Committee for Risk Assessment

RAC

Annex 2

Response to comments document (RCOM)

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

N-(2-nitrophenyl)phosphoric triamide

EC Number: 477-690-9
CAS Number: 874819-71-3

CLH-O-0000006850-73-01/F

Adopted

17 September 2020

ANNEX 2 - COMMENTS AND RESPONSE TO COMMENTS ON CLH PROPOSAL ON N-(2-NITROPHENYL)PHOSPHORIC TRIAMIDE

COMMENTS AND RESPONSE TO COMMENTS ON CLH: PROPOSAL AND JUSTIFICATION

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

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

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Substance name: *N*-(2-nitrophenyl)phosphoric triamide
EC number: 477-690-9
CAS number: 874819-71-3
Dossier submitter: Austria

TOXICITY TO REPRODUCTION

Date	Country	Organisation	Type of Organisation	Comment number
25.10.2019	France		MemberState	1
Comment received				
Based on available data, we agree with the classification Repr. 1B, H360FD.				
Dossier Submitter's Response				
Thank you for this support.				
RAC's response				
RAC agrees with the proposed classification in category 1B for effects on sexual function and fertility but not for effects on development. RAC proposes classification for effects on development in category 2 because the clear developmental effects were observed at a dose level also inducing maternal toxicity including most likely functional kidney effects. Therefore, it cannot be concluded that the observed developmental effects are not (at least in part) secondary, non-specific consequences of the maternal toxicity.				

OTHER HAZARDS AND ENDPOINTS – Specific Target Organ Toxicity Repeated Exposure

Date	Country	Organisation	Type of Organisation	Comment number
25.10.2019	France		MemberState	2
Comment received				
Based on available data, we agree with the classification STOT RE 2, H373 (kidney).				
Dossier Submitter's Response				
Thank you for this support.				
RAC's response				
Thank you for this support.				

OTHER HAZARDS AND ENDPOINTS – Hazardous to the Aquatic Environment

Date	Country	Organisation	Type of Organisation	Comment number
23.10.2019	Sweden		MemberState	3
Comment received				
p. 31-34; Toxicity to algae: The Swedish CA agrees with the proposed environmental classification; Aquatic chronic 3, H412. This is partly based on the algae ErC50 of 51.4 mg/L (nominal concentration). Based on the presented information, the initial exposure concentrations in the algae study were not maintained throughout the testing period. However, at the two highest nominal test concentrations (50 and 100 mg/L) the measured concentrations were close to 80% (79.8 and 81.3 %, respectively) of the nominal concentration at the end of the test. Therefore, it was argued that nominal concentrations could be used for calculating ErC50. The Swedish CA agrees with this and further concludes that if measured concentrations were used instead in the ErC50-calculation, the conclusion would have been the same.				
Dossier Submitter's Response				
Thank you for your comment. We agree that if measured concentrations were used instead of nominal for the ErC50-calculation the conclusion would have been the same. When using the geometric mean of the measured concentrations (table 26-p.33) the ErC50 would be 49.93mg/L (estimated via linear regression, R^2 = 0.98). This value is almost equal to the ErC50 based on nominal concentrations (= 51.4 mg/L) and does not alter the classification proposal.				
RAC's response				
RAC does not agree with the DS to base the E_rC_{50} on nominal concentrations. Test concentrations were not maintained throughout the testing period. At the lowest concentrations, 3.13 and 6.25 mg/L, no test item was found and the recovery rates for test concentrations 12.5 and 25.0 mg/L ranged from 7.8 to 81.3% of the nominal. In the two highest dose levels, 50 and 100 mg/L, the measured concentrations were close 80% (79.8% and 81.3%, respectively) of the nominal concentrations. For static tests, where the concentrations do not remain within 80 – 120% of nominal, the effects concentration should be expressed relative to the geometric mean of the measured concentration at the start and end of the test (CLP guidance and OECD 23 guidance document). Considering the test concentrations were not entirely maintained during the test as a whole, RAC prefers to express the effect concentrations as the geometric mean of the measured concentrations. RAC recalculated the E_rC_{50}, which resulted in a 72-hour E_rC_{50} of 49.8 mg/L based on geometric mean measured concentrations. This value is roughly the same as that provided by the DS, E_rC_{50} 51.4 mg/L (nominal) and is in line with the above reported E_rC_{50} of 49.8 mg/L (geometric mean), and does not alter the classification proposal for aquatic acute toxicity.				

Date	Country	Organisation	Type of Organisation	Comment number
25.10.2019	United Kingdom		MemberState	4
Comment received				
The algal growth inhibition study dated 2005 was conducted according to OECD Test Guideline 201 (1984). We note that the updated Test Guideline 201 (2006 and 2011) includes additional validity criteria to assess study controls. It would be useful for the DS				

to consider if these criteria were met to demonstrate the reliability of the controls. This is relevant because the study forms the basis of the classification proposal.

The DS considers that a valid NOErC could not be determined because the NOErC at the nominal concentration of 6.25 mg/L was below the limit of detection of 0.503 mg/L. We note that a geometric mean measured concentration of 1.227 mg/L for this treatment is included in the CLP report (Table 26-p.33). While the reliability of this endpoint is unclear we note that it is >1mg/L indicating no chronic classification is required. We also note that the online registration (ECHA, 2019) includes an nominal ErC10 of 22 mg/L. Based on data in Tables 25 and 26 of the CLH report, it appears a reliable ErC10 could be derived which is likely to be >1 mg/L. On this basis, we think the DS should present a 72-hour ErC10 based on a geometric mean measured dose-response curve.

Given this last point, we consider it would be useful to also present the ErC50 based on geometric mean measured concentrations although we recognise the endpoint is likely to remain >1 mg/L and not impact the classification proposal.

ECHA (2019) https://echa.europa.eu/registration-dossier/-/registered-dossier/5898 accessed 2019-10-10.

Dossier Submitter's Response

Thank you for your comments.

The DS evaluated the additional validity criteria for the controls in the updated test guideline 201 (2006 and 2011) – "the *mean coefficient of variation for section by section specific growth rate in the control cultures must not exceed 35%*" and – "*the coefficient of variation of average specific growth rate during the whole test period in replicate control must not exceed 7%*. Those validity criteria were met in the study.

As reported, at the nominal concentration of 6.25mg/L there is a high uncertainty of the analytical results (see table 25-p.32). Indeed an ErC10 of 9.99 mg/L can be estimated based on mean measured concentrations (table 26-p.33) via linear regression, however due to the mentioned uncertainty of the analytical results at this concentration range, the DS is of the opinion that this value is not reliable.

The nominal ErC50 of 51.4 mg/L is considered valid since the recovery rates at this concentration level are within the ±20% of the nominal at the start and the end of the test. When using the geometric mean of the measured concentrations (table 26-p.33) the ErC50 would be 49.93mg/L (estimated via linear regression, $R^2 = 0.98$). This value is almost equal to the ErC50 based on nominal concentrations and does not alter the classification proposal.

RAC's response

RAC agrees with the commenting MS that the validity criteria for the controls should be confirmed. Further, RAC would also like to point out that where a measured concentration at the end of the exposure period is absent or where a substance is not detected, the validity of the test should be reconfirmed (REACH Guidance). The DS states that the validity criteria were met in the study, albeit they did not provide data to verify this. As a result, RAC decided to check the validity of the algal test. The average number of cells per mL increased from 1100 to 597900 cells/mL, which is a factor of 54.4. This value exceeds the validity criterion for cell growth of the controls by a factor of at least 16 within three days. The mean coefficient of variation (CV) for section by section specific growth rate in the controls resulted in 17.7%. This value does not exceed the 35% limit. The CV of the average

specific growth rate during the whole test period in the replicate controls resulted in 1.4%. This value does not exceed the 7% limit. Consequently, RAC agrees with the DS that the validity criteria of the test were met.

Effect levels based on the nominal concentration, where analytical methods cannot quantify test concentrations, might result in an underestimation of the toxicity. Therefore, RAC recognizes the DS concerns with regard to the validity of using the NOEC value of 6.25 mg/L (nominal concentration) for classification purposes. According to the CLP guidance (I.4.1.a), where concentrations at the end of the test are below the analytical detection limit, such concentrations shall be considered to be half of that detection limit. In these cases, it is good practice to use half of the limit of detection to calculate a mean exposure concentration and final concentration. Taking this into account and the fact the study is valid, RAC considers that a calculated E_rC_{10} based on geometric mean measured concentrations, is adequate and valid for classification purposes. In this context, RAC disagrees with the DS that an E_rC_{10} value is not reliable.

RAC calculated the 72-hour E_rC_{10} value using geometric mean measured test concentrations and followed the current methodology described in OECD TG 23, and obtained a 72-hour E_rC_{10} of 29.0 mg/L. This value is above the E_rC_{10} values reported by the DS (above) and the ECHA dissemination website, 9.99 mg/L (mean measured concentration) and 22 mg/L (nominal concentration), respectively.

Subsequently, RAC considers that the 72-hour E_rC_{10} value of 29.0 mg/L is valid and can be used for classification purposes, and this will alter the long-term classification of the substance.
Aquatic chronic toxicity data on 2-NPT is available for one trophic level, algae. In absence of long-term toxicity data for fish and aquatic invertebrates, the surrogate method is applied as recommended in CLP regulation Annex I, 4.1.2.3. and Figure 4.4.1. The substance is considered not rapidly degradable and does not fulfil the criteria for bioaccumulating potential.

- Classification based on adequate chronic toxicity data. Algal testing resulted in a 72-hour E_rC_{10} of 29.0 mg/L. The E_rC_{10} is above 1 mg/L and the substance is not rapidly degradable. 2-NPT does not fulfil the criteria for chronic classification, based on Table 4.1.0 (b)(i).
- Classification based on surrogate data for other trophic levels. Two acute limit tests for fish and aquatic invertebrates resulted in $L(E)C_{50}$ values > 100 mg/L and the substance is not rapidly degradable. 2-NPT does not fulfil the criteria for chronic classification, based on Table 4.1.0 (b)(iii).

Overall conclusion: based on the available information, 2-NPT does not warrant Chronic classification.

With regard to comment on E_rC_{50} based on geometric mean measured concentrations, see RAC response to comment number 3.

Date	Country	Organisation	Type of Organisation	Comment number
25.10.2019	Belgium		MemberState	5

Comment received

Aquatic toxicity to algae:
A statistical significant effect was observed for biomass and growth rate at a nominal conc of 6.25 mg/L. However analytical results of the lowest test concentration (6.25 and 3.13 mg/L) after 72h were below the detection limit (0.503 mg/L). Following the CLP guidance I.4.1 where concentrations at the end of the test are below the analytical detection limit, such concentrations shall be considered to be half that detection limit, thus NOEC= 0.252 mg/L.

Based on the above and the fact that the substance is not rapidly degradable, a classification with Aquatic Chronic 2, H411 seems more appropriate.

Dossier Submitter's Response

Thank you for your comments.

As reported at the two lowest test concentrations of 3.13mg/L and 6.25mg/L, there is a high uncertainty of the analytical results (see table 25-p.32). Statistically significant effects compared to the control occurred at the concentration of 12.5mg/L for biomass and growth rate. Due to the fact that the NOEC cannot be exactly calculated and the high uncertainty in the two lowest test concentrations, the DS is of the opinion to use the reliable ErC50 and thus apply the proposed classification. Moreover, using LOD/2 would only refer to the 72h value as a valid measurement for 0h is available and the geomean would be created between these two values. Therefore the NOEC would not be simply LOD/2 as indicated by you. The geomean using LOD/2 of the working calibration function as 72h value would result in a NOEC of 1.227 (as stated in Table 26-p.33 in parantheses). But again this value is not regarded reliable in contrast to the ErC50 which can be clearly based on measured or nominal concentrations (please see also our response to comments 3 and 4 from SE and UK, respectively).

RAC's response

RAC agrees with the DS that the LOD/2 value of 0.252, as suggested by the MS cannot be used to derive the NOEC. As mentioned in comment 3, the LOD/2 can be used to calculate a mean exposure concentration where concentrations at the end of the test (e.g., a 72-hour exposure period) are below the analytical limit. In this case, 0.252 mg/L L from the working calibration function (LOD/2 = 0.503/2 = 0.252 mg/L) refers to the concentration after 72-hour exposure for a test concentration of 6.25 mg/L. The resulting geometric mean for exposure concentrations of 5.99 mg/L (0 hours) and 0.252 mg/L (72 hours) results in a value of 1.227 mg/L. This value is the geometric mean measured concentration for test concentration of 6.25 mg/L and not the NOEC.

See also RAC's response to comment 3 with regard to the E_rC_{50} and comment 4 with regard to the reliability of NOEC/E_rC10.

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Date	Country	Organisation	Type of Organisation	Comment number
25.10.2019	France		MemberState	6

Comment received

P29:
In table 26 in column "Results", there are typographical errors. For the fish acute toxicity test, LC50 of 100 mg/L is mentioned whereas a NOEC is set at >= 100 mg/L in the test description (p31). For the acute immobilisation test, the summary table describes an EC50 of 100 mg/L whereas a NOEC is also set at >= 100 mg/L in the test description (p31).

P32:
Regarding the static algae growth inhibition test, it is indicated that it was not possible to determine an exact NOEC. All the same, a NOEC might have been set at 12.5 mg/L.

Based on available data, we agree with the proposed classification Aquatic Chronic 3, H412.

Dossier Submitter's Response

Thank you for your comments.

Indeed there are typographical errors in table 26-p.29. In both tests, the fish acute toxicity and the acute immobilisation test, a limit test was performed with 100mg/L test item. Since no effects occurred in both tests the NOEC was set at >=100mg/L. Therefore in table 26-p.29 the mentioned LC50 of 100mg/L and EC50 of 100mg/L respectively are wrong and have to be corrected to NOEC of >=100mg/L.

At the nominal concentration of 12.5mg/L, there was a statistically significant inhibition of growth compared to the control. Even if the inhibiton was "only" 4.9% (table 26-p.33) it was a factor 10 higher than the inhibition observed in the next lower nominal concentration (6.25 mg/L). Therefore, this effect is not only statistically significant but also regarded ecotoxicologically relevant. Thus, the nominal concentration of 12.5mg/L was defined as LOEC.

RAC's response

It is not clear why the DS considers that the results of the fish acute toxicity and the acute immobilisation test should be corrected to reflect a NOEC endpoint instead of LC_{50} and EC_{50} values, respectively. Both tests methods are acute and 96h/48h-L(E)C_{50} values are reported. Therefore, RAC does not agree with the DS that the NOEC should be used. Taking into account that water solubility of 2-NPT is > 1 g/L and no effects occurred in both tests the LC_{50} and EC_{50} values should be set at > 100 mg/L.

See RAC's response to commnet 4 with regard to the reliability of NOEC/E_rC10.