's opinion and concludes on classification of propiconazole a **Skin Sensitizer Category 1 (without sub-categorisation); H317 (May cause an allergic skin reaction).**

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

Summary of the Dossier Submitter's proposal

The DS assessed sub-chronic toxicity studies of propiconazole in rat (oral, dermal and inhalation routes), rabbit (dermal route) and mouse (oral route). The DS concluded that liver is the main target organ of propiconazole, inducing weight increases, hepatocellular hypertrophy and, to a minor extension, necrosis and vacuolation. This hepatocellular toxicity was also accompanied with alterations in clinical chemistry. However, the DS did not consider the findings of toxicological significance and did not propose propiconazole to be classified for STOT RE.

Comments received during public consultation

One MSCA supported the 'no classification' proposed by the DS.

Assessment and comparison with the classification criteria

The three tables below summarises the main relevant findings in the repeated toxicity studies with propiconazole after oral, inhalation and dermal exposure, respectively.

Table: Summary of the repeated toxicity studies by oral route with propiconazole.

Method	Species Strain Nº/group Dose levels	Results	Reference
Repeated dose 28-day oral toxicity study in rodents Equivalent or similar to OECD TG 407 Gavage	Rat Tif:RAIf (SPF) 10 males and 10 females /group 0, 50, 150, 450 mg/kg bw/d	<u>50 mg/kg bw/d:</u> No histopathological changes Females: Absolute liver weight, liver weight/body weight ratio and liver weight/brain weight ratio 135%, 125% and 129% of control, respectively <u>150 mg/kg bw/d:</u> Minimal hypertrophy of hepatocytes (8/10 females and 4/10 males) Small focus organising necrosis in liver parenchyma (1/10 males) Females: Absolute liver weight, liver weight/body weight ratio and liver weight/brain weight ratio 158%, 144% and 152% of control, respectively Males: Absolute liver weight, liver weight/body weight ratio and liver weight/brain weight ratio 132%, 130% and 136% of control, respectively <u>450 mg/kg bw/d:</u>	DAR II A 5.3.1/01

		Minimal/moderate hypertrophy of hepatocytes (10/10 males and 10/10 females) Multiple recent areas of necrosis in liver parenchyma (3/10 females) Females: Absolute liver weight, liver weight/body weight ratio and liver weight/brain weight ratio 166%, 171% and 159% of control, respectively Males: Absolute liver weight, liver weight/body weight ratio and liver weight/brain weight ratio 137%, 148% and 146% of control, respectively **NOAEL males = 50 mg/kg bw/d** **LOAEL females = 50 mg/kg bw/d**	
Repeated dose 90-day oral toxicity study in rodents Equivalent or similar to OECD TG 408 Oral (diet)	Rat Tif:RAIf (SPF) 20 males and 20 females /group 0, 240, 1200 and 6000 ppm (nominal in diet) Males: 0, 15.9, 76.1, 461.7 mg/kg bw/d Females: 0, 16.8, 77.6, 480.9 mg/kg bw/d	<u>240 ppm:</u> No effects <u>1200 ppm:</u> Body weight: ↓ both sexes (males 2.4%, 4.7% and 2.5% at weeks 2, 4 and 12; females 1.8%, 4.9% and 7.4% at weeks 2, 4 and 12, respectively) <u>6000 ppm:</u> Body weight: ↓ both sexes (males 13.0%, 16.8% and 21.7% at weeks 2, 4 and 12; females 8.2%, 10.6%, 18.7% at weeks 2, 4 and 12, respectively) Histopathology: slightly ↑ haemosiderosis in spleen of 20/20 females **NOAEL = 240 ppm (males 15.9 and females 16.8 mg/kg bw/d)**	DAR II A 5.3.2/01
2-year chronic toxicity/ carcinogenicity study OECD TG 453 (1981) GLP Oral (diet)	Rat Sprague Dawley CD 80 males and 80 females 0, 100, 500, 2500 ppm. Males: 0, 3.60, 18.10, 96.46 mg/kg bw/d Females: 0, 4.57, 23.32, 130.63 mg/kg bw/d	<u>100 ppm:</u> No effects <u>500 ppm:</u> Reduced body weight gain and food utilization in females and lower adrenal weights in males Transient alterations in clinical chemistry <u>2500 ppm:</u> Reduced body weight gain and food consumption in both sexes Increased liver weights in both sexes (16% males, 19% females) and increased incidence of foci of enlarged liver cells in females (13/71 vs 1/70 in control)	DAR IIA 5.5/02

<table>
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<td></td>
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<td>Lower kidney weight and adrenal weight (with no histopathological associated changes)

Transient alterations in clinical chemistry.

NOAEL = 100 ppm (males 3.60 and females 4.57 mg/kg bw/d)</td>
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<tr>
<td>Repeated dose 90-day oral toxicity study in rodents

Equivalent or similar to EPA OPP 82-1/ OECD TG 408

GLP

Oral (diet)</td>
<td>Mouse

Crl:CD-1®(ICR) BR (Swiss)

20 males and 20 females /group

0, 20, 500, 850 (males only), 1450 (males only), 2500 ppm

Males: 0, 2.7, 65, 112, 194, 352 mg/kg bw/d

Females: 0, 3.1, 85, 434 mg/kg bw/d</td>
<td><u>Hepatic effects in males:</u>
<table>
<tr><td></td><td colspan="4">Dose (ppm)</td></tr>
<tr><td></td><td>500</td><td>850</td><td>1450</td><td>2500</td></tr>
<tr><td>Absolute and relative weight</td><td>↑</td><td>↑</td><td>↑</td><td>↑</td></tr>
<tr><td>Enlarged</td><td>0</td><td>0</td><td>14</td><td>20</td></tr>
<tr><td>Focal discoloration</td><td>0</td><td>2</td><td>5</td><td>6</td></tr>
<tr><td>Hypertrophy</td><td>4</td><td>14</td><td>20</td><td>20</td></tr>
<tr><td>Necrosis</td><td>2</td><td>4</td><td>8</td><td>12</td></tr>
<tr><td>Necrosis of individual cells</td><td>0</td><td>0</td><td>2</td><td>12</td></tr>
<tr><td>Necrosis of multi and/or individual cells</td><td>2</td><td>4</td><td>10</td><td>18</td></tr>
<tr><td>Vacuolation</td><td>6</td><td>2</td><td>3</td><td>10</td></tr>
<tr><td>Vacuolation of individual cells</td><td>0</td><td>0</td><td>0</td><td>6</td></tr>
<tr><td>Vacuolation multi and/or individual cells</td><td>6</td><td>2</td><td>2</td><td>16</td></tr>
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<u>Hepatic effects in females:</u>
<table>
<tr><td></td><td colspan="2">Dose</td></tr>
<tr><td></td><td>500 ppm</td><td>2500 ppm</td></tr>
<tr><td>Absolute and relative weight</td><td></td><td>↑</td></tr>
<tr><td>Enlarged</td><td>0</td><td>8</td></tr>
<tr><td>Focal discoloration</td><td>0</td><td>3</td></tr>
<tr><td>Hypertrophy</td><td>0</td><td>17</td></tr>
<tr><td>Necrosis</td><td>0</td><td>6</td></tr>
<tr><td>Necrosis of individual cells</td><td>0</td><td>1</td></tr>
<tr><td>Necrosis of multi and/or individual cells</td><td>0</td><td>6</td></tr>
<tr><td>Vacuolation</td><td>0</td><td>2</td></tr>
<tr><td>Vacuolation of individual cells</td><td>0</td><td>1</td></tr>
<tr><td>Vacuolation of multi and/or individual cells</td><td>0</td><td>3</td></tr>
</table>
Inconsistent clinical changes in males

Increased ALT and AST at 2500 ppm in females (severity not reported)

NOAEL males = 20 ppm (2.7 mg/kg bw/d)</td>
<td>DAR II A 5.3.2/03</td>
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<tr><td></td><td></td><td>NOAEL females = 500 ppm (85 mg/kg bw/d)</td><td></td></tr>
<tr><td>Repeated dose 90-day oral toxicity study in rodents

Equivalent or similar to EPA OPP 82-1/ OECD TG 408

GLP

Oral (diet)</td><td>Mouse

Crl:CD-1®(ICR) BR (Swiss)

40 males /group

0, 20, 500, 850, 1450 and 2500 ppm

0, 2.8, 71, 121, 199 and 360 mg/kg bw/d</td><td>Clinical chemistry alterations at 500 ppm and onwards (severity not reported)

<table><tr><td></td><td colspan="4">Dose (ppm)</td></tr><tr><td></td><td>500</td><td>850</td><td>1450</td><td>2500</td></tr><tr><td>Absolute and relative weight</td><td>↑</td><td>↑</td><td>↑</td><td>↑</td></tr><tr><td>Enlarged</td><td>0</td><td>14</td><td>21</td><td>40</td></tr><tr><td>Focal discoloration</td><td>0</td><td>6</td><td>8</td><td>8</td></tr><tr><td>Prominent lobular architecture</td><td>0</td><td>0</td><td>6</td><td>18</td></tr><tr><td>Hypertrophy</td><td>10</td><td>35</td><td>40</td><td>40</td></tr><tr><td>Necrosis</td><td>4</td><td>8</td><td>16</td><td>17</td></tr><tr><td>Necrosis of individual cells</td><td>0</td><td>1</td><td>23</td><td>29</td></tr><tr><td>Necrosis of multi and/or individual cells</td><td>4</td><td>9</td><td>31</td><td>34</td></tr><tr><td>Vacuolation</td><td>2</td><td>5</td><td>11</td><td>22</td></tr><tr><td>Vacuolation of individual cells</td><td>0</td><td>0</td><td>4</td><td>11</td></tr><tr><td>Vacuolation multi and/or individual cells</td><td>2</td><td>5</td><td>15</td><td>33</td></tr><tr><td>Mineralization</td><td>0</td><td>3</td><td>2</td><td>9</td></tr></table>
NOAEL = 20 ppm (2.8 mg/kg bw/d)</td><td>DAR IIA 5.3.2/04</td></tr>
<tr><td>2-year carcinogenicity study
OECD TG 451

GLP

Oral (diet)</td><td>Mouse

CD-1

64 males and 64 females

0, 100, 500 and 2500 ppm

Males: 0, 10, 49, 344 mg/kg bw/d

Females: 0, 11, 56, 340 mg/kg bw/d</td><td><u>500 ppm:</u>
Increased (123% of controls) interim absolute and relative liver weights (males)

Incidences of hepatocyte enlargement in 39/62 males

<u>2500 ppm:</u>

Reduced body weights (11-16%) and weight gain (20-38%) in both sexes

Increased terminal absolute (227% of controls) and relative (263% of control) liver weights (males)

Increased terminal absolute (151% of controls) and relative (175% of control) liver weights (males)

Incidences of hepatocyte enlargement (54/64 males and 43/64 females)

Hepatocyte vacuolation (39/64 females)

Inflammatory cell and chronic infiltration in 44/64 males</td><td>DAR IIA 5.5/03

DAR IIA 5.5/04</td></tr>
</table>

		Pigmented Kupffer cells in 44/64 males ALT, AST and ALP increases and cholesterol reductions **NOAEL (non-carcinogenic) = 100 ppm (males 10.04 mg/kg bw/d, males and females 10.79 mg/kg bw/d)**	
18-month study in CD-1 male mice OECD TG 451 GLP Oral (diet)	Mice CD-l (ICR) BR 80 males/group 0, 100, 500 and 850 ppm 0, 11, 59, 108 mg/kg bw/d	<u>500 ppm:</u> Reductions of cumulative mean body weight gain between weeks 13-50 (6.9-8.8%), but no significant changes in mean body weight or body weight gains beyond the first year Liver weight/body weight was increased by 13% at week 53 Hepatocellular hypertrophy in 6/10 animals by week 9 and in 28/50 animals at the end of the study Transient decreases in cholesterol levels <u>850 ppm:</u> Reductions of cumulative mean body weight gain between weeks 13-50 (5-15%), but no significant changes in mean body weight or body weight gains beyond the first year Liver weight/body weight was increased by 33% at week 9, by 29% at week 53 and by 20% at the end of the study Absolute liver weight was increased by 32% at week 9, by 11% at week 53 and by 19% at the end of the study Hepatocellular hypertrophy in 10/10 animals by week 9, in 8/10 by week 53 and in 29/50 animals at the end of the study Kupffer cell pigmentation in 11/50 animals at the end of the study Transient decreases in cholesterol and increases in sorbitol dehydrogenase activity levels **NOAEL (non-carcinogenic) = 100 ppm (11.0 mg/kg bw/d)**	DAR IIA 5.5/05
92-day study in dogs Equivalent or similar to OECD TG 409 Oral (diet)	Dog Beagle 4 males and 4 females /group 0, 50, 250 and 1250 ppm	No treatment related effects at any dose **NOAEL > 1250 ppm (35.28 mg/kg bw/d in males and 35.74 mg/kg bw/d in females)**	DAR IIA 5.3.2/02

	Males: 0, 1.34, 6.89 and 35.28 mg/kg bw/d Females: 0, 1.65, 7.56 and 35.74 mg/kg bw/d		
53-week study in dogs Equivalent or similar to OECD TG 452 Oral (diet)	Dog Beagle 5 males and 5 females /group 0, 5, 50, 250 ppm Males: 0, 0.17, 1.86, 8.43 mg/kg bw/d Females: 0, 0.19, 1.86, 8.86 mg/kg bw/d	No treatment related effects at any dose **NOAEL > 250 ppm (8.43 mg/kg bw/d in males and 8.86 mg/kg bw/d in females)**	DAR IIA 5.5/01
Two-generation reproduction study Draft OECD TG 418 Equivalent or similar to OECD TG 416 GLP Oral (diet)	Rat Charles River CD strain 15 males and 30 females 0, 100, 500 and 2500 ppm Males: 0, 8.4, 48.8 and 214.9 mg/kg bw/d Females: 0, 9.7, 43.7 and 242.9 mg/kg bw/d	<u>500 ppm:</u> Liver hypertrophy in males F_0 (13/15) and F_1 (5/15) Liver hypertrophy in females F_1 (15/30) Liver vacuolation in F_1 (8/15) males <u>2500 ppm:</u> ↓23% and ↓19% of total bodyweight gain in F_0 and F_1 parental females at the end of the study Severe incidence of liver hypertrophy in males F_0 (14/15), F_1 (15/15), F_{1b} (10/10), F_{2b} (10/10) Severe incidence of liver hypertrophy in females F_0 (29/30), F_1 (29/30), F_{1b} (8/10), F_{2b} (9/10) Severe incidence of liver vacuolation in F_0 (14/15) and F_1 (11/15) males and in F_1 (10/30) females **NOAEL parental toxicity: 100 ppm (8.4 mg/kg bw/d males and 9.7 mg/kg bw/d females)**	DAR IIA 5.6.1/01
Developmental toxicity OECD TG 414 GLP	Rat Crl:COBS CD (SD) BR VAF/PLUS) 30 females	<u>90 mg/kg bw/d:</u> Transient reduction in maternal food consumption and body weight gain but no significant differences by day 20	DAR IIA 5.6.2/01

Gavage Daily treatment on days 6-15 of gestation	0, 30, 90, 360/300 mg/kg bw/d The high dose was reduced to 300 mg/kg bw/d due to severe signs of maternal toxicity	<u>360 mg/kg bw/d:</u> During the first week of treatment: lethargy, ataxia and salivation, and signs of rales, prostration, hypothermia and bradypnea. The toxic signs decreased immediately following the lowering of the dose level to 300 mg/kg bw/d on the sixth day of dosing Transient reduction in maternal food consumption and body weight gain but no significant differences by day 20 **NOAEL maternal toxicity: 90 mg/kg bw/d**	
Teratology study Modified OECD TG 414 GLP Gavage Daily treatment on days 6-15 of gestation	Rat Crl:COBS CD (SD) BR VAF/PLUS 0, 300 mg/kg bw/d	<u>300 mg/kg bw/d:</u> ↓17% corrected maternal body weight gain in the period 0-20 days Clinical signs: ataxia, comatose, lethargy, prostration, salivation, altered respiration 2 deaths Transient (days 6-16) decrease in maternal food consumption but similar to control in periods 0-6 and 16-20 days) **LOAEL maternal toxicity: 300 mg/kg bw/d**	DAR IIA 5.6.2/02 Acceptable
Teratology study OECD TG 414/EPA OPP 83-3 GLP Gavage Daily treatment on days 7-19 of gestation	Rabbit New Zealand White 19 females 0, 100, 250, 400 mg/kg bw/d	<u>100 mg/kg bw/d:</u> One death for unknown cause <u>250 mg/kg bw/d:</u> Maternal food consumption reduced between 24-37% in the period between 7-21 days of gestation but not in the period between days 5-6 and 20-29 <u>400 mg/kg bw/d:</u> Maternal food consumption reduced between 34-57% in the period between 7-21 days of gestation but not in the period between days 5-6 and 20-29 Maternal bodyweight gain reduced by 89% in the period between 10-14 days of gestation and by 56% in the period between days 14-20 of gestation Increased incidence of stool variations (18/19 versus 11/19 in controls) Significant reductions in food consumption during the dosing period and increase afterwards until sacrifice **NOAEL maternal toxicity: 100 mg/kg bw/d**	DAR IIA 5.6.2/03

Table: Summary of the repeated toxicity studies by inhalation route with propiconazole.			
Method **Exposure**	**Species** **Strain** **Nº/group** **Dose levels**	**Results**	**Reference**
90-day, subchronic inhalation toxicity study Equivalent or similar to OECD TG 413 6 hours/day 5 days/week	Rat RAIf (SPF) 20 males and 20 females/group 0 (air), 10 mg/m^3 acetone (vehicle control), 21±2, 85±7 or 191±10 mg/m^3 (mean achieved concentration) Nose/head only	<u>21 mg/m^3:</u> Body weight: ↓ by 5.9%, 7.3% and 5.3% females at weeks 4, 8 and 12 respectively <u>85 mg/m^3:</u> Body weight: ↓ by 2.2%, 6.3%, 6.5% and 3.5% in males at weeks 1, 4, 8 and 12 respectively; 4.5% and 4.1% in females at weeks 4 and 8 respectively <u>191 mg/m^3:</u> Body weight: ↓ by 4.8%, 5.3%, 3.5% and 0.5% in males at weeks 1, 4, 8 and 12 respectively; 1.1%, 5.9%, 7.3% and 5.3% in females at weeks 1, 4, 8 and 12 respectively **NOAEC males: 21 mg/m^3** **LOAEC females: 21 mg/m^3**	DAR II A 5.3.3/02

Table: Summary of the repeated toxicity studies by dermal route with propiconazole.			
Method **Exposure**	**Species** **Strain** **Nº/group** **Dose levels**	**Results**	**Reference**
21-day study Equivalent or similar to OECD TG 410 Shaved skin of the back covered with a gauze and occlusive dressing intact and abraded skin 6 hours/day 5 days/week	Rabbit New Zealand White 10 males and 10 females/group 0, 200, 1000 and 5000 mg/kg bw/d	<u>200 mg/kg bw/d:</u> Slight skin irritation (no differences between intact and abraded skin) <u>1000 mg/kg bw/d:</u> Slight skin irritation (no differences between intact and abraded skin) Clinical observations: From day 4: sedation (10/10 males, 10/10 females). Days 15-21: ruffled fur, tremor, dyspnoea and diarrhoea (number of animals affected not reported) Histopathology: moderate acanthosis and hyperkeratosis of epidermis, chronic inflammatory infiltration in dermis (4/10 males, 4/10 females). Minimal or slight focal acanthosis of epidermis, slight chronic infiltration in dermis (5/10 males) <u>5000 mg/kg bw/d:</u> Slight skin irritation (no differences between intact and abraded skin)	DAR IIA 5.3.3/01

		Clinical observations: From day 4: ruffled fur (10/10 males, 10/10 females), dyspnoea (10/10 males, 10/10 females), tremor (10/10 males, 10/10 females), ataxia (10/10 males, 10/10 females) and sedation (10/10 males, 10/10 females) Body weight: ↓ by 8.6%, 8.7% and 12.5% on days 12, 15 and 19, respectively, in females Histopathology: marked acanthosis and hyperkeratosis of epidermis, chronic inflammatory infiltration and focal fibrosis in dermis (6/10 males, 9/10 females). Necrosis of epithelium, marked acanthosis and hyperkeratosis of epidermis, focal fibrosis and chronic infiltration in dermis (3/10 males, 1/10 females). Minimal or slight focal acanthosis of epidermis, slight chronic infiltration in dermis (1/10 males) **NOAEL systemic effects = 200 mg/kg bw/d** **LOAEL systemic effects = 1000 mg/kg bw/d**	
Equivalent or similar to OECD TG 410 GLP Shaved skin of the back covered with an occlusive dressing 4 weeks Weeks 1-3: 5 days/week Week 4: 7 days/week	Rat Hanlbm:WIST (SPF) 10 males and 10 females/group 0, 10, 100, 1000 mg/kg bw/d	<u>10 mg/kg bw/d:</u> No adverse effects observed <u>100 mg/kg bw/d:</u> No adverse effects observed <u>1000 mg/kg bw/d:</u> Blood clinical chemistry: protein ↑ 5.6% (F), globulin ↑ 8.2% (F), A/G ratio ↓ 4.5% (F), cholesterol ↑ 28.6% (F), chloride ↓ 2.3% (F) Organ weights: absolute liver weight ↑ 18.9% (M), 14.0% (F), liver weight relative to bw ↑15.1% (M), 9.5% (F) Histopathology: acanthosis (minimal) at application site (8/10 F compared to 1/10 F controls). **NOAEL systemic effects = 100 mg/kg bw/d** **LOAEL systemic effects = 1000 mg/kg bw/d**	dRAR B.6.3.3.1.2

Comparison with the criteria

The three tables above summarise the available information regarding the toxicity exerted by propiconazole after repeated exposures. The vast database contains information about sub-acute toxicity in rat by oral route and in rabbit by dermal inhalation route, sub-chronic toxicity in rat and mouse by oral route, sub-chronic toxicity in rat by inhalation route, sub-chronic and chronic toxicity in dogs by oral route, chronic toxicity in rat and mouse by oral route, a 2-generation reproduction study in rat and three developmental toxicity studies (two in rat and one in rabbit) and draws a toxicity profile where the main target organ is liver, with clinical alterations probably associated to hepatotoxicity, with reductions in bodyweight gain and with clinical symptoms at certain high exposures.

The clinical sings were reported in the teratology studies at 300 and 360 mg/kg bw/d in rat and in the subacute dermal toxicity in rabbit at doses of 1000 and 5000 mg/kg bw/d. RAC notes that Annex 1: 3.9.2.8 of the "Guidance on the Application of the CLP Criteria" excludes clinical observations that have toxicological importance but that do not, by themselves, indicate significant toxicity as effects considered for supporting classification as STOT RE. In addition, it is also remarkable that: i) the effects in teratology studies appear after exposures in the same order of magnitude of the reported LD_{50} and therefore a potential classification based on these clinical effects would cause a double-classification considering the Acute Toxicity Category 4; ii) the effects in sub-chronic dermal studies appear well above the limit for classification as STOT RE category 2 (600 mg/kg bw/d). In conclusion, **RAC does not consider that the clinical effects fulfil the criteria for classification as STOT RE.**

Several studies reported alterations in clinical chemistry that were likely associated to the hepatotoxicity. These alterations were of minor toxicological significance or appeared at concentrations above the limits for warranting classification as STOT RE 2. In addition, Annex 1: 3.9.2.8 of the "Guidance on the Application of the CLP Criteria" describes small changes in clinical biochemistry as example of effects that do not support classification as STOT RE. Therefore, in the case of propiconazole RAC does not consider the changes in clinical chemistry seen relevant for STOT RE classification.

Reductions in body weight were reported very often in the assessed studies (see the three tables above). The table below summarises the studies reporting these reductions in body weight and the lowest exposure for which such reduction was reported.

Table: Reductions in bodyweight reported after repeated exposures to propiconazole. Data were taken from the studies summarised in the three tables above. Bold text refers to those effects that appear at doses relevant for classification as STOT RE.

Study	Lowest reported dose (mg/kg bw/d) (except in inhalation studies)	Guidance value for STOT RE classification (mg/kg bw/d) (except in inhalation studies)
90-day oral toxicity in rat	460-481	10 ≤ C ≤ 100
2-year chronic toxicity in rat	18-23	1.25 ≤ C ≤ 12.5
2-year chronic toxicity in mouse	344-340	1.25 ≤ C ≤ 12.5
2-generation reproduction toxicity	214-243	4 ≤ C ≤ 40 (assuming 32 weeks of exposure in F_0)
Developmental toxicity in rats	**300**	**90 ≤ C ≤ 900**
Developmental toxicity in rabbits	**250**	**50 ≤ C ≤ 500**

90-day inhalation toxicity in rat	**21 mg/m³**	**20 ≤ C ≤ 200 mg/m³**
Sub-acute dermal toxicity in rat	1000-5000	60 ≤ C ≤ 600

The reductions in bodyweight found in the developmental toxicity in rats were reported concurrently with severe clinical signs and at doses in the same order of magnitude as the LD_{50} and are therefore not relevant for classification purposes. The reductions in the bodyweight in the developmental study in rabbits appear at concentration within the range of the classification criteria for Category 2. However, in this study the reductions in body weight were seen during the administration period and there were no statistically significant differences between the corrected body weight of all groups at the end of the study. The effects reported in the sub-chronic inhalation study could also be potentially relevant for classification in Category 2. However, in this study the effects in body weight were of relatively low severity and not dose-related, which significantly limits their toxicological importance. In addition, the already above stated Annex 1: 3.9.2.8 of the "Guidance on the Application of the CLP Criteria" recognises small changes in bodyweight gain as effects that do not fulfil the criteria for STOT RE classification. Therefore, RAC does not consider these changes in bodyweight as relevant for STOT RE classification of propiconazole.

Hepatotoxicity was consistently noted in most of the repeated toxicity studies. The main reported hepatic effects were liver weight increases, hypertrophy, vacuolation, necrosis and mineralization. The table below summarises the studies reporting hepatotoxicity and the lowest exposure for which such reduction was reported.

Table: Hepatotoxicity reported after repeated exposures to propiconazole. Bold text refers to those effects that appear at doses relevant for classification as STOT RE.

Study	Lowest reported dose (mg/kg bw/d)	Guidance value for STOT RE classification (mg/kg bw/d)
28-day oral toxicity in rat	**150**	**30 ≤ C ≤ 300**
90-day oral toxicity in rat	461/481	10 ≤ C ≤ 100
90-day oral toxicity in mouse (2 studies)	**65-71-85**	**10 ≤ C ≤ 100**
2-year oral toxicity in rat	96-130	1.25 ≤ C ≤ 12.5
2-year oral toxicity in mouse	49-55	1.25 ≤ C ≤ 12.5
18-month oral toxicity in mouse	59	2.2 ≤ C ≤ 22
2-generation reproduction toxicity in rat	**44-49**	**4 ≤ C ≤ 40 (assuming 32 weeks of exposure in F_0)**

Four of the different studies (90-day and 2-year oral toxicity studies in rat, and 2-year and 18-month oral toxicity studies in mouse) showed that the hepatotoxicity appeared at doses well above the respective limit dose for warranting classification as STOT RE 2. Therefore RAC does not consider these studies relevant for classification. However, the other three studies (28-day in rat, 90-day oral toxicity studies in mice and 2-generation reproduction toxicity in rat) showed hepatotoxicity at doses either warranting classification as STOT RE Category 2 or on the border for classification.

Annex 1: 3.9.2.8 of the "Guidance on the Application of the CLP Criteria" states that adverse effects relevant for classification would have to be toxicologically significant and affect the function or morphology of a tissue/organ. However, the effects reported at 150 mg propiconazole/mg bw/d in the 28-day oral toxicity study in rat were described as minimal hypertrophy of hepatocytes and small focus organising necrosis in liver parenchyma, while the

dose showing moderate hypertrophy and multiple areas of necrosis was 450 mg/kg bw/d, and hence above the limit for classification.

The two different 90-day oral toxicity studies in mouse yielded results compatible with STOT RE classification due to liver toxicity. However, the severity of hepatocellular necrosis was observed to be slight or very slight in all animals except in two individuals where was scored as moderate.

The 2-generation reproduction toxicity study in rat reported at 44-49 mg/kg bw/d liver hypertrophy in F_0 and F_1 in males affecting 93 and 33% of examined males and 50% of F_1 females. These alterations were not significant in other generations. The incidence and relevance of these alterations were not considered by RAC as toxicologically relevant for warranting classification. In the same study, liver hypertrophy was consistently reported in four different generations in almost 100% of male and female, although in this case these effects were found at 215-243 mg/kg bw/d, which is above the limits for warranting classification as STOT RE category 2.

In conclusion, RAC notes that the hepatotoxicity associated with repeated exposure of propiconazole either appears at concentrations above the cut-off values for warranting classification or when appearing below these limits, is seen with a severity and incidence not considered indicative of toxicologically relevant disturbances. Therefore RAC supports the DS's proposal and **does not propose classification of propiconazole for STOT RE**

RAC evaluation of germ cell mutagenicity

Summary of the Dossier Submitter's proposal

This hazard was not reviewed in the CLH-report.

Comments received during public consultation

One international non-governmental organization submitted a comment alerting about a few open publications on mutagenicity of propiconazole that were not included in the assessment of carcinogenicity by the DS. Similar comments were also submitted by the same international non-governmental organization in the carcinogenicity section. The DS replied that mutagenicity was included in the report only as supporting evidence. Please, see the DS's answer to this comment in the section of carcinogenicity.

Assessment and comparison with the classification criteria

Not applicable because this hazard was not reviewed in the CLH report.

RAC evaluation of carcinogenicity

Summary of the Dossier Submitter's proposal

One 2-year carcinogenicity study in mice reported incidences slightly above the contemporary historic controls of liver tumours in male CD-1 mice at dose levels of 107.8 and 344.3 mg/kg bw/d. However, the DS observed several factors reducing the relevance of this tumours for humans: i) the effect appeared only in male mice; ii) the effect did not appeared in rats; iii) the tumours appeared only at the end of the two years of exposure and at the maximum tolerable

dose (107.8 mg/kg bw/d) or higher (344.3 mg/kg bw/d); iv) another study showed no significant differences in incidences of liver carcinomas after 18 months of propiconazole exposure; v) a contemporary study showed relatively high spontaneous variability in incidences of hepatocellular adenomas and carcinomas in CD mice; vi) several studies showing a non-genotoxic mechanism involving CAR activation, induction of mitogenic hepatocyte proliferation and enlargement, hepatomegaly, and increase of xenobiotic metabolism, which is of less relevance for humans.

All these considerations reduced the concern of the liver tumours in mice and lead the DS to propose no classification of propiconazole for carcinogenicity.

Comments received during public consultation

Three MSCAs, one manufacturer/company and one industry/trade association supported the 'no classification' proposed by the DS.

One international non-governmental organization (NGO) commented that additional studies, not included in the CLH dossier, related to the potential carcinogenicity of propiconazole were found through a PubMed search and further commented that these publications according to them had far less risk of bias than the toxicity studies performed by Industry. The DS replied that CLP classification is mainly based on intrinsic properties of the substance through studies conducted in accordance with the EU test methods (Regulation 440/2008). The DS also replied that they had reviewed the published scientific literature prior to submission of the CLH proposal and that the classification proposal is based on studies which are considered adequate and reliable for classification purposes.The same NGO also commented that several papers describing a potential mutagenicity of propiconazole had not been taken into account and also questioned the validity of one of the studies where they suspected fraud and non-GLP compliance.

The DS replied to these comments clarifying that the incidences of hepatocellular adenomas in the available studies were 21 versus 40% and 2 versus 20% and that the incidence of hepatocellular adenomas in one study performed *ad hoc* in the same laboratory ranged between 6 and 18%, while this same parameter in another laboratory was essentially identical and ranged between 6 and 18.4%. Thus, according to the DS's opinion, incidences of liver adenomas in propiconazole treated males were statistically significantly increased compared to concurrent controls and slightly above the contemporary historical control range. The DS also clarified that the proposal of no classification for propiconazole is based on several factors that decrease the concern for human carcinogenicity, such as: tumours were found in one species and in males only; malignant tumours (carcinomas) were observed only at a dose level clearly exceeding the maximum tolerated dose at the end of the normal lifespan of mice; and, that propiconazole promoted formation of spontaneously occurring tumours only at high doses.

Regarding the suspicion of GLP fraud the DS consulted the Finnish GLP authorities, EFSA, ECHA and the EU Commission and according to the information from the GLP authority of the country where the testing laboratory is based, the laboratory conducting the 18-month mouse carcinogenicity study was included in their GLP monitoring program in the period of 1997-1999, although this particular mouse study was not audited in the inspections performed in 1996 and 1998.

The DS also reviewed both of the studies conducted on the request of regulatory authorities and studies published in the open scientific literature and found no *in vivo* animal studies showing a tumour profile that would differ from that described in the CLH report.

Finally, the DS reminded the commenter that no evidence of mutagenicity has been observed in the *in vivo* and *in vitro* mutagenicity assays usable for setting classification (those performed according to EU test methods; Regulation 440/2008) and that some of the open scientific literature does not consider propiconazole to act via genotoxic or mutagenic mechanism of action and that the increases in the mutation frequency in the liver is a consequence of oxidative stress.

Assessment and comparison with the classification criteria

2-year chronic toxicity and carcinogenicity study in Sprague Dawley CD rat (DAR IIA 5.5/02)

The main non-neoplastic findings were summarised in the table in the STOT RE section. Mortality rates at the end of the study were as follows: 38, 39, 40 and 31% for males and 53, 45, 45 and 33% in females of dose groups 0, 100, 500 and 2500 ppm, respectively. There was no evidence of treatment-related tumorgenesis.

2-year carcinogenicity study in CD-1 mouse (DAR IIA 5.5/03)

Mortality was high during the study; for males 41, 52, 50 and 64%, and for females 38, 31, 45 and 31%, in the dose groups of 0, 100, 500 and 2500 ppm, respectively. The highest mortality in all groups occurred during the second year of the treatment.

Food consumption of high dose (2500 ppm) males was significantly increased throughout the treatment. Despite this, the body weights and cumulative weight gain of this group remained significantly reduced throughout the study, indicating reduced food utilization. The same effect was apparent, but less marked in high dose females. Absolute body weights of high dose males remained 12-20% lower than controls throughout the study, suggesting that the maximum tolerated dose was exceeded. Cumulative weight gains of intermediate dose (500 ppm) males and females were reduced at the beginning of the study but recovered thereafter.

Histopathological analysis revealed signs of hepatotoxicity in both sexes and increased liver tumour incidence primarily in high dose males. No treatment-related effects were seen in the incidence and distribution of other tumour types, nor in organ weights or tissue histology. To confirm the original diagnosis, the observed tumour response and non-neoplastic changes of the liver were subsequently re-examined (DAR IIA 5.5/04). The morphology and biological behaviour of the liver tumours observed were evaluated according to new (at that time), diagnostic nomenclature and morphologic criteria. All liver specimens from males and females were examined for histopathological changes including neoplastic and non-neoplastic lesions. Proliferative hepatocellular lesions were classified as either foci of cellular alteration, benign (hepatocellular adenoma) or malignant (hepatocellular carcinoma) using the criteria published by the U.S. National Toxicology Program. The hepatocellular carcinomas were classified by degree of differentiation. In addition, all specimens of lung from males were examined for the presence of pulmonary metastasis from malignant hepatocellular neoplasms.

The table below summarises the neoplastic changes found in the carcinogenicity study in mouse. At interim sacrifice (53 weeks), the number of males with hepatocellular adenomas was slightly increased in the 500 ppm group and the number of males with adenomas and/or carcinomas was slightly increased in the high dose group (see table). All carcinomas were well differentiated and there was no evidence of pulmonary metastasis. There was no indication of neoplasia among the females at interim sacrifice.

For the terminal sacrifice (104 weeks) and decedent animals, the incidences of adenomas and/or carcinomas were significantly higher in high dose males than in controls (40% vs. 21% in controls and 80% vs. 51% in controls, respectively). The majority of this response was associated with an increased number of hepatocellular adenomas in high dose males. The slight increase in carcinomas was due to an increase in the number of well differentiated hepatocellular carcinomas relative to the control group. The incidences of moderately well and poorly differentiated carcinomas in treated groups were similar to controls. There was no significant difference in the morphologic appearance or biological behaviour of the carcinomas observed in the control as compared to the treated groups.

The number of adenomas and/or carcinomas was slightly, but not significantly, higher in high dose females compared to controls (9 vs. 6 in controls, number of mice with adenoma and/or carcinoma). No other indication of neoplasia was observed in females.

The total incidences of adenomas, carcinomas and of adenomas and carcinomas in males showed positive linear trend with dose, when all groups were evaluated (method by Peto *et al*., 1980). When the high dose group was excluded from the analysis, there was no evidence of a linear trend.

No treatment-related effect was seen in the incidence and distribution of other tumour types.

Table: Neoplastic changes in the carcinogenicity study in mouse. Incidences are based on re-examination of liver micropathology (DAR IIA 5.5/04).

	Dietary concentration of propiconazole (ppm)							
	Males				**Females**			
	0	**100**	**500**	**2500**	**0**	**100**	**500**	**2500**
Interim sacrifice								
No. animals examined	11	11	11	9	12	11	11	12
Adenoma [a]	1/1	0/0	4/4	4/3	0/0	0/0	0/0	0/0
Carcinoma well differentiated [a]	0/0	0/0	0/0	3/3	0/0	0/0	0/0	0/0
No. of mice with only adenoma	1	0	4	1	0	0	0	0
No. mice with at least one carcinoma	0	0	0	3	0	0	0	0
Adenoma + carcinoma	1	0	4	4	0	0	0	0
Terminal sacrifice and decedents								
No. examined	53	53	51	55	52	53	53	52
Adenoma [a]	25/18	18/11	20/15	68/35	6/5	0/0	2/2	13/8
Carcinoma well differentiated [a]	10/10	6/5	5/5	23/17	1/1	0/0	0/0	2/2
moderately well differentiated [a]	4/4	4/3	9/7	7/6	0/0	1/1	0/0	1/1
poorly differentiated [a]	3/3	2/2	3/3	3/2	0/0	0/0	0/0	0/0
No. mice with only adenoma	11 (21%)	7	9	**22* (40%)**	5	0	2	6
No. mice with at least one carcinoma	16	9	13	22	1	1	0	3
Adenoma + carcinoma	27 (51%)	16	22	**44** (80%)**	6	1	2	9
Total								
No. examined	64	64	62	64	64	64	64	64
No. mice with only adenoma	12 (19%)	7	13	**23* (36%)**	5	0	2	6

No. mice with at least one carcinoma	16 (25%)	9	13	25 (39%)	1	1	0	3
Adenoma + carcinoma	28 (44%)	16	26	**48*** (75%)**	6	1	2	9

[a] Total count of tumours/no. mice with tumours
* p ≤ 0.05, ** p ≤ 0.01, *** p ≤ 0.001 (pairwise Fisher' Exact Test)

In conclusion, in males the highest dose (2500 ppm) was above the maximum tolerated dose, based on increased mortality (64% compared to 41% in controls), decreases in absolute body weight (11-16% lower than controls), body weight gains (20-38% lower than controls), and evidence of hepatotoxicity. At this dose an increased incidence of liver tumours was observed (adenoma and/or carcinoma 80% vs. 51% in controls) at the end of the 104 weeks. The majority of this response was associated with an increased number of hepatocellular adenomas and the slight increase in carcinomas was primarily due to an increase in the number of well differentiated hepatocellular carcinomas. Signs of hepatotoxicity were observed also in high dose females, but there were no significant increases in liver tumour incidences.

18-months carcinogenicity study in CD-1 mouse (DAR IIA 5.5/05)

This study was conducted for additional mouse oncogenicity study since the high dose (2500 ppm) in the first study was considered to be excessively toxic. No significant clinical signs, behavioural changes or effects on survival were observed, although mortality was high, more than 60%, in all groups during the study.

The table below shows the incidences of neoplastic lesions found in liver (no other organ or tissues were microscopically assessed). A significant increase in adenomas and combined adenoma and carcinoma was found at the highest dose.

Table: Neoplastic findings in liver in 18-month study in mice.				
	Dietary concentrations of propiconazole (ppm)			
	0	**100**	**500**	**850**
Hepatocellular adenoma	1 (2%)	0	3	**10** (20%)**
Hepatocellular carcinoma	1	3	2	2
Total hepatocellular tumours	2 (4%)	3	5	**12** (24%)**

Spontaneous liver tumours in CD-1 mice

A contemporary study was conducted in the same laboratory to collect reference control data of in-life parameters and post-mortem findings, including the incidence of liver tumours from CD-1 mice over a period of 18 months under standard laboratory conditions. Five groups per sex with 80 mice per group were used. Incidences of hepatocellular adenoma, adenocarcinoma and combined adenoma + carcinoma ranged between 6 and 18%, between 8 and 16% and between 14 and 30%, respectively (table below).

Table: Incidences of liver tumours in 18-month reference study in CD-1 mouse.					
	Group 1	**Group 2**	**Group 3**	**Group 4**	**Group 5**
Number examined	50	50	50	50	50
Adenoma	9 (18%)	7(14%)	3 (6%)	4 (8%)	7 (14%)
Adenocarcinoma	6 (12%)	8 (16%)	5 (10%)	4 (8%)	7 (14%)
Combined: adenoma + adenocarcinoma	15 (30%)	12 (24%)	7 (14%)	7 (14%)	13 (26%)

The notifier also reported historical control data on CD-1 males from Charles River Laboratory showing a historical control range of 6.0-18.4% for hepatocellular adenomas and 0-12% for hepatocellular carcinomas in four studies with 199 control animals and a mean adenoma incidence of 10.8% with 12 studies with a total of 770 controls.
In conclusion, the incidences of adenomas (36% in the 2-year study at exposure level higher than the maximum tolerable dose and 20% in the 18-month study at exposure level below the maximum tolerable dose) were slightly above the contemporary historical control incidence range (6-18%) from the same laboratory, whereas the combined incidence of adenomas and carcinomas in the 18-month study (24%) was within the contemporary historical control range (14-30%).

Mechanism of action based on constitutive androsterone receptor (CAR) activation

Additional studies to investigate the MoA for the propiconazole-induced liver tumours in mice have been conducted. These studies were conducted to determine if propiconazole exert its liver carcinogenicity via the activation of the constitutive androsterone receptor (CAR), i.e. the phenobarbital mode of action (MoA). The conclusions from the studies are briefly summarised below. For detailed study descriptions, see Background document.

Study 1: The effect of propiconazole on drug metabolizing enzymes in the livers of Tif:RAIf rats and Tif:MAGf mice (DAR II 5.8.6/02)

Propiconazole caused clearly discernible changes in the ultrastructural organisation of hepatocytes and was an efficient inducer of xenobiotic metabolism in both rat and mouse. The profiles of liver enzyme induction in rat and mouse by propiconazole were found to be different in some respects, especially concerning activities of cytochrome P-450 enzymes, epoxide hydrolase, and glutathione S-transferase. The possible explanation to the observed differences in hepatic tumour formation between the two species and the sex differences noted in mice could be based on the observed differences in xenobiotic metabolism.

Study 2: Tumour promotion study with propiconazole in Tif:RAIf rat (DAR II 5.8.6/01).

The conclusion from this study was that propiconazole acts as a promoter of proliferative changes in rat liver at dietary concentrations of 2000 ppm.

Study 3: Assessment of hepatic cell proliferation in male CD-1 mice (DAR IIA/5.8)

Treatment with propiconazole at 850 and 2500 ppm for up to 60 days caused a prominent, time- and dose-related hepatomegaly. The liver enlargement was caused by a sharp and transient induction of hepatocellular proliferation and to a time- and dose-related increase in the severity of hepatocellular hypertrophy. In general, the temporal pattern of propiconazole induced hepatocyte proliferation was the same as for phenobarbital, suggesting that propiconazole is a phenobarbital-like mitogen in the male mouse liver.

Study 4: Effects on biochemical parameters in the liver following administration to male CD-1 mice (DAR IIA/5.8)

In this study, sub-chronic treatment of male mice with 850 and 2500 ppm propiconazole or 850 ppm phenobarbital caused strong and qualitatively similar induction effects on liver weight and biochemical liver parameters. Thus propiconazole was found to be a strong phenobarbital-type inducer of xenobiotic metabolising enzymes in the mouse liver.

Study 5: Cytochrome P450 2b, 3a and DNA-synthesis induction in cultured male CD-1 mouse hepatocytes (dRAR B6.8.2.2)

In this study it was concluded that 5 µM propiconazole resulted in the induction of Cyp2b10 mRNA levels, while Cyp3a11 mRNA was increased at 25 µM and both concentrations (5 and 25 µM) induced cell proliferation in mouse hepatocytes consistent with activation of CAR.

Study 6: Cytochrome P450 2B, 3A and DNA- synthesis induction in cultured male human hepatocytes (dRAR, B.6.8.2.3)

Phenobarbital and propiconazole induced CYP2B6 and CYP3A4 transcripts without affecting cell proliferation in human hepatocytes. This is consistent with species differences in CAR and PXR receptors between humans and rodents.

Study 7: CAR3 direct activation assay with mouse, rat and human CAR (dRAR, B.6.8.2.4)

This study showed that propiconazole is a direct CAR activator in mouse, rat and human and under the conditions of this assay the activation of rat CAR with propiconazole was strongest, whereas responsiveness of human CAR to propiconazole was much weaker than mouse and rat CAR. This suggests a quantitative difference between rodent CAR and human CAR with respect to their direct activation by propiconazole.

Overall discussion of mechanistic studies based on constitutive androsterone receptor (CAR) activation

According to Elcombe *et al*. (2014) the CAR-mediated pathway for induction of liver tumours consists in seven key events (depicted in a figure above). The table below overalls the experimental evidences for supporting such key events available for three differences species.

Table: Summary of evidences for the different key and associative events of a CAR-mediated induction of liver tumours in mice, rats and humans.

Key event	Mice	Rat	Humans	Study
CAR activation	YES	YES	YES	dRAR, B.6.8.2.4
Altered gene expression	Cyp 2b10 Cyp 3a11	Not determined	CYP 2B6 CYP 3A4	dRAR B6.8.2.2, dRAR, B.6.8.2.3
CYP induction	YES	YES	Not determined	DAR II 5.8.6/02, DAR IIA/5.8, dRAR B6.8.2.2
Increase liver weight	YES	YES	Not determined	DAR II A 5.3.1/01, DAR IIA 5.5/02, DAR II A 5.3.2/03, DAR IIA 5.3.2/04, DAR IIA 5.5/03, DAR IIA 5.5/05, dRAR B.6.3.3.1.2, DAR II 5.8.6/02, DAR II 5.8.6/01, DAR IIA/5.8, DAR IIA/5.8
Liver hypertrophy	YES	Not determined	Not determined	DAR IIA/5.8
Cell proliferation	YES	Not determined	**NO**	DAR IIA/5.8, dRAR B6.8.2.2, dRAR, B.6.8.2.3
Hepatic foci alteration	YES	YES	Not determined	DAR II 5.8.6/01, DAR IIA 5.5/02, DAR IIA 5.5/03

Tumour formation	YES	NO	Not determined	DAR IIA 5.5/02, DAR IIA 5.5/03, DAR IIA 5.5/05

The table above shows that all the key events in the development of liver tumours through CAR activation has been experimentally supported in mice and most of them in rat. RAC notes that one potential gap in the mechanistic information was the decrease of apoptosis as consequence of alterations in gene expression. The CLH dossier contains an Annex with a document submitted by the notifier with an assessment of the MoA for liver tumours induced by propiconazole using the framework developed by IPCS and ILSI/HESI. In this document it is stated that *"increased expression of pro-proliferative and anti-apoptotic gene Gadd45β"* was detected after exposures to 850 ppm propiconazole. However, there is no reference to support this information and the details were not contained in the CLH-report and consequently this information could not be assessed by RAC. Nevertheless, RAC considers that the overall picture of the available experimental information makes plausible that the mechanism of liver carcinogenesis induced by propiconazole in mouse was based on CAR-activation.

There were severe interspecies quantitative differences in the activation of CAR by propiconazole, specifically, 30 μM propiconazole is able to activate mouse, rat and human CAR by 60, 40 and 3-fold of solvent controls, respectively. These differences might be responsible of the fact that propiconazole failed to induce liver tumours in rat, while did it in mouse.

RAC notes two critical differences between mouse and humans. These differences were: i) the activation of human CAR is around 20 times lower than the activation of mouse CAR; and, ii) cell proliferation could not be detected through replicative DNA synthesis in human hepatocytes, while did in mouse hepatocytes. These two differences play in favour of lack of relevance of this CAR activation mechanism for humans.

Potential alternative mode of action for liver carcinogenesis induced by propiconazole

DNA reactivity and mutagenicity

Propiconazole was negative in wide array of *in vitro* and *in vivo* genotoxicity assays.

Peroxisome proliferation

Propiconazole produced little or no increase in lauric acid 12-hydroxylase activity and Cyp4a levels of protein in liver fractions of treated mice. Both of these markers are greatly increased by peroxisome proliferators, which suggest that propiconazole is not a peroxisome proliferator.

Aromatic Hydrocarbon Receptor P450 induction

Propiconazole did not produce a large increase in EROD activity nor an increase in Cyp1a protein levels in liver microsomes of treated mice. Both of these markers are greatly increased by aromatic hydrocarbon receptor activators, which suggest that propiconazole is not an aromatic hydrocarbon receptor activators.

Estrogenic stimulation

Propiconazole did not bind to the oestrogen receptors at most concentrations tested, and appeared to severely disrupt the assay at very high concentrations (10^{-3} M). Propiconazole was negative for estrogenic effects in an uterotrophic *in vivo* assay in the ovariectomized rat. In combination with the lack of effects on oestrogen-sensitive tissues in the wider toxicology database, the weight of evidence indicates that propiconazole does not show estrogenic potential.

RAC notes that all this information was cited in the CLH report as described in several references but that were no accessible to RAC.

Cytotoxicity and regenerative hyperplasia

The evidence does not support a finding of regenerative hyperplasia, which is the causal key event required for carcinogenesis to be produced as a secondary consequence of hepatotoxicity.

Cell proliferation caused by propiconazole was transient and can be contrasted with the sustained regenerative cell proliferation and development of long-term fibrosis seen with classical hepatotoxic agents that induce regenerative hyperplasia such as chloroform and carbon tetrachloride. As an example, an increase in liver cell proliferation was observed for up to 159 days of treatment in a study with chloroform in B6C3F1 mice, but no cell proliferation was observed beyond 7 days in the current studies with propiconazole.

In the *in vivo* mouse studies, a limited amount of hepatic necrosis (single cell or focal/multi-focal) plus chronic inflammatory cell infiltration were observed, which is in contrast with the pattern of effects seen with classic cytotoxic carcinogens that cause a diffuse necrosis in the liver that progressed to regenerative hyperplasia, as is the case of chloroform.

In conclusion, the weight of evidence shows that a MoA involving cytotoxicity and a subsequent sustained regenerative cell proliferation is not operative with propiconazole. RAC notes that the information cited in the CLH report regarding chloroform were no accessible to RAC.

Statins/altered cholesterol biosynthesis

Propiconazole was not designed to inhibit HMG-CoA reductase so this MoA is unlikely to be operating. Nevertheless, plasma cholesterol levels were decreased in mice by propiconazole treatment. The sites of action in the cholesterol synthesis and metabolism pathway that are theorized to cause this effect are thought to be different from the statins. Experiments with another triazole fungicide, cyproconazole, have shown that the effect of lower plasma cholesterol at a tumorigenic dose of 200 ppm was completely blocked in mice lacking the CAR receptor. CAR receptor activation has been shown to play a role in regulation of lipogenesis, β-oxidation of fatty acids, gluconeogenesis and cholesterol/bile acid metabolism. Therefore it is likely that an alteration in cholesterol metabolism is also a consequence of CAR activation by propiconazole.

Additional considerations for classification

The Guidance on the Application of the CLP Criteria establishes certain important factors which may be taken into consideration when assessing the overall level of concern. These factors are displayed and discussed in the table below.

Table: Some important factors which may be taken into consideration when assessing the overall level of concern of the propiconazole-induced tumours.	
Tumour type:	Liver tumours
Multi-site responses:	No (only liver)
Progression of lesions to malignancy:	Malignancy appeared only above the maximum tolerable dose
Reduced tumour latency:	No (the malignant tumours occurred at a later stage of the study)
Whether responses are in single or both sexes:	Single sex (males)
Whether responses are in a single species or several species:	Single species (mice)
Structural similarity to a substance(s) for which there is good evidence of carcinogenicity:	Not noted

Routes of exposure:	Oral (relevant for human)
Comparison of absorption, distribution, metabolism and excretion between test animals and humans:	Not known
The possibility of a confounding effect of excessive toxicity at test doses:	Carcinomas appeared in concurrence with liver toxicity, only adenomas were seen below the maximum tolerable dose.
Mode of action and its relevance for humans, such as cytotoxicity with growth stimulation, mitogenesis, immunosuppression, mutagenicity:	Potentials modes of action have been discussed above, but it is plausible that the MoA was through CAR activation, which is of low relevance for humans.

Comparison with the criteria

A substance can be classified as carcinogenic Category 1A when it is known to have carcinogenic potential for humans on the basis of human evidence. There is no information about the potential carcinogenicity of propiconazole for humans and therefore Category 1A is not supported.

A substance can be classified as carcinogenic Category 1B when it is presumed to have carcinogenic potential for humans on the basis of animal evidences, while Category 2 is reserved for substances suspicious to be carcinogenic on the basis of evidences not sufficiently convincing to classify as Category 1.

RAC notes that there are two different studies in mouse demonstrating that propiconazole is able to induce hepatocarcinogenicity, which in principle is enough to propose classification in Category 1B. However, RAC notes other factors that considerably reduce the level of concern regarding the propiconazole carcinogenicity for humans. These factors are:

- The tumours appeared only in one species (mice), tissue (liver) and sex (males);
- The incidence of hepatocellular carcinomas exceeded those of the controls only at doses clearly exceeding maximum tolerable doses in the 2-year carcinogenicity study causing 40% reduction body weight gain, while at the maximum tolerable dose in the 18-month carcinogenicity study the incidence of carcinomas were similar to that reported for control;
- There was no significant difference in the morphological appearance or biological behaviour of the carcinomas observed in the control when compared to the treated groups;
- No significant incidence of carcinomas was reported after 53 weeks of exposure at doses above the maximum tolerable dose, which suggest a long time of latency;
- The incidences adenomas (20%) at maximum tolerable dose in the 18-month carcinogenicity study was only slightly above the incidence of spontaneous adenomas (6-18%) reported in the same laboratory for a contemporary study;
- The incidences of carcinomas (4%) and combined adenomas plus carcinomas (24%) at maximum tolerable dose (in the 18-month carcinogenicity study) were within the incidences of spontaneous carcinomas (8-16%) and spontaneous adenomas plus carcinomas (14-30%) reported in the same laboratory for a contemporary study;
- Experimental evidence supporting a MoA for the induction of liver tumours in male mice attributable to CAR activation, with quantitative interspecies differences in response to CAR activation between mice and humans depicted in a figure above
- Low plausibility for other potential alternative MoA for liver carcinogenesis induced by propiconazole.

RAC also notes uncertainties in the available database, as the lack of information about how many independent hepatocyte cultures were used in the dRAR B6.8.2.2 (mouse) and specially in dRAR B6.8.2.2 (human) or the absence of data with CAR-knock-out mouse.

Nevertheless, the overall available information suggests that the liver tumours found in mice after exposure to propiconazole are not of concern for humans and RAC agrees with the DS that **no classification for carcinogenicity of propiconazole is warranted.**

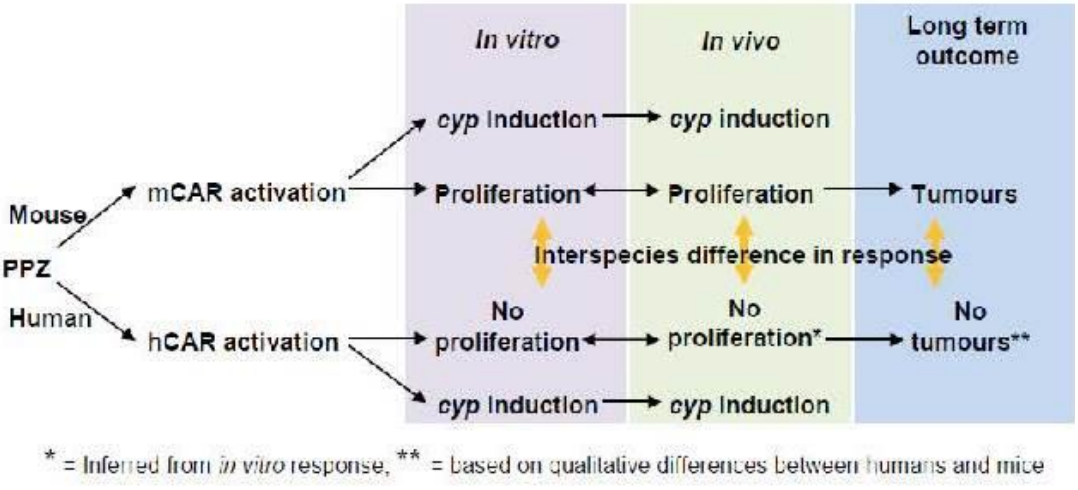

Figure: Summary of mouse-human differences in response to propiconazole.

RAC evaluation of reproductive toxicity

Summary of the Dossier Submitter's proposal

Sexual function and fertility

The DS proposed no classification of propiconazole for sexual function and fertility effects. It was based on a 2-generation reproduction study in rats showing negative results on mating, fertility, gestation, female and male fertility index and average of gestation length. The only effects on reproduction reported in this study were reduction in pup weights during the first generation and during the second generation reductions in litter size, number of viable pups delivered, pup survival and increased number of runt pups at the highest dose.

Developmental toxicity

The DS proposed classification of propiconazole for reproductive toxicity category 2 (H361d) on the basis of two different developmental studies in rat showing incidences of cleft palates higher than controls and historical control data but appearing always concurrently with maternal toxicity.

Comments received during public consultation

Three MSCAs supported the classification proposed by the DS.

One MSCA disagreed with the proposal of classification as category 2 and proposed category 1B considering that: i) the increased incidence of cleft palates in rats treated with propiconazole justifies classification for developmental toxicity; ii) there is no convincing evidence demonstrating that the sensitivity of humans is more similar to rabbits than rats; iii)

disagreement about a reduction of the concern by high maternal toxicity; iii) the incidences of cleft palate were observed in the two foetuses occur in different litters; iv) it cannot be excluded that some additional cases may be masked by the slightly increased post implantation loss and reduced number of viable foetuses in the developmental rat study; and, v) this rare malformation is commonly observed with other "conazoles". The DS agreed with the comments and leaved the final decision in RAC.

One MSCA requested a classification on fertility on the basis of oestrus cycle and anogenital distance. The DS replied that there are a few findings in the open scientific literature which add concern for reproductive toxicity of propiconazole and that were discussed in the CLH report. However, the DS chose not to propose classification for fertility for propiconazole because of the following reasons: i) there were no significant effects on fertility, fecundity or reproduction parameters in a two-generation reproduction study or when assessed, in studies published in open scientific literature; ii) increase of anogenital distance in male pups and reversible disruption of oestrus cycle may rather contribute to classification for developmental toxicity (reproductive development) than for fertility; iii) although all these effects suggest for disturbed steroidogenesis they may be considered as individual findings because when assessed, effects on anogenital distance or oestrus cycle have not been observed in other studies.

One MSCA requested more information for establishing a read-across with other triazoles. The DS did not consider it necessary since this information is already documented in the respective RAC opinions.

One manufacturer/company diminished the relevance of the cleft palate cases on the basis of low incidence, maternal toxicity, absence of embryo lethality and lack of evidences about if propiconazole-induced effects would result in functional deficiency in foetuses. The company also submitted a position paper requesting no classification for propiconazole together with another published historical control data and a third rat developmental study showing no cases of cleft palate. DS disagreed with the proposal of no classification considering that the incidence of cleft palate were above the historical control of the performing laboratory and noted that the third new study contains certain deviations from the OECD TG 414 that do not allowed the DS to assess the acceptability of the study. In addition, the DS is of the opinion that the negative findings of this study do not overrule the findings of other developmental toxicity studies.

Another MSCA highlighted that propiconazole is included in the Endocrine Disruptor Screening Program Tier 1 and therefore several other studies on endocrine properties are available and requested more discussion about the appropriate classification for developmental toxicity. The DS answered that endocrine disruption *per se* is not a hazard considered by the CLP Regulation. However, the disruption of endocrine receptors may form part of one or more MoA of a chemical considered by RAC under the Reprodutive toxicity hazard class.

An international NGO submitted a comment that classification in category 1 is needed on the basis of papers published in the open scientific literature on reproductive toxicity and on endocrine disruption.

Assessment and comparison with the classification criteria

Sexual function and fertility

In a two-generation reproduction study, propiconazole was administered in the diet (*ad libitum*) at concentrations 0, 100, 500 and 2500 ppm to groups of 15 male and 30 female Charles River CD rats. The main deviations to current OECD TG 416 were: oestrus cycle and sperm parameters were not determined, developmental landmarks of the offspring including parameters of sexual

maturation were not evaluated, food consumption was only determined during pre-mating period, only brain, ovary and testes weights were determined.

Statistical analysis of reproductive data (mating, fertility, gestation, female fertility and male fertility index, average gestation length) revealed no significant reductions in these parameters. Delivery and population data (the mean numbers of pups delivered, delivered viable, stillborn and partially cannibalized at birth, the numbers of survived pups during the lactation period) obtained for the groups of dams exposed to propiconazole were comparable to the control dams during both the F_{1a} and F_{1b} litters. In the F_{2a} litter, the number of pups delivered, delivered viable and surviving to lactation day 4, were significantly reduced for the 2500 ppm dams (8.1 in the treated animals versus 12.0 in control group). In the F_{2b} litter, the pup survival indices at lactation days 7, 14, and 21 were significantly reduced for the 2500 ppm group dams (6.2, 5.7 and 5.7 in exposed animals versus 7.7, 7.7 and 7.7 in control group, respectively).
The histopathological analysis revealed statistically significant increases in liver hypertrophy of males and females of all generations at the highest dose and in F_0 and F_1 males and females at 500 ppm (table below). F_0 and F_1 males and females showed liver vacuolation at 2500 ppm, while the incidence of this vacuolation was significant at 500 ppm only in F_1 males.

Table: Histopathological changes in liver of parental animals and progeny. Bolded figures highlight the statistically significant differences regarding the animals dieted with 0 ppm propiconazole.

	Dietary concentration of propiconazole (ppm)							
	Males				Females			
	0	100	500	2500	0	100	500	2500
Hypertrophy								
F_0	7/15	3/15	**13/15**	**14/15**	4/30	3/29	6/30	**29/30**
F_1	0/15	1/15	**5/15**	**15/15**	0/30	2/30	**15/30**	**29/30**
F_{1b}	2/10	1/10	2/10	**10/10**	1/10	1/10	2/10	**8/10**
F_{2b}	0/10	0/10	2/10	**10/10**	0/10	0/10	1/10	**9/10**
Vacuolation								
F_0	0/15	2/15	3/15	**14/15**	1/30	1/29	1/30	1/30
F_1	2/15	5/15	**8/15**	**11/15**	2/30	4/30	7/30	**10/30**
F_{1b}	0/10	0/10	0/10	0/10	0/10	0/10	0/10	0/10
F_{2b}	0/10	0/10	0/10	1/10	0/10	0/10	0/10	0/10

Absolute brain and testes (including epididymides) weights of the 2500 ppm group of F_{1a} males were significantly reduced in comparison to controls. Absolute testes weights and testes to brain weight ratios of 2500 ppm F_{2a} males were also significantly reduced compared to controls. The testes findings were not confirmed by microscopy, since histopathological examinations were only performed on F_{1b} and F_{2b} litters. A significant increase in the brain to body weight ratio was noted in the 2500 ppm F_{2b} males and a significant reduction in the brain weight was noted in the 2500 ppm F_{2b} females. RAC notes that the incidence of the above mentioned effects was not stated in the CLH report.

Development

Two-generation reproduction study in rats

In a two-generation reproduction study (see also 'Sexual function and fertility') propiconazole was administered in the diet (*ad libitum*) at concentrations 0, 100, 500 and 2500 ppm to groups of 15 male and 30 female Charles River CD rats.

In F_{1a} and F_{2a} litters, progeny of dams exposed to 2500 ppm had significantly lower body weights than controls on lactation days from 4, 7, 14 and 21 (table below). Body weights of 2500 ppm F_{1b} progeny were significantly lower than those of the control progeny on lactation days 14 and 21. Body weights of 2500 ppm F_{2b} progeny were significantly lower than the control progeny on

lactation days from 0, 4, 7, 14 and 21. The body weight data obtained for the 100 and 500 ppm progeny revealed inconsistent statistically significant reductions and increases in comparison to controls with equivocal biological significance.

Table: Effect of propiconazole on body weight of pups of different litters.					
		Mean pup body weight (g)			
		0 ppm	**100 ppm**	**500 ppm**	**2500 ppm**
F_{1a}	0	6.1	6.0	6.1	6.2
	4	8.9	8.8	9.2	**8.1****
	7	14.6	**13.0****	15.2	**12.3****
	14	28.2	**25.4****	**26.9***	**21.9****
	21	48.0/46.1	**44.1/42.5****	47.7/44.9	**35.8/34.7****
F_{1b}	0	6.1	6.0	**6.4****	6.1
	4	8.8	9.2	**9.5****	8.6
	7	13.4	14.5	**14.5***	13.3
	14	25.7	**27.7***	**28.0****	**22.9****
	21	44.3/40.1	46.1/43.5	47.7/43.5	**35.6/32.5****
F_{2a}	0	5.4	5.3	**5.7***	5.4
	4	8.5	**7.9****	8.3	**7.5****
	7	13.7	13.3	13.0	**10.8****
	14	25.6	25.4	25.2	**20.0****
	21	438./41.1	42.5/40.4	425./39.8	**31.5/30.2****
F_{2b}	0	5.8	**5.4****	**5.5****	**5.4****
	4	8.8	8.6	8.5	**7.3****
	7	14.6	14.7	13.8	**10.9****
	14	29.4	28.7	**26.8****	**21.9****
	21	48.8/46.2	48.6/46.6	**45.1/42.3****	**36.7/33.3****

Day 21, pup body weights are males/females ** statistically significant difference $p<0.01$, * $p<0.05$

Examination of the external structural development of the progeny did not reveal any statistically significant differences. Two 100 ppm F_{1a} anomalous pups (anurous and club limbs; cleft lip), one 2500 ppm F_{1a} anomalous pup (a partially opened left eyelid and a left eye which was smaller than normal), two 2500 ppm F_{1b} anomalous pups (a domed forehead [the brain was dilated] and eyes which appeared smaller than normal; a left eye which was enlarged with an opacity) and one 2500 ppm F_{1b} pup with an unopened eyelid (the eye was however not missing, and apparently of normal size) were obtained from dams treated with propiconazole. One stillborn 500 ppm F_{2a} pup exhibited agnathia and possible exencephaly (dam cannibalized top of head). One 100 ppm F_{2b} pup displayed clubbed limbs, shortened torso and a shortened tail. One 500 ppm F_{2b} pup kept its left eyelid closed (the left eye was shrunken in size). Number of runt pups was significantly increased in the 2500 ppm F_{2b} progeny.

<u>Rat developmental toxicity study (DAR IIA 5.6.2/01)</u>

The table in the STOT RE section summarises the maternal toxicity found in this study. Severe clinical signs were seen only at the highest dose of 360 mg/kg bw/d, which caused a reduction to 300 mg/kg bw/d. Despite maternal toxicity, all reproduction parameters remained similar in all groups (see table below). There were no significant differences in foetal weights between treated groups and controls. External and visceral examinations revealed one foetus in intermediate group to have cleft lip and cleft palate, micromelia and a club foot, and other foetus from different litter in the intermediate group to have cleft lip. In the high dose group one foetus had cleft palate, and another foetus from different litter had anasarca, cleft palate, hydromelia and protruding tongue. Significant increases in the incidences of short and absent renal papilla(e) and dilated ureters were detected in foetuses of the high dose group. Significant increases in the incidences of rudimentary ribs and non-ossified sternebrae were observed in foetuses of the high and intermediate dose groups.

Table: Summary of reproductive parameters and foetal findings.				
	Dietary propiconazole (mg/kg bw/d)			
	0	**30**	**90**	**360/300**
Number of pregnant females (% of mated)	23 (95.8)	21 (87.5)	22 (91.7)	22 (95.7)
Mean number of implantations	13.5	14.2	14.3	14.0
Mean number of Corpora Lutea	16.9	16.7	17.3	16.5
Number of viable litters examined	22	21	22	22
Viable foetuses per group	270	284	302	285
Mean no. viable foetuses per dam	12.3	13.5	13.7	13.0
Mean no. early resorptions per dam	1.1	0.7	0.5	1.0
Mean no. late resorptions per dam	0	0	0	0.1
Mean no. total resorptions per dam	1.1	0.7	0.6	1.1
% post-implantation loss	8.8	4.7	4.1	7.8
Foetal sex ratio (% males)	51.9	49.3	48.3	46.0
Mean foetal body weight (g)	3.5	3.5	3.5	3.5
External foetal findings				
Number of foetuses examined	270	284	302	285
Anasarca	0	0	0	1[b]
Cleft lip	0	0	**2[a]**	0
Cleft palate	0	0	**1[a]**	**2[b]**
Club foot	0	0	1[a]	0
Micromelia	0	0	1[a]	0
Visceral findings				
Number of foetuses examined	141	148	156	148
Renal papilla(e) short	32	27	40	**57****
Renal papilla(e) absent	4	4	8	**16****
Dilated ureter(s)	38	21	38	**63****
Protruding tongue	0	0	0	1[b]
Hydromelia	0	0	0	1[b]
Selected skeletal findings				
Number of foetuses examined	129	136	146	137
Lacrimal bone agenesis	0	0	1[a]	0
Rudimentary ribs	**0**	**1**	**4**	**53**
No. litters with foetuses with rudimentary ribs	**0/22**	**1/21**	**4/22***	**16/22****
Sternebrae not ossified	49	54	**83***	**99****

Excludes the skeletal malformation of rudimentary 13th thoracic ribs observed in 3 control foetuses, 1 low dose and 1 high dose foetus.
*Statistically different from control at $p<0.05$, ** $p<0.01$, [a] Same foetus cleft palate, cleft lip, lacrimal bone agenesis, micromelia, club foot , [b] Same foetus cleft palate, protruding tongue, hydromelia, anasarca.

In conclusion, a NOAEL of 30 mg/kg bw/d for foetal effect was established based on one cleft palate observed at 90 mg/kg bw/d and two cleft palates at 360/300 mg/kg bw/d (all in different litters). Moreover, an increased incidence of skeletal variations (rudimentary ribs and non-ossified sternebrae) were observed at 90 mg/kg bw/d and 360/300 mg/kg bw/d, and increased incidence of visceral variations (short and absent renal papilla(e) and dilated ureters) at 360/300 mg/kg bw/d.

In this study, the incidence of cleft palate in the intermediate dose group was 1/302 (0.33%) and in the high dose group 2/285 (0.70%). Cleft palate is a rare malformation in CD rats. According to data submitted by the registrant, the incidence of cleft palate in the performing laboratory was 0/5431 during 1983-1985, whereas in other laboratories the incidence was 4/25522, (0.016%) in 1983-1986. At the intermediate dose, maternal toxicity was moderately exhibited by transient decreases in body weight gain and food consumption during the first days of dosing. Although maternal toxicity was marked in high dose dams, there was no lethality and

no effect on corrected body weight gain or on any of the reproductive or foetal parameters examined. Thus, although cleft palates were observed at maternally toxic doses, treatment-related effect cannot be excluded.

Supplementary developmental toxicity in rat (DAR IIA 5.6.2/02)

The table in the STOT RE section summarises the main maternal effects of 300 mg propiconazole/kg bw/d that include clinical sings with two dead and reductions (17%) in maternal body weight gain. The table below summarises the results of the study regarding reproductive parameters and foetal findings.

Table: Summary of reproductive parameters and foetal findings.

	Dietary propiconazole (mg/kg bw/d)	
	0	**300**
Number of pregnant females/No. placed on study	155/178	131/189
Mean number of. implantations	14.5	14.2
Mean number of Corpora Lutea	16.9	17.0
Number of litters examined	155	158[a]
Number of viable foetuses per group	2122	2064
Mean number of viable foetuses/dam	13.7	**13.1***
Number of early resorptions	0.8	1.0
Number of late resorptions	0.1	0.1
Total number of resorptions	0.81	1.15
% post-implantation loss	5.8	8.6
Foetal sex ratio (% males)	49	50
Foetal weight - males (g)	3.569	**3.403****
Foetal weight - females (g)	3.387	**3.232****
Malformations / number of foetuses examined		
Spina bifida, gastrochisis, exencephaly, protruding tongue	1[b]/2122	0/2064
Agnathia	1/2122	0/2064
Filament tail	2/2122	0/2064
Cleft palate	0/2122	**2/2064[c]**

*Statistically different from control at p<0.05, **p < 0.001, [a] Three dams died before termination of the study
[b] Same foetus spina bifida, gastrochisis, exencephaly, protruding tongue, [c] Foetuses from different litters

In conclusion, oral administration of propiconazole at dose level of 300 mg/kg bw/d caused severe maternal toxicity (including premature death of two dams). Foetal and reproduction toxicity (decreased number of viable foetuses, decreased foetal weight, and slight increase in post-implantation loss) was also observed at the tested dose level of 300 mg/kg bw/d. In the treated group, **cleft palate was observed in 2/2064 foetuses (incidence 0.097%) from 2/158 litters**. According to historical control data submitted by the registrant cleft palate occurred sporadically during 1983 to 1985 in this rat strain at an incidence ranging from 0% (0/5431, this laboratory) to 0.016% (4/25522, other laboratories).

Supplementary developmental toxicity in rat

This study was submitted by Industry during the Public Consultation. It was performed with the objective to assess possible adverse effects of propiconazole on embryonic and/or foetal development. The study was conducted prior to Regulatory Test Guidelines and GLP, although following the principles of OECD TG 414. The major reported deviation was the dosing window, which was conducted from GD 6 to 15.

The dosage regime was 30, 100 and 300 mg/kg bw/d during days 6-15 of pregnancy. Dams of the highest dose reacted to the treatment by a marked reduction in body weight gain and food consumption and with mortality in three of the 25 dosed females (two on day 19 and one on day 20).

The gross examination of the foetuses did not reveal any treatment related malformation in any of the experimental groups. One case of hydrocephaly was reported in the 100 mg/kg bw/d group, which was within the historical control range. No pathological changes of the viscera were reported in any of the foetuses.

In both the 100 mg/kg bw/d and control groups, one case of irregularly sternum was reported. There was an increase in the number of un-ossified phalangeal nuclei of the fore and hind limbs in the 300 mg/kg bw/d group.

Developmental toxicity study in rabbit (DAR IIA 5.6.2/03)

On day 29, there were no statistically significant differences between the corrected body weights (minus uterus placentas and foetuses) of all groups. There were no abnormal necropsy findings.

The table below summarises the results of the study regarding reproductive parameters and foetal findings. No statistically significant differences were observed in the number of corpora lutea, number of implantation sites and number of viable or dead foetuses. Incidences of resorptions and abortions or early deliveries were significantly increased among dams of the high dose group. In the case of one high dose dam, the whole litter (ten pups) was resorbed early and there were no live pups at termination (the effect does not reach statistical significance if this dam is omitted from analysis). Five high dose dams were sacrificed prior to schedule because of early delivery abortion. In addition, one doe from the intermediate group aborted and one from the control group delivered early.

Foetal weights were not affected by the treatment. Only one foetus had malformations; foetus from the intermediate dose group had a cleft lip, umbilical hernia and hydronephrosis with hydroureter. Five foetuses had visceral variations: one control foetus (red area in left lung), two intermediate dose group foetuses (thick aorta and small gallbladder, other foetus had ovarian cysts) and two high dose group foetuses (the same litter) had coagulated blood above bladder. Since majority of the gross and visceral observations were limited to single intermediate-dose group foetus, without any dose-response, there were considered to be spontaneous in nature. Various skeletal variations were observed across all groups. Of these the incidence of fully formed 13th ribs, was significantly increased among the high dose group foetuses.

Table: Summary of reproductive parameters and foetal findings.

	Dietary propiconazole (mg/kg bw/d)			
	0	**100**	**250**	**400**
Number of pregnant does/ no. inseminated	15/19	18/19	17/19	18/19
Found dead/sacrificed	0	1	1	0
Aborted/Delivered early	1[a]	0	1[b]	**5*[c]**
Viable litters examined	14	17	15	12[d]
Mean no. Corpora Lutea	11.6	12.6	13.3	13.5
Mean no. implantations	8.4	9.4	10.0	9.2
Early resorptions: per group (mean per dam)	2 (0.1)	6 (0.4)	6 (0.4)	**17[e] (1.3)**
Late resorptions per group (mean per dam)	8 (0.6)	6 (0.4)	5 (0.3)	10 (0.8)
Total resorptions per group (mean per dam)	10 (0.7)	12 (0.7)	11 (0.7)	**27*(2.1*)**
Viable foetuses per group (mean per dam)	101 (7.2)	146 (8.6)	130 (8.7)	93 (7.2)
Dead foetuses per group (mean per dam)	6 (0.4)	1 (0.1)	9 (0.6)	0 (0)
Foetal sex ratio (% males)	55	49	50	41
Foetal weight (g), males/females	43.0/44.2	44.4/43.1	42.8/41.1	42.8/43.2
Gross malformations				
Number of foetuses examined[f]	101	146	130	93
Cleft lip	0	0	1[g]	0
Umbilical hernia	0	0	1[g]	0
Visceral malformations				

Hydronephrosis with hydroureter	0	0	1[g]	0
Selected skeletal variations				
Fully formed (13th) ribs	35	63	58	**63***
Rudimentary ribs	32	20	22	11
Floating rudimentary ribs	0	5	4	2
Wavy ribs	0	0	0	1

*Statistically different from control at $p<0.05$,
a delivery on day 29,
b abortion on day 21
c 3 dams aborted on day 26, one aborted on day 22 and one delivered on day 29
d one doe was pregnant but had no viable foetuses at terminal sacrifice, e one doe resorbed the whole litter, 10 pups.
f no. of foetuses examined for gross malformations, visceral malformations and skeletal variations
g same foetus cleft lip, umbilical hernia, hydoronephrosis with hydroureter, thick aorta and small gallbladder

In conclusion, following oral administration of propiconazole to pregnant female rabbits, a NOAEL for foetal effects of 250 mg/kg bw/d was established based on resorptions, abortions or early deliveries, and because of the increased incidence of fully formed 13th ribs at 400 mg/kg bw/d.

Other relevant information

A variety of studies on potentially endocrine disrupting effects of propiconazole have been published in the open scientific literature.

The effect of propiconazole exposure on reproduction and maturation of offspring has been studied by Goetz *et al*. (2007). The anogenital distance was significantly increased following exposure to 2500 ppm (144-174 mg/kg bw/d). Testes weights were increased on post-natal day 50 at 500 ppm (53 mg/kg bw/d) and on post-natal day 22 at 2500 ppm (205-413 mg/kg bw/d). Serum testosterone levels were increased at post-natal day 92 at 500 ppm and 2500 ppm. It was proposed that altered steroid homeostasis caused the observed increases in serum testosterone, anogenital distance and testes weights.

In another study (Taxvig *et al*., 2008), oral administration of propiconazole 50 mg/kg bw/d to pregnant Wistar rats from GD 7 to 21 caused a statistically significant increase in serum 17α-hydroxyprogesterone and a small but not significant increase in testosterone and no effect on progesterone or oestradiol levels or on anogenital distance.

Taxvig *et al*. (2008) also addressed the potential of propiconazole to affect male fertility through anti-androgenic effects using Hershberger assay. The serum concentration of follicle stimulating hormone was significantly increased at 150 mg/kg bw/d. It was concluded that propiconazole had no antiandrogenic effects.

Moreover, the study by Tully *et al*. (2006) revealed no effects of 150 g/kg bw/d on testes weights or histology, sperm morphology or motility or any of the serum hormones measured (testosterone, LH, FSH, oestradiol). In contrast, when Wistar male rats were treated with 4 mg/kg bw/d propiconazole from post-natal day 50 to 120, a significant increase in abnormal sperm tail morphology, increased seminal vesicle and vas deferens weight, and decreased serum estradiol levels were observed (Costa *et al*., 2015).

Oestrous cyclicity was disrupted on first two weeks after vaginal opening in rats exposed to 500 ppm propiconazole from gestation day 6 through gestation, parturition, and lactation (Rockett *et al*., 2006). No effects on anogenital distance was reported in this study at doses comparable to those employed in Taxvig *et al*. (2008). The oestrous cyclicity was later normalized. It was concluded that exposure to high concentrations of propiconazole adversely impacted the reproductive development of the female rat. The effects appeared to be either short term or reversible.

Comparison with the criteria

Sexual function and fertility

The only available study assessing the effects of propiconazole on fertility and sexual performance was a 2-generation study showing no effects on mating, fertility, gestation, female and male fertility index and average of gestation length. However, other effects in F_1 and F_2 generations were reported as reductions in body weight, hepatotoxicity, reductions in testes weight and reductions in mean number of live pups (only in F_2).

Reductions in body weight and hepatotoxicity were consistently reported in most of the repeated dose toxicity studies. It suggests that the effects found in F_1 and F_2 of this 2-generation study might be due to systemic toxicity rather than a direct effect on reproduction. Thus, RAC does not consider hepatotoxicity in the F_1 and F_2 generation to be relevant for classification.

A reductions in the weight of the testes was reported for the second litter of both generations. No histopathological assessment of the altered testes was available, but it is notable that the chronic toxicity studies did not report this effect and there were no alterations of sexual and reproductive performance, which makes the biological significance of this finding questionable. Taking into consideration these facts, RAC does not consider the effects on testes relevant for classification.

Finally, reductions in the mean number of live pups of F_{2b} were reported in different period of lactation, these reductions were statistically significant and ranged between 19 and 26%. However, RAC notes that in F_{2a} also reductions in the mean number of live pups were reported but in different periods of lactation and that such reduction were not reported for any of the F_1 litters.

RAC notes that there are several studies in the open scientific literature reporting impairments in serum testosterone levels, testes and foetus weight, anogenital distance, oestrus cyclicity and sperm quality, suggesting endocrine mediated effects. However, RAC also notes that such observations did not alter fertility in the 2-generation Guideline study,that the reported effects are reversible in some cases and finally, effects reported in individual studies were not further confirmed in others with similar approaches. Thus, RAC does not consider the effects reported in these studies to be consistent enough to warrant classification.

In conclusion, RAC supports the DS proposal for **no classification of propiconazole for fertility effects.**

Development

The two available developmental studies in rat reported cases of cleft palate. Cleft palate occurred in 1/302 (0.33%) pups at 90 mg/kg bw/d without significant maternal toxicity, and 2 cases were seen at 360/300 mg/kg bw/d, although at this dose together with severe maternal toxicity (clinical signs). In a second independent study cleft palate was again observed at 300 mg kg bw/d (also together with maternal clinical sings: 17% reduction in corrected maternal body weight gain and 2 mortalities) with an incidence of 2/2061 foetuses (0.097%) from 2/158 litters. According to the CLH report, cleft palate had not been seen previously in the performing laboratory (incidence 0/5431 during 1983-1985) and according to data submitted by the registrant the observed incidences are also above the historical control data of other laboratories during 1983-1986 (4/25522, 0.016%). RAC notes that cleft palate is a serious malformation that should be taken into consideration for classification purposes.

In addition to the cleft palate, other developmental effects were reported in the rat studies. These were skeletal variations (rudimentary ribs and non-ossified sternebrae) at 90 mg/kg bw/d and 360/300 mg/kg bw/d, and increased incidence of urinary tract variations at 360/300 mg/kg bw/d. These visceral findings appeared only at doses exerting maternal toxicity and might be attributable to a secondary consequence of it, while the skeletal findings appeared with both, maternal (360/300 mg/kg bw/d) and non-maternal (90 mg/kg bw/d) toxicity and following a dose-response pattern and therefore should be considered for classification.

Other reported developmental effects were resorptions, abortions, early deliveries and increased incidence of fully formed 13th ribs in rabbits exposed at 400 mg/kg bw/d. However RAC notes that these effects appeared at doses causing maternal body weight gain reductions of 89% and 56% in the periods between 10-14 and 14-20 days of gestation, respectively. RAC considers these effects as additional concerns for classification of developmental toxicity.

There is no information about the potential toxicity of propiconazole for humans and therefore Category 1A is not supported.

A substance can be classified as reproductive toxicant category 1B on the basis of animal studies providing clear evidence of an adverse effect on sexual function and fertility or on development in the absence of other toxic effects, or if occurring together with other toxic effects the adverse effect on reproduction is considered not to be a secondary non-specific consequence of other toxic effects.

Cleft palate is a severe malformation that can be induced by chemicals if the critical dose and timing of exposure are aligned. It has also been suggested that cleft palates could occur as a consequence of maternal toxicity. RAC notes that cleft palate appeared with low incidence, but in two independent studies and in different litters. RAC also notes that cleft palate appeared in the presence of severe maternal toxicity in two studies, but also at 90 mg/kg bw/d in the absence of relevant maternal toxicity and following a dose-response pattern (0.33% at 90 mg/kg bw/d and 0.70% at 300 mg/kg bw/d). These two facts (the appearance in two independent studies and the dose-response) speak against the cleft palates being chance findings and support their association with exposure to propiconazole. Furthermore, the increased incidence of cleft palates in rat has also been observed in response to exposure to other triazoles (e.g. cyproconazole and epoxiconazole).

The mode of action of propiconazole in the observed developmental alterations is not known, but the teratogenicity of triazoles is suggested to be related to altered embryonic retinoid acid catabolism, since abnormalities are confined to structures controlled by retinoid acid. There is no information showing that the mechanism is not relevant for humans and whether human sensitivity is more similar to rabbits (where no cases were reported) or to rats.

RAC noted that the cleft palate appeared only in rats and not in rabbit. However, RAC also notes that some cases might be masked by the post-implantation loss and the reduced number of viable foetuses in the rabbit study.

In conclusion, RAC considers increases in cleft palate incidences found in both rat developmental studies as of human relevance. The following findings also contribute to consider propiconazole as presumable developmental toxicant for humans: 1) skeletal variations at 90 mg/kg bw/d in rat study; and, 2) resorptions, abortions and early deliveries in rabbits exposed to 400 mg/kg bw/d.

RAC consequently proposes propiconazole to be classified as **reproductive toxicant category 1B H360D (May damage the unborn child).**

ENVIRONMENTAL HAZARD EVALUATION

RAC evaluation of aquatic hazards (acute and chronic)

Summary of the Dossier submitter's proposal

Propiconazole has currently the following classification as hazardous to the aquatic environment in Annex VI to CLP: Aquatic Acute 1 (H400) and Aquatic Chronic 1 (H410).

The current DS's proposal for consideration by RAC was Aquatic Acute 1 (H 400) with an M-factor of 1 and Aquatic Chronic 1 (H410) with a separate M-factor of 1. The proposal was based on the substance being not rapidly degradable, non-bioaccumulative and very toxic to aquatic invertebrates and fish regarding acute and chronic aquatic toxicity, respectively. Based on the available acute aquatic toxicity data for fish, aquatic invertebrates and algae, the lowest acute aquatic toxicity value is an EC_{50} of 0.51 mg/L for *Americamysis bahia,* which is between 0.1 and ≤1 mg/L leading to an M-factor of 1. Based on chronic aquatic toxicity data for fish, aquatic invertebrates and algae, the lowest chronic aquatic toxicity value is a NOEC of 0.068 mg/L for *Cyprinodon variegatus,* which is between 0.01 and ≤ 0.1 mg/L leading to an M-factor of 1 for this non-rapidly degradable substance. Consequently, the DS concluded that classification as Aquatic Acute 1, M-factor 1 and Aquatic Chronic 1, M-factor 1 is warranted.

The impurities were taken into consideration by the DS in the classification of this substance but none of them were found to be relevant for the classification.

Degradation

Propiconazole was not significantly hydrolysed when incubated at 70°C for up to 28 d at pHs 1, 5, 7, 9 and 13, and thus, it is considered hydrolytically stable under environmentally relevant conditions. Photolytic half-life was 249 d in a study following GLP principles and EPA subd. N, 161-2 guideline, and therefore, photolysis in water is not considered to be a major degradation pathway.

A ready biodegradation test (OECD TG 301B) resulted in 3% degradation (based on theoretical carbon dioxide) at day 28. On this basis, it is concluded that propiconazole is not readily biodegradable.

In biodegradation simulation studies, the DT_{50} values of propiconazole in aquatic water/sediment systems were 485-636 d for the whole system (Dir. 95/36/EC Annex II: 7.2.1.3.2 and guidelines for the approval of plant protection products, Part IV, 5-1BBA, Germany) and in surface water 78 d or higher (OECD TG 309). In the water/sediment study up to eight minor metabolites were detected with maximum concentrations not exceeding 5% of the applied radioactivity for any of them. In the surface water study, one major metabolite (1-(2,4-dichlorophenyl)-2-(1H-1,2,4-triazol-1- yl)ethanol) was found, reaching a maximum level of 41.8% of applied radioactivity in low dose test vessels (10 µg/L). Mineralisation was only a minor element of dissipation and degradation in the simulation studies. On this basis propiconazole is not considered to undergo rapid ultimate degradation.

Consequently, the DS concluded that propiconazole is considered not rapidly degradable for the purposes of classification.

Bioaccumulation

Propiconazole has a measured log Kow of 3.51-3.8 (EEC A.8, shake flask and HPLC methods), which is lower than the trigger value of 4 for substances with bioaccumulation potential according to the criteria in the CLP Regulation (EC 1272/2008).

A 28 d aquatic bioaccumulation study according to GLP principles and following OECD TG 305 is available. The steady-state whole fish bioconcentration factor for *Lepomis macrochirus* was 180 L/kg.

The DS concluded that on the basis of the bioaccumulation in fish study with BCFs less than 500 L/kg, propiconazole is considered not bioaccumulative for classification purposes.

Aquatic Toxicity

Acute and chronic aquatic toxicity data are available for the three trophic levels (fish, aquatic invertebrates and algae). The aquatic invertebrate *Americamysis bahia* was the most sensitive organism for acute aquatic toxicity and the fish *Cyprinodon variegatus* was the most sensitive organism for chronic aquatic toxicity.

As mentioned above in the degradation section, a major metabolite was observed in the biodegradation test with surface water. The metabolite was demonstrated to be less toxic than propiconazole, and therefore, it was not taken into account further in the classification proposal.

Table. Relevant aquatic toxicity data on propiconazole. The key study values triggering the classification are given in bold.

Method, test substance	**Test organism**	**Conditions**	**Endpoint**	**Toxicity value (mg/L)**	**Reference**
Acute toxicity to fish					
OECD TG 203 92/69/EEC C.1 EPA OPP 72-1	*Oncorhynchus mykiss*	Static nom	96 h LC_{50} 96 h NOEC	4.3 1.0	DAR IIA 8.2.1/02
OECD TG 203	*Leiostomus xanthurus*	Static mm	96 h LC_{50} 96 h NOEC	2.6 0.93	DAR IIA 8.2.1/01
Chronic toxicity to fish					
OECD Draft proposal (2002) OECD TG 229 EPA OPPTS 850.1500 OPPTS 890.1350	*Pimephales promelas*	Flow-through mm	235 d NOAEC (reproduction)	0.188	dRAR IIA 8.2.2.1/01
EPA OPPTS 72-4	*Pimephales promelas*	Flow-through mm	NOEC EC_{10} (wet length)	0.43 0.38	DAR IIA 8.2.2.1/04 dRAR 8.2.2.1/01

			EC_{20} (wet length) EC_{10} (length) EC_{20} (length)	0.47 0.49 0.68	
US EPA OPPTS 850.1500	*Cyprinodon variegatus*	Flow-through mm	NOEC (reproduction) EC_{10} (no. of eggs/d) EC_{20} (no. of eggs/d)	**0.068** 0.06 0.10 **Key study**	DAR IIA 8.2.2.1/02 dRAR 8.2.2.2/01
OECD TG 204	*Oncorhynchus mykiss*	Flow-through mm	21 d LC_{50} 21 d NOEC (lethal effects) 21 d NOEC (non-lethal effects	1.1 0.31 0.31 Supportive study	DAR IIA 8.2.2.1/03
OECD TG 229 OPPTS Guideline 890.1350	*Pimephales promelas*	Flow-through mm	21 d NOEC (no. of eggs/female) (based on visual observation by the Dossier submitter)	0.12	dRAR 8.2.2/02
Equivalent to OECD TG 229	*Pimephales promelas*	Flow-through nom	21 d NOEC (no. of eggs/female)	0.05	Skolness *et al.*, 2013
Acute toxicity to aquatic invertebrates					
OECD TG 202 EPA OPP 72-2 Under GLP conditions	*Daphnia magna*	Static nom	48 h EC_{50}	10.2	Grade, 1999a DAR IIA 8.2.4/01
Test method not specified Validity evaluated under Directives 98/8/EC and 91/414/EEC and EU Regulation 528/2012	*Americamysis bahia*	flow-through mm	96 h LC_{50}	**0.51** **Key study**	Hollister, 1981a DAR IIA 8.2.4/02
Modified ASTM, 2006 E-2455-06	*Lampsilis siliquoidea*	-	96 h EC_{50}	10.01	Bringolf, *et al.*, 2007

chronic toxicity to aquatic invertebrates					
US EPA 1975	*Daphnia magna*	flow-through mm	21 d NOEC	0.31 Supportive study	LeBlanc and Mastone,1981 DAR IIA 8.2.5/01
Method not speicified in the report	*Mysidopsis bahia*	flow-through mm	28 d NOEC	0.114 Supportive study	Hollister, 1981b DAR IIA 8.2.5
OECD TG 211 OCSPP Guideline 850.1000 OCSPP Draft 850.1300	*Daphnia magna*	semi-static mm	21 d NOEC (reproduction) 21 d NOEC (total body length)	0.73 0.37	Fournier, 2014 dRAR IIA 8.2.5.1/01
No standard guideline provided	*Daphnia magna*	-	21 d NOEC (development)	0.5	Kast-Hutcheson *et al.*, 2001
Toxicity to algae and cyanobacteria					
OECD TG 201 EPA OPPTS 850.5400 Commission Regulation (EC) No 761/2009 C.3 JMAFF Test Guidelines, 2-7-7	*Pseudokirchneriella subcapitata*	Static mm	72 h EbC_{50} 72 h ErC_{50} 72 h EyC_{50} NOErC NOEbC, NOEyC	1.6 9.0 1.0 0.46 0.13	Hoger, 2011 DAR IIA 8.2.6/01
ASTM, 1996 Vol 11.05	*Dunaliella tertiolecta*	-	96 h ErC_{50} 96 h NOErC	2.33 0.375	Baird and De Lorenzo, 2010
nom = nominal concentration (measured maintained within 80-120 %) mm = mean measured concentration					

Table. Relevant aquatic toxicity data on the major metabolite CGA091305.

Method, test substance	**Test organism**	**Conditions**	**Endpoint**	**Toxicity value (mg/L)**	**Reference**
OECD TG 203	*Oncorhynchus mykiss*	Static nom	96 h LC_{50}	24	dRAR 8.2.1/01
OECD TG 202	*Daphnia magna*	Static	48 h EC_{50}	110	Wallace, 2001b

		nom			dRAR 8.2.4.1/02
EPA OPPTS 850.5400	*Selenastrum capricornutum*	Static nom	72 h EbC_{50} 72 h ErC_{50}	9.6 19.1	Wallace & Woodyer, 2001 dRAR 8.2.6.1/05
nom = nominal concentration (measured maintained within 80-120 %)					

Fish

Two acute aquatic toxicity tests on fish were included in the CLH dossier, both carried out according to OECD TG 203. The lowest acute aquatic toxicity value for fish was an LC_{50} (96 h) of 2.6 mg/L for *Leiostomus xanthurus*.

Four chronic aquatic toxicity studies and one 21 d prolonged acute study on fish according to different standard guidelines are available. The lowest key study value, according to US EPA OPPTS 850.1500, resulted in a NOEC (reproduction) of 0.068 mg/L for *Cyprinodon variegatus*. In this study the effects of propiconazole on hatching success, survival, growth and reproductive success of first generation (F0) of sheepshead minnow and the hatching success, survival and growth of their progeny (F1) was studied for 100 d (95 d F0 exposure, 91 d post-hatch F0 exposure, five additional days to complete the F1 exposure).

Aquatic invertebrates

Two acute aquatic toxicity tests on aquatic invertebrates were provided. The lowest acute toxicity value is an LC_{50} (96 h) of 0.51 mg/L for the marine mysid *Americamysis bahia.* The test method was not specified but the method applied was claimed to be in conformity with international regulatory requirements for assessing the acute aquatic toxicity of chemicals to shrimps. The validity of the test has been evaluated under Directives 98/8/EC and 91/414/EEC and under EU Regulation 528/2012. The test was done prior to GLP requirements and the report did not include data concerning light conditions and no blank control was used. Nevertheless, as an extended explanation of the test conditions was included in the report, this test was considered by the DS to be reliable and the resulting LC_{50} as the lowest relevant acute value triggering the classification.

Regarding the chronic information on aquatic invertebrates, three chronic aquatic toxicity tests were included in the CLH dossier. The lowest reliable value was a NOEC (21 d) of 0.37 mg/L for body length for freshwater *Daphnia magna* obtained in a test carried out according to the OECD TG 211. 21 d NOEC values obtained in this study for survival, reproduction and total dry weight were 1.5, 0.73 and 0.73 mg/l, respectively. All results are based on mean measured concentrations. No adverse effect was observed for sex ratio. The other two chronic studies on *Daphnia magna* and *Mysodopsis bahia* resulted in slightly lower values (21 d NOEC of 0.31 mg/L and 28 d NOEC of 0.114 mg/l, respectively) but they were only used as supportive information for the classification due to the lower reliability of these studies.

Algae and aquatic plants

One reliable algal study with *Pseudokirchneriella subcapitata* carried out according to OECD TG 201 was included in the CLH dossier. The study resulted in an E_rC_{50} (72 h) of 9.0 mg/L and a NOE_rC (72 h) of 0.46 mg/L.

Other aquatic organisms (including sediments)

Information on other aquatic organisms was also included in the CLH dossier. A study on the sediment-dwelling phase of the midge *Chironomus riparius* was provided. The toxicity of propiconazole (purity 92.4%) was assessed in a full life cycle toxicity test according to GLP and following OECD TG 218 & 219 in a static test for 28 d in two exposure scenarios: spiked water and spiked sediment.
Based on the nominal concentrations, the values for spiked water are as follows:

- Emergence rate: EC_{50} 9.5 mg/L; NOEC 8.0 mg/L.
- Development rate: EC_{50} 35.5; NOEC 4.0 mg/L

Based on the nominal concentrations, the values for spiked sediment are as follows:

- Emergence rate: EC_{50} 123 mg/Kg; NOEC 25 mg/kg.
- Development rate: EC_{50} > 100 mg/kg; NOEC 50 mg/kg

The study is considered not relevant for the classification of propiconazole as it is not a pelagic test. However, the data indicate a low level of toxicity and was included as additional confirmatory information for the aquatic compartment.

Comments received during public consultation

Two MSCAs commented during the public consultation supporting the DS's proposal for the environmental classification and M-factors. In addition, one MSCA expressed general support for the DS's proposal for classification.

During public consultation, additional ecotoxicological information on synergistic effects and potential endocrine disruption effects in fish was provided. RAC evaluated this additional information. However, RAC notes that this new information does not change the classification proposed by the DS.

Assessment and comparison with the classification criteria

Degradation

Propiconazole is hydrolytically and photolytically stable, is not readily biodegradable (3% degradation) and shows slow ultimate degradation in water/sediment and surface water simulation tests (DT_{50} values of 485-636 d and 78 d, respectively). Therefore, RAC agrees with the DS's proposal that propiconazole is considered not rapidly degradable for the purposes of classification and labelling.

Bioaccumulation

Propiconazole has a measured log Kow of 3.51-3.8, which is lower than the trigger value of 4 for substances with bioaccumulation potential according to the criteria in the CLP Regulation (EC 1272/2008). Based on the information provided in the CLH dossier, the substance showed some surface active properties, (47.5 - 59.0 mN/m based on OECD TG 115), which could result in uncertainties in additional estimations such as Log Kow. However, any effect is expected to be low (criterion for surface active substances < 60 mN/m). Furthermore, an experimental BCF and chronic aquatic toxicity data are available.
The steady-state whole fish bioconcentration factor for *Lepomis macrochirus* was 180 L/kg (from a 28 d aquatic bioaccumulation study according to GLP principles and following OECD TG 305). Results were not expressed in relation to lipid normalisation. According to the provided

information, lipid content of experimental organisms ranged from 2.48 to 4.91%. Considering statistical differences of lipid content (20%) between treatment groups, a BCF of 284 L/kg based on the mean of the group with the lowest lipid content (3.17%) was re-calculated by RAC.

RAC agrees with the DS's proposal that propiconazole has a low potential for bioaccumulation based on a measured log Kow of 3.51-3.8 and an experimental lipid normalised BCF in fish of 284 L/kg.

Aquatic toxicity

The major metabolite (1-(2,4-dichlorophenyl)-2-(1H-1,2,4-triazol-1-yl)ethanol) identified in the degradation simulation test for surface water shows less toxicity than propiconazole, and therefore, the classification proposal is based only on propiconazole.

Reliable acute and chronic aquatic toxicity data on propiconazole is available for all three trophic levels.
Based on the available acute aquatic toxicity data for fish, aquatic invertebrates and algae, the lowest relevant acute aquatic toxicity value is a LC_{50} (96 h) of 0.51 mg/L for *Americamysis bahia.* This is below the classification threshold of 1 mg/L and in the range of $0.1 < L(E)C_{50} \leq 1$ mg/L leading to an acute M-factor of 1.

Based on the available chronic aquatic toxicity data for fish, aquatic invertebrates and algae, the lowest relevant chronic aquatic toxicity value is a NOEC of 0.068 mg/L for *Cyprinodon variegatus*. This is below 0.1 mg/L, which is the classification threshold for category Chronic 1 for non-rapidly degradable substances, and in the range of $0.01 < NOEC \leq 0.1$ mg/L) leading to a chronic M-factor of 1.

Additional long-term information on fish was provided during the public consultation. Skolness *et al.* (2013) showed potential endocrine activity of propiconazole on *Pimephales promelas* after 21 d exposure conditions similar to OECD TG 229. The cumulative number of eggs per female was significantly reduced at propiconazole concentrations of 1.0, 0.50, 0.05 and 0.005 mg/L. However, at the concentration of 0.05 mg/L no significant effect was observed. Hence, it was not possible to conclude that the effect observed at 0.005 mg/L was caused by exposure to propiconazole. Consequently, the 21 d NOEC was considered to be 0.05 mg/L. Neither fertility nor hatching success of the deposited eggs were affected by propiconazole. Endocrine disruption *per se* is of no relevance for classification according to the current EU system, whereas the observed effects on reproduction (number of eggs) are relevant. However, as the test followed the method of a screening assay, it is only used as supportive information by RAC, and other available long-term tests were considered to be of higher relevance. Therefore, the value considered by the DS (NOEC of 0.068 mg/L for *Cyprinodon variegatus*) was considered as the lowest relevant chronic value for classification.
RAC notes that there is acute information for the chronically most sensitive trophic level, *i.e.* fish, but not for the chronically most sensitive species (*Cyprinidon variegatus*), which has chronic values one order of magnitude lower than the other fish. Acute data on this species could potentially influence the acute M-factor. However, no effects on embryo hatching or post hatch survival were observed at 0.55 mg/L in the chronic Fish Full Life Cycle (FFLC) study. As this value is higher than the LC_{50} of 0.51 mg/L from the mysid shrimp study, which is used for the acute classification, it does not seem probable that further testing would provide relevant additional information.

Two additional studies on aquatic invertebrates were provided during the public consultation regarding synergistic effects. However, in the opinion of the RAC this new information does not affect the conclusions on the classification as proposed by the DS.

During the public consultation, a study following the guideline ASTM 1996 Vol. 11.05 on the alga *Dunaliella tertiolecta* was provided. The study resulted in a 96h E_rC_{50} of 2.33 mg/L and a 96 h NOE_rC of 0.375 mg/L. However, the reliability of the results could not be fully assessed based on the provided information, e.g. it was not mentioned whether the results were based on measured or nominal concentrations. Therefore, the study is only used as supportive information.

Furthermore, RAC noted that the biocides dossier PT 8 of propiconazole included a study on *Scenedesmus subspicatus* with an EC_{50} of 0.058 mg ai/L and a NOEC of 0.016 mg ai/L. However, the test was carried out with a propiconazole formulation, and the composition of that formulation has since changed. The *Pseudokirchneriella subcapitata* algae study with the active ingredient (propiconazole), which is included as the key algae study in the classification proposal, was performed at a later stage and was used for the biocides dossiers PT7 and PT9 of the substance. Consequently, RAC agrees with the DS that the earlier *Scenedesmus subspicatus* study is not relevant for the current classification proposal.

RAC noted that the dRAR of propiconazole included a *Xenopus laevis* study (OECD TG 231), which resulted in a 21 d NOEC of 0.056 mg/L for metamorphosis. RAC took the *Xenopus laevis* study into account as a supportive study since valid data for other species at the same trophic level shall also be considered, also according to the CLP guidance. RAC noted that it supports the DS's proposal for the environmental long-term (chronic) hazard classification and M-factor.

Conclusion on classification

Based on the above information, RAC agrees with the DS's proposal that propiconazole fulfils the classification criteria for **Aquatic Acute 1 (H400)** with an **M-factor of 1** and **Aquatic Chronic 1 (H410)** with an **M-factor of 1**.

Additional references

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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).