CLH report

Proposal for Harmonised Classification and Labelling

**Based on Regulation (EC) No 1272/2008 (CLP Regulation),
Annex VI, Part 2**

Substance Name: Tetrahydrofurfuryl alcohol (THFA)

EC Number: **202-625-6**

CAS Number: **97-99-4**

Index Number: **603-061-00-7**

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Part A.

Part A.

1 PROPOSAL FOR HARMONISED CLASSIFICATION AND LABELLING

1.1 Substance

Table 1: Substance identity

Substance name:	***Tetrahydrofurfuryl alcohol***
EC number:	***202-625-6***
CAS number:	***97-99-4***
Annex VI Index number:	***603-061-00-7***
Degree of purity:	***99.3%***
Impurities:	***Water and an unknown impurity***

1.2 Harmonised classification and labelling proposal

Table 2: The current Annex VI entry and the proposed harmonised classification

	CLP Regulation	**Directive 67/548/EEC (Dangerous Substances Directive; DSD)**
Current entry in Annex VI, CLP Regulation	Eye Irrit. 2 – H319	Xi; R36 SCL: Xi ≥ 10%
Current proposal for consideration by RAC	Repr. 2 – H361fd	Repr. Cat 3; R62-63
Resulting harmonised classification (future entry in Annex VI, CLP Regulation)	Eye Irrit. 2 – H319 Repr. 2 – H361fd	Xi; R36 Repr. Cat 3; R62-63 SCL: Xi ≥ 10%

1.3 Proposed harmonised classification and labelling based on CLP Regulation and/or DSD criteria

Table 3: Proposed classification according to the CLP Regulation

CLP Annex I ref	Hazard class	Proposed classification	Proposed SCLs and/or M-factors	Current classification [1)]	Reason for no classification [2)]
2.1.	Explosives	None		None	Not evaluated
2.2.	Flammable gases	None		None	Not evaluated
2.3.	Flammable aerosols	None		None	Not evaluated
2.4.	Oxidising gases	None		None	Not evaluated
2.5.	Gases under pressure	None		None	Not evaluated
2.6.	Flammable liquids	None		None	Not evaluated
2.7.	Flammable solids	None		None	Not evaluated
2.8.	Self-reactive subst/ mixtures	None		None	Not evaluated
2.9.	Pyrophoric liquids	None		None	Not evaluated
2.10.	Pyrophoric solids	None		None	Not evaluated
2.11.	Self-heating subst/ mixtures	None		None	Not evaluated
2.12.	Subst/mixtures which in contact with water emit flammable gases	None		None	Not evaluated
2.13.	Oxidising liquids	None		None	Not evaluated
2.14.	Oxidising solids	None		None	Not evaluated
2.15.	Organic peroxides	None		None	Not evaluated
2.16.	Subst/mixt. Corr. to metals	None		None	Not evaluated
3.1.	Acute toxicity - oral	None		None	Not evaluated
	Acute toxicity - dermal	None		None	Not evaluated
	Acute toxicity - inhalation	None		None	Not evaluated
3.2.	Skin corrosion / irritation	None		None	Not evaluated
3.3.	Serious eye damage / eye irritation	No change (not re-evaluated)		Eye Irrit. 2 – H319	
3.4.	Respiratory sensitisation	None		None	Not evaluated
3.4.	Skin sensitisation	None		None	Not evaluated
3.5.	Germ cell mutagenicity	**None**		**None**	**Not evaluated**
3.6.	Carcinogenicity	**None**		**None**	**Not evaluated**
3.7.	Reproductive toxicity	**Repr. 2-H361fd**	**None**	**None**	
3.8.	STOT –single exposure	None		None	Not evaluated
3.9.	STOT – repeated exposure	None		None	Not evaluated
3.10.	Aspiration hazard	None		None	Not evaluated
4.1.	Hazardous to the aquatic env.	None		None	Not evaluated
5.1.	Hazardous to the ozone layer	None		None	Not evaluated

[1)] Including specific concentration limits (SCLs) and M-factors [2)] Data lacking, inconclusive, or conclusive but not sufficient for classification

Labelling: Signal word: Warning
Hazard statements: H319; H361fd
Precautionary statements: not harmonised

Proposed notes assigned to an entry: none

Table 4: Proposed classification according to DSD

Hazardous property	Proposed classification	Proposed SCLs	Current classification [1)]	Reason for no classification [2)]
Explosiveness	None		None	Not evaluated
Oxidising properties	None		None	Not evaluated
Flammability	None		None	Not evaluated
Other physico-chemical properties	None		None	Not evaluated
Thermal stability	None		None	Not evaluated
Acute toxicity	None		None	Not evaluated
Acute toxicity – irreversible damage after single exposure	None		None	Not evaluated
Repeated dose toxicity	None		None	Not evaluated
Irritation / Corrosion	No change (not re-evaluated)		Xi; R36 SCL ≥ 10%	
Sensitisation	None		None	Not evaluated
Carcinogenicity	None		None	Not evaluated
Mutagenicity – Genetic toxicity	None		None	Not evaluated
Toxicity to reproduction – fertility	**Repr. Cat. 3; R62**	**None**	**None**	
Toxicity to reproduction – development	**Repr. Cat. 3; R63**	**None**	**None**	
Toxicity to reproduction –Effects on or via lactation	None		None	Not evaluated
Environment	None		None	Not evaluated

[1)] Including SCLs
[2)] Data lacking, inconclusive, or conclusive but not sufficient for classification

Labelling: Indication of danger: Xn
R-phrases: R36, R62, R63
S-phrases: S2, S36/37/39, S46

S2 and S39 were already harmonised with regard to Xi; R36 classification. S36/37 and S46 are also obligatory for substances classified as Repr. Cat. 3.

2 BACKGROUND TO THE CLH PROPOSAL

2.1 History of the previous classification and labelling

The harmonised classification of tetrahydrofurfuryl alcohol (THFA) has been inserted in the 3rd ATP of Directive 67/548/EEC (Directive 81/957/EEC) with the classification Xi; R36. No discussion of THFA classification occurred since then to our knowledge.

2.2 Short summary of the scientific justification for the CLH proposal

When administered to rats, THFA induced effects on sexual organs and function. Effects on testes, characterized by a testicular atrophy with impaired spermatogenic activity, were observed in the repeated-dose toxicity studies (28 and 90 days) and in the reproduction/developmental toxicity screening test. In this last test, no impaired reproduction was reported that could be explained by the fact that rodents produce sperm in numbers that greatly exceed the minimum requirements for fertility and that male exposure to THFA before mating was limited. However, it cannot be excluded that similar effects on testes in humans induce impairment of reproduction. A direct effect on testes (with impaired testosterone synthesis) or a disruption of the hypothalamus-pituitary-gonadal axis may be responsible for these effects. Furthermore, in the OECD 421 study, the mean estrous cycle and the gestation length were prolonged. The prolongation of estrous cycle could suggest a disruption of the hypothalamus-pituitary-gonadal axis but the degree of change was slight and considered not toxicologically significant. The effects on increased gestation length may be due to an impaired hormone synthesis and are considered not to be a secondary non-specific consequence of the other toxic effects.

When administered by oral route to rats, THFA induced effects on development. Increased incidence of resorption or mummification of fetuses was observed in the OECD 421 study and in the developmental study. This is associated with decreased total number of pups born, number of live pups on PND 0 and 4, and delivery and live birth index, and an increased number of dead pups on PND 0 in the OECD 421 study. The effects occurred in the presence of a maternal toxicity including decreased body weight with decreased food consumption and clinical signs. It is not known if the decreased body weight observed in females during gestation could be due to the lack of embryo/fetuses in these groups or due to a direct effect of THFA. In the range finding developmental study, decreased fetal weight was also reported in the absence of maternal toxicity.

In conclusion, a classification for reproductive toxicity category 2 is proposed for fertility endpoint considering that the effects on sexual function were not associated with fertility impairment in OECD 421 guideline study.

A classification for reproductive toxicity category 2 is also proposed for developmental endpoint considering that the effects were observed in a context of maternal toxicity (decreased body weight) and that the low level of information available from these preliminary studies does not allow to conclude on the potential link between maternal toxicity and developmental effects. Besides, uncertainties were raised by potential cannibalisation (abnormal behaviour of the dams or poor health status of the pups).

Finally, it is also important to note that no study reports were available and that the range finding developmental study and the reproduction/developmental toxicity screening test do not provide

complete information due to the relatively small numbers of animals per groups and selectivity of endpoints (for example, no skeletal examination was performed in the screening assay).

2.3 Current harmonised classification and labelling

2.3.1 Current classification and labelling in Annex VI, Table 3.1 in the CLP Regulation

The classification of tetrahydrofurfuryl alcohol is harmonised in Annex VI of CLP under the index number 603-061-00-7 as follows:

Table 3.1 (CLP)
Eye Irrit. 2 – H319

2.3.2 Current classification and labelling in Annex VI, Table 3.2 in the CLP Regulation

The classification of tetrahydrofurfuryl alcohol is harmonised in Annex VI of CLP under the index number 603-061-00-7 as follows:

Table 3.2 (67/548/EEC)
Xi; R36 (SCL ≥ 10%)

2.4 Current self-classification and labelling

No data available.

3 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

Available data show that tetrahydrofurfuryl alcohol has a CMR property, i.e. reproductive toxicity that is not currently harmonised and justify a harmonised classification and labelling according to article 36 of CLP.

Data from repeated dose toxicity are also relevant for assessment of the effects of THFA on fertility and are included in the CLH report in this aim but the classification for repeated dose toxicity *per se* is not further evaluated and not proposed for harmonisation.

Part B.

SCIENTIFIC EVALUATION OF THE DATA

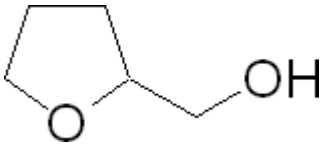
1 IDENTITY OF THE SUBSTANCE

1.1 Name and other identifiers of the substance

Table 5: Substance identity

EC number:	202-625-6
EC name:	tetrahydrofurfuryl alcohol
CAS number (EC inventory):	97-99-4
CAS number:	97-99-4
CAS name:	2-furanmethanol, tetrahydro-
IUPAC name:	tetrahydrofuran-2-ylmethanol
CLP Annex VI Index number:	603-061-00-7
Molecular formula:	$C_5H_{10}O_2$
Molecular weight range:	102.13 g/mol

Structural formula:



1.2 Composition of the substance

Table 6: Constituents (non-confidential information)

Constituent	Typical concentration	Concentration range	Remarks
Tetrahydrofurfuryl alcohol	99.3%	No data	THFA is expected to be essentially racemic. Chiral THFA was not found to be sold on the market.

Current Annex VI entry: the following harmonised classification applies:

According to table 3.2	According to table 3.1
Xi, R36 SCL ≥ 10%	Eye Irrit 2, H319

Table 7: Impurities (non-confidential information)

Impurity	Typical concentration	Concentration range	Remarks
Water	0.1%	No data	No harmonised classification
Unknown impurity	0.6%	No data	

No additives

1.2.1 Composition of test material

Composition of test materials is given in the description of each study when available.

1.3 Physico-chemical properties

Table 8: Summary of physico - chemical properties

Property	Value	Reference	Comment (e.g. measured or estimated)
State of the substance at 20°C and 101,3 kPa	liquid	OECD, 2005	
Melting/freezing point	< -80°C	OECD, 2005	Not specified
Boiling point	177.7°C	OECD, 2005	Measured
Relative density	1.0544 at 20°C	OECD, 2005	
Vapour pressure	1.86 hPa at 25°C	OECD, 2005	Measured
Surface tension	37 mN/m at 25°C	Merk index	
Water solubility	250 g/L at 20°C	OECD, 2005	Measured
Partition coefficient n-octanol/water	-0.11 at 20°C	OECD, 2005	Measured
Flash point	75°C	OECD, 2005	No flammable classification required as flash point > 55°C but data are presented here for information only as physical properties are not proposed for harmonisation in this dossier.
Flammability	Lower flammable limit in air: 1.5% (by volume) Upper flammable limit in air: 9.7% (by volume)	Merk index	No flammable classification required as flash point > 55°C but data are presented here for information only as physical properties are not proposed for harmonisation in this dossier..
Explosive properties	Explosive under influence of flame. Lower explosive limit: 1.5% Upper explosive limit: 9.7% At 22.2 to 50°C.	OECD, 2005	Method used to perform the test not provided Data from different MSDS indicate that above 65-75°C explosive vapour/air mixtures may be formed. Not enough data at this stage to conclude about explosive classification but data are presented here for information only as physical properties are not proposed for harmonisation in this dossier
Self-ignition temperature	282°C	OECD, 2005	
Oxidising properties	No data	No data	No data

			Physical properties are not proposed for harmonisation in this dossier.
Granulometry	Not applicable	Not applicable	Not applicable
Stability in organic solvents and identity of relevant degradation products	No data	No data	No data
Dissociation constant	No data	No data	No data
Viscosity	6.24 mPa.s at 20°C	OECD, 2005	

2 MANUFACTURE AND USES

2.1 Manufacture

Tetrahydrofurfuryl alcohol is produced by catalytic hydrogenation of furfuryl alcohol in a closed reactor tank followed by distillation. Residual non-reacted raw material and the substance are recovered from the reactor tank and applied for re-distillation and/or incineration.

2.2 Identified uses

The major uses of tetrahydrofurfuryl alcohol are as follows (OECD, 2005):

- Intermediates for industrial raw materials (30-50%: esterification products as a counter compound with various carboxylic acids)
- Solvents for fats, waxes, resins in organic synthesis
- Solvents for dyes for leather, chlorinated rubber and cellulose esters; solvent-softener for nylon; vegetable oils; coupling agents
- Plasticizer for synthesis of lysine, paints and varnish ingredient
- Solvents for specialty uses as nail cleaning agents and paints strippers; replacement for chlorinated solvents; crops sprays; water-based paints; dyeing and finishing of textiles and leathers; intermediate in pharmaceutical applications.
- Coupling solvents for pesticides and textile auxiliaries

3 CLASSIFICATION FOR PHYSICO-CHEMICAL PROPERTIES

Not evaluated in this dossier.

4 HUMAN HEALTH HAZARD ASSESSMENT

4.1 Toxicokinetics (absorption, metabolism, distribution and elimination)

No available information on toxicokinetics of THFA (OECD, 2005).

4.2 Acute toxicity

Not evaluated in this dossier.

4.3 Specific target organ toxicity – single exposure (STOT SE)

Not evaluated in this dossier.

4.4 Irritation

Not evaluated in this dossier.

4.5 Corrosivity

Not evaluated in this dossier.

4.6 Sensitisation

Not evaluated in this dossier.

4.7 Repeated dose toxicity

4.7.1 Non-human information

4.7.1.1 Repeated dose toxicity: oral

Several repeated-dose toxicity studies were performed with THFA. Although contacts with the lead registrant under REACH for THFA occurred (between June and October 2010) in order to obtain available relevant data and study reports, only a position paper was provided (Piccirillo, 2010). Therefore, all the data above were issued from the IUCLID obtained in the context of the HPV program of OECD (SIAM 20, 19-21 April 2005). As study reports were not available, this explains that some details are missing.

THFA was administered by gavage to 4 groups of 5 male and 5 female rats at dose levels of 10, 40, 150 and 600 mg/kg bw/day for 28 days (MHLW, 2004). Concurrently, control groups (5 males and 5 females) were exposed to the concurrent vehicle (distilled water). Five further animals per sex, in the control and highest dose groups, were killed after a 14-day recovery period.

No mortalities related to treatment were observed. The animals showed signs of neurotoxicity: increased locomotor activity in females at 150 mg/kg/day, followed by decreased locomotor activity and adoption of a prone position in both sexes at 600 mg/kg/day. Decreased grip strength of the hindlimb was also observed in males at 600 mg/kg/day. Body weight gain was suppressed in males at 600 mg/kg/day and associated with decreased food consumption. Food consumption was also decreased in females at the highest dose only in the first week of the administration.

Several parameters were modified at 600 mg/kg/day: decreased urinary pH in males, variation in haematological dosages (decrease in mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, leukocyte and platelet counts and prolongation of prothrombin time in males and females; decrease in reticulocyte count in males and haemoglobin concentration in females) and in biochemical parameters (decrease in alkaline phosphatase, total protein, albumin, total bilirubin and calcium in both sexes; decrease in lactate deshydrogenase, triglyceride and sodium and increased blood urea nitrogen in males). At 150 mg/kg/day, only significant decrease in total protein was observed in males. After the recovery period, effects on haematological parameters were no more observed and only a significant decrease in calcium is noted in males and females at 600 mg/kg/day.

Variations in organ weights were also reported: relative decreases in thymus weights in both sexes, in pituitary weights and kidneys weights in females at 600 mg/kg/day. Decreases in other absolute organ weights including testes and epididymides were noted in males (see table) but were not statistically significant when considering relative organ weight. After the recovery period, changes in organ weights were also observed at the highest dose with a greater magnitude for reproductive organs in males although their relative weight was not significant.

Table 9: Body weight and weight of reproductive organs in males

Dose (mg/kg/day)	0	10	40	150	600
Examination after the 28-day exposure					
Body weight (g)	357±30	348±28 (-3%)	362±43 (+1%)	326±16 (-9%)	290±28** (-19%)
Testes wt (g)	3.50±0.33	3.17±0.28 (-10%)	3.49±0.33 (-)	3.21±0.20 (-8%)	2.78±0.24** (-21%)
Epididymides wt (g)	0.85±0.05	0.89±0.11 (+5%)	0.83±0.08 (-2%)	0.78±0.03 (-8%)	0.68±0.06** (-20%)
Examination after the 14-day recovery period					
Body weight (g)	420±10	-	-	-	355±39** (-15%)
Testes wt (g)	3.33±0.12	-	-	-	2.47±0.52** (-26%)
Epididymides wt (g)	1.05±0.07	-	-	-	0.79±0.09** (-25%)

** $p<0.01$

At necropsy, thymus, spleen and testes were the main target organs. At the highest tested dose, atrophy of the thymus was observed in both sexes and atrophy of the red pulp and inflammation of the capsule in the spleen was reported only in males. Higher incidence of extramedullary hematopoiesis was observed in the control group than in the treated groups (severity -/+ in 0, 2, 1, 3, 5 animals out of 5 at 0, 10, 40, 150, 600 mg/kg/j and severity ++ in 5, 3, 4, 2, 0 animals out of 5 at 0, 10, 40, 150, 600 mg/kg/j respectively). Slight necrosis of the seminiferous tubular epithelium of the testes was also observed from 150 mg/kg/day (2/5 males at 150 mg/kg/day and all males at 600 mg/kg/day) and associated with a decrease in the ratio of the spermatid to Sertoli cell counts at the highest dose. The effects on the testes were also present after the recovery period (5/5 males with slight necrosis at the highest tested doses compared to 0/5 males in the control group).

Four groups of 15 male and 15 female rats were fed with THFA in diet at dose levels of 0, 1000, 3000 and 10,000 ppm (approx. equivalent to 0, 70, 215 and 720 mg/kg/d in males and 0, 90, 275 and 925 mg/kg/d in females, considering the default values for body weights and food consumption reported in the TGD on Risk Assessment) for 90 days (TSCA, 1991). Slightly decreased body weight gain was observed at 1000 ppm but was only significant at 3000 and 10,000 ppm (no values given and affected sexes not specified). At 10,000 ppm significantly decreased absolute and relative testes weights and testes to brain weight ratio were reported (no values given) and associated with moderate testicular degeneration in 14 animals. These animals exhibited complete loss of spermatogenic activity and their seminiferous tubules were partially to completely lined with a single layer of Sertoli cells. Tubules were also reduced in size. No more details were available in the OECD IUCLID on the magnitude of the effects or on the occurrence of other effects.

Groups of 20 male and 20 female rats were fed with THFA in diet at dose levels of 0, 500, 1000, 5000, 10,000 ppm (approx. equivalent to 0, 35, 70, 360 and 720 mg/kg/d in males and 0, 45, 90, 460 and 925 mg/kg/d in females, considering the default values for body weights and food consumption reported in the TGD on Risk Assessment) for 90 days (TSCA 1992a). Significantly decreased body weight gains were observed in males at 1000 ppm in weeks 5, 6, 7 and 8-13 and at 5000 and10,000 ppm during weeks 1-13 and in females at 10,000 ppm during weeks 8-13 (no values given). Several organ weights were statistically decreased in males, including brain, liver, adrenal glands, seminal vesicles, epididymides, prostate and testes (no values given). When relative weights were considered, relative liver weights were significantly decreased in all groups of treated males, brain and kidney relative weights were significantly increased and epididymides relative weights significantly decreased from 5000 ppm. Testes relative weights were also significantly decreased at 10,000 ppm. In females, the absolute brain weight was significantly decreased at 10,000 ppm. Relative kidney weights were significantly increased at 5000 ppm, relative liver weights from 5000 ppm and relative ovaries weight at 10,000 ppm. However, no information on histopathological examination is available. Additionally, changes in biochemical parameters in males from 1000 ppm and in haematological parameters in females were observed (dose not given).

Four groups of 4 male and 4 female Beagle dogs were fed with THFA in diet at dose levels of 0, 1000, 3000 and 6000 ppm (approx. equivalent to 0, 44, 131 and 262 mg/kg/d in males and 0, 62, 186 and 372 mg/kg/d in females considering the default values for body weights and food consumption reported in the TGD on Risk Assessment) for 90 days (TSCA, 1991). Significant decreased body weight gains were observed in 2 male and female animals in the 6000 ppm group. However, it is considered in the study report that "*it is difficult to conclude if this is truly a test related effect, since the animals were housed together in groups of four throughout the study. When dogs are grouped housed, generally one or two animals will dominate the others; thus limiting the intake of water and food of the non-aggressive animals*". Testes weights were significantly lower in all male treated groups (no values given). This is associated with severe testicular atrophy in all males at 6000 ppm and decreased spermatogenic activity, interpreted as a prodromal sign of atrophy, at 3000 ppm. There was also occasional prostatic atrophy in the 6000 ppm group. However, the test report considers that "*it is difficult to conclude whether or not the lower tested weights and atrophy are the result of test compound administration or sexually immature dogs. Because it appears randomization was not done and therefore the larger, older male dogs were assigned to the untreated control group, it is believed these findings in the group males are associated with sexual immaturity."* No more details were available in the OECD IUCLID on the

magnitude of the effects or on the occurrence of other effects. Even if the relevance of the testicular effects is doubtful, the findings were similar to those observed in the repeated-dose toxicity studies in rats. In absence of additional information on the age and weight of the animals assigned to each group at the beginning of the study, it is not clear whether testicular effects may be attributed to sexual immaturity at the highest doses due to study design or whether THFA may induce sexual immaturity or exert its toxicity on the male reproductive tract. Besides, decreased spermatic activity was also observed at the dose of 3000 ppm where no difference in body weight gain compared to controls was reported.

Groups of 4 male Beagle dogs were fed with THFA in diet at dose levels of 0, 200, 400 and 800 ppm (approx. equivalent to 0, 9, 17 and 35 mg/kg/d, considering the default values for body weights and food consumption reported in the TGD on Risk Assessment) for 90 days in a testicular maturation study (TSCA, 1991). One animal at 200 and 400 ppm exhibited a decrease in absolute and relative testes weights with little to no spermatogenesis. Sexual immaturity was reported as the probable cause for these findings but no details were given in the OECD IUCLID to justify this hypothesis. In addition, no dose-response relationship was observed since normal testicular development was noted at the highest dose. The doses used in this study were however lower that in the previous study.

Table 10: Summary of oral repeated-dose toxicity studies

Guideline	**Route**	**duration of study**	**Species Strain Sex no/group**	**dose levels frequency of application**	**Results**	**LO(A)EL**	**NO(A)EL**	**Reference**
Guideline for the 28-day repeated dose toxicity test in mammalian species (Chemical Substances Control Law of Japan)	Oral gavage	28 days + 14-day recovery period (control and highest dose)	Rat Crj:CD(SD)IGS 10/sex (control and high dose) 5/sex (other doses)	0, 10, 40, 150, 600 mg/kg/d Daily	- Clinical signs - ↓ bw gain in ♂ - Changes in haematological and biochemical parameters - Atrophy of the thymus - Necrosis of seminiferous tubular epithelium of the testes	150 mg/kg/d	40 mg/kg/d	MHLW, 2004 (In OECD, 2005)
No data	Oral diet	90 days	Rat Strain unknown 15/sex/group	0, 1000, 3000, 10 000 ppm	- ↓ bw gain - Testicular degeneration with loss of spermatogenic activity	3000 ppm	1000 ppm	TSCA, 1991 (In OECD, 2005)
No data	Oral diet	90 days	Rat Strain unknown 20/sex/group	0, 500, 1000, 5000, 10 000 ppm	- ↓ bw gain - ↓ testes and epididymides weights - Changes in haematological and biochemical parameters	1000 ppm	500 ppm	TSCA, 1992a (In OECD, 2005)
No data	Oral diet	90 days	Beagle dogs 4/sex/group	0, 1000, 3000, 6000 ppm	- ↓ bw gain - Testicular atrophy and ↓ spermatogenic activity	3000 ppm	1000 ppm	TSCA, 1991 (In OECD, 2005)
No data	Oral diet	90 days	Beagle dogs 4 males	0, 200, 400, 800 ppm	- Testicular effects but no clear dose-response relationship	n.a	n.a	TSCA, 1991 (In OECD, 2005)

4.7.1.2 Repeated dose toxicity: inhalation

THFA was administered by whole body inhalation to groups of 14 male and 10 female rats at concentration levels of 0, 50, 150 and 500 ppm for 13 weeks (at least 65 exposures), 6 hours per day, and five days per week (TSCA, 1995a). After 34 exposures, 4 males per group were killed for assessment of spermatogenic endpoints. The remaining 10 animals per sex per group were killed after 65 exposures. No treatment-related mortality was observed. The predominant clinical finding was intermittent dose-related whole-body spasma, which were observed frequently in all exposed groups. Hyperactivity was also noted in a dose-related manner one hour after exposure in all exposed groups. Occasional incidence of hypoactivity and excessive grooming were observed for a few animals of each sex at the highest concentration. At this same concentration, wet yellow urogenital matting and a low incidence of salivation were noted. Mean body weight gains decreased several times in males throughout the study leading to decreased mean body weights of 9.2 and 13.3 % lower in the mid and high exposure groups, respectively than in controls. This is associated with decreased food consumption. No changes in body weight or food consumption were reported in females. Variation in haematological parameters consisted in decreased platelet and haemoglobin means (both sexes at the highest concentration at weeks 3 and 13) and decreased MCH values (males at week 3 and in both sexes at study week 13 at 500 ppm).

At interim and terminal sacrifice, increased incidence of morphologically abnormal sperm was observed at the highest tested concentration (no further details given on the type of abnormalities). Changes in organ weights were also reported (no values given): decreased absolute and relative prostate weight (mid and high exposure groups), decrease in absolute seminal vesicle weights and in absolute and relative epididymides weights (high exposure group). The only microscopic lesion suggestive of a test material related effect was mild multifocal atrophy of the testes in a single male at 500 ppm.

Table 11: Summary of the repeated-dose toxicity study by inhalation

Guideline	Route	duration of study	Species Strain Sex no/group	dose levels frequency of application	Results	LO(A)EL	NO(A)EL	Reference
No data	Whole body inhalation	13 weeks	Rat Strain unknown 14 males and 10 females/group	0, 50, 150, 500 ppm	- ↓ bw gain in males - Clinical signs - Hematological changes - Variation in organ weights - Abnormal sperm	n.a	n.a	TSCA, 1995a and OECD, 2005

4.7.1.3 Repeated dose toxicity: dermal

Undiluted THFA was dermally applied 5 days per week for 90 days to 4 groups of 17 male and 12 female rats at dose levels of 0, 100, 300 and 1000 mg/kg bw/day (TSCA, 1995b). The concurrent control group received 0.9% saline on a comparable regiment at a dose volume equivalent to the highest dose level. After 37 applications (occlusive wrap/binder), 5 males per groups were killed for assessment of spermatogenic endpoints. The remaining 12 animals per sex per group were killed following 13 weeks of treatment (65 applications). All animals survived. No test article related clinical signs were observed. Lower mean body weights and body weight gains were observed in both sexes at 1000 mg/kg bw/day (no values given). Mean food consumption, hematology and serum chemistry parameters and organ weights were unaffected by treatment. Adverse effects on spermatogenesis were found at 300 and 1000 mg/kg bw/day, including a

decrease in mean number of sperm in the testis and in mean sperm production rate (no values given). Additionally, a decreased mean percentage of motile sperm was noted at the highest tested dose (no values given). However, no lesion was reported in the testis, epididymis, seminal vesicles, vas deferens, prostate, or coagulating gland or other organs.

Table 12: Summary of the dermal repeated-dose toxicity study

Guideline	**Route**	**duration of study**	**Species Strain Sex no/group**	**dose levels frequency of application**	**Results**	**LO(A)EL**	**NO(A)EL**	**Reference**
No data	Dermal	13 weeks	Rat Strain unknown 17 males and 12 females/group	0, 100, 300, 1000 mg/kg bw/day	- ↓ bw gain - ↓mean number of sperm in the testis and mean sperm production rate - ↓ mean percentage of motile sperm	300 mg/kg/d	100 mg/kg/d	TSCA, 1995b and OECD, 2005

4.7.1.4 Repeated dose toxicity: other routes

Not evaluated in this dossier.

4.7.1.5 Human information

Not evaluated in this dossier.

4.7.1.6 Other relevant information

Not evaluated in this dossier.

4.7.1.7 Summary and discussion of repeated dose toxicity

The main effects induced by THFA in repeated-dose toxicity studies in rats and dogs are localized in the male reproductive system, and especially in the testes. The findings includes decreased testes weights associated with necrosis of the seminiferous tubular epithelium/testicular degeneration and/or an impaired spermatogenesis (decrease in the ratio of the spermatid to Sertoli cells counts, loss of spermatogenic activity, decrease in mean number of sperm and in mean sperm production rate). Although these effects were clearly observed in the rats, the significance of the testicular findings is doubtful in dogs since they could be due to sexual immaturity.

The other target organs are the thymus (atrophy) and the spleen (atrophy of the red pulp, decreased extramedullary hematopoiesis and inflammation of the capsule in the spleen) and suggest that THFA could have effects on hematological and/or immunological systems. Effect on the hematological system is supported by the changes in haematological parameters.

In addition to these effects, decreased body weight gains associated with reduced food consumption were observed in all studies at the highest doses, in particular in the male rats. In the 90-day study in rats exposed to THFA by gavage, clinical signs that could indicate neurotoxicity were also reported (increased/decreased locomotor activity, prone position, decreased grip strength).

4.7.1.8 Summary and discussion of repeated dose toxicity findings relevant for classification according to DSD

Not evaluated in this dossier. Data from repeated dose toxicity are relevant for assessment of the effect of THFA on fertility and are included in the CLH report in this aim but the classification for repeated dose toxicity *per se* is not further evaluated and proposed for harmonisation.

4.7.1.9 Comparison with criteria of repeated dose toxicity findings relevant for classification according to DSD

Not evaluated in this dossier.

4.7.1.10 Conclusions on classification and labelling of repeated dose toxicity findings relevant for classification according to DSD

Not evaluated in this dossier.

4.8 Specific target organ toxicity (CLP Regulation) – repeated exposure (STOT RE)

Not evaluated in this dossier.

4.9 Germ cell mutagenicity (Mutagenicity)

Not evaluated in this dossier.

4.10 Carcinogenicity

Not evaluated in this dossier.

4.11 Toxicity for reproduction

4.11.1 Effects on fertility

4.11.1.1 Non-human information

In an OECD guideline 421 "Reproduction/Developmental Toxicity Screening Test", 5 groups of rats (n=12/group) received THFA by oral gavage at dose levels of 0, 15, 50, 150 and 500 mg/kg/day (Hirata-Koizumi, 2008). Doses were selected on the basis of a 14-day dose-finding study where rats received THFA at dose levels of 50, 100, 200, 500 or 1000 mg/kg/day.

Males were dosed once daily for 47 days, beginning 14 days before mating and throughout the mating period and females were dosed once daily from 14 days prior to mating and throughout the mating and gestation periods, to day 4 of lactation (total administration period: 42-52 days).

No substance-related clinical signs of toxicity were detected at 15 and 50 mg/kg. Increase and decrease in locomotor activity were observed in 10/12 males and 11/12 females in the 150 mg/kg group (mainly in the first half of the administration period) and in all animals of the 500 mg/kg group (mainly in the first half of the administration period in both sexes and also in the second half of the administration period in males). Vaginal hemorrhage was observed during the late gestation period in 1/11 pregnant female at 150 mg/kg and 2/12 pregnant females at 500 mg/kg, which did not deliver their pups or experienced total litter loss.

Treatment also resulted in a decreased parental body weight. In the 500 mg/kg/day male group, body weight was significantly reduced on day 7 and from day 21 to the end of dosing period. In females, significant reduction of body weight was found on day 20 of gestation at 150 mg/kg/day and on day 14 and 20 of gestation at 500 mg/kg/day. Body weight gain during the whole period of administration in males and during the gestation period in females was significantly decreased in the 150 and 500 mg/kg groups (see table 13). This is associated with transient decreased food consumption in males (at day 21 at 50 mg/kg/day; day 7 at 150 mg/kg/day; days 0, 7 and 21 at 500 mg/kg/day) and in females (at gestation days 14 and 20 at 150 mg/kg/day; premating day 0, gestation days 0, 14, 20 at 500 mg/kg/day).

Table 13: Variation of body weight gains

	Doses				
	0 mg/kg/d	15 mg/kg/d	50 mg/kg/d	150 mg/kg/d	500 mg/kg/d
Males (N = 12)					
Body weight during administration (g)					
Day 0	393 ±17	394 ±17	393 ±14	392 ±17	392 ±16
Day 7	422 ±23	420 ±18	421 ±16	419 ±22	**400 ±18* (-5%)**
Day 14	448 ±28	441 ±21	445 ±18	444 ±24	424 ±21
Day 21	470 ±28	459 ±29	469 ±19	466 ±24	**443 ±19* (-6%)**
Day 28	492 ±31	482 ±22	488 ±21	482 ±21	**458 ±22* (-7 %)**
Day 35	516 ±34	506 ±24	510 ±25	491 ±22	**472 ±28** (-9 %)**
Day 42	536 ±38	524 ±29	523 ±28	505 ±21	**482 ±31** (-10 %)**
Day 46	550 ±40	532 ±29	533 ±27	513 ±21	**489 ±32** (-11%)**
Body weight gain	157 ±29	136 ±19 (-13%)	140 ±25 (-11%)	**122 ±16* (-22%)**	**98 ±23** (-38%)**
Females (N = 12)					
Body weight during premating (g)					
Day 0	236 ±15	234 ±13	232 ±14	235 ±16	234 ±14
Day 7	249 ±14	244 ±13	241 ±14	243 ±20	242 ±15
Day 14	265 ±18	255 ±15	252 ±18	260 ±21	256 ±16
Body weight gain	29±10	21±7 (-28%)	20±10 (-31%)	25±9 (-14%)	22±10 (-24%)
Body weight during gestation (g)					

Day 0	275 ±23	266 ±19	261 ±18	259 ±20	262 ±20
Day 7	317 ±24	304 ±25	300 ±23	301 ±21	297 ±18
Day 14	357 ±23	339 ±26	335 ±27	332 ±21	**322 ±20* (-10%)**
Day 20	438 ±23	422 ±31	411 ±34	**373 ±27** (-15%)**	**320 ±20** (-27%)**
Body weight gain	164±9	156±15 (-5%)	150±18 (-9%)	**114±20* (-30%)**	**58±8** (-64%)**
Body weight during lactation (g)					
Day 0	343 ±19	327 ±28	321 ±26	308 ±17	
Day 4	361 ±22	351 ±34	341 ±28	306	
Body weight gain	18±12	24±13 (+33%)	20±9 (+11%)	3 (-83%)	

* Significantly different from the group control (P< 0.05)

** Significantly different from the group control (P< 0.01)

Organ weights analysis revealed a decreased absolute pituitary weight at 150 mg/kg/d and above in both sexes and a decrease in absolute and relative weight of the thymus, testes and epididymides in males at 500 mg/kg/d. In addition, changes in kidney weight were detected in males and females. In the thymus, the incidence of atrophy was significantly increased at 500 mg/kg/d in males. In the spleen, the incidence of capsule inflammation was significantly increased at 500 mg/kg/d in both sexes and the grade of extramedullary hematopoiesis was significantly decreased at 150 mg/kg/d and above in females. Significant increases in the incidence of seminiferous tubular atrophy and hyperplasia of interstitial cells in the testes and cell debris and decreased sperm in the lumen of epididymides were also detected in males of the 500 mg/kg/d group. Reproductive effects in males are summarized in Table 14.

Table 14: Reproductive parameters in males: organ weights and histopathological findings

			0 mg/kg/d	15 mg/kg/d	50 mg/kg/d	150 mg/kg/d	500 mg/kg/d
No. of males			12	11	12	12	12
Weights of reproductive organs in males							
Testes	Absolute weight (g)		3.41±0.50	3.18±0.83	3.52±0.29	3.40±0.45	**1.77±0.44***
	Relative weight (g)		0.63±0.11	0.60±0.15	0.66±0.07	0.66±0.10	**0.36±0.09***
Epididymides	Absolute weight (g)		1.40±0.20	1.30±0.30	1.38±0.15	1.26±0.17	**0.87±0.15***
	Relative weight (g)		0.26±0.04	0.24±0.05	0.26±0.03	0.24±0.04	**0.18±0.03***
Histopathological findings in reproductive organs in males							
No. of males			12	5	5	5	12
Testes							
Atrophy of seminiferous tubules		+	0	0	0	1	**4**
		++	1	0	0	0	**7**
		+++	0	0	0	0	**1**
Hyperplasia of interstitial cells		+	1	0	0	0	**9**
		++	0	0	0	0	**1**
Epididymides							
Decrease in sperm		+	0	0	0	1	**3**
		++	1	0	0	0	**8**

	+++	0	0	0	0	**1**
Cell debris in lumen	+ ++	1 0	0 0	0 0	1 0	**3** **9**

* Significantly different from the group control (P< 0.01)
+: slight, ++: moderate, +++: severe

Several reproductive parameters were impaired. The mean estrous cycle was significantly prolonged at 500 mg/kg/d. The copulation index, precoital interval and fertility index were not significantly different between the control and THFA-treated groups. All pregnant females at 500 mg/kg/d and 2/11 females at 150 mg/kg/d did not deliver any pups. In these females, total early resorption (1/2 female at 150 mg/kg/d and 12/12 females at 500 mg/kg/d) or mummification of all fetuses (1/2 female at 150 mg/kg/d) were found in the uterus. In the 150 mg/kg/d group, the remaining nine pregnant females began to deliver on days 24-25 of gestation but 5 did not have any pups the next morning. Cannibalisation by the dams was hypothesised. The gestation length in the 150 mg/kg/d group was significantly prolonged. The gestation index was significantly decreased at 150 mg/kg/d and above. The results are summarized in the table below.

Table 15: Reproductive findings

	0 mg/kg/d	15 mg/kg/d	50 mg/kg/d	150 mg/kg/d	500 mg/kg/d
No of pairs	12	12	12	12	12
Estrous cycle (day)[a]	4.3±0.6	4.0±0.1	4.1±0.3	4.5±0.6	**4.8±0.5***
Copulation index (male/female)[b]	100/100	91.7/91.7	100/100	100/100	100/100
No. of pair with successful copulation	12	11	12	12	12
Precoital interval (day) [a]	2.7±1.2	2.5±1.4	2.9±1.2	2.3±1.4	3.7±2.7
Fertility index[c]	100	90.9	100	91.7	100
No. of pregnant females	12	10	12	11	12
No. of pregnant females with parturition	12	10	12	9	0
Gestation length (day) [a]	22.6±0.5	22.7±0.5	22.9±0.3	**24.7±0.7***	
Gestation index[d]	100	100	100	**36.4***	**0***
No. of dams delivering live pups	12	10	12	4	0

[a] Values as the mean ± SD
[b] Copulation index (%) = no. of copulated rats/no. of pairs x 100
[c] Fertility index (%) = no. of pregnant females/no. of pairs with successful copulation x 100
[d] Gestation index (%) = no. of dams with live pups/no. of pregnant females x 100
* Significantly different from the control group

The developmental findings are the following: a significantly decreased total number of pups born, number of live pups on post-natal days (PND) 0 and 4, and delivery and live birth index, and an increased number of dead pups on PND 0 were found at 150 mg/kg/d. At 500 mg/kg/d, no pups were obtained. No effects were observed in the number of corporea lutea and implantations and the implantation index. There was no significant difference in the sex ratio of live pups.

Table 16: Developmental findings

	0 mg/kg/d	15 mg/kg/d	50 mg/kg/d	150 mg/kg/d	500 mg/kg/d
No. of pregnant females	12	10	12	11	12
No. of corporea lutea[a]	17.7±2.1	16.5±2.7	17.8±1.5	16.4±2.0	17.0±2.8
Implantation index[a, b]	88.8±7.4	93.5±7.4	90.7±8.0	84.5±13.1	87.9±23.7
No. of implantation sites[a]	15.6±1.3	15.3±1.9	16.1±1.8	13.7±2.1	14.5±3.7
No. of litters	12	10	12	4	0
Delivery index[a, c]	95.3±7.1	94.7±6.2	91.9±5.9	**46.4±14.0***	
Total no. of pups born[a]	14.8±1.6	14.5±2.1	14.8±1.7	**7.0±1.4***	
Live birth index[a, d]	100±0	100±0	98.8±2.8	**43.1±29.3***	
No. of live pups on PND 0[a]	14.8±1.6	14.5±2.1	14.6±1.8	**3.0±2.2***	
No. of dead pups on PND 0[a]	0	0	0.2±0.4	**4.0±2.2***	
Sex ratio of live pups (male/female)	86/92	72/73	82/93	6/6	
Viability index on PND 4[a, e]	98.9±2.6	99.3±2.1	97.7±3.5	26.7±46.2[f]	
No. live pups on PND 4[a]	14.7±1.6	14.4±2.1	14.3±2.0	**1.3±2.3***	

[a] Values as the mean ± SD
[b] Implantation index (%) = no. of implantation sites/no. of corporea lutea x 100
[c] Delivery index (%) = total no. of pups born/no. of implantation sites x 100
[d] Live birth index (%) = no. of live pups on PND 0/total no. of pups born x 100
[e] Viability index on PND4 (%) = no. of live pups on PND 4/no. of live pups on PND 0 x 100
[f] Value given in the publication, however after recalculation, the viability index on PND 4 should be about 43%
* Significantly different from the control group

No significant difference between the control and THFA-treated groups was seen concerning the body weight of pups on PND 0 and 4. No significant difference in the incidence of pups with internal and external malformations was found.

Table 17: Summary of the OECD 421 study

Route of exposure	**Test type Method Guideline**	**Species Strain Sex no/group**	**Exposure Period**	**Doses**	**Critical effect**	**NO(A)EL Parental**		**NO(A)EL F1**		**NO(A)EL F2**		**Reference**
						m	f	m	f	m	f	
Oral gavage	OECD 421	Rat (Crj: CD (SD)IGS M/F 12/sex/group	Males: From 14 days before mating and throughout the mating period Females:	0, 15, 50, 150, 500 mg/kg/d	Parental: ↓ bw, clinical signs, effects on thymus, spleen and testes, ↑ gestation length, ↑	NOAEL = 50 mg/kg/day						Hirata-Koizumi, 2008

Route of exposure	Test type Method Guideline	Species Strain Sex no/group	Exposure Period	Doses	Critical effect	NO(A)EL Parental		NO(A)EL F1		NO(A)EL F2		Reference
						m	f	m	f	m	f	
			from 14 days prior to mating and throughout the mating and gestation periods, to day 4 of lactation		resorption, ↑ oestrous cycle <u>Pups</u>: ↑ mortality							

4.11.1.2 Human information

No data.

4.11.2 Developmental toxicity

4.11.2.1 Non-human information

In a range finding developmental study, groups of 8 pregnant rats received THFA orally during gestation day 6 to 15 at concentration levels of 0, 10, 50, 100, 500 and 1000 mg/kg/day (TSCA, 1992b).

No maternal mortality or abortions was recorded. Maternal clinical signs, such as impaired mobility, decreased muscle tone of hindlimbs, absence of pain response of hindlimbs, and exophthalmus of both eyes, were observed at 1000 mg/kg/day. Other transient clinical signs included lacrimation of eyes, dried red material around one eye, and/or dried red material around nose.

Treatment also resulted in a decreased maternal body weight gain, noted during gestation day 8 and 15 for groups receiving 1000 and 500 mg/kg/day. The body weight gain continued to decrease in these groups throughout the remainder of the study, with increased statistical significance. This change was associated with decreased food consumption, firstly observed at gestation day 7 and persisted for the most part throughout the remainder of the study.

One hundred percent incidence of early resorptions was observed at 500 and 1000 mg/kg/day and decreased fetal weight was found at 100 mg/kg/day (value not given). Females at 0, 10, 50 and 100 mg/kg/day did not exhibit any early or late resorptions. Thus, fetuses of litters at these dose levels were all viable.

Although not significant statistically, 5 of 124 (4%) fetuses (4 of 8 litters) exhibited an external malformation (filamentous tail) at 100 mg/kg/day. No historical control data was presented in the publication and no relevant information was found in the literature concerning the spontaneous incidence of this type of malformation in rats.

Therefore, the NOAELs for maternal and developmental toxicity were considered to be 100 and 50 mg/kg/day.

Table 17: Summary of the range-finding developmental study

Route of exposure	Test type Method Guideline	Species Strain Sex no/group	Exposure Period	Doses	Critical effects dams fetuses	NO(A)EL maternal toxicity	NO(A)EL Teratogenicity Embryotoxicity	Reference
Oral gavage	No stated	Rat female 8/dose group	Day 6-15 of gestation	0; 10; 50; 100; 500 and 1,000 mg/kg bw/day;	Dams: clinical signs, ↓ bw, resorptions Fetuses: ↓ bw	100 mg/kg bw/day	50 mg/kg bw/day	TSCA 1992b (In OECD, 2005)

4.11.2.2 Human information

No data

4.11.3 Other relevant information

No data

4.11.4 Summary and discussion of reproductive toxicity

THFA induced some parental effects including changes in locomotor activity and inhibition of body weight gain associated with reduced food consumption. The decreased body weight observed in females during gestation in both studies could be due to the lack of embryo/fetuses in these groups and decreased food consumption could be due to the decreased nutritional requirement accompanied with embryonic/fetal loss. However, it cannot be excluded that decreased body weight gain could be due to a direct effect of THFA since this finding was also observed in some repeated-dose toxicity studies at high dose levels.

In the OECD 421 study, the target organs reported in the parental generation are the thymus, the spleen, the testes and/or the epididymides. Effects in the spleen (incidence of capsule inflammation increased and grade of extramedullary hematopoiesis decreased) and in the thymus (atrophy) suggest that THFA could affect the hematological and/or immunological parameters.

In the testes, seminiferous tubular atrophy and hyperplasia of interstitial cells were observed. The following hypothesis is proposed: THFA might impair the synthesis of testosterone leading to an increased LH (luteinizing hormone) levels via negative feedback. This hypothesis is supported by the hyperplasia of interstitial cells which is known to develop with increased LH. Reduced pituitary weight was observed in males and females and could indicate a disruption of the hypothalamus-pituitary-gonadal axis. This is also suggested by the prolonged estrous cycle, however, the degree of change in estrous cycle was slight and most females showed 4 to 5 day estrous cycles. It is unknown whether the disruption of the hypothalamus-pituitary-gonadal axis is secondary to a direct effect on testes or whether the effects on the testes are secondary to a hormonal disruption. It is also noted that an effect on testosterone levels may have an impact on the body growth of males, and *vice versa*.

Despite these effects, no effects on reproductive parameters were noted. This could be explained by the fact that rodent males produce sperm in numbers that greatly exceed the minimum requirements for fertility (sperm production could be reduced up to 90 % without affecting fertility in Sprague-Dawley and Wistar rats). Besides, in the OECD 421 study, exposure of male is limited to two weeks before mating and it is probably not sufficient to affect spermatogenesis and fertility.

THFA also induced effects on the development of rats. In the reproduction/developmental toxicity screening test, total embryonic loss was noted at 500 mg/kg/day. At 150 mg/kg/day, most females delivered (8/11 pregnant females). Besides, only about half of the dams had pups the next day after parturition and the total number of pups born markedly decreased. This could be due to cannibalism but may also mask potential resorptions. Cannibalism can reflect either an abnormal behaviour of the dams (possible neurotoxicity of THFA) or behaviour of the dams resulting from a poor health status of the pups (health status of the missing pups unknown). Among the pups born at 150 mg/kg around half of them were dead and half of the pups born alive died before PND 4. In the range-finding developmental study, 100 % incidence of early resorptions was observed at 500 and 1000 mg/kg/day and decreased fetal weight was found at 100 mg/kg/day. Concerning the incidence of external and internal malformations, there was no difference between control and treated groups in both studies. However, in the OECD 421 study, no skeletal examination was performed and effects on development could not be evaluated at the higher dose since no litter was obtained. In the second study, although not significant, filamentous tail was observed in 5 of 124 fetuses (4 of 8 litters) at 100 mg/kg/day.

4.11.5 Comparison with criteria

The effects observed fulfilled the criteria for reproductive toxicity category 2 (*suspected human reproductive toxicant*) set in the CLP regulation:

"Substances are classified in Category 2 for reproductive toxicity when there is some evidence from humans or experimental animals, possibly supplemented with other information, of an adverse effect on sexual function and fertility, or on development, and where the evidence is not sufficiently convincing to place the substance in Category 1. If deficiencies in the study make the quality of evidence less convincing, Category 2 could be the more appropriate classification. Such effects shall have been observed 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 the other toxic effects."

When administered to rats, THFA induced effects on sexual organs and function:

- Effects on testes, characterized by a testicular atrophy with impaired spermatogenic activity, were observed in the repeated-dose toxicity studies (28 and 90 days) and in the reproduction/developmental toxicity screening test. In this last test, no impaired reproduction was reported that could be explained by the fact that rodents produce sperm in numbers that greatly exceed the minimum requirements for fertility and that male exposure to THFA before mating was limited. However, it cannot be excluded that similar effects on testes in humans induce impairment of reproduction.
  Furthermore, even if the effects on reproductive organs in males were generally reported in the presence of decreased body weight, the following findings are not considered secondary to a general toxicity:
  - Slight necrosis of the seminiferous tubular epithelium of the testes in 2/5 males at 150 mg/kg/day [28-day oral study in rats] in absence of effect on body weight at this dose,

- Adverse effects on spermatogenesis at 300 and 1000 mg/kg bw/day, including a decrease in mean number of sperm in the testis and in mean sperm production rate [90-day dermal study in rats] in absence of effect on body weight at these doses,
- Significantly decreased relative testes weights at 10,000 ppm (≈ 720 mg/kg/d) [90-day oral in rats],
- Decreased epididymides relative weights from 5000 ppm (≈ 360 mg/kg/d) and decreased testes relative weights at 10,000 ppm (≈ 720 mg/kg/d) [90-day oral study in rats],
- Decreased prostate (150 and 500 ppm) and epididymides relative weights (500 ppm) [90 day inhalation study in rats],
- Decreased relative testes and epididymides in males at 500 mg/kg/d [OECD 421 study].

The effects on relative weight of reproductive male organs indicate that the severity of the decrease in these organs is more important that the general body weight decrease. These effects on male reproductive organs are therefore considered not to be a secondary non-specific consequence of the other toxic effects.

- In the OECD 421 study, the mean estrous cycle and the gestation length were prolonged. The prolongation of estrous cycle could suggest a disruption of the hypothalamus-pituitary-gonadal axis but the degree of change was slight and considered not toxicologically significant.

The effects on increased gestation length may be due to an impaired hormone synthesis and are therefore considered not to be a secondary non-specific consequence of the other toxic effects.

When administered by oral route to rats, THFA induced effects on development:

- Increased incidence of resorption or mummification of fetuses was observed at 150 and 500 mg/kg/day in the OECD 421 study and at 500 and 1000 mg/kg/day in the developmental study. This is associated with decreased total number of pups born, number of live pups on PND 0 and 4, and delivery and live birth index, and an increased number of dead pups on PND 0 in the OECD 421 study.

- In the range finding developmental study, decreased fetal weight was also reported.

The effects occurred in the presence of a maternal toxicity including decreased body weight with decreased food consumption and clinical signs. In the CLP regulation, it is written that *"developmental effects which occur even in the presence of maternal toxicity are considered to be evidence of developmental toxicity, unless it can be unequivocally demonstrated on a case-by-case basis that the developmental effects are secondary to maternal toxicity."* In this case, it is not known if the decreased body weight observed in females during gestation could be due to the lack of embryo/fetuses in these groups or due to a direct effect of THFA. However, in the range finding developmental study, decreased fetal weight was reported in the absence of maternal toxicity. "*Moreover, classification shall be considered where there is a significant toxic effect in the offspring, e.g. irreversible effects such as structural malformations, embryo/foetal lethality, significant post-natal functional deficiencies"*. As the main developmental effect observed in the developmental/reproduction studies was a high incidence of resorption, it is considered that this finding fulfill the criteria for classification set in the CLP regulation.

In conclusion, a classification for reproductive toxicity category 2 is proposed for fertility endpoint considering that the effects on sexual function were not associated with fertility impairment in OECD 421 guideline study.

A classification for reproductive toxicity category 2 is also proposed for developmental endpoint considering that the effects were observed in a context of maternal toxicity (decreased body weight) and that the low level of information available from these preliminary studies does not allow to conclude on the potential link between maternal toxicity and developmental effects. Besides, uncertainties were raised by potential cannibalisation (abnormal behaviour of the dams or poor health status of the pups).

Finally, it is also important to note that no study reports were available and that the range finding developmental study and the reproduction/developmental toxicity screening test do not provide complete information due to the relatively small numbers of animals per groups and selectivity of endpoints (for example, no skeletal examination was performed in the screening assay).

4.11.6 Conclusions on classification and labelling

In conclusion, based on the effects observed in reproduction/developmental toxicity screening test and in the range finding developmental study, a classification H361fd "*suspected of damaging fertility or the unborn child for reproductive toxicity*" is proposed.

As no reproductive toxicity study is available by inhalation or dermal routes, it is proposed not to specify route of exposure in the hazard statement.

4.12 Other effects

Not evaluated in this dossier.

5 ENVIRONMENTAL HAZARD ASSESSMENT

Not evaluated in this dossier.

6 OTHER INFORMATION

Information considered in this report was collected by a literature search performed on 30 September 2010.

Contacts with the lead registrant under REACH for THFA occurred between June and October 2010. They were requested to provide relevant data and study reports that were available to them. Only a position paper that review the available data was provided (Piccirillo, 2010). The position paper is attached in the CLH dossier for information.

No registration dossier is available for THFA on 08/08/2011.

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