Committee for Risk Assessment

RAC

Opinion

proposing harmonised classification and labelling
at EU level of

Phosmet (ISO); *S*-[(1,3-dioxo-1,3-dihydro-2*H*-isoindol-2-yl)methyl] *O,O*-dimethyl phosphorodithioate

EC Number: 211-987-4
CAS Number: 732-11-6

CLH-O-0000001412-86-113/F

Adopted

3 June 2016

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3 June 2016

CLH-O-0000001412-86-113/F

OPINION OF THE COMMITTEE FOR RISK ASSESSMENT ON A DOSSIER PROPOSING HARMONISED CLASSIFICATION AND LABELLING AT EU LEVEL

In accordance with Article 37 (4) of Regulation (EC) No 1272/2008, the Classification, Labelling and Packaging (CLP) Regulation, the Committee for Risk Assessment (RAC) has adopted an opinion on the proposal for ha*rmonised classification and labelling (CLH) of:*

Chemical name:	**Phosmet (ISO); *S*-[(1,3-dioxo-1,3-dihydro-2*H*-isoindol-2-yl)methyl] *O,O*-dimethyl phosphorodithioate**
EC Number:	**211-987-4**
CAS Number:	**732-11-6**

The proposal was submitted by **Spain** and received by RAC on **31 July 2015.**

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

PROCESS FOR ADOPTION OF THE OPINION

Spain has submitted a CLH dossier containing a proposal together with the justification and background information documented in a CLH report. The CLH report was made publicly available in accordance with the requirements of the CLP Regulation at ***http://echa.europa.eu/harmonised-classification-and-labelling-consultation/*** on **30 September 2015**. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by **16 November 2015**.

ADOPTION OF THE OPINION OF RAC

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

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

The RAC opinion on the proposed harmonised classification and labelling was adopted on **3 June 2016** by **consensus**.

Classification and labelling in accordance with the CLP Regulation (Regulation (EC) 1272/2008)

	Index No	International Chemical Identification	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors	Notes
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Pictogram, Signal Word Code(s)	Hazard statement Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	015-101-00-5	phosmet (ISO); *S*-[(1,3-dioxo-1,3-dihydro-2*H*-isoindol-2-yl)methyl] *O,O*-dimethyl phosphorodithioate	211-987-4	732-11-6	Acute Tox. 4 * Acute Tox. 4 * Aquatic Acute 1 Aquatic Chronic 1	H302 H312 H400 H410	GHS07 GHS09 Wng	H302 H312 H410		M=100	
Dossier submitter's proposal	015-101-00-5	phosmet (ISO); *S*-[(1,3-dioxo-1,3-dihydro-2*H*-isoindol-2-yl)methyl] *O,O*-dimethyl phosphorodithioate	211-987-4	732-11-6	**Retain** Aquatic Acute 1 Aquatic Chronic 1 **Add** Acute Tox. 4 STOT RE 1 **Modify** Acute Tox. 3 **Remove** Acute Tox. 4 *	**Retain** H400 H410 **Add** H332 H372 (nervous system) **Modify** H301 **Remove** H312	**Retain** GHS09 **Add** GHS06 GHS08 **Modify** Dgr **Remove** GHS07	**Retain** H410 **Add** H332 H372 (nervous system) **Modify** H301 **Remove** H312		**Retain** M=100 **Add** M=10	
RAC opinion	015-101-00-5	phosmet (ISO); *S*-[(1,3-dioxo-1,3-dihydro-2*H*-isoindol-2-yl)methyl] *O,O*-dimethyl phosphorodithioate	211-987-4	732-11-6	**Retain** Aquatic Acute 1 Aquatic Chronic 1 **Add** Repr. 2 Acute Tox. 4 STOT SE 1 **Modify** Acute Tox. 3 **Remove** Acute Tox. 4 *	**Retain** H400 H410 **Add** H361f H332 H370 (nervous system) **Modify** H301 **Remove** H312	**Retain** GHS09 **Add** GHS06 GHS08 **Modify** Dgr **Remove** GHS07	**Retain** H410 **Add** H361f H332 H370 (nervous system) **Modify** H301 **Remove** H312		**Retain** M=100 **Add** M=100	
Resulting Annex VI entry if	015-101-00-5	phosmet (ISO); *S*-[(1,3-dioxo-1,3-dihydro-2*H*-isoindol-2-	211-987-4	732-11-6	Repr. 2 Acute Tox. 4 Acute Tox. 3	H361f H332 H301	GHS08 GHS06 GHS09	H361f H332 H301		M=100 M=100	

agreed by COM		yl)methyl] *O,O*-dimethyl phosphorodithioate			STOT SE 1 Aquatic Acute 1 Aquatic Chronic 1	H370 (nervous system) H400 H410	Dgr	H370 (nervous system) H410			

GROUNDS FOR ADOPTION OF THE OPINION

RAC general comment

Phosmet is an insecticide and acaricide used as an active substance in plant protection products, included in the Annex I of Regulation (EU) No. 540/2011, with an existing Annex VI entry. An application for the renewal of phosmet as an active substance for plant protection products has been submitted, including new studies assessing the potential of phosmet to affect oestrogen, androgen or thyroid hormone systems.

Since TC C&L did not reach a final agreement on Phosmet classification (issues for discussion: acute oral and inhalation toxicity), it was handed over to ECHA (29 May 2008).

Phosmet was discussed at EFSA, by EPCO experts' meeting for mammalian toxicology (EPCO 33) in September 2005, and at the PRAPeR[1] 86 Experts' Meeting for mammalian toxicology in March 2011, and it was concluded that the toxicological equivalence of the current technical material has not been fully demonstrated according to the Guidance Document on the Assessment of the Equivalence of Technical Materials of Substances Regulated under Council Directive 91/414/EEC (SANCO/1059/2003). Namely, acute and subacute oral studies showed similar results, but the whole acute toxicity profile has not been assessed.

It is considered that in technical phosmet two impurities of toxicological concern are present, phosmet oxon and isophosmet, and their quantities differ between technical materials produced at different production plants.
Phosmet has been produced by the applicant at three different plants: Stauffer Chemical Company, Zéneca in the USA, Teckhem in Mexico, and General Química, S.A in Spain, the latter being a current producer in the EU.

The majority of toxicological studies provided were carried out with the technical material from Stauffer Chemical Company (no longer manufactured or used), which contains both isophosmet and phosmet oxon as impurities (the latter is also shown to be rat metabolite).
New technical material (produced by General Química) has a lower content of phosmet oxon ($\leqslant$ 0.8 g/kg) and isophosmet ($\leqslant$0.4 g/kg) compared to the older source (Stauffer), but has three new impurities (<1%), as well as another impurity (which was only detected qualitatively in the old technical material) quantified up to 2%. The identity of these other impurities was not available to the RAC Rapporteurs, and a toxicological profile is not presented in DAR or CLH report.

In order to compare the new and old sources of technical material, an acute oral study in rats and repeated toxicity studies in dogs were performed, showing a 2-times higher LD_{50} in rats with new compared to the old source (but in different rat strains), and rather comparable NOELs in repeated toxicity testing in dogs (see table below).

[1] Unit Pesticide Risk Assessment Peer Review Unit

Table *Comparison between new and old source of Phosmet technical*

Species	Study	Source (purity)	Results
Wistar Rat*	acute oral toxicity*	General Química (95.4%)	LD_{50}= 230 mg/kg (151-347 mg/kg)
Sprague Dawley Rat**		Stauffer (96.1%)	LD_{50}=113 mg/kg (98-130 mg/kg)
Dog	28-day oral	General Química (97.0%)	NOEL=1.5 mg/kg bw /day
Dog	90-day oral	Stauffer (98%±0.5%)	NOEL=1.9 mg/kg bw /day

Reference: *Navarro Aragay, 1998, **McCabe , 1978

HUMAN HEALTH HAZARD EVALUATION

RAC evaluation of acute toxicity

Summary of the Dossier Submitter's proposal

Acute oral toxicity

Three studies are presented in the CLH report, of which two are considered by the Dossier Submitter (DS) as acceptable (McCabe, 1978; Navarro Aragay, 1998), and one study acceptable only as additional information, due to deficient data reporting (Meyding, 1966).

Additionally, acute oral LD_{50} values are presented for two impurities considered relevant, isophosmet and phosmet oxon.

In an **acute oral toxicity study in Sprague-Dawley rats (McCabe, 1978)**; pre-guideline study similar to OECD Test Guideline (TG) 401; doses from 60 to 175 mg/kg bw, material source: Stauffer), LD_{50} values were calculated to be:

- 113 mg/kg bw (101-127 mg/kg bw) in males and
- 113 mg/kg bw (98-130 mg/kg bw) in females.

Most of deaths occurred within first two days, and clinical signs of toxicity, observed at all doses, included depression, tremors, salivation, dyspnea, exophtalmus, chromodacryorrhea, and stains around the ano-genital region. Necropsy revealed red lungs, dark livers, red fluid in the intestine and red spots on the urinary bladder and the intestine.

In an **acute oral toxicity study in Wistar rats (Navarro Aragay, 1998)**; OECD 401 study; doses from 70 to 560 mg/kg bw, material source: General Química), LD_{50} values were calculated to be:

- 230 mg/kg bw (151-347 mg/kg bw) in males and
- 230 mg/kg bw in females (LD_{50} calculated for males was administered by gavage to 6 females with 3/6 deaths).

Mortality occurred between the first and the second day after dosing, and clinical signs of toxicity, observed at ≥100 mg/kg bw, included prostration and chromodacryorrhea, with a dose-related intensity. Necropsy findings were not reported.

In an **acute oral toxicity study in male S/D rats and male S/W mice (Meyding, 1966)**; pre-guideline study, without information on clinical signs, body weights and necropsy findings; doses in rats: 100 – 500 mg/kg bw; doses in mice: 10 – 100 mg/kg bw, material source not reported), LD_{50} values were calculated to be:

- 245 mg/kg bw (161-367 mg/kg bw) in male rats and
- 50.1 mg/kg bw (34.4-73.0 mg/kg bw) in male mice.

Mortality occurred within first two posttreatment days.

Acute oral toxicity of impurities:

- LD_{50} value of isophosmet was 171 mg/kg bw in female Sprague-Dawley rats
- LD_{50} value of phosmet oxon is stated to be 46-50 mg/kg bw (according to Californian Environmental Protection Agency, Spencer, 2003).

The DS concluded that although it is not clear whether the difference in acute oral toxicity between old (Stauffer) and new (General Química) material is due to the differences in impurities or to other reasons (e.g. different rat strain used), both reported LD_{50} values (113 and 230 mg/kg bw) fall within the dose range for classification as **Acute Tox. Cat 3** (50< ATE ≤300 mg/kg bw), **H301 (Toxic if swallowed),** according to CLP. Mice data support this conclusion.

Acute inhalation toxicity

Two acute inhalation toxicity studies are presented in CLH report, both in Charles River CD rats: pre-guideline, OECD TG 403 similar study by **Leong (1977)**, with significant deviations, and **Mould (1995)** study, performed according to US EPA 81-3 guideline (equivalent to OECD TG 403).

In the study by **Leong (1977),** no mortality was observed, and only an increase of the activity and eye squint were observed during exposure.

However, there were significant deviations in the study design (temperature, relative humidity, and oxygen concentration in gas chamber not recorded, gross necropsy not performed) and maximal vapour concentration attained was only 0.152 mg/L, during 4 h of whole body exposure (from the revised DAR, vol. 3, B6: "*The vapours of the test material were generated by passing air at the rate of 20 liters/minute through the coarse crystals contained in a fritted bottom gas bubbler. The vapours emerging from the bubbler were introduced into the chamber without dilution.*").

According to the notifier, technical phosmet is an amorphous, crystalline agglomerate with a negligible vapour pressure (6.5×10^{-5} Pa at 25 °C), while the generation of an atmosphere of respirable particles with an MMAD/GSD recommended by the corresponding testing guidelines was not possible without destroying the test substance. Therefore, an acute inhalation toxicity test in rats was performed with Imidan 70 WP, a wettable powder formulation containing 70% (w/w) phosmet (Mould, 1995), and the LC_{50} value was calculated both for the product and active subtance, phosmet.

In the study by **Mould (1995)**, doses of 0, 0.61, 0.66 and 3.69 mg Imidan 70 WP/L were applied to 5 rats per gender per dose during 4 h (head only) as aerosol (70.2% of phosmet), with MMAD±GSD of particles ranging from 1.61±2.00 µm to 2.38±1.95 µm [which is in line with OECD TG 403 (update 2009) that recommends for aerosols "mass median aerodynamic diameters (MMAD) ranging from 1 to 4 µm with a geometric standard deviation (σg) in the range of 1.5 to 3.0"].

All animals at the highest dose level were killed *in extremis* already during exposure, while in surviving animals clinical signs typical for cholinesterase inhibition were observed. Necropsy in dead animals showed dark or red lungs.

Based on analytically determined concentration of phosmet in the exposure chamber, the LC_{50} was determined to be:

- 1.6 mg/L for the product, and
- 1.12 mg/L for active substance (phosmet).

The DS concluded that although the generation of toxic atmosphere with technical phosmet may be difficult to achieve, results obtained with formulated Imidan 70 WP warrant classification of phosmet as **Acute Tox. Cat. 4 - H332** (Harmful if inhaled) according to CLP (LC_{50} lies within 1.0< ATE ≤5.0 mg/L range for dusts).

Acute dermal toxicity

Two acute dermal toxicity studies are presented in CLH report, one in New Zealand rabbits (McCabe, 1978) and one in Wistar rats (Dos Santos, 1998).

Study by **McCabe (1978)**, was performed in New Zealand rabbits as a "limit test" at single dose level of 5000 mg/kg bw. It is a pre-guideline study similar to OECD TG 402 (dose of 5000 mg/kg bw was used instead of 2000 mg/kg bw; 3 instead of 5 animals were used per gender per dose).

One day before treatment, the abdominal area was clipped, skin was abraded (but without "disturbing the derma"), and material applied with a protective binder for 24 h and then washed and rewrapped with gauze for 3 days (it is not mentioned whether the substance was moistened before the application).

One female rabbit died on 5th day after the treatment, with mild depression and salivation, clamped jaws around caging, and haemorrhagic changes in pancreas and intestines on necropsy. Other animals were free of signs of toxicity.

An LD_{50} >5000 mg/kg bw was proposed based on this study.

In the study by **Dos Santos (1998)**, similar to OECD TG 402, test substance diluted in corn oil was applied at a single dose of 1000 mg/kg bw to 5 Wistar rats per gender (no information about the use of gauzes). One day before the treatment, dorsal area was clipped and substance was applied for 24 h.

Body weights of treated and control rats were very similar and there were no deaths, clinical signs, cutaneous reactions or macroscopic abnormalities during 14 days observation period.

An LD_{50} >1000 mg/kg bw was proposed based on this study.

The DS concluded that available data does not warrant classification for acute dermal toxicity of phosmet.

Comments received during public consultation

Two MSCA supported proposed classification.

Industry argued that phosmet should not be classified for inhalation toxicity since technical phosmet has no potential for aerosol formation and possess only a low potential for vapour formation (technical phosmet is an amorphous crystalline agglomerate with low vapour pressure, and does not contain respirable or inhalable particles).

Assessment and comparison with the classification criteria

Acute oral toxicity

RAC supports the conclusion of the DS that according to the CLP Regulation, phosmet should be classified in Category 3 for acute oral toxicity. Namely, LD_{50} values for both sources of technical material (113 mg/kg bw in case of Stauffer and 230 mg/kg bw in case of General Química) lie within 50< ATE ≤300 mg/kg bw range that corresponds to Category 3 for acute oral toxicity.

Regarding toxicicity profiles of two impurities stated to be relevant, isophosmet and phosmet oxon, RAC is of the opinion that these impurities are not expected to significantly influence acute toxicity of phosmet, especially in the case of technical material form a new source (General Química). The LD_{50} of isophosmet (171 mg/kg bw in rats) is within the same toxicity range as phosmet (50< ATE ≤300 mg/kg bw), and although phosmet oxon is more toxic than phosmet (LD_{50} values of phosmet oxon of 46-50 mg/kg bw lie within 5 < ATE ≤ 50 mg/kg bw, corresponding to Category 2 for acute oral toxicity), its concentration in new material is stated to be ≤ 0.8 g/kg (i.e. ≤0.08%). In addition, phosmet oxon seems to be a rat metabolite, rapidly formed with a maximum of 20%-25% of the applied dose observed after 1 hour, according to *in vitro* study in rat liver microsomes (Hassler, 2006) and supported by limited evidence from *in vivo* studies (McBain *et al.* 1968, Ford *et al.* 1966; these studies, however, are not considered scientifically valid due to analytical problems with metabolites identification in rats' urine).

Acute inhalation toxicity

RAC supports the conclusion of the DS that according to CLP Regulation phosmet should be classified in Category 4 for acute inhalation toxicity based on the results from acute inhalation toxicity study with phosmet formulation as wettable powder, Imidan 70 WP (LC_{50} of 1.12 mg/L lies within 1.0< ATE ≤5.0 mg/L range for dusts that corresponds to Category 4 for acute inhalation toxicity).

RAC disagrees with Industry's opinion that any significant exposure to phosmet via the inhalation route under normal use conditions is not expected.

In the OECD 39 Guidance Document on Acute Inhalation Toxicity Testing (July 21, 2009) it is stated that "acute inhalation testing is not required if the physical form of a test article, as it is marketed or used, precludes any human inhalation exposure (*e.g.*, solid metal block, non-friable granules, composite elastic materials)", and in the Guidance on the Application of the CLP Criteria Guidance to Regulation (EC) No 1272/2008 on classification, labelling and packaging (CLP) of substances and mixtures (Version 4.1, June 2015) it is pointed out that regarding assessment of inhalation toxicity of substances for which inhalation exposure is not expected under realistic conditions "specific problems may arise with respect to classification and labelling, as **these substances are tested in a form** (*i.e.* specific particle size distribution) **that is different from all the forms in which these substances are placed on the market and in which they can reasonably be expected to be used"**.

Technical phosmet is used for production of the plant protection products - wettable powders with different content of phosmet: Imidan 50 WP (50%) and Imidan 70 WP (70%). Therefore, generation of phosmet aerosol could be expected during production of plant protection products (also illustrated by an Industry statement provided during Public Consultation: "*During the manufacturing of plant protection products containing phosmet as active substance, grinding of technical phosmet is performed in a closed automated system resulting in a complete lack of worker exposure to non-formulated ground technical phosmet by the inhalation route*") and during the use of these products (e.g. for Imidan 70 WP, formulation used for acute inhalation toxicity study, MMAD±GSD of particles ranged from 1.61±2.00 μm to 2.38±1.95 μm). RAC also

points out that lowering health risks by good occupational hygiene measures does not affect classification and labelling of the substance, since the C&L process is hazard and not risk-based.

RAC is of the opinion that in the absence of an adequate inhalation study with aerosol of technical phosmet, the results of acute inhalation study with exposure to phosmet formulation in the form of wettable powder could be used for classification purposes.

According to OECD 39 Guidance document on acute inhalation toxicity testing (2009) (p 36-37), "a vehicle may also be considered to enhance the dustiness of solid test articles (powders). The kind and concentration of vehicle should not interfere with the outcome of the study with regard to the airborne test article's analytical stability or toxicity. Ideally, the vehicle selected should be non-toxic with water being given first preference. When a vehicle other than water is used, a vehicle control group should only be used when historical inhalation toxicity data are not available. If a concurrent vehicle control is to be avoided, historical data should show that the vehicle does not interfere with the outcome of the study."

Uncertainty regarding this study is that it is not stated either in CLH report or in revised DAR were the co-formulants of Imidan 70 WP used in vehicle control group. Nevertheless, other ingredients of wettable powder formulation are inert filling materials which are not expected to be toxicologically relevant (from the DAR, p 339: "It is not expected that different amount of co-formulants have an impact on the toxicity of the formulations as can be seen from acute oral and dermal toxicity data obtained with the active substance and the formulated products Imidan 70 WP and Imidan 50 WP, thus the toxicity of the products is given by the active substance Phosmet."), and, indeed, clinical signs in exposed animals were typical for organophosphate poisoning (e.g. lethargy, salivation, tremors).

Phosmet concentration in the chamber atmosphere was measured by gas chromatography, and used in the LC_{50} calculation. Also, adequate particle size distribution was achieved.

Acute dermal toxicity

RAC agrees with the DS that available data (LD_{50} >5000 mg/kg bw in rabbits) does not warrant classification for acute dermal toxicity of phosmet.

Conclusion

Following a comparison of the available acute oral, dermal and inhalation LD_{50} and LC_{50} values with the classification criteria, RAC supports the conclusion of the Dossier Submitter that according to CLP Regulation phosmet should be classified in following categories:

- **Acute Tox. Cat. 3 - H301: Toxic if swallowed** (acute oral LD_{50} values of 113 and 230 mg/kg bw in rats are in 50< ATE ≤300 range, with supporting mice data of LD_{50} of 50.1 mg/kg bw);

- **Acute Tox. Cat. 4 - H332: Harmful if inhaled** (acute inhalation LC_{50} value of 1.12 mg/L in rats is within 1.0< ATE ≤5.0 mg/L range for dusts);

- No classification for acute dermal toxicity is proposed (LD_{50} >5000 mg/kg bw in rabbits is greater than ATE of 2000 mg/kg bw).

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

Summary of the Dossier Submitter's proposal

Eight repeated dose toxicity studies are described in CLH report, 6 with oral exposure (feeding studies) and 2 with dermal exposure.

Critical value for cholinesterase inhibition (considered as biologically relevant) was defined as **>20% decrease** at the moment of sacrifice (either compared to pre-treatment value in the same animal or to control values).

28 day dietary range-finding study (feeding) in B6C3F1 mice (Jones, 1981)

This is a non-guideline (range-finding) study, in which phosmet was given in diet at concentrations of 0, 5, 15, 50, 150 or 500 ppm to 10 mice/gender/dose, with mean daily intake of phosmet of 0, 1.2, 3.8, 12.0, 25.7 and 62 mg/kg bw/day, respectively.

Plasma cholinesterase (ChE) inhibition (compared to control group) was observed at 62 mg/kg bw/day (however, it is not clear how the comparison was performed since values were invalid in control group due to quality control problems and are not presented in CLH report).

Red blood cell (RBC) ChE inhibition (compared to control group) was observed at 12.0 mg/kg bw/day (-30% in males and -24% in females, compared to control), and showed to be dose-dependent.

Statistically significant inhibition of brain ChE was observed at highest dose, 62 mg/kg bw/day, in females (-15.7%, compared to control).

Other adverse effects included statistically significant decrease in platelet count in males (-19.3%) and females (-21.4%) and mean corpuscular volume in females (-2.8%) at the highest dose, an increase in relative weight of liver in both genders at mid and highest dose, and relative weight of kidney in females at the highest dose. On necropsy, there were no apparent treatment related findings.

Decrease in feed consumption observed at 62 and 25.7 mg/kg bw/day was suggested to be the result of feed impalatability. Decreased body weight was also observed at 62 and 25.7 mg/kg bw/day, and an unexplained decrease in body weight in males (7.7%-8.0%) during first two weeks of treatment was observed at 3.8 mg/kg bw/day. At 62 mg/kg bw/day a decrease of activity was observed in all animals, emaciation in 2/10 animals (both in males and females), listless in 10/10 males and 4/10 females, and tremors in one female. No mortality was observed in any treatment group.

A **NOAEL of 3.8 mg/kg/day** for male and female mice was proposed based on RBC cholinesterase inhibition at 12.0 mg/kg bw/day.

28 day dietary range-finding study (feeding) in Beagle dogs (Brown, 2003)

This is a non-guideline (range-finding) study, in which phosmet was given in diet at concentrations of 0, 1.5, 3.0 and 6 mg/kg bw/day to 3 dogs/gender/dose (mean daily intakes: 0, 1.6, 3.2 and 6.4 mg/kg bw/day for males and 0, 1.6, 3.4 and 6.4 mg/kg bw/day for females).

Plasma ChE inhibition was observed at 6 mg/kg bw/day (-23.0% in males and -34.6% in females, compared to pre-treatment values in the same animals).

RBC ChE inhibition was observed at 3 mg/kg bw/day (-21% in males and -24% in females), and was more pronounced at 6 mg/kg bw/day (-71.4% in males and -77.6% in females, compared to pre-treatment values in the same animals).

Brain ChE inhibition above 20% was observed at 6 mg/kg bw/day (-41% in males and -55% in females, compared to controls).

Changes in feed consumption, body weight and organ weight were not observed, and no clinical signs or macroscopical abnormalities at necropsy related to phosmet exposure were noted. Haematology was not assessed. No mortality was observed in any treatment group.

A **NOAEL of 1.5 mg/kg/day** for male and female dogs was proposed based on RBC cholinesterase inhibition of >20% at 3.0 mg/kg bw/day (-21% in males and -24% in females).

90 day dietary study (feeding) in Albino rats (Johnston, 1962)

This is a pre-guideline study, with significant deviations when compared to OECD TG 408 (no ophthalmological examination, sensory reactivity to stimuli and Functional Observational Battery (FOB) investigated, some haematology parameters and some organ histopatology examinations are missing, all biochemistry parameters and urinalysis are missing).

Phosmet was given in diet at concentrations of 0, 20, 100 and 500 ppm to 2 groups of 15 rats/gender/dose, with mean daily intake of phosmet of 0, 2, 10 and 50 mg/kg bw/day, respectively.

Plasma ChE inhibition above 20% (compared to control group) was observed at 10 mg/kg bw/day at week 11, both in males (-23%) and females (-28%), and was observed earlier (week 3) and to higher degree at 50 mg/kg bw/day (-62% in males and -63% in females).

RBC ChE inhibition above 20% (compared to control group) was observed at 10 mg/kg bw/day at week 2, both in males and females (-44%), and complete inhibition both in males and females was found at 50 mg/kg bw/day at the same sampling time (in group 2).

Brain ChE inhibition above 20% (compared to control group) was observed at 10 mg/kg bw/day at the end of the study (13 weeks), both in males (-42%) and females (-39%), and was more pronounced at 50 mg/kg bw/day (-75% in males and -82% in females).

One death occurred (one male at 50 mg/kg bw/day in week 13). No substance-related clinical signs were observed. Feed consumption was decreased in males dosed at 50 mg/kg bw/day (up to 18%), and at 10 mg/kg bw/day (decrease up to 30%). Mean body weights were decreased in males dosed at 50 mg/kg bw/day (7.8% at week 11 and 15.4% at week 14).

Haematological parameters were within the normal range for all dose groups, except increased white cell counts in one male at 50 mg/kg bw/day and one male at 10 mg/kg bw/day at week 9, which was not considered as treatment-related.

There were no pronounced changes in the relative organ weights of the treated animals, except for a slight decrease in the mean relative prostate weight (23.4% in group 1 and 10.6% in group 2) and mean absolute prostate weight (26.9%) in group 1 at 50 mg/kg bw/day, and a slight increase of the relative kidney weight of females at mid and high dose of group 1 (3.8% and 6.4% respectively).

On necropsy, slight liver cell damage was noted in males at 50 mg/kg bw/day and in one male at 2 mg/kg bw/day.

A **NOAEL of 2 mg/kg/day** for male and female rats was proposed based on RBC cholinesterase inhibition of >20% at 10 mg/kg bw/day (-21% in males and -24% in females).

90 day dietary study (feeding) in Beagle dogs (Johnston, 1962)

This is a pre-guideline study, with significant deviations from OECD TG 409. Ophthalmological examination, food consumption, sensory reactivity to stimuli and biochemistry parameters were

not determined, some haematology parameters, organ weights (epididymides and parathyroid) and some histopathology data are missing.

Phosmet was given in diet at concentrations of 0, 10, 75 and 563 ppm to 4 dogs/gender/dose, with mean daily intake of phosmet of 0, 0.25, 1.88, 14.1 mg/kg bw/day, respectively (however, it was not reported whether all food was eaten).

Four-week recovery of cholinesterase inhibition (week 14-18) was investigated in one male and one female dog from the highest and mid dose groups.

Plasma ChE inhibition above 20% (compared to control group) was observed only on week 10 at 14.1 mg/kg bw/day (-27.9%, males and females analysed together).

RBC ChE inhibition above 20% (compared to control group) was observed already at the lowest dose, 0.25 mg/kg bw/day, on weeks 4 and 10 (-22% and -23.6%, respectively), on week 6 at mid dose (-20.4%), and already on week 1 at the highest dose (-33.5%). At the highest dose, inhibition was almost complete on week 13 (-99.7%).

Brain ChE inhibition was observed only at the highest dose (-92.3% in males and -98.1% in females).

ChE recovery:

- regeneration of plasma ChE was essentially complete within 2 – 4 weeks;
- RBC ChE returned to 20–40% of pre-treatment values in 4 weeks (high dose group).

There were no mortalities, and no treatment-related clinical signs were observed.

Weight loss was observed in one male dog at 1.88 mg/kg bw/day (-12.8% on week 14), and in one female dog at 0.25 mg/kg bw/day (-6.5% on week 13).

There were no treatment-related findings in haematology, clinical chemistry, urinalysis or on necropsy. An increase in the relative weight of kidneys (12.2% in males and 60% in females) and adrenals (19.4% in males and 10.5% in females) was observed at high dose, and of kidneys in mid dose males (32.8%). However, it was pointed out that organ weights were determined only for one animal/gender/dose, and high variability of organ weights was observed.

A **NOAEL of 1.88 mg/kg bw/day** for male and female dogs was proposed based on cholinesterase inhibition at the highest dose (observed for plasma, RBC and brain ChE). RBC ChE inhibition above 20% observed at the mid and low dose was not considered relevant since it was not dose-related.

2 year dietary study (feeding) in Albino rats (Johnston, 1966) – not acceptable

This is pre-guideline study, similar to OECD TG 451, but with deviations (some haematology and clinical chemistry parameters are missing, urinalysis was only performed qualitatively, pathology/histopathology of certain organs is missing).

The main drawback, however, is high mortality (up to 84%) across all dose groups and not dose-related, which is considered by the study authors to be related to respiratory infection and not to the treatment.

Phosmet was given in diet at concentrations of 0, 20, 40 and 400 ppm corresponding to 0, 2, 4 and 40 mg/kg bw/day, to 25 rats/gender/dose.

Noticeable decrease of the plasma ChE (≥34% in males and ≥50% in females) and RBC ChE activity (≥76% in males and ≥72% in females) was observed at 40 mg/kg bw/day throughout the treatment period. Brain AChE activity was also decreased at sacrifice (-65% in males and -82% in females). At this dose level, moderate liver vacuolation was also observed.

A **NOAEL of 4 mg/kg/day** was established for male and female rats, but the study was not considered acceptable for further evaluation.

2 year dietary study (feeding) in Beagle dogs (Johnston, 1966)

This is pre-guideline study, similar to OECD TG 451, but with deviations (some haematology and clinical chemistry parameters are missing, urinalysis was only performed qualitatively, pathology/histopathology of certain organs is missing).

Phosmet was given in diet at concentrations of 0, 20, 40 and 400 ppm to 3 dogs/gender/dose, corresponding to 0, 0.5, 1 and 10 mg/kg bw/day.

Plasma ChE was not inhibited above 20% in any treated group.

RBC ChE inhibition above 20% compared to controls was noted at 10 mg/kg bw/day (-61.2% to -85.4% during the study; males and females analysed together).

Brain ChE showed inhibition in dose–related manner, with values of -9.05%, -15.23% and -57.53% observed and lowest, mid and high dose, respectively (compared to controls, with males and females analysed together).

There was no mortality. Only one male dog from the high dose group was sacrificed at week 52 due to its poor condition which was not considered to be treatment-related (based on autopsy findings). No treatment-related changes in body weight was observed.

Clinical findings noted during the course of the study did not follow dose-related pattern (including body temperature, respiration rate, heart rate, ophthalmological examination) "with the exception of lacrimation" (no further data on lacrimation is provided either in CLH report or in the DAR).

Haematological values were within the normal ranges, except for isolated cases of elevated sedimentation rates in several dogs. Clinical chemistry also did not show marked changes. Two isolated cases (at week 78) of very high SGOT (serum glutamic-oxalacetic transaminases) and SGPT (serum glutamic-pyruvic transaminases) were found in controls and low dose group.

Necropsy did not reveal treatment-related effects.

A **NOAEL of 0.5 mg/kg bw/day** for male and female dogs was proposed based on brain cholinesterase inhibition of -15.23% observed at 1 mg/kg bw/day.

21 day dermal study in New Zealand White rabbits (Henwood, 1988)

Study is performed according to US EPA guideline 82-2 (corresponds to OECD TG 401). Phosmet was administered to 5 New Zealand White rabbits /gender/dose, at concentrations of 0, 10, 100 and 1000 mg/kg bw/day for 6 hours/day and 5 days/week using semi-occlusive dressing. Purity of the test substance was 71.2%.

Plasma ChE inhibition above 20% compared to control group was observed at the highest dose, 1000 mg/kg bw/day, both in males (-25.4%) and females (-35.0%), but the difference was statistically significant only for females.

RBC ChE inhibition above 20% compared to control group was observed at the highest dose, both in males (-24.5%) and females (-22.1%), but the difference was statistically significant only for males.

Brain ChE was not measured.

No mortality was observed in treated group. In control group, one male was found dead on day 4 and one female was sacrificed in moribund condition on day 17.

No treatment-related clinical signs of phosmet toxicity were observed.

Treated rabbits at all dose levels exhibited dermal irritation during the study, but its severity did not appear to be dose or gender-related.

No test substance-related effects on body weights, feed consumption, or haematology parameters were observed. Male rabbits at all dose levels had statistically significant lower absolute and relative kidney weights, which was not considered as treatment-related since it was only observed in males, no clear dose-related pattern was observed, and there were no associated remarkable macroscopic and microscopic findings. Necropsy of other organs did also not reveal treatment-related effects.

A **NOAEL of 100 mg/kg bw/day** in rabbits was proposed based on RBC cholinesterase inhibition of higher then 20% observed at 1000 mg/kg bw/day.

21 day dermal study in Sprague Dawley rats (Hilaski, 1999)

The study is performed according to US EPA guideline 82-2 (corresponds to OECD TG 401). Phosmet was administered to the skin of 10 Sprague-Dawley rats/gender/dose, at concentrations of 0, 15, 22.5, and 60 mg/kg bw/day for 6 hours/day and 5 days/week, with a gauze bandaging secured with non-irritating tape. Purity of the test substance was 71.2%.

In addition, a second (expanded) control group of 20 animals/gender was run immediately after the first control group, under the same experimental conditions, in order to increase the control database for cholinesterase inhibition since no historical control data were available.

No mortality was observed. There were no effects on body weight gain, nor marked effects on feed consumption. Clinical signs, dermal reactions or changes in haematology parameters were not considered as treatment-related.

Plasma ChE values were statistically significant decreased in females dosed at 60 mg/kg bw/day (the highest dose) compared to concurrent control group (-37.5%) on day 22. When compared to second expanded group controls there was a significant decrease at 22.5 mg/kg bw/day and 60 mg/kg bw/day in males (-33.3% at both doses) on day 22.

RBC ChE was statistically significantly decreased in females at the highest dose (60 mg/kg bw/day) on day 8 (-28.6%) and non-significantly on day 22 (-21.43%), when compared to concurrent controls.

Brain ChE was statistically significant decreased in males (-35.8%) and females (-61.2%) at the highest dose (on day 22) compared to first control group. The differences were confirmed as statistically significant (and above 20%) when compared to second expanded control group. In females, statistically significant decrease was observed at 15 mg/kg bw/day and 22.5 mg/kg bw/day, but the differences were either lower than 20% (at 15 mg/kg bw/day) or slightly above 20% (-20.4%, at 22.5 mg/kg bw/day), and not significant when compared to second expanded control group (in females dosed at 22.5 mg/kg bw/day difference to second expanded control group was -3.97%).

The necropsy showed statistical increase in brain weight in high-dose males, and increase in brain weight and decrease in liver/brain weight in low-dose females. Brain weight changes did not show a dose-response, and brains in high-dose males and females were histologically normal. Therefore, the finding were not considered as treatment-related.

At the highest dose tested distended uterus was observed.

A **NOAEL of 22.5 mg/kg bw/day** for male and female rats was proposed based on brain cholinesterase inhibition observed at 60 mg/kg bw/day in males and females, since brain ChE inhibition slightly above 20% found in females at 22.5 mg/kg bw/day was not observed when values were compared to second expanded control group.

For repeated dose toxicity the DS also took into account other relevant studies, including carcinogenicity, reproductive toxicity and neurotoxicity studies, and summarised main findings from these studies in Tables 29 and 30, (pages 55-57 of CLH report).

Neurotoxicity studies with phosmet

Two acute neurotoxicity oral studies in rats (Cappon, 1998a and 1998b), one subchronic (90 day) oral study in rats (Cappon, 1999) and two acute delayed neurotoxicity studies in hens (Sprague, 1982 and Johnson, 1997) are described in CLH report.

In two acute neurotoxicity studies (Cappon, 1998a and 1998b) phosmet was given in a single dose by gavage to Sprague-Dawley rats.

In a **dose range-finding acute neurotoxicity study of phosmet in rats** (Cappon, 1998a) reductions of ChE activity >20% were observed at 9 mg/kg bw for plasma ChE and at 6 mg/kg bw for RBC ChE. Clinical signs of organophosphate toxicity (whole body tremors, gait alterations, salivation) were observed at 36 mg/kg bw.

A NOAEL of 3 mg/kg bw was proposed based on RBC ChE inhibition.

In **an acute neurotoxicity study of phosmet in rats** (Cappon, 1998b) ChE inhibition in plasma, RBC and brain was observed at 22.5 mg/kg bw (highest dose), with no observed specific treatment-related changes within the functional observational battery, locomotor activity measurements or neuropathological examination.

NOAEL of 22.5 mg/kg bw was proposed for acute neurotoxicity, and of 4.5 mg/kg bw for ChE inhibition.

Dietary subchronic 90-day neurotoxicity study of phosmet in rats (Cappon, 1999) was performed in accordance to US EPA OPPTS 870.6200 criteria, in 32 Sprague-Dawley rats/gender/dose, at dose levels of 0, 25, 50 and 150 ppm (equivalent to 0, 1.5, 2.7 and 9.4 mg/kg bw/day in males, and 0, 1.6, 3.1 and 11.0 mg/kg bw/day in females).

No test article-related clinical signs, effects on body weight, food consumption, FOB evaluations or total motor activity were observed at any concentration. Also, no effects on absolute and relative weights of brain and brain regions were found, and there were no treatment-related neuropathological lesions.

EFSA established a NOAEL of 25 ppm based on significant ChE inhibition (>20%) at 50 and 150 ppm, although in females ChE activity decrease in the olfactory region (-36%), brainstem region (-21%) and in hippocampus (-33%) was observed already at 25 ppm (1.6 mg/kg bw/day).

An **acute delayed neurotoxicity study with Imidan technical in adult hens** (Sprague, 1982) was performed in White Leghorn hens according to US EPA 81-7 guideline (corresponds to OECD TG 418), with deviations, namely neuropathy target esterase (NTE) activity was not performed and histopathology examination was considered not valid.

Although acute oral toxicity of phosmet was induced, reduced ChE activity in plasma (-55%) and brain (-45%) was found ≥65.6 mg/kg bw, and doses of 200 and 2050 mg/kg bw produced neurotoxicity (alterations in the appearance, behaviour and motor coordination impairment), no clinical signs indicative for delayed neurotoxicity in form of weakness or incoordination with a delayed onset occurred in any of phosmet treated hens.

An **acute delayed neurotoxicity study in the domestic hen** (Johnson 1997) was performed in Lohmann Brown hens according to US EPA 81-7 guideline (corresponds to OECD TG 418).

Oral administration by gavage of a single dose of 600 mg/kg bw resulted in clinical signs (unsteadiness, subdued behaviour, recumbencey and weakness) for 4 days after dosing.

Inhibition of brain AChE (-63%) was also seen at 48 h. However, phosmet treatment did not induce any clinical signs of delayed neurotoxicity (assessed as clinical locomotor ataxia seen 10-20 days later). There were no biologically relevant or significant reductions in NTE in brain or spinal cord 24-48 hours after dosing, and no histopathological evidence of specific acute delayed neurotoxicity was found.

According to described animal data and lack of reported human cases of delayed neurotoxicity after exposure to phosmet (Poisindex, 2016 and open literature search), it was concluded that phosmet does not induce delayed neurotoxicity.

A study in humans is also described in CLH report. This was a randomised double blind ascending single oral dose study with phosmet to determine the no effect level on plasma and RBC cholinesterase activity (Cameron, 1999).

Single oral dose of 1.0, 2.0 and 4.0 mg/kg bw in 27 men and of 2.0 mg/kg bw in 9 women was applied, without adverse events considered to be related to the test compound.

Statistically significant inhibition of RBC ChE was observed already at 1 mg/kg bw (approximately -4.5% 24 h after dosing).

For repeat-dose toxicity the DS also took into account other relevant studies, including carcinogenicity, reproductive toxicity and neurotoxicity studies.

Regarding repeated toxicity assessment (STOT RE), based on the findings from repeated-dose studies, carcinogenicity, reproductive toxicity and neurotoxicity studies in different species, the DS concluded that adverse effects of phosmet consisting of brain and erythrocite cholinesterase inhibition and some clinical signs associated with neurotoxicity (primarily cholinergic effects) were observed after oral and dermal repeated dose exposure, below the cut-off values for classification as STOT RE 1 and at lower dosages for single exposure (dose levels that trigger STOT RE 1 or STOT RE 2 classification, applying Haber's rule for study duration shorter or longer than 90 days, are shown in Tables 29 and 30 in CLH report).

The DS therefore proposed to classify phosmet as **STOT RE 1 (H372)**: Causes damage to nervous system through prolonged or repeated exposure.

The DS also pointed out that the EFSA Panel on Plant Protection Products and their Residues (PPR) provided a scientific Opinion (EFSA, 2013) in which phosmet was grouped in the acute and chronic cumulative assessment groups (CAGs; identification of pesticides to be included in the CAGs based on their toxicological profile regarding effects on thyroid or nervous system) for the nervous system, based on neurochemical endpoints, i.e., inhibition of brain AChE, establishing chronic LOAEL of 1 mg/kg bw/day.

In addition, an association between environmental exposures to organophosphate pesticides and neurodevelopmental/neurobehavioural effects in humans has been recently suggested (EFSA 2013).

Regarding neurotoxicity studies, it was concluded that phosmet is a neurotoxic compound since it inhibits acetylcholinesterase (AChE), which is a class of the enzymes that catalyses the hydrolysis of the neurotransmitting agent acetylcholine (ACh).

Apart from inhibition of AChE and some cholinergic effects observed after phosmet dosing, no other signs of neurotoxicity were observed.

The DS was of the opinion that these neurotoxic effects are covered by the proposed classification of phosmet for acute oral toxicity and repeated dose toxicity after oral and dermal exposure.

Comments received during public consultation

Two MSCA supported the classification proposed by the DS, while one Industry commenter argued against classification stating that "inhibition of cholinesterase should be only considered as relevant for STOT-RE classification if noted in conjunction with additional adverse effects on the nervous system, e.g. clinical signs of neurotoxicity, microscopical finding in the nervous system" which were not observed in the key long-term repeated dose toxicity studies. Also, it was pointed out that there are no phosmet-related health hazards which would have to be specifically addressed by a STOT RE (or STOT SE) classification for effects on the nervous system and which would not be covered by the existing classification for acute effects.

Assessment and comparison with the classification criteria

The DS based their proposal for STOT RE classification primarily on statistically significant RBC and/or brain cholinesterase inhibition of >20%, considered as biologically relevant.

The cut-off value of 20% ChE inhibition was applied by the DS in line with recommendation from the WHO/FAO JMPR[2].

This value is also stated as a cut-off value for adverse effects of organophosphates in ECHA Guidance on the Biocidal Products Regulation: "statistically significant inhibition by 20% or more represents a clear toxicological effect and any decision to dismiss such findings should be justified."

It is recognised that this value does not separate adverse from non-adverse effects, but, according to OPP's (the Office of Pesticide Programs, US EPA) experience, "difference between pre- and post-exposure of 20% or more in enzyme levels is nearly always statistically significant and would generally be viewed as biologically significant" (US EPA, 2000).

RAC's overview of studies in which ChE inhibition was measured and/or clinical observation was performed, is shown in the tables in the Supplemental information section (see Background Document).

It could be observed that in studies other than acute, clinical signs typical for organophosphate poisoning were rarely described, despite inhibition of RBC and brain AChE up to 100% (Johnston, 1962, 90-day study). The ChE inhibition in an oral 28-day study in mice at 62 mg/kg bw/day (Jones, 1981; table in the Supplemental information section) was accompanied by clinical signs such as decreased activity, emaciation, listlessness and tremor. Also, clinical signs typical for organophosphate poisoning (e.g. shaking, piloerection, salivation, unsteady gait) were observed in teratogenicity studies in rats (Hodge, 1991) and rabbits (Pinto, 1991, Moxon, 1991) at 15 mg/kg bw/day in pregnant females (no ChE activity measured). However, in the Jones (1981) study and in the teratogenicity studies the time of onset of these symptoms was not stated.

Namely, it is unclear whether the symptoms related to cholinergic inhibition appeared following repeated exposure or already as a response to the first exposure. Even regarding ChE inhibition, evidence of accumulation of effects with repeated dosing is limited. Comparing acute neurotoxicity studies and studies of longer duration in rats, some cumulative inhibition of RBC ChE could be observed, both in males and females, at 3 and 9 mg/kg bw/day (see table below),

[2] Report of the Joint Meeting of FAO Panel of Experts on Pesticide Residues in Food and the Environment and the WHO Core Assessment Group on Pesticide Residues, held in Rome, 20–29 September 2004: "The Meeting has previously defined criteria for the assessment of cholinesterase inhibition (Pesticide residues in food—1997 evaluations. Part II. Toxicological and Environmental. World Health Organization, WHO/PCS/98.6, Geneva, 1998); these apply equally to the setting of ADIs and ARfDs. For inhibition of acetylcholinesterase activity, a specific cut-off (20%) is used routinely to differentiate between adverse and non-adverse effects."

but not accompanied with clinical symptoms of organophosphate poisoning. On the other hand, at higher dose of 23 mg/kg bw/day, inhibition observed in acute neurotoxicity study (Cappon, 1998b) was at the same level as an inhibition observed in studies of longer duration (Cappon, 1999, Chang *et al.,* 1991).

Assessment of adverse effects following repeated exposure to cholinesterase inhibitors could be complicated by the development of tolerance to organophosphate toxicity, observed both in humans and animals. It is shown that the tolerance is mediated, at least in part, by a homeostatic decrease in the density of cholinergic receptors (Costa and Manzo, 1998). This mechanism compensates for the prolonged increase in acetylcholine levels and protects against acute organophosphate toxicity, but a balance of neuronal connections could be altered and higher brain functions might be compromised (e.g. as shown for memory functions in rats) (McDonald *et al.*, 1988, Bushnell *et al.*, 1993). These functions were not tested in acute and 90-day neurotoxic studies with phosmet in rats.

Although RAC acknowledges that potential neuropsychological adverse effects of chronic, low level exposure to organophosphates in humans (without overt signs of acute poisoning) cannot be excluded (e.g. Pilkington *et al.*, 2001), these studies are not specific for phosmet but refer to organophosphates as a group, and suffer from well-known limitations, inherent to population studies.

Taking into account these uncertainties, in the opinion of RAC.the animal studies with phosmet did not show strong enough evidence to warrant STOT RE classification.

On the other hand, clinical symptoms typical for organophosphate exposure were observed after single oral exposure at a dose lower than those which triggered acute toxicity classification (below 50< ATE ≤300 mg/kg bw range that corresponds to Category 3 for acute oral toxicity proposed for phosmet) (see table below). Namely, in an acute neurotoxicity study in rats (Cappon, 1998a), whole body tremors, gait alterations and salivation were observed after single oral exposure at 36 mg/kg bw, lasting up to 6 h after dosing.

Although these symptoms were of limited duration, they are indicative of acute poisoning with a cholinesterase inhibitor, which has been described for phosmet also in humans at sub-lethal exposures (e.g. Gallagher, 1990, CDC Report, 1999).

In accordance to CLP Regulation which states that STOT SE is defined as "specific, non-lethal target organ toxicity arising from a single exposure to a substance", and includes "all significant health effects that can impair function, both reversible and irreversible", RAC proposes **STOT SE 1, H370 (nervous system)**, based on animal data (Cappon, 1998a) showing morbidity resulting from single exposure at a dose below the Guidance value of 300 mg/kg bw for Category 1, and below dose range of 50< ATE ≤300 mg/kg bw corresponding to acute oral toxicity Category 3 proposed for phosmet. Human poisoning cases support this conclusion.

A distinction between the central and peripheral nervous system is not proposed in the hazard statement, since adverse effects of acetylcholine accumulation due to OP-related inhibition of ChE were observed at cholinergic neuroeffector junctions (muscarinic effects), skeletal neuromuscular junctions and autonomic ganglia (nicotinic effects), as well as in the central nervous system (Roberts and Reigart, 2013).

Specific concentration limits for STOT SE are not proposed since the phosmet dose that induces STOT SE Category 1 is not clearly more than one magnitude below the guidance values for this category (i.e. <30 mg/kg bw for the oral single exposure study).

RAC evaluation of germ cell mutagenicity

Summary of the Dossier Submitter's proposal

In the CLH report, 11 *in vitro* assays and 6 *in vivo* assays are presented, evaluating gene mutation, chromosomal aberrations and DNA damage as endpoints. They are summarised in the following tables.

Table *In vitro genotoxicity assays of phosmet*

Assay/Guideline	System/Dosage	Results/Comments	Reference/ Acceptance
Bacterial reverse mutation test OECD TG 471 GLP: yes	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537 156-2500 μg/plate ±S9	**Positive**: slight, dose-related ↑ of revertants in TA 100 strain, + and – S9 Toxicity not observed (limited solubility of Phosmet); 4 strains were used instead of 5	Majeska, 1986 Acceptable
Bacterial reverse mutation test OECD TG 471 GLP: no	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538, and *E. coli* WP2 hcr 10-5000 μg/plate ±S9	**Positive**: dose-related ↑ of revertants in strain TA 100, + and - S9 - non-significant but dose-related ↑ in the number of revertants in WP2 hcr strain with S9	Shirasu *et al.,* 1979 Acceptable
Bacterial reverse mutation test non-guideline, scientific publication GLP: no	*S. typhimurium* strains and *E. coli* WP2, without S9 doses not reported	**Positive** for strain TA 100 without S9 No information on batch or purity	Shirasu *et al.*, 1984 As additional information due to lack of information
Bacterial reverse mutation test non-guideline, scientific publication GLP: no	*S. typhimurium* TA97, TA98, TA100, TA1535, and TA1538 10-1000 μg/plate ±S9	**Positive** for TA100 and TA97, + and - S9 Toxicity was not observed; Positive control results not reported; 4 concentrations tested instead of 5	Vlcková *et al.*, 1993 Acceptable
***Saccharomyces cerevisiae* gene mutation assay** non-guideline, scientific publication GLP: no	*Saccharomyces cerevisiae* D7 strain 0.5-2.5%	**Positive**: dose-related ↑ of revertants; adenine locus mitotic crossing- over at two highest dose levels No data about positive control, number of replicates, incubation time; no S9; results not confirmed in independent experiment	Vlcková *et al.*, 1993 Not acceptable - lack of information
***In vitro* mammalian gene mutation assay**	L5178Y mouse lymphoma cells	**Positive** without S9 for gene mutations at the	Hertzel, 1986

OECD TG 476 GLP: yes	20-100 μg/plate no S9 4–40 μg/plate with S9	thymidine kinase locus, at cytotoxic concentrations (average relative growth ≤21%) **Negative** with S9	Acceptable
***In vitro* mammalian chromosome aberration assay** OECD TG 473 GLP: yes	L5178Y mouse lymphoma cells 40-100 μg/ml no S9 8-40 μg/ml with S9	**Negative** + and – S9 ↑ of chromosomal aberrant cells found in one culture without S9 at highest dose (100 μg/ml) cell growth inhibition of 80% 100 methaphases measured instead of 200 per concentration; additional experiment without S9 was not done with continuous treatment until sampling at a time equivalent to about 1.5 normal cycle lengths	Snyder, 1986a Acceptable
***In vitro* mammalian chromosome aberration assay** non-guideline, scientific publication; GLP: no	Culture of lymphocytes of human peripheral blood 0.01-20 μg/ml no S9	**Positive**: significant ↑ (but of low magnitude, according to CLH report) of chromosomal aberrations at all doses Test substance purity and evaluation criteria not given	Kurinnyi & Pilinskaya, 1977 Not acceptable - lack of information
Rec assay with *Bacillus subtillis* non-guideline, scientific publication; GLP: no	Recombination wild (H17) and deficient (M45) strains of *B. subtilis* 20-2000 μg/plate	**Negative**: Phosmet did not affect DNA repair	Shirasu *et al.*, 1979 Acceptable with restrictions
***In vitro* sister chromatid exchange assay in mammalian cells** OECD TG 473 GLP: yes	L5178Y mouse lymphoma cells, + and - S9 40-100 μg/ml no S9 8-40 μg/ml with S9	**Questionable**: statistically significant ↑ at all doses, + and - S9, but without clear dose-response and without doubling of SCE frequency Toxicity observed at the highest doses (±S9)	Snyder, 1986a Acceptable
***In vitro* DNA breaks study** guideline not available; GLP: no	Diploid human fibroblast cells 0.25-1 mg/ml, + and – S9	**Negative**: Phosmet did not induce DNA strand breaks (it did not slow DNA nucleoids sedimentation) No information was provided on dose selection or cytotoxicity; higher doses not used because of test substance insolubility	Snyder, 1986b Acceptable with restrictions

+ and – S9 with and without S9; SCE sister chromatid exchange

Several *in vivo* studies are available: two mammalian micronucleus tests, and two studies assessing unscheduled DNA synthesis (UDS) in hepatocytes have been conducted.

Another two *in vivo* studies, published as scientific literature (chromosomal aberrations in mice and DNA alkylation in mice), as well as one study of chromosomal aberrations in workers producing organophosphate insecticides (Kiraly *et al.*, 1979) were included as additional information for assessment purposes.

Table *In vivo genotoxicity assays of phosmet*

Assay/Guideline	System/Dosage	Results/Comments	Reference/ Acceptance
In vivo **mammalian micronucleus test** US EPA 84-2, 84-4; similar to OECD TG 474 GLP: yes	Bone marrow cells from male and female CD-1 mice 17 mg/kg by gavage, single dose (0-45 mg/kg bw in range-finding study) Positive control: cyclophosphamide	**Negative**: no significant ↑ of micronucleated PCEs Cell toxicity: small ↓ of % of PCEs among total erythrocytes at 24h (11% in males, 6% in females) Positive control: appropriate response	Gibbs, 1986 Acceptable
In vivo **mammalian micronucleus test** OECD TG 474 GLP: no	Bone marrow cells from Swiss male mice Imidan 500 PM (Phosmet: 499 g/kg) 13.86 mg Imidan 500 PM/kg bw/day, 2 consecutive days, i.p. Positive control: cyclophosphamide	**Negative**: no significant ↑ of micronucleated PCEs Positive control: appropriate response Only one dose level was tested; PCE/NCE ratio in treated group was slightly greater than in negative control group	Pestana, 1999 Acceptable
In vivo **mammalian chromosome aberration assay** pre-guideline GLP: no	Bone marrow cells from white nonlinear male mice 0, 5, 10, 20 and 50 mg/kg bw, by gavage, single dose Positive control: not used	**Positive**: ↑ of chromosomal aberrations only at 20 mg/kg bw* Purity of test substance not indicated; treatment of control animals not stated; no evaluation criteria stated for aberrations	Kurinnyi, 1975 Not acceptable – lack of information
In vivo **UDS assay** guideline not available GLP: yes	Alpk APfSD rat hepatocytes 32 and 50 mg/kg bw, by gavage, single dose Positive control: DMH	**Negative**: no DNA damage with subsequent DNA repair Positive control: appropriate response	Mackay, 1996 Acceptable
In vivo **UDS assay** OCDE TG 486 GLP: yes	Sprague-Dawley rat hepatocytes Single doses: 54 and 180 mg/kg bw, gavage (2 h time point)	**Negative**: no DNA damage with subsequent DNA repair Positive control: appropriate response	Proudlock, 1998 Acceptable

	54 and 108 mg/kg bw, gavage (14 h time point) Positive control: dimethylnitrosamine or 2-acetylaminofluorene		
***In vivo* DNA alkylation assay** guideline not available GLP: no	Liver and kidney from AB Jena/Halle male mice 20 mg/kg bw, single dose, i.p. (liver and kidney sampling at 6 and 24h)	**Negative**: no DNA adducts (N-7 methylation of guanidine) induced in mouse liver or kidney	Dedek *et al.*, 1984 As additional information

PCEs polychromatic erythrocytes; i.p. intraperitoneal application

* Statically significant ↑ of chromosomal aberrations at 20 mg/kg bw was found (2±0.46% cells with aberrations *vs.* 0.9±0.29% in controls); however at 5, 10 and 50 mg/kg bw no relevant increase was reported.

The DS concluded that phosmet showed genotoxic effect in *in vitro* studies, observed in the *Salmonella* test and in *in vitro* mammalian gene mutation assay.

In vivo studies (mammalian micronucleus tests, UDS assays, and DNA alkylation assay) showed negative results. In Kurinnyi (1975) study (*in vivo* mammalian chromosome aberration assay) the increase in chromosome aberrations was not dose-dependent and the study was not considered acceptable. The DS pointed out that phosmet reaches bone marrow according to toxicokinetics data and cell toxicity observed in two *in vivo* micronucleus tests (small reduction in the number of PCEs after 24 hours in phosmet-treated males and females in Gibbs (1986) study and an increase in the PCE/NCE ratio in phosmet-treated groups in Pestana (1999) study, compared to negative controls).

Therefore, the DS proposed **no classification for genotoxicity** of phosmet, based on the weight of evidence approach.

Comments received during public consultation

Three MSCA supported proposed classification, with one MSCA had questions regarding the DS justification that phosmet is able to reach bone marrow in *in vivo* micronucleus test and on influence of phosmet water solubility on its ability to reach DNA binding sites in *in vivo* DNA alkylation assay.

Assessment and comparison with the classification criteria

RAC agrees with the DS conclusion that phosmet showed genotoxic effect in *in vitro* assays.

Regarding *in vitro* **gene mutation**, phosmet was clearly positive in bacterial reverse mutation tests, while in *in vitro* mammalian gene mutation assay it was positive only at concentrations with rather high cytotoxicity.

Phosmet was positive in all four presented bacterial reverse mutation tests, out of which three were of acceptable quality, although with some deviations from OECD TG 471 (Majeska, 1986, Shirasu *et al.*, 1979 and Vlcková *et al.*, 1993). Positive results were found in *S. typhimurium* strains TA100 and TA97, both with and without metabolic activation, and in *E. coli* WP2 hcr strain with metabolic activation. A positive result was also found for *Saccharomyces cerevisiae* gene mutation assay (D7 strain) without metabolic activation, but this study was published as a non-

guideline, scientific publication, with significant deficiencies in data reporting (Vlcková *et al.*, 1993).

Phosmet was also positive in *in vitro* mammalian gene mutation assay without metabolic activation but negative with metabolic activation (Hertzel, 1986). However, an increase in gene mutations at the thymidine kinase locus in L5178Y mouse lymphoma cells was observed at two highest concentrations, 80 and 100 μg/plate, which showed high cytotoxicity (average relative cell growth of 21% and 16%, respectively). It has been recognised that in a mouse lymphoma assay caution is needed in interpreting positive results obtained at doses inducing reduction in growth (cell survival) greater that 80%, since although the evaluation of cells treated at these levels of cytotoxicity can result in greater test sensitivity, it also bears an increased risk of non-relevant positive results (ICH, 2011).

On the other hand, phosmet was negative in an *in vitro* assay evaluating **chromosomal aberrations** (Snyder, 1986a). RAC agrees with the DS that this test showed negative result both with and without metabolic activation. Namely, an increase of cells with chromosomal aberrations was found in one culture without metabolic activation at the highest dose (100 μg/ml) at which cell growth was inhibited by 80%.

RAC agrees with the DS that the Kurinnyi (1977) chromosomal aberrations assay is not considered acceptable for hazard assessment due to significant deficiencies in methodology and data reporting.

Two additional, non-guideline *in vitro* tests evaluating **DNA damage,** namely DNA repair in *Bacillus subtillis* assay (Shirasu *et al.*, 1979) and DNA breaks study in diploid human fibroblasts (evaluating DNA sedimentation rate after exposure to phosmet, Snyder *et al.*, 1986b), showed negative result. Regarding *in vitro* sister chromatid exchange assay in mammalian cells by Snyder *et al.* (1986a), a statistically significant increased frequency of sister chromatid exchanges (SCE) was found at all doses, with and without metabolic activation, although in the assay with metabolic activation cytotoxicity higher that recommended could reduce the validity of the findings according to ICH (2011) (more than 50% growth reduction at all phosmet concentrations).

RAC agrees with the DS that acceptable *in vivo* mutagenicity assays in mammals showed negative result for genotoxicity of phosmet.

Two mammalian micronucleus tests (Gibbs, 1986 and Pestana, 1999), and two studies assessing unscheduled DNA synthesis (UDS) in hepatocytes (Mackay, 1996 and Proudlock, 1998) were clearly negative.

RAC agrees with the DS that Kurinnyi (1975) study is not considered acceptable for hazard assessment due to significant deficiencies in methodology and data reporting.

During Public Consultation a question was raised regarding the unclear justification that phosmet reaches bone marrow, as a target tissue in mammalian micronucleus tests (Comment number 4). RAC is of the opinion that the justification in CLH dossier was unclear. However, bone marrow is a well perfused tissue, and for chemicals present in the blood it is expected that also the bone marrow is accessible (ECHA Guidance on the Biocidal Products Regulation, 2015). Toxicokinetic data in the CLH report showed systemic availability of phosmet after oral exposure. Oral absorption in rats was shown to be fast and almost complete (based on urinary excretion of 75% to 89% at 96h after dosing, cage wash radioactivity recovery and radioactivity recovery in tissues, Fisher, 1989), and wide distribution in tissues was observed (Ford, 1964). In addition, as pointed out in the CLH report, small decrease in the PCEs among total erythrocytes at 24h was observed (11% in males, 6% in females) at single oral dose applied in micronucleus test, suggesting phosmet-induced bone marrow toxicity.

RAC points out that although the results of Pestana (1999) were clearly negative and the study was performed according to OECD TG 474, Imidan 500 PM formulation with 499 g/kg of phosmet was used in this study (2 consecutive daily doses of phosmet with 6.92 mg/kg bw/day), and no cytotoxicity was observed. It is questionable, therefore, whether high enough dose of phosmet was tested in this study.

An epidemiological study evaluating chromosomal aberrations in workers producing organophosphate insecticides (Kiraly *et al.*, 1979) is also briefly described in CLH report.

In workers producing Safidon 40 WP (assumed to contain 40% phosmet), gaps and isogaps (achromatic lesions) chromatid-type aberration were moderately increased in lymphocytes of the peripheral blood. Differences in frequency of other types of chromosome aberrations, including breaks and isobreaks, were not observed between workers and control group. There are number of uncertainties in this study, primarily lack of information regarding the co-formulants in Safidon 40 WP, and potential confounders, such as exposure to other pesticides/chemicals in phosmet-exposed workers, smoking habit, previous diseases and treatments, and age of workers. Furthermore, only 50 metaphases/person were examined, number of workers in groups varied (data not shown), and no statistical evaluation was performed. The DS also points out that "according to recent guidelines (OECD TG 473) gaps and isogaps - which are regarded as achromatic lesions according to recent definitions - should be recorded separately but generally not included in the total aberration frequency."

In conclusion, RAC is of the opinion that due to the uncertainties in this study it is not consider as relevant for classification purposes.

To summarise, increased frequency of **chromosomal aberrations** was neither found in *in vitro* (Snyder, 1986a) nor in *in vivo* assays (Gibbs, 1986). These results support that the lack of relevance of the epidemiological study (Kiraly *et al.*, 1979).

Regarding **DNA damage**, negative results were found in an *in vitro* DNA repair assay with *Bacillus subtillis* (Shirasu *et al.*, 1979) and a DNA breaks study in diploid human fibroblasts (Snyder *et al.*, 1986b), while positive response was observed in the sister chromatid exchange assay in mouse lymphoma cells (Snyder, 1986a). Nevertheless, this result was not confirmed in *in vivo* tests, namely UDS assays (Mackay, 1996, Proudlock, 1998) and DNA alkylation assay (Dedek *et al.*, 1984).

For the endpoint **gene mutation** positive results were found in bacterial reverse mutation tests and in one mammalian cells assay when tested without S9 mix (Hertzel, 1986). Two *in vivo* UDS assays (Mackay, 1996, Proudlock, 1998) are available, showing negative results for phosmet. However, this test system is not considered to be highly sensitive for "shortpatch repair" (1-3 bases), which are detected with much lower sensitivity than "longpatch repair" (20-30 bases), and therefore the potential ability of a substance to induce point (frameshift) mutations *in vivo* cannot be excluded. Nevertheless, the fact that *in vitro* results for gene mutation primarily rely on bacterial reverse mutation tests and gene mutation in mammalian cells was positive only at cytotoxic phosmet concentrations without metabolic activation, lowers the concern that phosmet could induce gene mutation *in vivo*.

Conclusion

Considering the overall weight of evidence, RAC agrees with the DS that phosmet **does not warrant classification for germ cell mutagenicity**, since there is no reliable evidence from human data on phosmet that would trigger classification in Category 1A, and although phosmet showed some positive effects in *in vitro* genotoxicity assays, the results were not sufficient to trigger classification in Category 1B or 2.

RAC evaluation of carcinogenicity

Summary of the Dossier Submitter's proposal

Two 2-year dietary carcinogenicity studies are presented in CLH report, one in Sprague-Dawley rats and one in B6C3F1 mice.

2-year combined dietary chronic toxicity/oncogenicity study in Sprague-Dawley rats (Chang et al., 1991)

The study was performed according to US EPA guideline 83-5, which corresponds to OECD 453 guideline.

Phosmet was given in diet at concentration of 0, 20, 40, 200, and 400 ppm (equivalent to 0, 1.1, 1.8, 9.4, and 23 mg/kg bw/day in males and 0, 1.1, 2.1, 10.9, and 27 mg/kg bw/day in females) to 70 Sprague-Dawley rats per gender for the control group, 60 rats per gender for phosmet-treated groups at 20-200 ppm, and 20 rats per gender for phosmet dose level of 400 ppm (20 control animals per gender and 10 animals per gender from each treatment group were terminated after 12 months). Dose of 400 ppm was applied only in chronic part (12-month duration).

Serum and RBC ChE activity was measured in 10 rats/gender/group after 6, 12, 18 and 24 months, and brain ChE activity in 10 rats/gender/group at interim sacrifice and in those designated for clinical chemistry at 24 months termination.

Findings:

Survival: no treatment-related deaths were observed, with survival rates in treatment groups similar to or higher than in control animals (see table below). Male control rats showed the lowest survival rate, which was considered by the notifier to be associated with the "fat rat" syndrome of obesity. Nevertheless, according to historical data, the survival rate in control group in the phosmet study was within the range typically seen in contemporaneous chronic studies with Sprague-Dawley rats (Charles River Laboratories, data originate from 24 chronic studies performed in six different laboratories between 1991 and 1997; Table 38, CLH report p 81).

There were no specific clinical signs associated with phosmet treatment.

Body weight gain was reduced throughout the study in males and females at 400 ppm, and in females at 200 during the early phase of the study (week 4). Body weights did not differ significantly between groups at final sacrifice (week 104).

Food consumption was unaffected by treatment.

A plasma ChE activity decrease (>20%) was observed in males and females already at 20 ppm after 18 months (non-significant), and statistically significant decrease was observed after 6 months (first time point measured) in males at 200 ppm (-24%) and 400 ppm (-40%), and at 40 ppm in females (-24%), and persisted till the end of study.

RBC ChE activity was statistically significant decreased in males and females at 40 ppm, 200 and 400 pmm (-19% in both genders low dose, >20% in both gender in the mid and high doses).

Brain ChE activity was significantly reduced in males and females at 200 ppm (-20% and -27%, respectively, after 12 months) and at 400 ppm (-34% and -43%, respectively, after 12 months).

On necropsy absolute and relative kidney weights were statistically significant reduced in females at 400 ppm.

Histopathological examination showed increased incidence of fatty liver change in both males and females at 200 ppm and 400 ppm. The severity of this finding was increased in males at 200 ppm. No changes of serum cholesterol or triglycerides were observed.

The incidence, type and severity of tumours in the treatment groups was comparable to the concurrent control group and in line with that expected for Sprague-Dawley rats.

Table *Main findings in 2-year phosmet dietary study in Sprague-Dawley rats*

	Males					**Females**				
Dose (ppm)	**0**	**20**	**40**	**200**	**400**	**0**	**20**	**40**	**200**	**400**
Dose (mg/kg bw/d)	***0***	***1.1***	***1.8***	***9.4***	***23***	***0***	***1.1***	***2.1***	***10.9***	***27***
Number of animals	70	60	60	60	20	70	60	60	60	20
Survival at 12 months (%)	93	85	90	93	90	96	93	95	97	100
Survival at 24 months (%)	20	24	32	38	-	32	35	32	42	-
Body weight at sacrifice (% of Control)	-	110	104	104	NA	-	102	92	102	NA
Fatty liver change										
incidence (%)	37	60	53	72*	75*	14	18	23	35*	20*
severity (%)										
Gr 1&2	27	32	42	32	30	11	15	17	27	20
Gr 3	7	25	8	18	30	3	2	3	5	0
Gr 4&5	3	3	3	22	15	0	2	3	3	0

NA not applicable; Gr Gradus; *Statistically different from control by Fisher's exact test with Bonferroni correction (P<0.05); Dose of 400 ppm was applied only in chronic part of the study (12-month duration)

The DS concluded that phosmet toxicity in the study was mainly manifested by statistically significant reduction in brain (at 200 ppm and 400 ppm) and red blood cells cholinesterase activities (at and above 40 ppm). An increased incidence of liver fatty changes was observed from 200 ppm.

The DS pointed out that the results of this study are supported by chronic/carcinogenicity study in rats with phosmet by Johnston (1966) in which also not carcinogenic effects could be observed (reported under repeated dose toxicity studies).

2-year dietary oncogenicity study in B6C3F1 mice (Katz et al., 1984)

The study was performed according to US EPA guideline 83-2, which corresponds to OECD TG 451 guideline.

Phosmet was given in the diet at concentrations of 0, 5, 25 and 100 ppm (equivalent to 0, 1, 4 and 14 mg/kg bw/day in males and 0, 1.2, 5 and 18 mg/kg bw/day in females) to 60 B6C3F1 mice per gender per group. An interim sacrifice was performed at 12 month, with 10 mice/gender/group.

Findings:

The survival rate was reported to be not affected by phosmet treatment. However, no data on mortality are presented in CLH report or in revised DAR.

Body weights: After the first 3 treatment months, the body weights of the high dose males were slightly but significantly higher compared to controls (8% at final sacrifice, 104 week). Body weights of the high dose females were also slightly elevated compared to controls from months 2 to 18 of treatment (up to 17%), and were similar at final sacrifice. The DS did not consider these differences as biologically significant.

Feed consumption: Reduced consumption was observed in high dose males, intermittently during the first 9 months of the study (up to 17%), and in high dose females during the first 3 months (up to 13%). Since no concomitant body weight reduction was found, it was suggested that the reduced feed consumption for the high dose reflected increased food efficiency and/or reduced spillage.

Clinical signs: Convulsions were observed in male mice when animals were handled or within a short period of time after they were returned to their cages. The incidence was significantly higher only in the high dose group. It was pointed out that spontaneous, handling-induced convulsions in B6C3F1 and related strains of mice were described in the open literature and have been associated with extended individual housing of the test animals (Takemoto *et al.*, 1975, King *et al.*, 1955, Serota *et al.*, 1986). In the present study animals were individually caged. It was also stated that cholinergic convulsions, as life-threatening events, are expected to be associated with mortality in at least some of the test animals. According to CLH report (no data presented), the convulsions observed in the study were not correlated with brain cholinesterase inhibition or with unscheduled (early) deaths, and there was no evidence of typical signs of cholinergic poisoning (e.g. diarrhoea, salivation, lacrimation, miosis, coma). It was, therefore, concluded that convulsions observed in the study are not treatment-related.

No treatment-related haematological changes were observed.

Plasma ChE activity was significantly reduced, >20%, in top dose males and females, throughout the study.

RBC ChE activity in males and females no treatment related changes were observed.

Brain ChE activity was statistically significantly decreased after 12 months of treatment at all doses, by 22% to 31% in males and 28% to 34% in females. At the termination of the study, no significant changes were observed in males, while in females values were significantly reduced at mid (-14%) and high dose (-22%).

Organ weights: At interim sacrifice (12 months) increase in the mean relative liver weight in males at 100 ppm was observed (12%).

Histopathology

Non-neoplastic findings: In males at the highest dose (100 ppm), possibly treatment-related increase in incidence of mild vacuolar degeneration of the liver was observed.

No further treatment related finding have been observed.

Neoplastic findings: Frequency of hepatic adenomas was significantly increased in male mice dosed at 100 ppm (45% *vs.* 22%), and frequency of Harderian gland adenomas was increased in male mice dosed at 5 ppm and 100 ppm, but not in 25 ppm males. These findings are shown in the table below.

Other types of liver tumours (lymphoma, hemangiosarcoma, various types of sarcomas) and Harderian gland tumours (lymphoma, papillary cyst adenoma) that occurred at low frequency and without dose-response, are not presented in the Table below.

Table *Main findings in 2-year phosmet dietary oncogenicity study in B6C3F1 mice*

	Males				**Females**			
Dose (ppm)	**0**	**5**	**25**	**100**	**0**	**5**	**25**	**100**
Dose (mg/kg bw/d)	***0***	***1***	***4***	***14***	***0***	***1.2***	***5***	***18***
Number of animals	60	60	60	60	60	60	60	60
Body weight at sacrifice (% of control)	-	103	97	108	-	93	100	98
Vacuolar degeneration, individual hepatocytes	3/49 (6)	1/50 (2)	2/50 (4)	16/50 (32)	0/49	0/50	0/48	0/50
Neoplastic changes [N (%)]								
Liver								
Hepatocellular adenoma	13/60 (22)	10/60 (17)	14/60 (23)	27/60 (45)	6/60 (10)	4/60 (7)	5/59 (8)	11/60 (18)
Adenocarcinoma	0/60	0/60	0/60	1/60 (2)	0/60	0/60	0/59	0/60
Hepatocellular carcinoma	13/60 (22)	11/60 (18)	11/60 (18)	14/60 (23)	5/60 (8)	4/60 (7)	3/59 (5)	9/60 (15)
Pulmonary metastasis of hepatocellular carcinoma	2/60 (3)	4/60 (7)	2/60 (3)	3/60 (5)	2/60 (3)	0/60	0/59	0/60
Harderian gland								
Adenoma	3/60 (5)	7/60 (12)	4/59 (7)	9/60 (15)	1/59 (2)	0/15	3/14 (21)	2/59 (3)
Adenocarcinoma	0/60	2/60 (3)	0/60	0/60	0/60	1/15 (7)	0/14	1/59 (2)

Increased incidence of Harderian gland adenomas was not considered treatment-related, since no dose-response was observed.

Regarding hepatic adenomas, study authors pointed out that the biological significance of this observation is questionable, since:

- the distribution of hepatocellular carcinomas and the incidence of pulmonary metastasis of malignant liver tumours were similar in all male dose groups;
- phosmet treatment did not appear to affect the period of latency for the development of liver tumours;
- the mean survival rate of mice with hepatocellular tumours were comparable to controls (i.e. 668 days for high dose males *vs.* 630 days for controls);
- benign tumours were increased only in one dose group of one sex; and
- tumour incidence of hepatic tumours is considered within the normal background incidence for this strain of mice.

In support of the last statement, historical data on hepatic adenoma incidence were presented in the CLH report, namely Katz *et al.* (1986) and Haseman *et al.* (1999).

Katz *et al.* (1986); addendum I: Historical control data on the incidence of hepatocelullar adenoma and hepatocelullar carcinoma in B6C3F1 mouse generated in a 2-year study.

The same strain of mice was used for the study, purchased from the same supplier, housed in the same laboratory at approximately the same time and with the same principal pathologist. Sixty B6C3F1 mice of each gender were used.

Following results were obtained (data from CLH report Table 45):

	Incidence (affected/total, %)	
Tumor type	**Male**	**Female**
Hepatocellular adenoma(single or multiple)	25/60 (42%)	9/60 (15%)
Hepatocellular carcinoma (single or multiple)	10/60 (17%)	3/60 (5%)
Either hepatocellular adenoma or carcinoma.	31/60 (52%)	11/60 (18%)

It was concluded that although the number of incidence of hepatocellular adenomas and incidences of hepatocellular adenomas combined with carcinomas at 100 ppm (Katz *et al.*, 1984) was significantly increased, it was not significantly different to the incidences in the historical control group (45% *vs.* 42% for adenomas and 58% *vs.* 52% for total adenomas and carcinomas in males exposed to 100 ppm phosmet and historical control, respectively). Additionally, it was pointed out that the incidence of adenomas in males of the historical control (42%) was significantly higher in the concurrent control group of Katz *et al.* (1984) study (22%).

Further historical control data (Haseman *et al;* 1999) are presented, showing the incidence of hepatocellular adenoma in male mice collected from 400 long term carcinogenicity studies with the same strain of mice (B6C3F1) conducted by NTP (National Toxicological Program and the National Cancer Institute) over 25 year period.

Conclusions for 2-year mouse study

In this study, liver was shown to be the target organ with observed hepatic degenerative changes and increased incidence of liver tumours.

In the opinion of the DS, although prevalence of liver adenomas in males of the highest dose group was slightly higher than in the historical control data (Katz *et al.*, 1986), liver tissue in B6C3F1 mice is known for its high spontaneous tumour rate in this animal strain. This is also reported in the Guidance on the Application on the CLP Criteria (ECHA, November 2013). Based on the fact that the tumours occurred in the highly sensitive B6C3F1 strain of mice, the DS considered that the significant increase in liver cell adenomas in the high dosed males is of uncertain biologic significance and of questionable relevance.

Conclusion on phosmet carcinogenicity

In the opinion of the DS, although the mode of action for induction of the tumours has not been clarified, the tumours occurred in the highly sensitive B6C3F1 strain of mouse. Phosmet is non-genotoxic, and no increase in liver tumours was seen in exposed rats. Therefore, the finding is most likely to have been specific to the strain and species tested.

The DS proposed no classification for carcinogenicity of phosmet, since

- There is no epidemiological evidence regarding the carcinogenicity of phosmet to humans, so Category 1A is not appropriate.
- Although evidence for carcinogenic response was found in mice, the tumour type found was largely sex-specific and there are significant doubts about the relevance to humans. In addition, phosmet is not genotoxic. Therefore, Category 1B is considered inappropriate.
- No other tumour type was detected in mice and no increase in liver tumours were seen in exposed rats, and a robust genotoxicity/mutagenicity database confirms that phosmet is

neither genotoxic nor mutagenic *in vivo*. These finding do not justify classification of phosmet into Category 2.

In the CLH report, it is stated that at the ECB TC C&L it was agreed not to classify phosmet for carcinogenicity, and the EFSA Peer Review of phosmet (EFSA, 2011) did not include a proposal of classification regarding the carcinogenic potential of this substance, but it is mentioned that "the experts could not agree on the proposed classification as carcinogenic with the application of R40".

Comments received during public consultation

Two MSCA supported no classification for carcinogenicity, with one MSCA providing comments regarding reliability of NTP historical controls (Haseman *et al.*, 1999), potential influence of increased body weights on tumour incidence in mice, and corrections for table with mice tumour data in CLH report.

Assessment and comparison with the classification criteria

RAC agrees with the DS that **2-year carcinogenicity study in Sprague-Dawley rats** (Chang *et al.*, 1991) showed no carcinogenic effect of phosmet.

Although interpretation of the results is affected by low survival rate in all groups, including controls (20-42% survival at the end of the study, not treatment-related), RAC considers that this study can be used for the assessment of carcinogenicity of phosmet. According to OECD TG 453, in order for a negative test to be acceptable, survival in each group should be no less than 50 per cent at 24 months for rats. However, according to the UK comment in the Reporting Table of the EFSA Peer Review Report on phosmet (2006), if half the test animals in the highest dose groups survive for 90% of the intended period (i.e. if survival falls below 50% after week 94 in 104 week study), "the number of animals receiving test material for the majority of the lifespan should be adequate to detect carcinogenic activity" and "the study is not considered to be unduly compromised". In the study of Chang *et al.* (1991) survival started to fall below 50% after week 91, and in the high dose groups it fell below 50% after week 97 in males and week 99 in females. According to historical data, survival in control group in this study was within the range typically seen in contemporaneous chronic studies with Sprague-Dawley rats (at the lower bound of the range of data provided by Charles River Laboratories, which originates from 24 chronic studies performed in six different laboratories between 1991 and 1997; see Table 38, CLH report).

At EPCO Expert Meeting 33 (as quoted in EFSA Scientific Report 2006) study limitations were noted, but it was concluded that the study is adequate for the assessment of carcinogenicity in rats. Also, this study was accepted by US EPA during the Interim Registration Eligibility Decision for Phosmet (IRED) in 2001 and WHO (JMPR, 1994, 1998), with a conclusion that phosmet is not carcinogenic in rat.

RAC agrees with the DS opinion that phosmet was not carcinogenic in a **2-year dietary oncogenicity study in B6C3F1 mice (Katz *et al.*, 1984)**.

The incidence of harderian gland tumours did not follow a clear dose-response (see table below), and even at the highest dose it was similar to the occurrence expected for this mouse strain (mean incidence of 11% in male and 8% in female B6C3F1 mice, with higher incidence of adenomas compared to carcinomas (Brayton, 2009).

The hepatocellular carcinoma incidence in males did not follow a dose-related pattern, while in females at the highest dose the incidence was lower than in male control (15% vs. 22%, respectively). In addition, in control females the incidence was slightly higher than the historical control data in Katz *et al.* (1986) (8% vs. 5%, respectively).

Regarding liver adenomas, RAC agrees with the DS that the increased incidence of this type of tumour in B6C3F1 mice strain does not provide evidence strong enough for carcinogenicity classification, taking into account historical data and ECHA CLP Guidance 2015 recommendations regarding liver tumours in B6C3F1 mice. In addition, as also pointed out by one MSCA during PC, a slight increase in body weights (8%) in top dose males could be related to increased incidence of liver adenomas, since it is considered that mice liver tumours are within the group of tumours most strongly associated with an increase in body weight (Haseman *et al.*, 1998).

Regarding hepatocellular adenomas, comparison with historical control data (same strain and same breeder as in animals used in main study) reported by Katz *et al.* (1986) showed that incidences of liver adenomas in males and females at the high dose in the main study (Katz *et al.*, 1984) were only 3% higher than in historical controls. No dose-response pattern was observed. RAC considers this higher incidence than in historical controls not sufficient to trigger classification for carcinogenicity for a tumour known to have a high rate of spontaneous incidence (ECHA, CLP Guidance 2015).

Haseman *et al.* (1999), although comprising data from long term carcinogenicity studies with the same strain of mice conducted by NTP (National Toxicological Program and the National Cancer Institute), covers 25-year period and is, therefore, not considered by RAC as an acceptable historical control.

In ECHA CLP Guidance 2015, the liver tumours in B6C3F1 mice are given as an example for tumours with a high spontaneous incidence, and it is stated that "where the only available tumour data are liver tumours in certain sensitive strains of mice, without any other supplementary evidence, the substance may not be classified in any of the categories."

In line with justification given above, and acknowledging that:

- There are no human data showing phosmet carcinogenicity (rendering Cat. 1A inappropriate),
- Phosmet was not carcinogenic in rats and no treatment-related effect on incidence on any other tumour type in mouse study was found (rendering Cat. 1B classification inappropriate),
- Phosmet was not mutagenic in vivo, and
- In mice, the only treatment effect (liver tumours) was observed only at the highest dose, for a tumour type with a high spontaneous incidence, with an increase slightly above adequate historical control data (rendering Cat. 2 inadequately justified, in line with ECHA CLP Guidance),

RAC proposes **no classification for carcinogenicity** for phosmet.

RAC evaluation of reproductive toxicity

Summary of the Dossier Submitter's proposal

Six studies evaluating the reproductive toxicity of phosmet are presented in CLH report:

- 2-generation study in CD rats (dietary study, Meyer & Walberg, 1990)

- Prenatal developmental toxicity in Alpk APfSD rats (gavage study, Hodge, 1991)
- Prenatal developmental toxicity in New Zealand White rabbits (gavage study, Pinto, 1991)
- Prenatal developmental toxicity in New Zealand White rabbits (gavage study, Moxon, 1991)
- Prenatal developmental toxicity in CD rats (Staples *et al.*, 1976)
- Study of the effect of phosmet on embryogenesis in Wistar rats (Martson & Voronina, 1976).

Three of them were guideline studies (Meyer & Walberg, 1990, Hodge, 1991, Moxon, 1991), one was a range-finding study (Pinto, 1991), and two were scientific publications (Staples *et al.*, 1976, Martson & Voronina, 1976).

2-generation study in CD rats (Meyer & Walberg, 1990)

The study was performed according to US EPA Guideline 83-4 and OECD TG 416.

Phosmet was administered in the diet to 25 rats/gender/dose levels of 0, 20, 80 or 300 ppm (equivalent to 0, 1, 4.2 and 16.4 mg/kg bw/day in males and to 0, 1.8, 7.3 and 25.5 mg/kg bw/day in females), through two generations (P0, P1) with two litters (F1a, F1b; F2a, F2b) per generation.

P0 parents yielded F1a litter and, after weaning of the F1a pups, F1b litter. Pups from F1a litter were sacrificed after weaning [postnatal day (PND) 21]. F1b litters were culled on PND 4 to 8 pups (approximately 4 males and 4 females per litter). Culled pups were necropsied, while the surviving pups were weaned at PND 21, and either continued on treatment as P1 parental animals (25 animals per gender from each group) yielding F2a and F2b pups, or necropsied.

Parental toxicity

In parental generation in both genders, signs of toxicity were observed at 80 and 300 ppm. At 300 ppm they included significant decreases in body weight, body weight gain and feed consumption during growth, gestation and lactation periods, clinical signs (impaired general health status, dehydration in P0 females and chromorhinorrea in P1 females), statistically significant reduction (>20%) of plasma and RBC ChE, significant absolute and relative organ weight reductions, and liver impairment (mild to moderate centrilobular hepatocellular vacuolisation in P1 males).

At 80 ppm, toxicity signs included decreased body weight gain on certain days during growth, gestation and lactation, clinical signs (dehydration in P0 females), significant decreases of relative weight of liver and adrenals in P0 females and of spleen in P1 females and statistically significant reductions (>20%) in RBC cholinesterase activities.

Reproductive toxicity

At 300 ppm, a statistically significant reduction in fertility indexes in P0 to generate F1b litters and in P1 to generate F2a and F2b litters was observed (see table below). Mating index was statistically significantly decreased in P1 for F2b litters. Significant reduction of the absolute weight of testes in P1 males and of the relative weight of ovaries in P0 females was observed. Significant decrease in the number of total born pups delivered was observed in F1b, F2a and F2b litters.

At 80 ppm, a statistically significant reduction of fertility indexes were observed from 80 ppm in P0 to generate F1b litter.

Developmental toxicity

At 300 ppm, a significant reduction of the numbers of total born pups delivered and live born pups per litter (postpartum day 0) in F1b, F2a and F2b litters was observed. On postpartum day

21, there was a significant decrease in live pups per litter in F1a, F2a and F2b litters, and in the pup body weights in all generations.

Table (taken from CLH report Table 57): Summary of reproductive indices

Males																
	P0 (1st mating for F1a)				P0 (2st mating for F1b)				P1 (1st mating for F2a)				P1 (2st mating for F2b)			
	Doses (ppm)															
Males	**0**	**20**	**80**	**300**	**0**	**20**	**80**	**300**	**0**	**20**	**80**	**300**	**0**	**20**	**80**	**300**
Number of males	24[e]	25	25	25	23	25	25	25	25	25	25	25	25	25	25	25
Cohabited	24	25	25	25	23	25	25	25	25	25	25	25	25	23	25	25
Positive Mating Sign	24	25	24	25	23	25	22	20	24	23	22	18	22	22	18	**14***
Mating index (%) [a]	100	100	96	100	100	100	88	80	96	92	88	72	88	96	72	**56***
Fertile	22	24	18	21	22	22	**17***	**15***	22	21	15	**12****	19	20	15	**9***
Fertility index (%) [b]	92	96	72	84	96	88	**68***	**60***	88	84	60	**48****	76	87	60	**36***
Females																
Number of females	25	25	25	25	25	25	25	25	25	25	25	25	25	25	25	25
Cohabited	25	25	25	25	25	25	25	25	25	25	25	25	25	25	25	25
Positive mating sign [c]	25	25	24	25	25	25	22	20	24	23	22	18	22	24	18	**14***
Mating index (%)	100	100	96	100	100	100	88	80	96	92	88	72	88	96	72	**56***
Delivered	23	24	18	21	24	22	**17***	**15***	22	21	15	**12****	19	21	15	**9***
Without positive mating signs	0	0	1	0	2	0	2	1	4	0	1	2	0	1	0	0
Fertility index (%)	92	96	72	84	96	88	**68***	**60***	88	84	60	**48****	76	84	60	**36***
Gestation index (%) [d]	92	96	75	84	96	88	77	75	92	91	68	67	86	88	83	64
Number of litters with <6 pups	0	1	0	1	1	1	3	1	1	1	1	0	0	1	1	2
Mean gestation length (days)	21.9	21.9	21.8	22.1	22	21.8	21.7	22	21.8	22	21.8	21.9	21.9	21.8	21.7	22
Pups																
No. of pups/litter (total born) [f]	14.1	14.6	15	14	15.1	14.8	13.4	**12.6***	14.1	14.2	14.3	**11.8***	15.3	14.6	14.9	**11.0***
No. live pups/litter (day 0)	13.2	14.5	14.7	13.6	14.7	14.2	12.9	**11.9***	13.8	13.6	13.9	**11.5***	14.8	14.1	13.9	**9.6****
No. live pups/litter (day 21)	13.6	14.2	14.4	**11.9***	7.9	7.7	7.2	6.9	13.5	12.8	13.5	**9.4****	7.8	7.7	7.7	**5.2***
Pup weight on day 0 (g)	6.4	6.3	6.1	6.2	6.4	6.2	6.4	6.3	6.4	6.2	6.3	6.0	6.4	6.4	6.4	6.2
Pup weight on day 21 (g)	44.5	42.4	41.3	**33.2****	49.4	49.1	50.8	**41.8****	42.2	44.3	41.8	**33.8****	56.4	54.5	53.1	**33.0****

a The percentage of animals in a dose group that had positive mating signs or whose females became pregnant.
b The percentage on animals in a dose group that had live born pups
c Females that delivered without positive mating signs are included
d The percentage of sperm-positive mating resulting in the birth of live pups: a female that delivered offspring was considered to have been sperm-positive even if sperm was not detected.
e Male No. 110 was cohabited on 6/13/88, however, he died on 6/16/88. It is not included in the summary of reproductive indices.
f Number of live pups on day 4 plus all pups either dead or missing from day 0 to day 4.
* Significantly different from the 0 ppm dose level, $p<0.05$, two-tailed
** Significantly different from the 0 ppm dose level, $p<0.01$, two-tailed

The DS concluded that a NOAEL of 20 ppm (1 - 1.8 mg/kg bw/day) could be proposed based on the adverse effects observed at 80 ppm which included statistically significant reduction (>20%) of RBC ChE in P0 and P1 parental animals of both sexes, clinical signs (dehydration in P0 females), relative organ weight reductions in females of P0 (liver and adrenals) and of P1 (spleen) and decrease in body weight gain during growth, gestation and lactation periods (statistically significant in P0 males during growth (days 7, 21 and 112), and during lactation in P0 females (1st mating) on day 14).

A NOAEL of 20 ppm (about 1 - 1.8 mg/kg bw/day) was proposed based on statistically significant reduction in fertility indexes observed from 80 ppm, and lower mating indexes also observed from 80 ppm. However, the DS pointed out that these effects were observed in the presence of parental toxicity.

A NOAEL of 80 ppm (about 4.2 - 7.3 mg/kg bw/day) was proposed based on reduced number of total born pups delivered, impaired pup survival and reduced pup body weights at 300 ppm. Again, it was pointed out that developmental toxicity effects occurred only at a dose level which was toxic to the parental animals.

Teratogenicity studies

Prenatal developmental toxicity in Alpk APfSD rats (Hodge, 1991)

In this guideline study (US EPA 83-3, corresponding to OECD TG 414), phosmet was given by oral gavage to 24 female Alpk APfSD rats/gender/dose, at dose levels of 0, 5, 10 and 15 mg/kg bw/day, from days 7-16 of gestation. On day 22 of gestation, all females were killed and their uteri and foetuses examined for the signs of developmental toxicity.

Clinical signs of toxicity typical for organophosphate poisoning were observed in the dams at the highest dose of 15 mg/kg bw/day (shaking, piloerection and salivation), and at all doses there were signs of urinary incontinence, which showed an increase at 15 mg/kg bw/day.

Body weight gain was statistically significantly decreased at 15 mg/kg bw/day during dosing (47.5% compared to controls), and, to a lesser extent, at 10 mg/kg bw/day (11.9%).

Food consumption was significantly reduced at 15 mg/kg bw/day.

There were no compound-related effects regarding macroscopic abnormalities, intrauterine survival, sex ratio, post-implantation losses, and major, minor or variant external/visceral or skeletal defects.

Mean foetal weight at 15 mg/kg bw/day was reduced (3.7%), although the value (4.89 g) was within historical control range (4.87-5.25 g), with no adverse effect on litter weight.

It was concluded that no indications of embryo/foetotoxicity or teratogenicity were noted up to (and including) the high dose level, 15 mg/kg bw/day.

A NOAEL of 5 mg/kg bw/day for maternal toxicity was proposed based on clinical signs of organophosphate poisoning and decreased food consumption and body weight gain. The NOAEL of 15 mg/kg bw/day (highest dose level) was proposed for developmental toxicity including teratogenicity.

Prenatal developmental toxicity in New Zealand White rabbits (Pinto, 1991)

In this range-finding study phosmet was administered by gavage to 10 female New Zealand White rabbits/dose, at 0, 5, 10 and 15 mg/kg bw/day.

At 15 mg/kg bw/day clinical signs of organophosphate toxicity were observed (shaking, constricted/dilated pupils, salivation, high stepping gait) and one female died.

Body weight gain and food consumption were not affected by treatment.

A statistically significant reduction (22.8%) in mean litter weight was observed at 15 mg/kg bw/day.

Non-significant reductions of mean litter weight (-19.9%) at 10 mg/kg bw/day, and uterus weight at mid and high doses (11.1% and 18.9% respectively) were observed.

There were no substance-related external abnormalities.

It was concluded that the dose of 15 mg/kg bw/day is maternally toxic, and that slight indications of impairment in foetal development were noted ≥10 mg/kg bw/day. Phosmet did not cause external malformations or variations in this range-finding study.

Prenatal developmental toxicity in New Zealand White rabbits (Moxon, 1991)

In this guideline study (US EPA 83-3, corresponding to OECD TG 414), phosmet was given by oral gavage to 20 New Zealand White rabbits/gender/dose, at dose levels of 0, 2, 5 and 15 mg/kg bw/day, from days 7-19 of gestation. On day 30 of gestation, all females were killed and their uteri and foetuses examined for the signs of developmental toxicity.

At 15 mg/kg bw/day, two animals died, one with signs of salivation, diarrhoea, mucus on faeces and significant weight loss, and animals that survived had reduced body weight gain and signs typical for organophosphate poisoning (unsteady gait, salivation and increased breathing rate).

At 5 mg/kg bw/day, there were body weight gain reductions on certain days during the treatment and overall body weight gain was 13% lower compared to controls.

The number of foetuses with minor skeletal defects (areas of reduced or lacking ossification) was statistically significantly increased at 5 and 15 mg/kg bw/day. There was no evidence for teratogenicity.

It was concluded that phosmet is not teratogenic in the rabbit. Slight foetotoxicity was seen only in presence of maternal toxicity at the highest dose (15 mg/kg bw/day).

A NOAEL for maternal toxicity of 5 mg/kg bw/day was proposed, based on mortality, clinical signs of organophosphate poisoning and decrease in body weight gain observed at 15 mg/kg bw/day.

A NOAEL for development of 5 mg/kg bw/day was proposed based on slight foetotoxicity manifested by areas of reduced or lacking ossification at 15 mg/kg bw/day.

Prenatal developmental toxicity in CD rats (Staples et al., 1976)

In this non-guideline study (scientific publication), phosmet was neither teratogenic nor foetotoxic after administration through diet to CD rats at any dose level. Maternal effects were observed at doses ≥22 mg/kg bw/day, as a decrease in food consumption and body weight gain.

NOAEL of 10 mg/kg bw/day for maternal toxicity, and the NOAEL ≥29 mg/kg bw/day for development was established.

Study of the effect of phosmet on embryogenesis in Wistar rats (Martson & Voronina, 1976)

In this non-guideline study (scientific publication), phosmet application (by gavage) induced embryo and foetotoxicity at single high dose (30 mg/kg bw) and at 1.5 mg/kg bw/day throughout pregnancy. Malformations (hydrocephaly, hypognathia, general edema and dislocation of extremities) were also noted, either after single or repeated exposure.

There were no adverse effect at 0.06 mg/kg bw/day.

No data on maternal toxicity were available.

It was concluded that phosmet embryo/feototoxicity and teratogenicity at the dose of 1.5 mg/kg bw/day (exposed throughout the embryogenesis period), but that these data has to be taken with precaution.

General conclusions on reproductive toxicity

The DS considered that the **fertility** effects observed at 300 ppm and incidentally at 80 ppm, without gross lesions or histopathological changes in relevant organs and no dose-related effects on spermatogenesis, are likely to be a secondary, non-specific consequence of general toxicity observed at these dose levels.

Since no effects providing sufficient evidence of impaired fertility were observed in the absence of parental toxicity, no classification for fertility effects is proposed.

Regarding **developmental** toxicity the DS proposed no classification since the effects observed were not sufficiently severe and occurred at doses in presence of maternal toxicity.

The DS also pointed out that at TC C&L meeting in Arona, 15-16 May 2007, it was agreed not to classify phosmet for effects on fertility, and that there are no concerns about developmental toxicity.

Comments received during public consultation

Two MSCA supported proposed no classification for reproductive toxicity.

Assessment and comparison with the classification criteria

2-generation study in CD rats (dietary study, Meyer & Walberg, 1990)

Parental Toxicity

Survival rate in all groups was high (92-100%), and reported not to be affected by phosmet treatment.

In CLH report it was stated that at 300 ppm "impaired general health status including clinical signs indicative for an organophosphorous ester" were observed. However, to RAC's opinion clinical signs of toxicity described (dehydration, chromorhinorrhoea, dacriorrhoea, thin hair, swollen eyes and pinna) do not represent typical clinical feature of poisoning with cholinesterase inhibitor (e.g. ataxia, twitching, fasciculations, miosis, salivation, diarrhoea, lacrimation, muscle weakness, depressed responses, tremors). Dacriorrhoea (lacrimation), which could be a sign of organophosphate poisoning, was reported only for P0 females, and was not accompanied by other typical symptoms.

Body weight gain was decreased during growth (pre-mating) period at 300 ppm in both males and females of P0 and P1 generation (see figures below), with the difference in cumulative (overall) body weight gain on day 56 of approximately -10% in males and females of P0 generation, and -6% in males of P1 generation. At day 168 overall body weight gain was 4% lower in P0 males. At 80 ppm, a decrease in body weight gain during growth period was observed only in P0 males (10% lower cumulative body weight gain on day 56, and 6% on day 168, compared to controls). Food efficiency was not affected.

During gestation, body weight gain at 300 ppm was lower in P1 females at both 1st and 2nd mating (cumulative body weight gain 79% and 76% of control, respectively) and drastically lower in P0 females at 2nd mating (cumulative body weight gain of -144% of control during the last week of gestation). Food efficiency was decreased in these groups (see figures below). Body weight gain at lower doses was similar to controls.

During lactation, negative overall body weight gain was observed in all groups including controls (-1 g), but was more pronounced in females dosed at 80 ppm, and especially at 300 ppm, with lower food efficiency in P0 females after 1st mating, and in P1 females after 1st and 2nd mating (see figures below).

Statistically significant reductions of plasma ChE (>20%) were observed at 80 ppm and 300 ppm in P0 females, and reductions in RBC ChE were observed at 80 ppm and 300 ppm in P0 and P1 males and females, with dose-related increase (see table below).

In males dosed at 300 ppm, statistically significant decrease in absolute organ weights was found for testes (-10.1% in P0 and -6.9% in P1 males) and spleen (-7.9% in P1 males). However, no clear dose-relationship was observed, except for testes in P0 males. Treatment-related effects on spermatogenesis or histopathology of testes were not observed. Mild to moderate centrilobular hepatocellular vacuolisation was observed in 10 out of 25 P1 males.

In P0 and P1 females dosed at 300 ppm a decrease in absolute and relative (to whole body) organ weights were observed for several organs - spleen, liver, kidneys, adrenals, heart and ovaries, among which dose-related, statistically significant decrease was observed for relative weights of heart (-12%), kidneys (-12%), liver (-12%) and spleen (-23% and -26%) in P0 and

P1 females. Regarding absolute weight of ovaries, statistically significant, dose-related decrease was found for P0 females (-20.6%).

In females dosed at 80 ppm, dose-related decrease in relative weights of organs was observed for heart (-8% in P0, -4% in P1), kidneys (-5% in P0 and P1), liver (-6% in P0 and P1) and spleen (-9% in P0, -14% in P1), although statistically significant was only a decrease in relative weight of liver in P0 and spleen in P1.

Histopathological changes in these organs were not described, either at 300 ppm or 80 ppm.

Developmental Toxicity

A decrease in total number of born pups and live born pups was found at 300 ppm, a dose at which maternal toxicity was present. There were no marked differences in pups' weight on PND 0, but at the time of weaning (PND 21), pups' body weight at a top dose was decreased compared to control (up to 41% decrease; however during the 3rd pre-weaning week, direct exposure of pups to phosmet could be expected due to consumption of mother's pelleted feed by pups).

In pups, no biologically relevant soft tissue anomalies or malformations were observed, and there was no increase in external pup anomalies at any dose level.

In conclusion, RAC agrees with the DS that no findings indicating developmental toxicity were observed, since developmental toxic effects (decrease in total number of born pups and live born pups) was found only at the highest dose, 300 ppm, which also induced maternal toxicity, i.e. reduction in body weight gain, especially during pre-mating period and even greater decrease during gestation (reduction of cumulative body weight gain up to 24%), as well as a decrease in absolute and relative weights of number of organs. Marked inhibition of RBC ChE (up to -81% compared to control values) was also found.

Fertility

Mating, fertility and gestation indices were reduced at 300 ppm and 80 ppm. Reduced mating index was found for P0 parental animals at 2nd mating, and P1 parental animals at 1st and 2nd mating (lower than in controls for up to 32% at 300 ppm and up to 16% at 80 ppm), with clear dose-related pattern observed in P0 parental animals at 2nd mating, and P1 parental animals at 1st mating.

Fertility index was reduced in P0 and P1 parental animals after 1st and 2nd mating for up to 40% at 300 ppm and up to 16% at 80 ppm, with clear dose-response for P1 parental animals after 1st mating. According to RAC analysis, for females dosed at 80 ppm statistically significant decrease in fertility index was observed for P0 at 2nd mating (Pearson chi square = 6.64, Fisher's exact P = 0.023) and for P1 at 1st mating (Pearson chi square = 5.09, P = 0.024)].

Gestation index was reduced at 300 ppm and 80 ppm in P0 and P1 females after 1st and 2nd mating (up to 25% at 300 ppm and up to 24% at 80 ppm), with clear dose-related decrease in P0 females after 2nd mating and P1 females after 1st mating.

Regarding the fertility effects observed at mid and high dose (80 and 300 ppm), i.e. decrease in mating, fertility and gestation indices, it is questionable whether this can be considered as a consequence of general toxicity of phosmet, as considered by the DS.

At 300 ppm, a decrease in body weight gain (cumulative lower body weight gain up to 24%) and food efficiency during gestation were observed, as well as decrease in absolute and relative organ weights of several organs (e.g. up to 26% reduced relative spleen weight). At 80 ppm, however, no significant clinical signs of phosmet toxicity were observed, body weight gain was not strongly affected either during pre-mating period or during gestation, and relative organ weight was only slightly reduced (-4% to -8% for heart, kidneys and liver; -9% and -14% for spleen) without

histopathological changes. The most prominent effect of phosmet was a reduction of plasma and RBC ChE (up to -59% for RBC ChE), without overt clinical signs of organophosphate poisoning.

RAC regards the maternal toxicity at 80 ppm as not severe enough to explain the fertility effects as a consequence of general toxicity.

RAC recognises that changes in fertility indices were not accompanied by treatment-related morphological changes in reproductive organs, including spermatogenesis. However, it could be hypothesised that observed decreases in fertility indices are caused by behavioural changes, as a consequence of potential neurotoxic effects of phosmet.

It has been shown that repeated exposure to cholinesterase inhibitors could lead to the development of tolerance to organophosphate toxicity, protecting against acute organophosphate poisoning, but inducing more subtle changes in higher brain functions (as described in STOT RE). Specific tests for neurotoxicity were not performed in this study, and neurotoxicity testing in acute and 90-day neurotoxicity studies was limited to FOB (Functional Observation Battery), covering home cage, handling, open field, sensorimotor and neuromuscular and physiological observations, and mean ambulatory and total motor activity counts as locomotor activity outcomes. No higher brain functions, such as memory, were tested. Possible neurobehavioral changes leading to mating and fertility impairment can therefore not be ruled out. In addition, it has been recognised that cholinergic pathways are important for male fertility, including sperm transport through the duct system or motor activity of mammalian testicular capsule (Gupta, 2011, da Silva Júnior *et al.*, 2013).

Consequently, classification for fertility is proposed.

Teratogenicity studies

In these studies the lowest maternal NOAEL identified was 5 mg/kg bw/day in rats and rabbits, based on clinical signs of organophosphate poisoning and decreased food consumption and body weight gain in the prenatal developmental toxicity study in Alpk APfSD rats (Hodge, 1991) and on mortality, clinical signs of organophosphate poisoning and decrease in body weight gain in the prenatal developmental toxicity study in New Zealand White rabbits (Moxon,1991) In the rabbit decrease in maternal body weight gain during pregnancy observed at 5 mg/kg bw/day is not considered to be severe enough to justify this dose for LOAEL setting in this species.

Significant Embryo/foetotoxicity indicating teratogenicity was only observed at dose levels inducing also maternal toxicity.

One study of the effect of phosmet on embryogenesis in Wistar rats (Martson & Voronina, 1976), in which signs of embryo/feototoxicity and teratogenicity were found, is considered not relevant for assessment since no data on purity of phosmet and on maternal toxicity are available.

RAC conclusion on reproductive toxicity of phosmet

Fertility

Since there is no evidence that phosmet adversely affects fertility in humans, Category 1A is not justified.

According to CLP Regulation, classification in Category 1B is largely based on data from animal studies providing *clear evidence* of an adverse effect on sexual function and fertility in the absence of other toxic effects, or if occurring together with other toxic effects the adverse effect on reproduction is considered not to be a secondary non-specific consequence of other toxic effects.

Substances are classified in Category 2 for reproductive toxicity when there is some evidence from humans or experimental animals, but the evidence is not sufficiently convincing to place

the substance in Category 1. Such effects shall have been observed in the absence of other toxic effects, or if occurring together with other toxic effects the adverse effect on reproduction is considered not to be a secondary non-specific consequence of the other toxic effects.

In the rat 2-generation study, mating index, fertility index and gestation index were reduced at 80 ppm and 300 ppm in both generations and at both mating time points in a generally dose-dependent manner. Nevertheless, parental toxicity was noted at 300 ppm and, to a certain degree, also at 80 ppm dose level (reduction of plasma and RBC ChE, although without overt clinical signs of organophosphate poisoning). Observed parental effects, however, are not considered serious enough to explain fertility adverse effects solely as a consequence of general toxicity of phosmet.

Available data for phosmet does not allow determination of mechanisms responsible for observed changes in fertility indices and an assessment of their relevance for humans. Although treatment-related morphological changes in reproductive organs, including spermatogenesis, were not found, there is a possibility that observed fertility effects are caused by changed rats' behaviour as a consequence of neurotoxic effects of phosmet, or via disturbances in cholinergic pathways involved in fertility functions. These mechanisms cannot be either proven or completely dismissed since assessment of neurobehaviour and specific physiologic fertility functions that are regulated by cholinergic pathways were not reported in this study.

In light of these uncertainties, RAC proposes **Category 2** for adverse effects on fertility for phosmet.

RAC does not propose specific concentration limits (SCLs) for adverse effect on fertility since the main effects found in the 2-generation study appeared at a dose level which is within the range for medium potency group (ED_{10} >4 mg/kg bw/day, and <400 mg/kg bw/day), for which GCLs are applied.

Calculation is given in the Supplemental information section (see Background Document).

Developmental toxicity

Since there is no human data indicating developmental toxicity of phosmet, and animal data from 2-generation study in rats and teratogenicity studies in rats and rabbits do not indicate significant embryo/foetotoxicity in the absence of clear signs of maternal toxicity, no classification for developmental toxicity of phosmet is proposed.

ENVIRONMENTAL HAZARD EVALUATION

RAC evaluation of aquatic hazards (acute and chronic)

Summary of the Dossier Submitter's proposal

Phosmet is currently included in Annex VI to the CLP Regulation. The DS proposed to modify the existing environmental hazard classification by adding a separate chronic M-factor of 10 based on the long-term NOEC (21 d) of 0.00078 mg/L for *Daphnia magna* and the substance being considered rapidly degradable.

Degradation

The following items were discussed and concluded by the DS in the CLH report.

Photodegradation in water: US EPA guideline 161-2

Photolytic DT_{50}= 4.5 days (pH 5). As the processes of photolysis and hydrolysis occur together, and the results of the two are aggregated in the test, photolysis DT_{50} was calculated after taking the hydrolysis losses away from the whole. This resulted a photolytic DT_{50} = 4.5 d at pH 5.

Hydrolysis was studied by Chang, 1987 (US EPA guideline 161-1) at three pH values in sterile buffered solution at 25 °C. Phosmet was demonstrated to be rapidly hydrolysed at all pHs tested with half-lives of less than 8 days. The half-lives were DT_{50}: 180 h at pH 5, DT_{50}: 7.8 h at pH 7 and DT_{50}: 4.5 min at pH 9.

The major degradation products determined at pH 5 (day 11, 25 °C) in the organic phase were phthalamic acid (PAA, 34.3% 14C), phthalic acid (PA, 8.8% 14C), N-hydroxymethyl phthalimide (PiMOH, 2.6% 14C), phthalimide (Pi, 9.8% 14C) and N-methyl phthalimide (2.1% 14C). The aqueous phase contained mainly O,O-dimethyl phosphorodithioic acid (79.4% of original phosmet) with a small amount of O-methyl phosphorodithioic acid (4.1%).

According to McBain *et al.* (1973, no guideline), hydrolysis in buffered water resulted in half-life equal to DT_{50}: 225 h at pH 5, 18 h at pH 7 and DT_{50}: <10 min at pH 9. The amount of phthalimide (Pi) at the end of the study (28 days) was 21% and PAA is the likely degradation product of Pi.

After 4 months incubation at pH 9 the main hydrolysis product is phthalamic acid (PAA). Phthalimide (Pi) is rapidly degraded (0.6% at 5 hours after the start of the treatment).

At pH 7: the main metabolites (>10%) are phthalamic acid (PAA; after 50 days 72%), phthalimide (Pi; 21.2% after 7 days) and hydroxymethyl phthalamic acid (PaAMOH, 25% after 5 days). Smaller amounts of phthalic acid (PA 6% after 5 days) were also identified.

At pH 5: the main metabolites are PAA (34.3%), Pi (9.8%) and phthalic acid (8.8%). At the end of the study 43.9% phosmet was detected. N-hydroxymethyl phthalimide (PiMOH) was identified at low levels.

Ready biodegradability was tested according to OECD TG 301D, a closed bottle test with activated sludge microorganisms (Kelly and Paterson, 2003). The tested phosmet concentration was 3.62 mg/L this concentration was not toxic to sewage treatment micro-organisms.

Biodegradation after 28 days = 19.5% → Not readily biodegradable.

Degradation in a water–sediment system was studied in a laboratory simulation test at 20 °C with sandy and sandy silt loam type natural sediments (Lynn *et al*., 2003). The degradation of ^{14}C-phosmet in aquatic systems was studied under aerobic conditions in two water/sediment systems. The pH increased during the test, so the conditions do not represent a worst case, as phosmet hydrolyses faster at high pHs.

Overall, the study demonstrated that phosmet degraded/dissipated rapidly (both hydrolysis and partitioning to sediment): water phase DT_{50} = 0.246 days (mean); total system DT_{50} = 0.505 days (mean).

Degradation products were similar as in hydrolysis: phthalamic acid, phthalic acid and N-hydroxymethyl phthalimide. DS stated that ecotoxicities of these metabolites are not significant, so the degradation products shall not be classified as hazardous to the aquatic environment. Toxicity results of degradation products have not been included into the CLH report.

Degradation products and pathways of ^{14}C-phosmet were studied according to OECD TG 308 – Aerobic and anaerobic transformation in aquatic sediment systems – by Kidd *et al*. (2005), see Figure 4 in the CLH report.

Aerobic and anaerobic transformation in soil (OECD TG 307) was studied by Lynn *et al*. (2003). DT_{50} / DT_{90} in three soils, under aerobic conditions showed rapid aerobic degradation: loamy sand (5.0 / 16.7 days), silt loam (2.7 / 8.85 days) and sandy loam (1.65 / 5.5 days).

Due to the results of all the abiotic and biotic degradation studies the DS concluded that phosmet can be considered as rapidly degradable in the environment, however no justification on the degradants has been included in the CLH report regarding the fulfilment of the relevant requirements of the CLP Regulation.

Bioaccumulation

Phosmet has a log Kow of 2.95 (at pH 7.25 ºC) which is below the cut-off value of log $K_{ow} \geqslant 4$, so no potential for bioaccumulation is expected, in relation to the CLP criteria. No experimental BCF was established. The metabolites have a log Kow between – 0.26 and 1.26 (calculated by KOWWIN version 1.66).

Toxicity

In addition to the three taxonomic groups of fish, Daphnia and algae, the toxicity toward several other aquatic and sediment dwelling organisms have been studied, and daphnids proved to be most sensitive both in acute and chronic tests.

Acute toxicity – lowest EC_{50} results of fish, invertebrates and algae:

- Fish (*Lepomis macrochirus*): LC_{50} = 0.07 mg/L (Acute toxicity to fish: Guide*, 1975);
- Invertebrates (*Daphnia magna*) EC_{50} = 0.00211 mg/L (OECD TG 202);
- Invertebrates (*Gammarus pseudolimnaeus*); EC_{50} = 0.0024 mg/L (Aquatic toxicity to crustaceans and insects: Guide*, 1975);
- Algae growth inhibition (static) (*Raphidocelis subcapitata,* former name: *Selenastrum capricornutum*): ErC_{50} (24h) = 1.2 mg/L, (OECD TG 201).

*Guide of Committee on Methods of toxicity tests with aquatic organisms (1975)

Chronic toxicity (lowest NOEC values):

- Fish early life stage (*Oncorhynchus mykiss*) NOEC (96d) = 0.0032 mg/L (mm) (EPA-FIFRA 40 CFR 72-4);
- Invertebrates (*Daphnia magna*) NOEC (21d) = 0.00078 mg/l (EPA-FIFRA 40 CFR 72-4);
- Algae growth inhibition (static) (*Raphidocelis subcapitata*, former name: *Selenastrum capricornutum*): NOE_rC = 0.36 mg/l (mm); E_bC50 (48h) = 0.51 mg/l; NOE_bC = 0.14 mg/l (mm) (OECD TG 201).

Data on the ***toxicity of the degradation products/metabolites*** were not included in the CLH report but some results of ecotoxicity studies were provided by the DS following a comment during Public Consultation (see more about these results in the paragraph of 'Additional key elements').

Comments received during public consultation

Two MSCA agreed with the proposed environmental classification.

Another MSCA questioned the designation of phosmet as rapidly degradable and stressed that further information is required including clarification of primary and ultimate DT_{50} values and evaluation of ecotoxicity studies on the degradants. They referred to the ECHA guidance on the application of the CLP criteria (version 4.1, June 2015), particularly to section 4.1.3.2.3.2

outlining the decision scheme that may be used as general guidance to facilitate the decision in relation to rapid degradability of phosmet in the aquatic environment.

Assessment and comparison with the classification criteria

Phosmet is photodegradable, but rapid photolysis is usually not relevant for CLP purposes.

Phosmet is considered not readily biodegradable, because it does not fulfil the criterion of biodegradation of >70% within 28 days measured by OECD TG 301D (equivalent to EC method C.4-E).

Phosmet is considered rapidly degradable in the aquatic environment, due to **primary** biotic and abiotic degradation (based on the disappearance of the parent compound) in a water–sediment simulation test, where the DT_{90} values for two different types of sediments were ca. 3 and 0.3 days, which are well below the >70% within 28 days criterion.

Data on primary degradation can only be used for demonstrating rapid degradation under CLP when it is satisfactory demonstrated that the degradation products should not be classified as hazardous to the aquatic environment.

Some toxicity data of degradation products/metabolites are available in the literature, but the DS has not collected and included such data into the CLH Report, however they provided some information on degradants' toxicity following a comment during PC. The additionally provided information on the metabolites toxicity (O,O-dimethylphosphoric acid, O,O-dimethylphosphorodithioic acid potassium salt, phthalamic acid) revealed EC_{50} >100 mg/L, NOEC = 100 mg/L and for phthalic acid a NOEC = 6.04 mg/L, which are toxicity values not implying classification. However, not all degradation products' toxicities are available, whilst the additional ecotoxicity results were derived from just one species (*Daphnia magna*).

To investigate the toxicity of degradants further, additional QSAR investigations were undertaken; they indicate generally low aquatic toxicity for most key degradants, with the exception of some ECOSAR predictions for PiMOH (algae), PI (algae) and O,O-dimethylphosphorodithioic acid (Daphnia). Furthermore, there is information for some of the key degradants in the ECHA dissemination site (see http://echa.europa.eu/information-on-chemicals/registered-substances) as these substances have been registered under REACH. The majority of the disseminated information on aquatic toxicity is of low reliability and/or indicates low aquatic toxicity. A separate report on these findings is attached to the current ODD.

As discussed earlier, the results of the environmental simulation tests with ^{14}C-phosmet ('Degradation in water–sediment systems', OECD TG 308 and 'Aerobic and anaerobic transformation in soil' – OECD TG 307, both studies from Lynn *et al*. 2003) are available and did show rapid primary degradation. However, the DS did not evaluate ultimate degradation based on $^{14}CO_2$ production and did not directly compare these results to the CLP criteria. So, the DS proposal to consider phosmet as 'rapidly degradable' is not consistent with the decision scheme of the CLP Guidance Version 4.1., paragraph 4.1.3.2.3.2., page 496, shown below:

"*A substance is considered to be not rapidly degradable unless at least one of the following is fulfilled:*

a. It is demonstrated to be readily biodegradable;

RAC note: The substance was shown to be not readily biodegradable (OECD TG 301D)

b. It is demonstrated to be ultimately degraded in a surface water simulation test;

RAC note: Water–sediment studies resulted in slow ultimate degradation calculated from the produced $^{14}CO_2$ percentages: 22% and 29% CO_2 (<70%) on the day 30th (OECD TG 308).

c. It is demonstrated to be primarily degraded biotically or abiotically e.g. via hydroysis, in the aquatic environment with a half-life <16 days (corresponding to a degradation of >70 % within 28 days), ***and*** *it can be demonstrated that the degradation products do not fulfil the criteria for classification as hazardous to the aquatic environment*;

RAC note: Phosmet degraded/dissipated rapidly, but valid toxicity results are not available for all relevant degradants.

The same Guidance continues by saying that

"*When these preferred data types are not available rapid degradation may be demonstrated if one of the following criteria is justified:*

d. The substance is demonstrated to be ultimately degraded in an aquatic sediment or soil simulation test with a half-life of < 16 days (corresponding to a degradation of >70 % within 28 days) …"

RAC note: Water–sediment and soil test with ^{14}C phosmet resulted in less than 70% radioactive $^{14}CO_2$ within 28 days.

Consequently, RAC concludes that phosmet cannot be considered as rapidly degradable in the environment for CLP purposes, unless the degradants prove to be non-hazardous to the aquatic environment.

After discussions in the RAC plenary, RAC concluded that the information currently provided on the ecotoxicity of the degradation products (some of which are classifiable) is not adequate to consider phosmet as rapidly degradable.

In summary phosmet should be classified as follows:

Aquatic Acute 1 (H400),
based on the lowest aquatic acute toxicity value from the results of three trophic levels:
Daphnia magna EC_{50} = 0.00211 mg/L < 1 mg/L.

Acute M factor = 100
based on the criterion of: 0.001 mg/L < EC_{50} = 0.00211 mg/L $\leqslant$ 0.01 mg/L.

Aquatic Chronic 1 (H410),
based on the lowest aquatic chronic toxicity value from the results of three trophic levels:
Daphnia magna NOEC = 0.00078 mg/L < 0.1 mg/L.

Chronic M factor = 100

based on the criteria of: 0.0001 mg/L < NOEC=0.00078 mg/L $\leqslant$ 0.001 mg/L and not rapidly degradable in the environment.

RAC evaluation of hazards to the ozone layer

Summary of the Dossier Submitter's proposal

DS did not propose any classification due to the lack of data.

Comments received during public consultation

No comments have arrived for this end point.

Assessment and comparison with the classification criteria

Any substances having an Ozone Depleting Potential (ODP) greater or equal to the lowest ODP (i.e. 0.005) of the substances currently listed in Annex I to Regulation (EC) No 1005/2009 should be classified as hazardous to the ozone layer (category 1).

Phosmet has an ODP = 0.

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ANNEXES:

Annex 1 The Background Document (BD) gives the detailed scientific grounds for the opinion. The BD is based on the CLH report prepared by the Dossier Submitter; the evaluation performed by RAC is contained in 'RAC boxes'.

Annex 2 Comments received on the CLH report, response to comments provided by the Dossier Submitter and by RAC (excluding confidential information).