Committee for Risk Assessment

RAC

Annex 2

Response to comments document (RCOM)

to the Opinion proposing harmonised classification and labelling at EU level of

phenmedipham (ISO); methyl 3-(3-methylcarbaniloyloxy)carbanilate

EC Number: 237-199-0
CAS Number: 13684-63-4

CLH-O-0000001412-86-297/F

Adopted

20 September 2019

ANNEX 2 - COMMENTS AND RESPONSE TO COMMENTS ON CLH PROPOSAL ON PHENMEDIPHAM (ISO);METHYL 3-(3-METHYLCARBANILOYLOXY)CARBANILATE

COMMENTS AND RESPONSE TO COMMENTS ON CLH: PROPOSAL AND JUSTIFICATION

Comments provided during public consultation are made available in the table below as submitted through the web form. Any attachments received are referred to in this table and listed underneath, or have been copied directly into the table.

All comments and attachments including confidential information received during the public consultation have been provided in full to the dossier submitter (Member State Competent Authority), the Committees and to the European Commission. Non-confidential attachments that have not been copied into the table directly are published after the public consultation and are also published together with the opinion (after adoption) on ECHA's website. Dossier submitters who are manufacturers, importers or downstream users, will only receive the comments and non-confidential attachments, and not the confidential information received from other parties.

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Substance name: phenmedipham (ISO);methyl 3-(3-methylcarbaniloyloxy)carbanilate
EC number: 237-199-0
CAS number: 13684-63-4
Dossier submitter: Finland

GENERAL COMMENTS

Date	Country	Organisation	Type of Organisation	Comment number
12.02.2019	France	<confidential>	Company-Manufacturer	1
Comment received				

10. Health hazards -Read-across justification

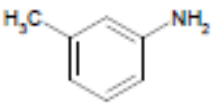
We agree that the classification proposal should only be based on the data on phenmedipham itself, but not with the proposal that this should be supported by read-across from desmedipham and their assumed common metabolites. We disagree with the statement that the chemical structure, chemical properties, breakdown products and toxicological profiles of desmedipham and phenmedipham are similar since it is known that even within the same compound classes between compounds with similar structures differences in their toxicological profiles can occur. From the metabolic pathways shown in the table under this chapter it is obvious that the metabolic pathways show differences so that this does not support the stated similarity of both molecules. Some of the endpoints appear to be similar, like the hematological effects, however, depending from the study type and species, the potency and reference doses are different.

Aniline formation

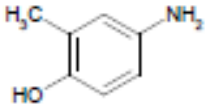
In the CLH report based on read-across it is stated that formation of aniline may happen in the metabolism of phenmedipham, like it is suggested for desmedipham. However, the statement that in the metabolism of desmedipham as one of the metabolites aniline may happen was not confirmed in the ADME studies since aniline was not detected in them. This is also unlikely as stated in the CLH report for desmedipham due to the fact "that in the metabolic pathway of desmedipham, the first metabolite

of the PC ring radiolabelled form was PMC (phenyl methyl carbamate), which was supposed to convert to aniline and then further rapidly to 4-aminophenol and at last acetylated to 4-acetaminophenol. However, aniline was not detected in metabolism studies in rat." Based on the quick conversion to 4-aminophenol apparently no aniline is occurring at measurable amounts if at all which is demonstrated by the fact that no aniline was detected in the ADME studies with desmedipham.

It is stated in the CLH report that "There are slight differences in the substances formed during metabolism between the two substances. The first step of metabolism pathways seems to be slightly different. The –NHCOO- group in between the aromatic rings is metabolised in the first step to –NH2 and HO- in phenmedipham and to –NHCOOCH3 and HO- for desmedipham. However, both substances are suggested to produce compounds which have aromatic amine structure. Some of the identified metabolites are common for both substances such as 3-aminophenol and various acetamidophenols. Phenmedipham is also suggested to produce acetamidocarboxylic and salicylic acids, which are not identified in the toxicokinetic studies of desmedipham. Not detected in the studies but phenmedipham is also suggested to produce aniline."
Like we disagree with the statement that aniline occurs as metabolite of desmedipham as explained before, **we especially disagree with the speculation that phenmedipham could produce aniline.** This is impossible based on the structure of phenmedipham and since it was not demonstrated for desmedipham this speculation can even not be supported by read-across from desmedipham. This is confirmed by our experts with their statement " There is no indication that aniline is formed from phenmedipham. The corresponding counterpart of methyl 3-hydroxyphenylcarbamate (MHPC), which was detected as a major plant metabolite, is m-toluidine (3-methylaniline) and not aniline. M-toluidine was not detected in the ADME nor in livestock metabolism studies, however 4-amino-o-cresol and other possible subsequent metabolites (3-methylacetanilide, 3-aminobenzoic acid) - therefore m-toluidine was proposed as an intermediate metabolite (see structures below).



m-toluidine



4-amino-o-cresol

The formation of aniline from all these metabolites is not reasonable."

Thus, we conclude that neither from desmedipham nor from phenmedipham production of aniline in the metabolic pathway is evident.

Genotoxicity

In the table "Toxicology comparison of desmedipham and phenmedipham" under genotoxicity it states "Negative Ames, Negative OECD 476, Positive OECD 473 (2), OECD 474 (+/-), negative OECD 483 (exposure not shown)". We agree with the statement that "phenmedipham is not genotoxic in vivo.", but the statement in the table that "exposure is not shown" is not correct since in a special study bone marrow exposure was demonstrated as also acknowledged later in the CLH report. Results of a special study on bone marrow exposure are available. Bone marrow exposure was clearly

demonstrated in a mouse quantitative whole body autoradiography study ([redacted]). Thus, the available data provide evidence for the presence of the test substance systemically and in the bone marrow. Furthermore, test substance was detected systemically and in the bone marrow in toxicokinetic studies: In an ADME study ([redacted]), mean plasma levels of methylphenyl-labelled phenmedipham, which were measured 96 hours after administration of a single dose of 1000 mg/kg bw, were 23.6 and 41.0 µg/g in males and females, respectively. Maximum individual levels of up to 68.9 µg/g were reached in females. In bone marrow, mean levels of 1.65 and 2.24 µg/g were measured in males and females, respectively, with maximum individual levels of up to 4.8 µg/g in females.
In addition, toxicity to the bone marrow was shown in the micronucleus test: The guidance states that a decrease in the ratio of polychromatic to normochromatic erythrocytes is sufficient evidence of bone marrow exposure. In one of the micronucleus studies (Kallesen, 1985), phenmedipham was tested at an oral gavage dose of 15,000 mg/kg bw (>7X the currently recommended limit dose). This dose caused a reduced polychromatic erythrocyte count (PCE) which, according to the study director, was evidence that the dose affected erythropoiesis. There was no effect of treatment on micronucleus induction in this study.
These data support the line of evidence that the bone marrow was exposed sufficiently at this high dose level in the micronucleus test as was also stated by the external genotoxicity expert, Bhaskar Gollapudi (see Appendix 1) :
"Collectively, the above lines of evidence clearly indicate that bone marrow was exposed to the test substance in the mouse micronucleus tests on phenmedipham. Since phenmedipham was tested up to a dose level of 15,000 m/kg bw (more than 7X the limit dose for these studies), it is reasonable to conclude that the substance was tested at high enough doses to result in sufficient exposure of bone marrow. The available data to support bone marrow exposure in phenmedipham micronucleus studies are rather exemplary."
We do not agree to the statement that no 'exposure was shown' for the test according to OECD483 in the table "Toxicology comparison of desmedipham and phenmedipham". It is concluded that in the dominant lethal assay in which phenmedipham did not cause chromosomal aberrations, sufficient exposure of the spermatogonia to phenmedipham must have occurred. Based on ADME study results the compound concentrations in testes were 3.0 µg/g tissue which is corresponding to 0.004% of dose as measured 96 hours after administration of 1,000 mg/kg bw of methylphenyl labelled phenmedipham. Since in this study a dose of 15,000 mg/kg bw was administered a much higher concentration in the testes tissues than 3.0 µg/g can be expected. Therefore a sufficiently high exposure can be assumed for this extremely high dose so that the validity of this test is supported by the exposure data.

Based on the above information, also the presentation of the OECD 474 results for phenmedipham as +/- is not correct. The tests were negative and exposure has been shown.

Overall, we agree to the CLH conclusion that there is no evidence of a genotoxic potential which would warrant classification.

Under 10.10.1 Carcinogenicity the CLH report states: Increased incidences of neoplasms in phenmedipham treated rats were observed in two studies. The incidence of endometrial stromal sarcoma was increased at 34 mg/kg bw/day (6% vs. 2% in controls, historical control range of the performing laboratory 0-4%) in the study by [redacted] B.6.5.1/06 M-145589-01-1. In the study (2004 M-240148-01-1 B.6.5.1/07), the incidence of adenomas in the pars distalis of the pituitary in male Wistar rats showed a dose-dependent increase that was found to be statistically significant (time-to-tumour method) at 118 mg/kg bw/day (38% vs. 14% in controls, all animals).

We disagree with the proposed carcinogenicity cat. 2 classification due to the endometrial stromal sarcoma incidences. The incidences for this finding in the Reno, 1980 study in SD rats are shown in the following table:

Overview on endometrial stromal sarcoma incidences

104-week necropsy	Females				HCD[a]	HCD[b]	HCD[c]
Dose (ppm)	0	20	100	500			
N=	50	50	49	50			
Uterine findings							
Endometrial stromal sarcoma: Incidence	1	0	2	3	12/2275		
%	2.0	0	4.1	6.0	0-4.0	0-6	0-18

[a] Historical control data from Covance (formerly Hazleton, Vienna, Va (104-week studies in SD rats between 1980 and 1990
[b] Historical control data from Walsh, K. et al, 1994
[c] Historical control data from a catalogue from Charles River with HCD of Sprague-Dawley rats from studies between 1994 and 1996

It can be seen that the incidence in the control group with 2 % is rather at the higher end of the HCD range so that apparently this batch of animals had a higher background incidence of this finding. This puts the incidence at the highest dose in perspective as this is likely probably not above the HCD of this animal batch. A publication about HCD of endometrial stromal sarcoma incidences in Wistar rats at that time period gives a HCD range of 0-6 % ([redacted]). In a catalogue from [redacted] with HCD of Sprague-Dawley rats from studies between 1994 and 1996 incidences of a maximum of 18 % is given. The published ranges show that the normal variability range in rats of the same and of a different rat strain is higher than the limited HCD in the report. Thus, it seems that in general this finding has a higher background incidence than the limited HCD set given in the report, so that the incidences in the study are regarded as covered by the available HCD data.

Most importantly, a trend test (Exact Cochran-Armitage test from SAS test) which was performed with the incidences in this study (see table above) did not prove a positive trend. This is a very powerful test to check incidences on a dose-related trend and much better than others, like chi-squared test.
The report states:

"The Exact One-sided (increasing linear trend, indicated by a low p-value) and two-sided (increasing or decreasing linear trend, indicated by a low p-value) Cochran-Armitage Trend Tests are displayed.

One-sided Pr <= Z 0.0693
Two-sided Pr >= |Z| 0.1220
The two results from the Exact Test lead us to conclude, that there is no trend associated with the results, i.e. the probability of incidence='Yes' doesn't increase with larger doses. "

Thus no evidence of a dose-related treatment effect exists so that this test proves a lack of a treatment effect on these tumor incidences.

Incidence of adenomas in the pars distalis of the pituitary in male Wistar rats

We disagree with the statement in the CLH report that the incidence of adenomas in the pars distalis of the pituitary in male Wistar rats in the long-term rat study from [redacted] (M-240148-01-1) is regarded as dose-related.

An overview of the incidences of adenomas in the pars distalis of the pituitary, including HCD is given in the following table:

Pituitary, pars distalis	Dose (ppm)				Historical data 1995-2006 (%)
	0	100	500	2500	
Males					
Adenoma (%)	14	14	24	38	12.0 – 45.0
Adenocarcinoma (%)	0	0	0	0	0.0 – 1.9
Focal hyperplasia (%)	12	16	6	16	na
Females					
Adenoma (%)	46	66	52	52	45-0 – 76.7
Adenocarcinoma (%)	6	0	2	4	0.0 – 12.0
Focal hyperplasia (%)	28	20	20	10	na

Na not available

It can be seen that no increased tumor incidence of pituitary adenomas is seen in females and only a slight and spurious increase of the incidences in males. These findings are due to variability and not to treatment. Especially the fact that the incidence of these tumors was not affected in females speaks against a treatment relationship. It is more likely that the incidences are due to a high variability of this finding which is known from other strains, too. This is especially seen in the HCD which were provided from the same laboratory which conducted the study and which demonstrate a high variability of this finding in males between the studies from 12 to 45 % in males and of 45 to 76.7 % in females, despite the fact that all parameters (strain, diet, husbandry etc.) were the same. Also no dose-related increase in the incidence of precursors, like focal hyperplasia or of adenocarcinoma incidences of the part distalis was seen which however would be expected in case of a treatment relationship. Also the fact that the total incidence of benign and malignant tumors was not increased speaks against a treatment relationship, see following overview:

	Incidences							
	Males				Females			
Dose (ppm)	0	100	500	2500	0	100	500	2500
No. of animals	50	50	50	50	50	50	50	50
No. of animals with tumors	31	33	36	35	40	43	42	41
No. of animals with single tumors	17	22	21	24	20	20	22	25
No. of animals with multiple tumors	14	11	15	11	20	23	20	16
No. of animals with benign tumors	30	29	34	35	35	47	41	41
No. of animals with malignant tumors	7	3	5	2	10	7	12	10
No. of animals with metastasising tumors	1	2	2	1	4	3	5	8

Total number of tumors	51	50	56	51	65	78	66	63
Total number of benign tumors	44	47	50	48	53	68	54	53
Total number of malignant tumors	7	3	6	3	12	10	12	10
Total number of metastasising tumors	1	2	1	1	4	4	5	8
% Animals with tumors	62	66	72	70	80	86	84	82
% Animals with single tumors	34	44	42	48	40	40	44	50
% Animals with multiple tumors	28	22	30	22	40	46	40	32
% Animals with benign tumors	60	58	68	70	70	94	82	82
% Animals with malignant tumors	14	6	10	4	20	14	24	20
% Animals with metastasising tumors	2	4	4	2	8	6	10	16

We disagree with the statement in the CLH report that "although no definite conclusion can be drawn it is plausible that the occurrence of these tumours is hormonally related, namely by disturbed homeostasis of the hypothalamus-pituitary-gonad (thyroid) axis".

The alleged hormonal association of the aforementioned tumors is not supported by the toxicology data for phenmedipham, since based on the toxicology studies with PMP there is no evidence of an endocrine potential. The cited effects on some hormone sensitive organs (e.g. decreases in uterus, prostate and thymus weights and increases in adrenals, testes and ovaries weights in rats) do not support a hormonal relationship due to treatment since they were mostly not dose-related and/or secondary to body weight effects or toxicity and a hormonal impact on such different organs from different sexes and thus different hormonal mechanisms appears rather implausible. This is discussed as follows.

Testicular findings

It is stated that the reduced incidences of testicular seminiferous tubular atrophy, of spermatozoa absent degenerate spermatogenic cells in ducts could be associated with the changed incidence of pituitary adenomas. However, the incidences of pituitary adenomas are not regarded as affected by treatment so that a relationship is unlikely. The incidences of the aforementioned non-neoplastic findings were not increased but decreased so that a direct effect of the treatment can be excluded, especially since no other effects in the testes or in the epididymides, especially no negative effects or damages were seen. Also no such findings occurred in the interim sacrifice after 52 week which would be expected if they were directly associated with the treatment. Furthermore, no similar findings occurred in the other long-term studies.

Mammary acinar hyperplasia

Also the association between the reduced incidences of the mammary acinar hyperplasia with the changed incidence of pituitary adenomas is not obvious. The incidences of this finding with 13/30, 8/38, 11/35 and 5/31 for controls and the doses 100, 500 and 2500 ppm, respectively do not show a clear dose- and thus treatment-related effect. Also other findings in the mammary gland which would indicate a negative effect on this organ were not seen and especially negative effects, like increased tumor incidences did not occur. Also in other studies no mammary gland findings indicating a negative impact were noted.

Organ weights of endocrine organs in short-term studies

The DRAR, 2016 suggests that some weights of endocrine organs appear to be changed and thus indicate some endocrine effects in some of the phenmedipham studies. In the following text it is analyzed whether such organs are affected in a dose-related manner in the different studies with phenmedipham.

Pituitary weight:

In a subchronic rat study ([redacted] 1986b, M-145380-02-1) with doses of 0, 150, 500 and 1500 ppm, the slightly decreased pituitary weights in males only (do not show a distinct dose response, the decreased relative pituitary weight at the highest dose is due to a higher body weight in this group as compared to the controls. The following tables give an overview of the pituitary weights of males in subchronic and chronic studies:

Subchronic studies

Study	Pituitary weight (males)	Doses (ppm)											
		0	50	150	400	500	800	1000*/ 1200	1500	3000	5000	10000	20000
Subchronic studies													
[redacted], M-146380-02-1 (SD)	absol (g)	0.013		0.013		0.011			0.010**				
	rel (%)	0.00276		0.00265		0.00231*			0.00203***				
[redacted] 1986a, M-146355-02-1 (SD)	absol (g)	0.0116			0.0107		0.0116	0.0121					
	rel (%)	0.0022			0.0021		0.0023	0.0024					
[redacted] 1981, M-145614-01-1 (F344)	absol (g)	0.008	0.009			0.007					0.008		
	rel (%)	0.003	0.003			0.002					0.003		
[redacted] 2002 M-211096-01-1(Wistar rats)[b]	absol (g)	0.009						0.009		0.008		0.006*	0.005***
	rel (%)	0.0017						0.0019		0.0017		0.0016	0.0013

(SD) = Sprague-Dawley rats
F344 = Fischer 344 rats
a 1000 ppm in study of Foulon, 2002
b Absolute weight reductions due to very high body weight reductions at ≥ 3000 ppm
* $p<0.05$, ** $p<0.01$, *** $p<0.001$

Chronic studies

Study	Pituitary weight (males)	Doses (ppm)			
		0	60	250	1000
Chronic studies					
[redacted] 1980 M-145589-01-1 (SD)	absol (g) week 52	0.0104	0.0102	0.0100	0.0116
	rel (%) week 52	0.0019	0.0017	0.0017	0.0020
	absol (g) week 104	0.0837	0.0445	0.0386	0.0524
	rel (%) week 104	0.0181	0.0082	0.0070	0.0111

This overview demonstrates that in other subchronic toxicity studies with similar doses and also in a long-term study no effects on the pituitary weights were observed. The slightly decreased pituitary weights in the [redacted] 2002 study were caused by severe body weight effects of the very high doses used

in this study as can be seen in the relative weights which did not show such an effect. If it would be assumed that phenmedipham would have such an effect, this should be especially seen in the long-term studies with longer exposure duration, however, this was not the case. Thus, overall from the apical studies, no evidence of an effect on the pituitary weight was obvious. Therefore it is concluded that this pituitary weight change was a spurious chance finding due to variability.

Uterus weight:

In a subchronic rat study ([redacted], 1986a, M-146355-02-1) with doses of 0, 400, 800 and 1200 ppm, the uterus weight reduction is regarded as a spurious chance effect due to high variability and the very high control weight, as compared with uterus weights from other subchronic rat studies. Most importantly, in other subchronic toxicity studies with similar or even higher doses no effects on the uterus weights occurred ([redacted] 1986b, M-146380-02-1; [redacted] 2002, M-211096-01-1). Therefore this was a spurious chance finding due to variability.

An overview of the uterus weights in different studies is given in the following tables:

Subchronic studies

Study	Uterus weight	Doses (ppm)										
		0	150	400	500	800	1000	1200	1500	3000	10000	20000
Subchronic studies												
[redacted] 1986a, M-146355-02-1 (SD)	absol (g)	0.820		0.661		0.563*		0.606*				
	rel (%)	0.309		0.245*		0.212*		0.238*				
[redacted] 1986b, M-146380-02-1 (SD)	absol (g)	0.69	0.87		0.68				0.77			
	rel (%)	0.24	0.32		0.24				0.28			
[redacted] 2002 M-211096-01-1 (Wistar rats)[a]	absol (g)	0.584					0.808			0.533	0.398**	0.393*
	rel (%)	0.224					0.312			0.215	0.184	0.186

(SD) = Sprague-Dawley rats
a Absolute weight reductions due to very high body weight reductions at ≥ 3000 ppm
* $p<0.05$, ** $p<0.001$

Chronic studies

Study	Uterus weight	Doses (ppm)			
		0	100	500	2500
Chronic/ carcinogenicity study					
[redacted], 2004, M-240148-01-1 (HanWistar)	absol (g) week 52	0.848	0.664	0.777	0.915
	rel (%) week 52	0.3271	0.2630	0.2864	0.3794
	absol (g) week 104	0.797	0.651	0.854	0.760
	rel (%) week 104	0.2382	0.1938	0.2587	0.2764

It is obvious from the overview that the other studies do not confirm any treatment-related effect on the uterus weights. The slightly decreased uterus weights in the [redacted], 2002 study were caused by severe

body weight effects of the very high doses used in this study as can be seen in the relative weights which did not show such an effect. If phenmedipham would have an effect, this should be especially seen in the long-term studies with longer exposure duration, however, this was not the case. In addition, the ovary weights were not affected in any study. If it would be assumed that an endocrine effect was responsible for the uterus weight changes, also the ovary weights would be assumed to be changed, however, this was not the case, also not in the study from Everett et al, 1986a as shown in the following table:

Study	Ovary weight	Doses (ppm)			
		0	400	*500*	1200
[redacted], 1986a, M-146355-02-1 (SD)	absol (g)	0.0933±0.0015	0.0935±0.0230	0.0836±0.0100	0.0912±0.0186
	rel (%)	0.0353±0.0060	0.0351±0.0089	0.0314±0.0043	0.0358±0.0081

Thus, overall from the whole study package of apical studies, no evidence of an effect on the uterus or ovary weights was obvious.

Prostate weight:

The DRAR, 2016 remarks that in a subchronic rat study ([redacted] 2002, M-211096-01-1) with doses of 0, 1000, 3000, 10000 and 20000 ppm, the prostate weights were reduced. The doses used in this dose range finding study were extremely high and caused very strong body weight reductions in all doses, but more severely from 3000 ppm on. The mentioned prostate weight reductions were clearly the consequence of this severe body weight effect and thus were only seen at such doses, i.e. from 3000 ppm on.
An overview of the prostate weights in this and in other studies is given in the following table:

Subchronic studies

Study	Organ	Doses (ppm)							
		0	150	500	1000	1500	3000	10000	20000
Subchronic studies									
[redacted] 2002 M-211096-01-1(Wistar rats)	Terminal body weight (g)	490.1			488.3		449.0	375.9***	343.2***
	Prostate weight absol (g)	0.676			0.526*		0.526*	0.376***	0.334***
	Prostate weight rel (%)	0.138			0.107		0.118	0.098**	0.099**
	Testes weight (g)	3.733			3.871		3.785	3.818	3.821
	Rel. Testes weight (%)	0.769			0.809		0.859	1.016***	1.121***
[redacted] 1986b, M-146380-02-1 (SD)	Prostate weight absol (g)	0.77	0.90	0.82		0.85			
	Prostate weight rel (%)	0.16	0.18	0.17		0.17			

* p<0.05, **p<0.01, ***p<0.001

It can be seen that the prostate weight reductions occurred at doses which also caused severe body weight reductions which is the main reason for this finding. If an endocrine effect would have been behind this finding also the testes weights should have been affected which was not the case since the absolute testes weights were not affected by treatment, with an increase of the relative weights as % of the body weight as an indirect arithmetical consequence of the severe body weight reductions.

Overall, these examples show that there is no evidence of an endocrine or hormonal influence on the discussed findings. If changes occurred they were not treatment-related but the consequence of variability in a wide variation range. We therefore conclude that the discussed tumor incidences were not affected by treatment or treatment-related hormonal effects.

An absence of a hormonal influence is furthermore supported by the results of ToxCast and Tox21 activity data for phenmedipham. The results from these systems did not show an agonistic or antagonistic effect on the androgen receptor neither an agonistic or antagonistic effect on the estrogen receptor.

Therefore, a hormonal induction of the tumor incidences by disturbed homeostasis of the hypothalamus-pituitary-gonad (thyroid) axis as stated in the CLH report is not supported by the aforementioned study data. As demonstrated, there is no evidence of an endocrine potential of phenmedipham. The overview of results in hormone sensitive organs (uterus, prostate, adrenals, testes and ovaries) shows that if changes occurred they were mostly not dose-related and/or secondary to body weight effects or toxicity so that a hormonal impact on such different organs from different sexes and thus different hormonal mechanisms appears rather implausible.

ECHA note – An attachment was submitted with the comment above. Refer to public attachment PMP_ECHA commenting_task force_sanitized.pdf

Dossier Submitter's Response

Thank you for your comment. We agree that aniline was not detected in metabolism studies in rat. Aniline was proposed as a intermediate metabolite for desmedipham (desmedipham dRAR B6.1.1/03, B6.1.1/06). Aniline is very transient in nature and may rapidly and completely converted to more polar products. Based on the results from the toxicokinetic study, phenmedipham is metabolised to compounds which have aromatic amine structure. It is well known that aromatic amines have potential to induce haematological effects (e.g. 3-aminotoluene). The classification proposal is based on the data on phenmedipham itself supported by read-across.

FICA thanks the commenter (company/manufacturer) for the very detailed comments. Regarding the in vivo micronucleus studies we agree that the wording on the bone marrow exposure is not the most consistent. While in one test there was no obvious PCE/NCE indicating that exposure might not have occurred, the study conducted in 1985 seems to provide evidence that at high dose there was exposure as demonstrated by the change in PCE/NCE ratio. As you rightly point out, also the toxicokinetic distribution studies show that bone marrow exposure to the substance or its metabolite does occure. Concerning the carcinogenicity findings we agree that they are very borderline and their proposed homonal origin is not clearly demonstrated with the available information.

RAC's response
RAC does not agree with the read-across from desmedipham. There is a relatively rich data base on phenmedipham itself, and the toxicological similarity between the two substances is not sufficient for desmedipham to make a meaningful contribution to the assessment of phenmedipham. As to genotoxicity, in this case it serves only as background information for the carcinogenicity assessment. The RAC assessment is focused on the tumour incidences. Nevertheless, the available data do not raise a significant concern about genotoxicity of phenmedipham. RAC agrees that the increase of the incidence of endometrial stromal sarcomas above the concurrent control in study B.6.5.1/06 is rather weak. In addition, there was no increase in these tumours at higher doses in three other rat carcinogenicity studies. Therefore, endometrial stromal sarcomas are not considered to warrant classification. The incidence of pituitary tumours in male rats (B.6.5.1/07) was statistically significantly increased above concurrent controls and there appears to be a dose-response relationship. This indicates that the tumours might be treatment-related. However, taking into account the benign nature, the high background incidence, the lack of preneoplastic lesions and occurrence in only one sex of one species, RAC agrees that classification is not warranted. The analysis of organ weights is appreciated. As there is no robust mode of action (MoA) information, RAC retains the default assumption of human relevance of any observed tumours.

Date	Country	Organisation	Type of Organisation	Comment number
11.02.2019	Germany		MemberState	2
Comment received				
Our comments refer to phenmedipham. Table 13 (page 27): Please correct the incidences of pars distalis focal hyperplasia in female rats. Currently, the table contains the incidences of pars distalis adenocarcinoma in female rats.				
Dossier Submitter's Response				
Thank you for pointing that out!				
RAC's response				
Thank you, noted.				

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Denmark		MemberState	3
Comment received				
We propose to make it clear that the CLH was only open for carcinogenicity, reproductive toxicity and specific target organ toxicity on human health. Hence, other endpoint such as skin sensitisation and genotoxicity classification has not been taken into consideration. This is important as often the harmonized classification after renewals of pesticides are considered to cover all toxicological endpoints.				

Dossier Submitter's Response
Thank you for your comment. We have followed ECHA´s instructions in order to make clear in the CLH report which hazards classes are opened for public consultation.
RAC's response
Noted.

Date	Country	Organisation	Type of Organisation	Comment number
15.02.2019	France		MemberState	4
Comment received				
FR, page 2: The relevant impurities 3-aminophenol and 3-methylaniline have a maximal content at 1 g/kg in the active substance phenmedipham, according to EFSA conclusion 2018;16(1):5151. The maximum content of 1 g/kg (0.1%) should be added in table 3 for 3-aminophenol and 3-methylaniline.				
Dossier Submitter's Response				
Thank you for your comment.				
RAC's response				
Noted.				

CARCINOGENICITY

Date	Country	Organisation	Type of Organisation	Comment number
11.02.2019	Germany		MemberState	5
Comment received				
The classification Carc. 2, H351 is proposed for phenmedipham based on endometrial stromal sarcomas (malignant) in female SD rats (study 1980) and pituitary adenomas of pars distalis (benign) in male Han Wistar rats (study 2004). The incidence of endometrial stromal sarcoma is increased in female SD rats, where the high dose group (500 ppm) incidence of 6 % is outside the historical control range (0 – 4 % from 1983 - 1990). It is noted that part of the historical control data (HCD) extends to more than 5 years from the conduct of the study (1980). So the HCD should be used with caution. The female survival rate was reduced in the high dose group compared to control (63 % vs. 78 %). This could be interpreted that either the MTD was exceeded which would result in no classification or that the incidences of endometrial stromal sarcomas would be higher than 6 % if the dead female rats had lived longer (Kaplan Meier method) which would result in category 2. For conclusion the time of death would be relevant. Thus, we would appreciate if the dossier submitter could deliver information on the time of death for female rats. However, no increased incidences of endometrial stromal sarcoma were observed in further studies with SD rats up to 1000 ppm (studies 1988b and 1988c). A statistically significant increase of pituitary adenomas of pars distalis were observed in the high dose group (2500 ppm) of male Wistar rats. There were no differences in the incidences in females. The incidence of 38 % at 2500 ppm for males is within the historical control range (19 – 45 % from 2001-2006), but the incidence of the concurrent control group of 14 % is lower than the control range. However, based on HCD it seems to be that there is a high spontaneous incidence of pituitary adenoma in Wistar rats (especially for females) as also stated for F344 rats and SD rats in the CLP Guidance (Version 5.0, 2017) and therefore the increased incidence of pituitary adenoma may not be reliable evidence of treatment related carcinogenicity. Furthermore, at 2500 ppm methemoglobin was increased. Did the dossier submitter consider whether this hematotoxic effect might enhance the spontaneous tumorigenesis in the pituitary gland based on a disturbed delivery of oxygen? A short discussion by dossier submitter would be appreciated.				

From our point of view, as discussed above the increased incidences of endometrial stromal sarcomas in female SD rats and pituitary adenomas of pars distalis in male Han Wistar rats are considered as borderline evidence between category 2 and no classification.

Dossier Submitter's Response

FICA thanks Germany for their comments. We agree that the tumour findings are borderline at best. The time of death of the high dose females was in most cases between weeks 81-105. We have not considered tumorigenesis relating to hematotoxic effects or hypoxia.

RAC's response

Endometrial sarcomas (B.6.5.1/06)
RAC does not consider the slight difference in survival to significantly affect the tumour indicence. The maximum tolerated dose (MTD) was not reached at 500 ppm. As there was no significant increase in the incidence of endometrial sarcomas above concurrent controls in study B.6.5.1/06 and no increase at higher doses in three other rat carcinogenicity studies. RAC does not consider the endometrial sarcomas to contribute to classification.

Pituitary adenomas (B.6.5.1/07)
RAC notes the statistically significant increase above concurrent controls and the apparent dose-response relationship, which indicate that the increase might be treatment-related. However, taking into account the benign nature of the pituitary tumours, the high background incidence, the lack of preneoplastic lesions and occurrence in only one sex of one species, RAC concludes that no classification is appropriate.

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Netherlands		MemberState	6

Comment received

NL agrees with the Dossier Submitters proposal for a Carc. 2 (H351) classification, though considering this as a borderline case.
An increased incidence of endometrial stromal sarcoma was observed in one of the rat carcinogenicity studies. The total incidences are low (i.e. 1/50 (2%), 0/50 (0%), 2/49 (4.1%) and 3/50 (6%) at control, low, mid and high dose group, respectively), though being increased in a dose-related manner and outside the historical control range (0-4.0%). It is noticed that historical control data were collected after this study, which reduces their applicability.
In another rat carcinogenicity study, a statistically significant increased incidence of adenoma of the distal part of the pituitary gland was observed. This was noticed at the high dose group (38% vs. 14% in controls) only and in one sex (male). No malignant tumour type was noted in this tissue. In addition, a comparison with relevant historical control data (19-45%, average 31.6%) further reduces the relevancy of this finding.
Overall, it is considered that the data provide limited evidence for carcinogenicity.

Dossier Submitter's Response

FICA thanks the Netherlands for their comment. We agree that the findings make only a borderline case for classification.

RAC's response

Please see response to comment no. 5.

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Denmark		MemberState	7
Comment received				
We agree that the pituitary adenomas in Wistar male rats and the endometrial stromal sarcoma in Sprague Dawley female rats are treatment related and warrant classification as Carcinogenicity 2, H351: Suspected of causing cancer. The CLH report mentions some genotoxicity studies and in the introductory of section 10.10.1 Phenmedipham is considered not genotoxic. However, the genotoxic potential has not been fully covered by the tests. In one micronucleus test sufficient exposure of the target tissue was not demonstrated and the other was inconclusive. A new micronucleus test was included in the RAR but again sufficient exposure of the bone marrow was not demonstrated. Mice are not the most sensitive species for phenmedipham according to the toxicological dossier, therefore, a test in rats might be more feasible and here bone marrow is considered reached. See the RAR vol. 3 B6 or the EFSA conclusion (EFSA Journal 2018;16(1):5151). The genotoxic potential was therefore considered a datagap in the EFSA conclusion. It is therefore not feasible to refer MoA considerations to these studies.				
Dossier Submitter's Response				
FICA thanks Denmark for their support and comments. We agree that in some studies bone marrow exposure was incompletely demonstrated.				
RAC's response				
RAC does not see a sufficient indication that the endometrial sarcomas in study B.6.5.1/06 are treatment-related: there was no statistically significant increase in this study and no increase in three other rat carcinogenicity studies using higher top doses (1 000 ppm or 2 500 ppm vs 500 ppm in B.6.5.1/06). As to the pituitary adenomas in study B.6.5.1/07, these might be treatment-related considering the significant increase above concurrent controls and the apparent dose-response relationship. However, taking into account the benign nature of the pituitary tumours, the high background incidence, the lack of pre-neoplastic lesions and occurrence in only one sex of one species, RAC does not consider this finding sufficient for classification. As to genotoxicity, here it serves only as background information for the carcinogenicity assessment. Nevertheless, the available data do not raise a significant concern about genotoxicity of phenmedipham.				

Date	Country	Organisation	Type of Organisation	Comment number
15.02.2019	France		MemberState	8
Comment received				
FR, page 29-30: We support the classification Carc. 2 H351 proposed by the MSCA based on the increased incidence of endometrial stromal sarcoma and pituitary adenoma observed in the 2-year studies in rats. It is also noted that, although not assessed in the CLH dossier, a genotoxic potential for phenmedipham was not excluded during the peer-reviewed assessment of the substance under Regulation (EC) No 1107/2009.				

Dossier Submitter's Response
FICA thanks France for their comments and support.
RAC's response
Please see response to comment no. 7.

TOXICITY TO REPRODUCTION

Date	Country	Organisation	Type of Organisation	Comment number
12.02.2019	France	<confidential>	Company-Manufacturer	9
Comment received				

Under point 10.11.4 Adverse effects on development, the CLH report states "We conclude that there is some evidence for developmental toxicity of phenmedipham and therefore classification for Category 2 is appropriate."

We disagree with this conclusion:

Discussion of 'runts'

It is proposed to classify PMP based on an occurrence of runts in a rat developmental toxicity study ([redacted] 1988). In this study, doses of 0, 150, 450 or 1350 mg/kg body weight/day between days 6 and 15 post coitum were administered. This was associated with slightly reduced food consumption at 1350 mg/kg during the whole of the dosing period, minimally reduced food consumption at 150 and 450 mg/kg during the first five days of dosing and a slight dose-related retardation of body weight gain during the dosing period. There were no treatment-related effects on fetal parameters up to the highest dosage tested, 1350 mg/kg body weight/day. Effects on maternal parameters, although minimal, were observed down to the lowest dosage tested, 150 mg/kg bw, but there was no evidence of embryotoxicity or teratogenicity up to and including the highest dosage tested, 1350 mg/kg body weight/day. There was only one runt at 150 and 450 mg/kg bw each and two in the 1350 mg/kg bw group so that no dose response is obvious and thus no treatment effect. Based on the fetal and litter incidences no dose-relationship is obvious as shown in the following table:

Dose (mg/kg bw)	Litters	No. of fetuses	Runt - fetus number (sex)	Litter number	Fetal incidence (%)	Litter incidence (%)
0	24	293	-	-	-	-
150	23	250	331 (m)	33	0.40	4.35
450	21	241	430 (f)	68	0.41	4.76
1350	25	289	148 (f) 149 (f)	76 76	0.69	4.00

There was no dose-relationship, furthermore, it appeared to be associated with a lower average weights of the whole litter. The following table gives an overview of the litter weights of the litters with runts and their weights in comparison with the litter weights of the litters without runts:

Dose (mg/kg bw)	Litters	No. of fetuses	Runt - fetus no. (sex)	Litter number	Runt weight (g)	Average 'runt' litter weights (g)[a]	Other litter weights (g)b
0	24	293	-	-		-	4.6 (4.4-5.3)
150	23	250	331 (m)	33	2.5	4.2	4.6 (4.1-5.0)
450	21	241	430 (f)	68	2.2	4.2	4.7 (4.2-5.2)
1350	25	289	148 (f) 149 (f)	76 76	2.6 2.4	3.8	4.6 (3.8-5.1)

[a] average weights of litters with 'runts'
[b] average weights of males/females (range)

It can be seen that runts occurred as an isolated finding in litters which had lower litter weights compared to the other litters without runts which had litter weights comparable to the controls. Therefore, this was an artificial isolated phenomenon due to variability. Furthermore, the expression 'runt' is very subjective since it is often used for small pups when objective criteria were not found ([redacted] 2003). However, this is not a malformation. As a definition often a fetus is defined 'runt' if the weight is half of the average litter weight. (ECETOC, monograph 31). If this is applied here, it can be seen from the table above that the weights of all fetuses called 'runts' were more than half of the average 'runt litter' weights. Therefore in this study per definition no 'runt' occurred so that this cannot be used to warrant classification. Also in the commenting phase UK denies an association with treatment, it states: "Regarding the developmental toxicity study by [redacted] (1988), the maternal toxicity appears to be more severe than 'very slight' as reported in B.6, since the corrected body-weight gain was reduced by 17% and 36% at 450 and 1350 mg/kg bw/d, respectively. Given this and the absence of a dose-response relationship (1, 1, 1 affected litters at 150, 450, 1350 mg/kg bw/d, respectively, with one small female in the control group), it seems unlikely that the occurrence of runts was a specific developmental effect of phenmedipham."
This finding was also not repeated in another rat study or in two rabbit studies that used equivalent or higher doses.

It was stated in the CLH report that PMP leads to a shift of the sex ratio in the Allen, 1988 study.

The following table gives an overview of the % males and females in the Allen, 1988 study (M-145693-01-1):

Sex ratio of fetuses	Dose (mg/kg bw)			
	0	150	450	1350
Males (%)	46.4	46.8	49.0	51.6
Females (%)	53.6	53.2	51.0	48.4

The table shows that a spuriously slightly higher percentage of males versus females was noted at the highest dose, however the percentages are close to the expected 50 % for both sexes so that this ratio does not appear to be abnormal, further it was not statistically significant. The main reason for this spurious change was the rather low % of males and respectively higher % of females in the control group which made the increase in % males and decrease in % females artificially looking higher. This was mainly due to variability.

That this was no treatment effect is confirmed by the pilot study which was performed in the same laboratory as a dose range finding study for the main study by [redacted] 1988 (M-145777-01-1). The data of the pilot study are summarized in the following table:

Sex ratio of fetuses	Dose (mg/kg bw)	
	0	1000
Males (%)	46.3	40.7
Females (%)	53.7	59.3

The table shows that in this study no increase in % males was observed, just the opposite, the % of males was distinctly reduced and that of the females correspondingly increased although the highest dose was close to the one in the main study. This confirms that there was no increasing or decreasing effect on the percentages of the sexes, but that the observed changes are only due to a high variability.
In addition to this, also no effect on the sex ratios were seen in the other developmental toxicity study with phenmedipham in rats ([redacted] 1989 (M-146392-01-1). The following table gives an overview of the sex ratio of the fetuses in this developmental toxicity study in rats:

Sex ratio of fetuses in rat developmental toxicity studies	Dose (mg/kg bw)			
Svendsen et al, 1989 (M-146392-01-1)	0	625	1250	2500
Males (%)	48.5	46.6	48.8	45.7
Females (%)	51.5	53.4	51.2	54.3

It is obvious that also this study does not confirm an increase in the ratio of males versus females since no increase in the percentage of male fetuses occurred in this study. Thus, the statement in the CLH report that the ratio of female fetuses was reduced and that this is evidence of an endocrine mechanism ([redacted] 1988) is not supported by the other studies with phenmedipham.

Other reproduction aspects

It is concluded that there is no concern for either reproductive or developmental toxicity in the data provided. Although the multi-generation studies did not include all the parameters given as examples in the data requirements of possible effects on male and female reproductive impairment, we consider that an argument can be made that the information provided is sufficient to reach a conclusion. This should include information from the other available repeated-dose toxicity studies; for example, sperm parameters were measured in the 52 and 104 week rat studies ([redacted] 1998b) and no adverse effects were found. Furthermore, there were no effects on fertility indices in any of the multi-generation studies, and so it can be concluded that phenmedipham exposure did not adversely affect fertility in rats when tested at doses that were toxic to the dams.

ECHA note – An attachment was submitted with the comment above. Refer to public attachment PMP_ECHA commenting_task force_sanitized.pdf

Dossier Submitter's Response

Thank you for your comments. Yes, slight to moderate maternal toxicity was evident in all phenmedipham treated groups in the developmental toxicity study RAR B. 6.6.2/02. However, individual data of dams does not show that maternal toxicity (or litter size) would correlate with occurance of runts (CLH report p. 44-45) in this study and runts were reported also in phenmedipham treated group (1000 mg/kg bw/day) of a preliminary range finding study whereas no runts were seen in control litters (see also response to comment

number 10.). Therefore the data does not show that occurrence of runts in phenmedipham treated groups would be secondary non-specific consequence of maternal toxicity.
With regard to your argumentation that this finding would be an artificial isolated phenomenon due to variability and associated with a lower average weights of the whole litter we note the following. In the table included in your comments the mean pup weight of litters where runts occurred is compared to mean pup weight of all litters of the dosage group. The means are calculated from male and female pups together and weights of runts are included. We note that since male pups are generally heavier than female pups, the sexes should be treated separately in this comparison. Moreover, weights of runts should be excluded from the calculation of mean litter weights. Using individual animal data of the study report we calculated mean weights of other male and female pups (runts not included) in litters where runts occurred.

Table. Mean weights of "runt litters" compared to all litters (g) ± s.d

Dose (mg/kg bw/day)	0	150	450	1350
litter/dam		33	68	76
runt weight (g)		2.5	2.2	2.6
				2.4
"runt litter" male weight (g),		**4.7 ± 0.1** [a)]	4.5 ± 0.2	4.3 ± 0.1
"runt litter" female weight (g)		4.4 ± 0.2	**4.1 ± 0.3** [a)]	**3.8 ± 0.2** [a)]
"runt litter" males/females combined weight		4.5 ± 0.2 [a)]	4.3 ± 0.3 [a)]	4.0 ± 0.3 [a)]
all litters male weight (g)	4.7 ± 0.2	**4.7 ± 0.3** [b)]	4.9 ± 0.3	4.7 ± 0.3
all litters female weight (g)	4.5 ± 0.2	4.5 ± 0.3	**4.5 ± 0.3** [b)]	**4.5 ± 0.3** [b)]
all litters weight males/females combined	4.6 ± 0.2	4.6 ± 0.3	4.7 ± 0.3	4.6 ± 0.3

a) runt/runts are not included, b) runt/runts are included

The table shows that mean weights of female pups in mid and high dose "runt litters" were lower than mean weights of females of other litters, whereas mean weight of male pups in low dose "runt litter" was comparable to that of mean weights of males in other litters (in addition to runt pup there were only two male pups in this litter). We agree that all pups defined as runts in the study report do not have body weights half of their littermates. Alltogether, although there are uncertainties which RAC should carefully consider, we consider that the data does not show that occurrence of runts in phenmedipham treated groups in this study would be secondary non-specific consequence of maternal toxicity.
Sex ratios. The proportion of female fetuses was statistically significantly reduced in the high dose group compared to control group (48.4% and 53.7% at high dose and controls, respectively, $p<0.05$, Fischer´s Exact test, CLH report page 44, Table 19). However, we note that proportions of female fetuses (53.7%) and male fetuses (46.4%) in the control group (as well as in the low dose group) are outside the historical control range of the performing laboratory (females 45.4-51.8%) suggesting that this finding may be spurious due to high variability in sex ratios.

RAC's response

Foetal weight distribution shows that the abnormally small foetuses cannot be explained by variability. Foetal weight distribution was approximately normal with a mean of 4.6 g, and the lowest foetal weight in the control group was 3.3 g. 'Runts' were rare in historical controls. Maternal toxicity might appear high when expressed as corrected bw gain (reduction by 37 % at the top dose), but absolute numbers are less remarkable (reduction by 5-6 grams). However, taking into account the very low incidence, the very shallow dose-

response curve and lack of this finding in study B.6.6.2/01 up to 2 580 mg/kg bw/d, RAC agrees that classification is not warranted.

RAC agrees that the altered sex ratio is a spurious finding due to variability.

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Netherlands		MemberState	10

Comment received

NL agrees with the 'no classification' for effects on sexual function and fertility and the 'no classification' for effects on/via lactation.

With respect to adverse effects on development, the uncertainties are noticed. Runts occurred in one rat teratogenicity study (including its preliminary dose range finding study). A second rat teratogenicity study (using higher dose levels) or the generation studies did not reveal such effect.
The runts occurring with the low dose dams may be attributed to the treatment, though this may not be the case for the runts occurring with the high dose dams given the large litter size and the maternal toxicity in these dams (negative corrected body weight gain). On the other hand, it is also noticed that the control group also included a small fetus, thereby reducing the concern of this finding.
It may therefore be questioned whether the data can be considered sufficient for classification.

Dossier Submitter's Response

Thank you for your comments. We agree with your argumentation that occurrence of runts in one study only is limited evidence for developmental toxicity classification. However, we note that in a preliminary range finding study where 5 pregnant females were exposed to 1000 mg/kg bw/day of phenmedipham there were two runts in one litter in the phenmedipham-treated group (one male of 2.6 g and one female of 3.0 g, both with hydrocephaly and the male also had brachygnathia, fetus incidence 3.7%, litter incidence 20%), whereas no runts were seen in the four control litters. Since the data, neither in the preliminary or in the main study, does not allow to conclude that these findings would be secondary non-specific consequence of maternal toxicity we consider category 2 appropriate. Moreover, ossification effects in the main study (RAR B. 6.6.2/02) cause additional concern developmental toxicity.

RAC's response

RAC agrees that no classification is warranted for effects on fertility and on/via lactation.

Regarding the preliminary PNDT study, both the small foetuses were malformed and as malformed foetuses usually have lower weight, it is not clear whether this finding is equivalent to 'runts' in the main study.

The occurrence of 'runts' in the main study cannot be sufficiently explained by maternal toxicity, nor by variability. However, taking into account the very low incidence, the very shallow dose-response curve and lack of this finding in study B.6.6.2/01 up to 2 580 mg/kg bw/d, RAC proposes no classification.

The ossification effects (slightly reduced incidence of non-ossified metatarsalia) are not considered relevant for classification.

ANNEX 2 - COMMENTS AND RESPONSE TO COMMENTS ON CLH PROPOSAL ON PHENMEDIPHAM (ISO);METHYL 3-(3-METHYLCARBANILOYLOXY)CARBANILATE

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Denmark		MemberState	11

Comment received

Development:
We agree that observations of runts in all treatments groups and absence in control groups of the rat developmental study and the preliminary limit test in rats should be considered treatment related and not in the presence of severe maternal toxicity. Hence, classification as Repr. 2; H361d is warranted.
In addition, we think that fertility should also be considered.

Fertility:
We do not agree with non-classification for fertility. As the current toxicological dossier on phenmedipham cannot provide the necessary evidence to exclude effect on fertility, we propose to read-across to desmedipham which is structural similar and possess similar toxicity. A newer study has been provided for desmedipham in which sperm parameters were included and effect on sperm conts observed. Therefore, we would propose to classify phenmedipham on this basis as Repr. 2: H361f.

The generation studies of phenmedipham are of older date and were not conducted according to newer OECD guidelines. They lack information on sensitive parameters including such as sperm related endpoints which were affected by the structural and toxicological analogue desmedipham.
Various sperm parameters have been investigated to some extent in the phenmedipham dossier (but not in the generation studies with different treatment periods) without a clear picture emerging. However, that histopathological examinations (e.g. 2 yr rat study by XXXX 1988b) are not as reliable as sperm counting (sperm number, sperm morphology, and sperm motility) when evaluating potential effects on male reproduction. This is also stated in OECD 43 guidance on assessment of reprotoxicity: "Again, information on the other sperm parameters and histopatholgy should be considered in the overall interpretation. Testicular lesions of sufficient magnitude will be reflected in the sperm counts, but changes in sperm counts should not be discounted in the absence of histological lesions."
Furthermore, according to the same guidance (OECD 43), the fertility index cannot be used as a measure of unaffected sperm: "A reduction in sperm count may not result in reduced fertility, particularly in rodent studies. This is due to the fact that rats and mice have a tremendous excess of spermatozoa in their ejaculates, and as such sperm counts have to be reduced by as much as 90% to affect fertility."

If classification on fertility is not considered warranted by read-across we would propose at least to make clear that the specific phenmedipham data were insufficient to conclude on (datagap).

Dossier Submitter's Response

Thank you for your support regarding developmental toxicity classification. We agree that RAC should carefully consider whether the available data including deviations from guidelines and other deficiencies is sufficient to conclude on fertility effects for phenmedipham. However, we do not consider the reported sperm effects with desmedipham sufficient for classification. Therefore we do not consider classification of phenmedipham on the basis of read across from desmedipham appropriate.

RAC's response
Development Regarding the preliminary study, both the small foetuses were malformed and as malformed foetuses usually have lower weight, it is not clear whether this finding corresponds to the 'runts' in the main study. The occurrence of 'runts' in the main study cannot be sufficiently explained by maternal toxicity. However, taking into account the very low incidence, the very shallow dose-response curve and lack of this finding in study B.6.6.2/01 up to 2 580 mg/kg bw/d, RAC proposes no classification. Fertility The studies with phenmedipham do not report any adverse effects on sexual function and fertility. RAC notes that several sensitive parameters introduced into the OECD TG 416 in 2001 were not investigated for phenmedipham. However, a decision whether this constitutes a data gap is beyond the mandate of RAC. RAC does not consider desmedipham and phenmedipham to be sufficiently similar for read-across of toxicological endpoints (the metabolites are not identical or occur in different proportions, and there are differences in haematotoxicity, thyroid toxicity and developmental toxicity). In addition, RAC agrees with the DS that classification of desmedipham for fertility is not warranted.

Date	Country	Organisation	Type of Organisation	Comment number
15.02.2019	France		MemberState	12
Comment received				
FR, page 40 and 48: There was numerous deficiencies highlighted in the reproductive studies. Indeed, information related to the most sensitive end-points (such as sperm-related endpoints which were affected by the structural analogue desmedipham) is missing. Thus, we are of the opinion that no firm conclusion can be drawn regarding the need for classification for fertility of desmedipham. Regarding classification for developmental toxicity, we agree with the classification proposal of phenmedipham as Repr. 2 H361d based on the high incidence of runts in rats in all treatment groups.				
Dossier Submitter's Response				
Thank you for your comments and support. We agree that RAC should carefully consider whether the available data including deviations from guidelines and other deficiencies is sufficient to conclude on fertility effects for phenmedipham.				
RAC's response				
Fertility The studies with phenmedipham do not report any adverse effects on sexual function and fertility. RAC notes that several sensitive parameters introduced into the OECD TG 416 in 2001 were not investigated for phenmedipham. However, a decision whether this constitutes a data gap is beyond the mandate of RAC. Development Taking into account the very low incidence of 'runts', the very shallow dose-response curve and lack of abnormally small foetuses in study B.6.6.2/01 up to 2 580 mg/kg bw/d, RAC concludes that no classification is appropriate.				

OTHER HAZARDS AND ENDPOINTS – Specific Target Organ Toxicity Repeated Exposure

Date	Country	Organisation	Type of Organisation	Comment number
12.02.2019	France	<confidential>	Company-Manufacturer	13
Comment received				

Under 10.13.3 the CLH report concludes that "Overall, in conclusion, based on haemotoxic effects seen in repeated dose toxicity studies in mice, rats and dogs, classification of phenmedipham for STOT-RE 2 ("H373: May cause damage to organs (blood) through prolonged or repeated oral exposure") is proposed. "

We disagree with this conclusion. The major targets after phenmedipham treatment in rats are slight to moderate changes in red blood cell (RBC) parameters, leading to hemolytic anemia, increased activities of the liver and spleen - the organs mainly involved in the turnover of red blood cells - and compensatory medullar hematopoiesis. As another finding minimal to moderate methemoglobinemia was seen. For these effects in the conducted studies clear thresholds were determined. The most important studies for an assessment of hematological effects and the established NOAELs are discussed in this statement and the mechanism of hemosiderin formation and methemoglobinemia.

Hemolytic anemia:
Under normal conditions at the end of the lifetime of an erythrocyte (approximately 120 days) the erythrocytes are removed from the arterioles and get into the sinus of the spleen where old erythrocytes are destroyed. The erythrocyte fragments are then phagocytized by macrophages mainly in spleen, liver bone marrow and other organs. The iron released from hemoglobin is re-utilized whereas bilrubin and further heme catabolism products are excreted via bile, urine and feces. The released proteins transfer into the pool of amino acids which are also re-utilized. Anemia-causing compounds lead to a disturbance of this cycle after high doses. After a temporary higher hemolysis, like after a hemolytic anemia situation, this catabolism pathway can be temporarily overwhelmed so that an increased formation of iron-containing hemosiderin occurs which can lead to an increased deposition of hemosiderin in the reticulo-histiocytary system (RHS). Mainly spleen, but also liver and kidneys can be affected. Even larger amounts of hemosiderin deposits in tissues do not cause symptoms or impact organ functions. These findings are reversible once the erythrocyte turnover rate returns to the normal physiologic situation.

Methemoglobinemia:
Under normal physiological conditions methemoglobin occurs at approximately 1 % of the hemoglobin content. Methemoglobin, unlike hemoglobin, is not able to bind oxygen. The underlying mechanism is the oxidation of the Fe (II) in the hemoglobin molecule to Fe (III), either spontaneously or by chemicals with such an effect. The enzyme methemoglobin reductase reduces the Fe (III) to Fe (II) again to a normal situation. High doses of chemicals with a methemoglin-inducing effect can lead to higher methemoglobin formation which is reversible upon cessation of exposure to such a chemical.

Evaluation of hematological effects in studies with phenmedipham

52-week rat study (Everett et al., 1987)

Groups of 20 male and 20 female Sprague-Dawley rats were dosed with phenmedipham (98.5 %) via the diet at concentrations of 60 (low dose), 250 (middle dose) and 1000 ppm (high dose) for 52 weeks. Groups of 20 males and females each received untreated diet and acted as controls. The animals were ordered ca. 4 weeks old, weighing ca. 85 g (males) and 60 g (females). The animals were acclimatised for 15 days before treatment. Viability during the study was checked twice per day. All animals were examined for clinical signs daily, and all animals were palpated at least once a week. Each animal was weighed at weekly intervals starting from the week before the start of treatment until week 13 and thereafter at 4-weekly intervals. Food consumption was recorded once each week using the same schedule as with weighing the animals. Blood samples were taken from 10 males and 10 females per group for hematological and clinical chemistry investigations during weeks 13, 25 and 51 of dosing. The samples were taken from the same animals at the different time points. During weeks 25 and 51 of dosing, samples were taken also for urinalysis investigations from the same animals as those subjected to hematology sampling. After 52 weeks of dosing all animals were killed and autopsied. Histopathological examinations were initially carried out on all rats in the control and high dose groups and any premature decedents. Livers, kidneys and spleen of all animals were stained with Perls' Prussian Blue (PBR) and examined for hemosiderin deposition.

6 animal died, but not in a dose-related manner and, thus not due to treatment. As clinical signs scabby tail, encrustations around nose and eyes, alopecia, exophthalmia and immobile swollen limb joints were reported. They occurred with approximately equal incidence and severity in all groups during the study and were therefore not treatment-related. No statistically significant differences between the groups were noted in body weight gain or food consumption among either sex.

Hematologically, in the male and female high dose groups there were reductions in red cell parameters (hemoglobin (Hb), red blood cell count (RBC) and hematocrit (HCT)) at Weeks 13, 25 and 51. The intermediate dose group showed a reduction in HCT at all time points in the males and at Week 51 only in the females. Intermediate dose males also showed significant reductions in Hb levels at week 51. In high dose females Hb and HCT was decreased at all time points and RBC at weeks 13 and 51. At week 51 HCT was decreased in mid dose females also. During week 25 the low dose group females also showed reductions in Hb and HCT, but since the next higher dose of 250 ppm did not show an effect and is comparable to the control value, this is regarded as due to variability and not to treatment.

An overview of the hematological results during the different determination periods is given in the following tables:

Table 1: Hematological changes males – week 13

Group/Dose Level	Phenmedipham 52 Week study Males			
(ppm)		Hb	RBC	HCT
1	N	10	10	10
0	Mean	15.5	7.87	0.440
	SD	0.6	0.17	0.016
2	N	9	9	9
60	Mean	15.4	7.77	0.493
	SD	0.6	0.35	0.118
3	N	8	8	8
250	Mean	15.0	7.78	0.418
	SD	0.5	0.53	0.018**
4	N	10	10	10
1000	Mean	14.3	7.15	0.404
	SD	0.6***	0.40***	0.016***

**: Significantly different from controls, P<0.01
***: Significantly different from controls, P<0.001
N. Number of animals

Table 2: Hematological changes males – week 25

Group/Dose Level (ppm)	Phenmedipham 52 Week study Males			
		Hb	RBC	HCT
1 0	N	9	9	9
	Mean	15.2	7.82	0.413
	SD	0.7	0.36	0.016
2 60	N	10	10	10
	Mean	15.1	7.82	0.412
	SD	0.3	0.28	0.013
3 250	N	8	8	8
	Mean	14.6	7.54	0.389
	SD	0.9	0.35	0.023*
4 1000	N	10	10	10
	Mean	14.3	7.24	0.383
	SD	0.3**	0.26***	0.020**

**: Significantly different from controls, P<0.01
***: Significantly different from controls, P<0.001
N: Number of animals

Table 3: Hematological changes males – week 51

Group/Dose Level (ppm)	Phenmedipham 52 Week study Males			
		Hb	RBC	HCT
1 0	N	10	10	10
	Mean	15.3	8.18	0.432
	SD	0.6	0.36	0.016
2 60	N	10	10	10
	Mean	15.0	8.12	0.421
	SD	0.2	0.30	0.010
3 250	N	10	10	10
	Mean	14.6	7.93	0.404
	SD	0.6*	0.37	0.019***
4 1000	N	10	10	10
	Mean	14.1	7.42	0.392
	SD	0.3***	0.34***	0.015***

*: Significantly different from controls, P<0.05
***: Significantly different from controls, P<0.001
N: Number of animals

Table 4: Hematological changes females – week 13

Group/Dose Level	Phenmedipham 52 Week study Females			
(ppm)		Hb	RBC	HCT
1	N	10	10	10
0	Mean	15.3	7.31	0.423
	SD	0.5	0.30	0.016
2	N	10	10	10
60	Mean	15.4	7.60	0.432
	SD	0.4	0.17*	0.012
3	N	10	10	10
250	Mean	15.0	7.34	0.416
	SD	0.5	0.33	0.018
4	N	10	10	10
1000	Mean	14.5	6.81	0.402
	SD	0.8**	0.34***	0.022*

*: Significantly different from controls, P<0.05
**: Significantly different from controls, P<0.01
***: Significantly different from controls, P<0.001
N: Number of animals

Table 5: Hematological changes females – week 25

Group/Dose Level	Phenmedipham 52 Week study Females			
(ppm)		Hb	RBC	HCT
1	N	10	10	10
0	Mean	14.9	7.33	0.420
	SD	0.4	0.27	0.011
2	N	10	10	10
60	Mean	14.0	7.08	0.397
	SD	0.9**	0.46	0-029*
3	N	10	10	10
250	Mean	14.5	7.24	0.411
	SD	0.08	0.38	0.025
4	N	9	9	9
1000	Mean	14.1	6.88	0.392
	SD	0.7*	0.38	0.021***

*: Significantly different from controls, P<0.05
**: Significantly different from controls, P<0.01
***: Significantly different from controls, P<0.001
N: Number of animals

Table 6: Hematological changes females – week 51

Group/Dose Level	Phenmedipham 52 Week study Females			
(ppm)		Hb	RBC	HCT
1	N	9	9	9
0	Mean	14.8	7.50	0.412
	SD	0.5	0.31	0.014
2	N	10	10	10
60	Mean	14.6	7.66	0.409
	SD	0.4	0.24	0.012
3	N	10	10	10
250	Mean	14.4	7.37	0.397
	SD	0.08	0.46	0.019*
4	N	10	10	10
1000	Mean	13.7	6.96	0.378
	SD	0.4***	0.32**	0.015***

*: Significantly different from controls, P<0.05
**: Significantly different from controls, P<0.01
***: Significantly different from controls, P<0.001
N: Number of animals

No effects on clinical chemistry which would be related to treatment were noted among males during the experiment. Urinalysis did not show significant differences between the groups in pH, specific gravity and urinary volume among males throughout the whole study. A higher than expected incidence of positive blood pigment results in urine was noted among males, especially at 250 and 1000 ppm towards the end of the study. In several cases there was a corresponding presence of red blood cells in the microscopic examination. Significantly increased urinary volume was noted in mid and high dose animals at week 25, but not later. Specific gravity was correspondingly lowered in mid and high dose female groups at week 25. At the end of the experiment, several females were reported to have given positive blood pigment results.

Organ weight analyses showed an increase in spleen weights in males at 1000 ppm. In females a decrease in absolute kidney weights was noted at 1000 ppm. No gross pathology lesions attributed to treatment with phenmedipham were reported in the study report.

As described before, hemolytic anemia with secondary increase of hemoglobin turnover leads to an increased catabolism of hemoglobin to bilirubin and it is known that after a temporary higher hemolysis this catabolism pathway can be temporarily overwhelmed. In such a situation an increased formation of hemosiderin occurs which can lead to an increased deposition of hemosiderin in the reticulo-histiocytary system (RHS). Mainly spleen, but also liver and kidneys can be affected.

This was also seen in this study in which hemosiderin deposits were reported in some organs. High dose group males and females showed an increase in incidence and severity of Kupffer cell pigmentation in the liver, the pigment being haemosiderin (by PBR staining). The spuriously increased incidence of hemosiderin deposits in the low-dose (60 ppm) males is regarded as consequence of variability and not of treatment since the next higher dose of 250 ppm did not show a statistically significant effect and grade ++ incidence and overall incidence was lower again at this dose and without statistical significance. It can be seen that grade +/- and grade + incidences are not different from the control ones, only the grade ++ incidences are affected statistically significantly at the highest dose. The fact that no grade +++ findings occurred at any dose in males, only one at the high dose in females, speaks for a slight effect at the highest dose only, as seen by the increased incidences of grade ++ in males and females and of grade +++ incidences in females. Therefore, the overall incidences do not show a dose- and treatment-relationship in the dose range below 1000 ppm.

This is furthermore confirmed by the hemosiderin deposits in other organs which only had such findings at the highest dose of 1000 ppm. In the kidneys, only in high-dose males and females a slightly higher incidence and severity of proximal tubule cytoplasmic haemosiderin was seen. Also in the spleen which normally is the primary organ to be affected by consequences of hemolytic effects, only in the high dose males a slight increase of grade ++ hemosiderin deposit was seen, whereas grade +++ was even lower at the higher dose, and no effects of grade +/- or grade + in males and no effects in females were seen.

As explained before, these findings are a consequence of the hemolytic effects only secondary to them, and thus can only occur at doses at which also hemolytic anemia effects, as a prerequisite, are seen. Therefore, the incidence for the 60 ppm males in the liver is not related to treatment since no hematological effects occurred at that dose. Since only an overwhelmed hemoglobin catabolism after a higher degree of hemolytic anemia can lead to an increased deposition of hemosiderin in the reticulo-histiocytary system (RHS), this can be expected only at doses which cause a more severe hemolytic anemia. Such an effect occurred only after higher doses of phenmedipham, but not at 60 ppm.

An overview of the aforementioned findings is given in table 7.

Table 7: Histopathology changes – week 52

Lesions	Incidence of lesions (numeric)							
	Treatment (ppm)							
	Males				Females			
	0	60	250	1000	0	60	250	1000
Liver:	20	20	20	20	20	20	20	20
Haemosiderin negative (P.B.R.)	6	1	2	0*	1	2	0	0
Haemosiderin positive (P.B.R.)								
(Grade +/-)	7	5	3	0**	14	12	4**	0***
(Grade +)	6	7	9	2	5	5	14*	10
(Grade ++)	1	7*	6	18***	0	1	2	9**
(Grade +++)	0	0	0	0	0	0	0	1
Kidneys:	20	20	20	20	20	20	20	20
Haemosiderin negative (P.B.R.)	5	4	2	0*	0	0	0	0
Haemosiderin positive (P.B.R.)								
(Grade +/-)	11	12	7	0***	5	5	3	1
(Grade +)	2	3	6	2	11	14	11	4*
(Grade ++)	2	1	5	8	4	1	6	11*
(Grade +++)	0	0	0	10***	0	0	0	4
Spleen:	20	20	20	20	20	20	20	20
Haemosiderin positive (P.B.R.)								
(Grade +/-)	0	2	0	0	0	0	0	0
(Grade +)	4	1	5	3	4	9	7	3
(Grade ++)	9	15	13	16*	13	10	12	12
(Grade +++)	7	2	2	1*	3	1	1	5

* $p<0.05$, ** $p<0.01$, *** $p<0.001$

The described findings of hemosiderin deposition in the affected organs are sometimes reflected in the organ weights. In this study with phenmedipham only increases of the spleen weight at the highest dose in males were seen so that the hemosiderin deposition in the other organs apparently were so minimal that they were without an impact on the organ weights of liver and kidneys.

Other rat studies

Rat:

In order to compare the dose-response relationship regarding the hematological findings in the discussed 52-week study with other study results, a short overview of the hematological findings in other studies is given here.
The results of the comparison with the hematological findings in other relevant short- and long-term toxicology studies can be seen in the following two tables.

Table 8: Overview of the hematological results in short-term rat studies

Study	Doses	Main hematology and secondary effects	NOAEL methemo-globinemia	NOAEL RBC, Hb, MCG etc.	NOAEL hemosiderin deposition	Comment
13-week oral, dietary rat plus 4-week recovery [redacted] 1986b)	0-150-500-1500 ppm (m:9-18, 30-60, 90-181 mg/kg bw; f: 12-20, 38-64, 102-193 mg/kg bw	RBC,Hb, MCH decreased: ≥500 ppm; increased hemosiderin deposition liver and kidney: ≥500 ppm (m,f) and in spleen (f)	na	150 ppm (9-18 (m)/ 12-20 (f) mg/kg bw	150 ppm (9-18 (m)/ 12-20 (f) mg/kg bw	Hematological effects were mainly reversible within the 4-week recovery period
13-week oral, dietary rat ([redacted]., 1986a)	0-400-800-1200 ppm (m: 30-60-92 mg/kg bw; f:33-72-122 mg/kg bw;	RBC, Hb, MCH decreased: ≥400 ppm (m: week 6) ≥400 ppm (f: week 6, 13) 1200 ppm (m: week (13); increased hemosiderin deposition liver and kidneys: ≥400 ppm (m,f) and in spleen (f)	na	<400 ppm (<30 (m)/<33 (f) mg/kg bw	400 ppm (30 (m)/33 (f) mg/kg bw	
13-week oral dietary rat ([redacted] 1981)	0-50-500-5000 ppm (m: 3.5-35.4-366.5 mg/kg bw; f: 3.7-37.4-377.5 mg/kg bw	RBC, Hb, MCH decreased: ≥500 ppm (m, f: increased hemosiderin deposition and hematopoiesis in spleen: 5000 ppm (m, f)	na	50 ppm (3.5 (m)/ 3.7 (f) mg/kg bw	500 ppm (35.4 (m)/ 37.4 (f) mg/kg bw	

RBC= red blood cells Hb= hemoglobin, MCH=mean corpuscular hemoglobin, na= not applicable, m=males, f=females

Table 9: Overview of the hematological results in long-term rat studies

Overview of the hematological results in long-term rat studies						
Study	**Doses**	**Main hematology and secondary effects**	**NOAEL methemo-globinemia**	**NOAEL RBC, Hb, MCG etc.**	**NOAEL hemosiderin deposition**	**Comment**
52-week oral, dietary rat ([redacted] 1988a)	0-60-250-1000 ppm (m: 3.4-14.6-58.7 mg/kg bw; f: 4.6-18.7-78.1 mg/kg bw)	RBC, Hb, MCH decreased: males: ≥250 ppm (m: week 13, 25, 51), f: 1000 ppm (week 13, 25,51) Increased hemosiderin deposition in liver and kidneys, m: 1000 ppm, f:≥250 ppm	na	60 ppm (3.4 (m) 250 ppm, (18.7 (f) mg/kg bw)	250 ppm (14.6 (m) mg/kg bw/60 ppm (4/6 (f) mg/kg bw)	
52-week oral, dietary rat [redacted] 1987)	0-60-250-1000 ppm (m: 4.2-17-70 mg/kg bw; f: 5.1-20-84 mg/kg bw	RBC, Hb, MCH decreased: m: ≥250 ppm (m: week 13, f: week 13, 25); increased hemosiderin deposition in liver and kidneys: ≥250 ppm (m)	na	60 ppm (4.2 (m)/5.1 (f) mg/kg bw)	60 ppm (4.2 (m) mg/kg bw) /1000 ppm (84 (f) mg/kg bw	
104-week dietary carcinogenicity rat study [redacted] 1988c)	0-60-250-1000 ppm (m/f: 2.1-7.3 (60), 8.9-30 (250), 33.1-114.3 mg/kg bw;	RBC, decreased: ≥250 ppm (f) ; 1000 ppm: increased pigment deposition in liver (m,f) and kidneys (m), increased hematopoiesis in sternum (f not significant)	na	M: 1000 ppm (33.1-114.3 mg/kg bw); f: 60 ppm (2.1-7.3 mg/kg bw	250 ppm (m/f: 8.9- 30.0 mg/kg bw)	Dose in mg/kg bw based on min.-max. of both sexes
104-week dietary carcinogenicity rat study [redacted] 1988b)	0-60-250-1000 ppm (m: 3.1-12.5-50.1 mg/kg bw; f: 4.1-16.8-67.5 mg/kg bw)	No hematology done; 1000 ppm: m/f: increased pigmented macrophages and Kupffer cells in livers	na	na	250 ppm (12.5 mg/kg bw (m)/ 16.8 mg/kg bw (f)	No DART report available
104-week chronic toxicity and carcinogenicity oral rat study [redacted] 2004)	0-100-500-2500 ppm (52 weeks: m: 5.6-27.1-137.0 mg/kg bw; f: 7.6-35.0-196.0 mg/kg bw; 104 weeks: m: 4.6-23.6-117.6 mg/kg bw; f: 6.4-33.1-170.5 mg/kg bw	RBC, Hb, MCH decreased: ≥500 ppm; Met-Hb increased: m: ≥250 ppm (wk 13,26, f: 2500 ppm (≥wk 26); increased hemosiderin deposition liver and kidneys: ≥500 ppm (m,f) and in spleen (f)	100 ppm (4.6 (m) mg/kg bw/ 500 ppm (33.1 (f) mg/kg bw	100 ppm (4.6 (m)/ 6.4 (f) mg/kg bw)	100 ppm (4.6 (m)/ 6.4 (f) mg/kg bw)	

RBC= red blood cells Hb= hemoglobin, MCH=mean corpuscular hemoglobin, na= not applicable, m=males, f=females

It can be seen that all other studies revealed a similar toxicological profile of phenmedipham. Also in the other studies, besides effects on body weight and food consumption, the main target of phenmedipham in rats were hematological effects, mainly on red blood cell parameters. These effects included reductions of the RBC counts, of Hb and MCH.
Hemosiderin deposition was generally observed in the liver and in the spleen, in some cases also in the kidneys in rats. Increased liver and spleen weights and in some cases also kidney weight changes were noted. A 13-week study with a 4-week recovery phase ([redacted] 1986) showed that the changes in RBC parameters were almost completely reversible during the 4-week recovery period, as were also body weight changes and increases in hemosiderin deposition in liver and kidneys and partly spleen.

Occasionally, increased bilirubin concentrations were observed, also increased hemopoiesis in the bone marrow and increased reticulocyte counts in blood. The lowest NOAEL in all studies was at 50-60 -100 ppm, with ranges up to 500 ppm.

In a recent long-term study in rats ([redacted] 2004), which seems to be representative and important for this evaluation because of the much longer exposure time, three groups of 50 male and 50 female rats received phenmedipham orally, via the diet, at concentrations of 100, 500 or 2500 ppm (equivalent to doses of 4.6, 23.6 and 117.6 mg/kg bw/day for males and of 6.4, 33.1 and 170.5 mg/kg bw/day for females in the 104-week carcinogenicity phase, and of 5.6, 27.1 and 137.0 mg/kg bw/day for males and 7.6,35.0 and 196.0 mg/kg bw/day for the females in the 52-week toxicity phase). A control group received untreated diet. A further 20 male and 20 female rats were allocated to each group. These animals comprised the toxicity phase of the study and were sacrificed after 52 weeks of treatment. In this study, hematologically, low hematocrit, haemoglobin concentration and red blood cell counts were seen in males and females at 2500 ppm, with a slight effect in 500-ppm males. At 2500 ppm, there was a consistent trend towards increased reticulocyte count and there were increased incidences and degree of anisocytosis in males receiving 2500 ppm and of hyperchromasia in males receiving 2500 ppm and during Week 13 in females receiving 500 ppm and in week 13 and 26 in those receiving 2500 ppm. Slight, but consistently higher methaemoglobin levels were recorded in females and at some time points also in males at 2500 ppm. At 500 ppm slightly high methaemoglobin levels were evident in week 13 and 26 in males. Spleen weights were increased in 2500-ppm males. Liver weights were slightly increased in females at 2500 ppm and kidney weights were slightly decreased in males and females at 2500 ppm. Histopathologically, in the liver a slightly increased incidence of pigmented macrophages or pigmented Kupffer cells in males and females at 2500 ppm was seen.

The incidences of pigment were given in the report for macrophages and Kupffer cells (which are also macrophages) separately, they can be seen in the following tables.

Table 10: 104-week rat study: liver changes, week 52

Group/sex		1M	2M	3M	4M	1F	2F	3F	4F
Level (ppm)		0	100	500	2500	0	100	500	2500
Pigmented macrophages	Minimal	0	0	0	3	3	5	2	10
	Moderate	0	0	0	0	0	0	1	0
	Total	0	0	0	3	3	5	3	10 a
Pigment in Kuppfer cells	Minimal	0	0	1	5	0	0	0	4
	Total	0	0	1	5 a	0	0	0	4 a
n		20	20	20	20	20	20	19	19

Significant when compared with group 1: a- p< 0.05

n = number of animals examined

Table 11: 104-week rat study: liver changes, week 104

Group/sex		1M	2M	3M	4M	1F	2F	3F	4F
Level (ppm)		0	100	500	2500	0	100	500	2500
Pigmented macrophages	Minimal	2	1	3	18	8	8	14	20
	Slight	0	0	0	2	0	2	3	4
	Moderate	1	0	0	1	0	0	0	0
	Total	3	1	3	21 b	8	10	17	24 b
Pigment in Kupffer cells	Minimal	0	1	2	16	2	1	2	17
	Slight	0	0	0	5	0	0	0	4
	Total	0	1	2	21 b	2	1	2	21 b
n		50	50	50	50	50	50	50	49

Significant when compared with group 1: a- p< 0.05

n = number of animals examined

The tabular overviews clearly show that there were effects at the highest dose of 2500 ppm, but not at lower doses which also confirms that the spurious liver finding in 60 ppm males in the discusssed 52-week rat study is not treatment-related.

In the spleen a high incidence of congestion was seen in all male treated groups and in females at 500 or 2500 ppm with slightly increased incidences of haemosiderosis or extramedullary haemopoiesis in males and females at 2500 ppm.

Thus, the NOAEL for all effects, including hematology effects, was established at 100 ppm. Like the

other studies mentioned before, also this long-term study confirms clearly the NOAEL of 60 ppm in the 52-week study as a NOAEL for the hematological effects of phenmedipham.

Studies in other species:

An 8-week study in mice with oral administration revealed the same type of effects as seen in the rat studies. Again slight anemia was noted together with increased liver weight, and hemosiderin deposition in the liver. Also increased methemoglobin levels were observed, The highest observed methemoglobin values were 4.0 – 4.5 % at approximately 2000 mg/kg bw/day in mice (compared to 2.3 % in controls) and an increase in hepatic extramedullary hematopoiesis. The NOAEL in mice is 125 mg/kg bw/day and, thus, much higher than in the rat.
Furthermore, these methemoglobin levels are approximately in the range of the level of 4% which is regarded as maximum non-adverse level ([redacted] 2005, Food and Chemical Toxicology 43 (2005) 1569-1593).
Also in the dog decreases in red blood cell parameters were seen together with extramedullary hematopoiesis, again indicating slight anemia.Transient hematological changes were also observed in the 2-year dog study, but at rather high doses only, e.g. the NOAEL in the 2-year dog study is higher than 28 mg/kg bw/day. Thus, in summary, the toxicological profile in these species were similar to the one in rats. Mostly moderate and reversible (within 4 weeks) hematological changes in red blood cell parameters, liver and spleen weight changes (sometimes also kidneys), hemosiderin deposition in spleen, liver and kidneys, and increased medullar and/or extramedullar hematopoiesis were seen, with very high NOAELs. Also a slight increase in methemoglobin levels (up to 4%) was noted, but adverse or non-reversible health effects as a result of the slight methemoglobinemia could not be identified.

Overall conclusion on the hematological findings after phenmedipham

The most important outcome of this analysis is that all rat studies and studies in other species show that the hematological effects were not so severe that they could be regarded as findings of 'significant' or 'severe' toxicity. Also the methemoglobin levels were in ranges which cannot be defined as adverse. The findings have a NOAEL of 60 ppm or even higher than 60 ppm. In the other studies mostly NOAELs were 100 ppm, ranging up to 500 ppm. A recent 104-week chronic toxicity and carcinogenicity study with a thorough evaluation of all relevant hematological parameters over many timepoints of the long-term study confirmed a NOAEL of 100 ppm for all hematological parameters, including hemosiderin formation and deposition.

ECHA note – An attachment was submitted with the comment above. Refer to public attachment PMP_ECHA commenting_task force_sanitized.pdf

Dossier Submitter's Response

Thank you for your comments. We agree that the main target organ of toxicity in experimental animals following repeated oral administration is blood. At the dose levels approximately equal to STOT RE 2 guidance values haematotoxic effects stayed rather slight or moderate (e.g. MetHb levels were below 4 %) and in most of cases were reversible Although the severity of the haemotoxic effects represents a borderline case, multiple less severe and dose-related effects with regenerative capacity involving several organs were observed consistently in oral repeated dose toxicity studies at the dose levels approximately equal to the STOT-RE 2 guidance values. We consider these effects sufficient for classification.

RAC's response

RAC agrees that the haematotoxicity-related effects below the guidance values (GVs) are rather weak and do not meet the criteria for classification.

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Netherlands		MemberState	14
Comment received				
NL agrees with the Dossier Submitter that this is a borderline case. The haematopoietic system is clearly the target tissue, as observed in multiple studies in rat, mouse and dog. Such effects are also observed with the structurally related chemical desmedipham. However, none of the individual studies fulfill the criteria for a STOT RE classification. Effects at dose levels relevant for classification were only slight to moderate, and in some cases reversible. It is noticed that in the 90-d rat study B.6.2.3.2/06, a 17% decrease in Hb-levels was found at a dose level of 214 mg/kg bw/d. However, even at the higher dose level of 658 mg/kg bw/d the decrease in Hb-level is still below 20%. The Dossier submitter proposes to apply the criterion as described in CLP Annex I 3.9.1.4 ("Assessment shall take into consideration not only significant changes in a single organ or biological system but also generalised changes of a less severe nature involving several organs"). However, one can question the validity of applying this criterion as the various adverse effects concern specific effects (i.e. effects on the haematological system) and not generalized changes involving several organs. In conclusion, although the data point towards the haematological system as the primary target, we consider the severity of the effects to be insufficient for classification for STOT RE cf. the current criteria for classification, and the Dossier Submitters proposal for a cat. 2 is not supported.				
Dossier Submitter's Response				
Thank you for your comment. We agree that the severity of the haemotoxic effects is not high and represents a borderline case for classification.				
RAC's response				
RAC agrees that the haematotoxicity-related effects below the GVs are rather weak and do not meet the criteria for classification. As to the 17 % decrease in Hb at 3 000 ppm after 4 weeks in the rat study B.6.3.2/06, the effect did not follow Haber's rule (the reduction after 12 weeks was 13 % and study B.6.5.1/07 shows that the effective dose does not decrease with time for up to 2 years). Therefore the default GV for a 90-day study of 100 mg/kg bw/d is considered more appropriate, leaving the dose of 1 000 ppm with a maximum Hb reduction of 8 %. The criterion of the CLP Regulation, Annex I, 3.9.1.4 is exemplified by the following criteria for haemolytic anaemia in the CLP guidance: • Marked increase of haemosiderosis in the spleen, liver or kidney in combination with other changes indicating significant haemolytic anaemia (e.g. a reduction in Hb at ≥ 10 %) in a 28-day study, • Significant increase in haemosiderosis in the spleen, liver or kidney in combination with microscopic effects like necrosis, fibrosis or cirrhosis. Neither of these two criteria is met for phenmedipham.				

Date	Country	Organisation	Type of Organisation	Comment number
13.02.2019	Denmark		MemberState	15
Comment received				
We agree that phenmedipham affects blood parameters in such an extent that classification				

STOT RE 2 (blood) is warranted.
Dossier Submitter's Response
Thank you for your support.
RAC's response
RAC considers the haematotoxicity-related effects below the GVs rather weak and not meeting the criteria for classification.

Date	Country	Organisation	Type of Organisation	Comment number
15.02.2019	France		MemberState	16
Comment received				
FR, page 54: The classification as STOT RE H373 (blood) proposed by the MSCA is supported.				
Dossier Submitter's Response				
Thank you for your support.				
RAC's response				
RAC considers the haematotoxicity-related effects below the GVs rather weak and not meeting the criteria for classification.				

OTHER HAZARDS AND ENDPOINTS – Hazardous to the Aquatic Environment

Date	Country	Organisation	Type of Organisation	Comment number
15.02.2019	United Kingdom		MemberState	17
Comment received				
Phenmedipham (EC: 237-199-0; CAS: 13684-63-4) Please can you confirm if an ErC10 (dry weight) endpoint is available for the Myriophyllum spicatum study? We note that nearly 20% inhibition was observed at the quoted NOErC and wonder if a statistically based EC10 might be more appropriate given the steep toxicity profile? In addition, are measurements of test item concentrations in sediment available to support the use of water phase concentrations which declined over the study period? This is important to consider exposure routes.				
Dossier Submitter's Response				
Thank you for your comments. Inhibition of growth rate for total plant dry weight was 19.8% at geometric mean measured NOErC value of 12.8 µg/L. Growth rate ErC_{10} (dry weight) value of 4.837 µg/L has been calculated in the original *Myriophyllum spicatum* study report, however, that ErC_{10} value was not further reported in the study and dRAR since the control coefficient of variation of this parameter was higher than the respective effect level. Thus, EC_{10} endpoint for growth inhibition (dry weight) is not considered reliable. The lowest valid 14 d EC_{10} value of 20.8 µg/L was determined based on growth rate (fresh weight). Analytical results are only available from fresh and aged media (water phase) so test item concentration measurements are not available for sediment. The pH of the test solution was purposely decreased to 6.5 in order to prevent the degradation of phenmedipham as much as possible, because phenmedipham is hydrolytically very unstable in alkaline medium. At				

pH 6.5 DT_{50} value of 3.3 days was observed and at pH 7 DT_{50} ranged from 3 hours to 1 day (dRAR B.8.2.1.1/03, 2003, dRAR B.8.2.1.1/04, 2004 & dRAR B.8.2.1.1/05, 2015). We think that the observed loss of test item during the *Myriophyllum spicatum study* occurred mainly because of hydrolytic degradation of phenmedipham. Shoots of *Myriophyllum spicatum* were exposed via water phase and, thus we consider that water phase is relevant exposure route.
RAC's response
RAC takes note of the fact that a reliable EC_{10} dry weight cannot be obtained although it cannot check the raw data. RAC agrees with the DS and considers that the *Myriophyllum* test is adequate for classification for various reasons (although exposition via sediment cannot be totally discarded): the substance is an herbicide acting only via the foliage of emerged weeds and *Myriophyllum* has been demonstrated to be the most sensitive acute species, application of the test substance is done via the water column and the substance mainly disappears because of hydrolysis.

Date	Country	Organisation	Type of Organisation	Comment number
11.02.2019	Germany		MemberState	18
Comment received				
page 3, point 2.1 Proposed harmonised classification and labelling (Table 6): We support the proposal of classification for environmental hazards as Aquatic acute 1 (H400), Aquatic chronic 1 (H410) and acute/chronic M-factor of 10.				
Dossier Submitter's Response				
Thank you for your support.				
RAC's response				
Thank you.				

Date	Country	Organisation	Type of Organisation	Comment number
15.02.2019	France		MemberState	19
Comment received				
FR: We agree with the proposed classification: Aquatic Acute 1 (H400) with an acute M-factor of 10 and Aquatic Chronic 1 (H410) with a chronic M-factor of 10. However, since the metabolite m-toluidine is 21 d NOEC value of 0.00478 mg/L for Daphnia magna, it appears to be more toxic than the parent, could you please indicate if this value should be considered for chronic classification.				
Dossier Submitter's Response				
Thank you for your support. Long-term aquatic hazard classification is based on flow-through 21 d NOEC value of 0.005 mg/L of phenmedipham for *Daphnia magna.* The degradation product m-toluidine was slightly more toxic compared to phenmedipham for aquatic invertebrate *Daphnia magna* with 21 d NOEC value of 0.00467 mg/L and 21 d EC10 value of 0.00478 (NOEC value at page 95 of CLH dossier should refer to NOEC value of 0.00467 mg/L). Chronic toxicity of degradate m-toluidine was considerable lower for aquatic macrophyte *Lemna gibba* (7d NOEC 3.05 mg/L). No chronic toxicity test of m-toluidine is available for fish, but acute toxicity of m-toluidine was much higher for aquatic invertebrate *Daphnia magna* (48h EC50 0.1 mg/L) compared to acute toxicity for fish (96 h LC50 93.3 mg/L). Thus, aquatic invertebrates are considered to be the most sensitive taxa for degradate m-toluidine as well as for the parent substance phenmedipham. Toxicity of the parent substance phenmedipham and degradate m-toluidine is within the				

same order of magnitude for aquatic invertebrates, and both toxicity values would result in the same classification of Aquatic Chronic 1 with chronic M-factor of 10. In this case, we prefer to classify phenmedipham according to the lowest toxicity value for the parent substance (21 d NOEC 0.005 mg/L).
RAC's response
RAC agrees with the DS in using Phenmedipham data for classification since for the parent there is full a data set whereas for the metabolite there is no chronic data for fish. The lowest acute endpoint corresponds to a test with Phenmedipham with Myriophyllum EC_{50} = 0.0519 mg/L whereas the lowest acute value for the metabolite is 0.1 mg/L for Daphnia (there is no data for the metabolite with M. spicatum). In the case of chronic toxicity the lowest value both for parent and metabolite corresponds to Daphnia magna. The endpoint in both cases is almost the same. NOEC = 0.005 mg/L for parent and NOEC = 0.00467 and EC_{10} = 0.00478 mg/L for *m*-toluidine.

PUBLIC ATTACHMENTS
1. PMP_ECHA commenting_task force_sanitized.pdf [Please refer to comment No. 1, 9, 13]