EUROPEAN CHEMICALS AGENCY

Helsinki, 21 February 2019

Addressee: [redacted]

Decision number: TPE-D-2114460735-44-01/F
Substance name: Benzoic acid, 2-hydroxy-, mono-C14-18-alkyl derivs., calcium salts (2:1)
EC number: 931-276-9
CAS number: NS
Registration number: [redacted]
Submission number: [redacted]
Submission date: 01/02/2018
Registered tonnage band: Over 1000

DECISION ON A TESTING PROPOSAL

Based on Article 40 of Regulation ((EC) No 1907/2006) (the REACH Regulation), ECHA examined your testing proposal(s) and decided as follows:

Your testing proposal for Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.; test method OECD TG 443) with the registered substance is rejected and you are requested to perform:

Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.; test method OECD TG 443), in rats, by oral route, with the analogue substance Magnesium, bis(2-hydroxybenzoato-o1,o2)-, ar,ar'-di-c14-18-alkyl derivs. (List No 931-371-5) specified as follows:

- **Ten weeks premating exposure duration for the parental (P0) generation;**
- **Dose level setting shall aim to induce systemic toxicity at the highest dose level;**
- **Cohort 1A (Reproductive toxicity);**
- **Cohort 1B (Reproductive toxicity) with extension to mate the Cohort 1B animals to produce the F2 generation.**

You have to submit the requested information in an updated registration dossier by **1 March 2021**. You also have to update the chemical safety report, where relevant.

The reasons for this decision are set out in Appendix 1. The procedural history is described in Appendix 2 and advice and further observations are provided in Appendix 3.
The results of the extended one-generation reproductive toxicity study on the analogue substance (Magnesium, bis(2-hydroxybenzoato-O1,O2)-, ar,ar'-di-C14-18alkyl derivs., List No 931-371-5), as requested above, shall be used for read-across to the registered substance, as explained in the separate decision for this analogue, with communication number TPE-D-2114460734-46-01/F.

Appeal

This decision can be appealed to the Board of Appeal of ECHA within three months of its notification. An appeal, together with the grounds thereof, has to be submitted to ECHA in writing. An appeal has suspensive effect and is subject to a fee. Further details are described under: http://echa.europa.eu/regulations/appeals.

Authorised[1] by Ofelia Bercaru, Head of Unit, Hazard Assessment C4

[1] As this is an electronic document, it is not physically signed. This communication has been approved according to ECHA's internal decision-approval process.

Appendix 1: Reasons

The decision of ECHA is based on the examination of the testing proposals you submitted and scientific information submitted by third parties.

Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.)

a) Examination of the testing proposal

Pursuant to Article 40(3)(d) and (c) of the REACH Regulation, ECHA may reject a proposed test and require the Registrant to carry out other tests in cases of non-compliance of the testing proposal with Annexes IX, X or XI.

The basic test design of an extended one-generation reproductive toxicity study (Cohorts 1A and 1B, without extension of Cohort 1B to include a F2 generation, and without Cohorts 2A, 2B and 3) is a standard information requirement as laid down in column 1 of 8.7.3., Annex X of the REACH Regulation, whereas column 2 defines when the study design needs to be expanded.

The information on this endpoint is not available for the registered substance but needs to be present in the technical dossier to meet the information requirements. Consequently there is an information gap and it is necessary to provide information for this endpoint.

You have submitted a testing proposal for an extended one-generation reproductive toxicity study according to OECD TG 443 with the registered substance and ECHA requested your considerations for alternative methods for this information requirement. ECHA notes that you provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed.

However, when considering the case history, ECHA understands that you originally intended to use the test results of the analogue substance, Magnesium, bis(2-hydroxybenzoato-o1,o2)-, ar,ar'-di-c14-18-alkyl derivs. (List No 931-371-5) to read across to the registered substance (calcium salt).

Therefore, ECHA has analysed the information provided in the dossiers of the registered substance and of its analogue substance (magnesium salt) in light of the requirements of Annex XI, Section 1.5.. ECHA concludes that the conditions for read-across are met for these substances as the structures, compositions, and manufacturing processes are similar, the only difference lying with the non-toxic metal cation (Ca^{2+} vs. Mg^{2+}). Furthermore, the available data show that the physico-chemical and toxicological properties of the two substances are very similar. Therefore, ECHA accepts the plausibility of the read-across.

ECHA notes that the solubility of the analogue magnesium salt is higher (OECD TG 105: 3.63 mg/L at 20 °C) than that of the registered substance, the calcium salt (OECD TG 105: 1.91 mg/L at 20 °C), indicating that the analogue magnesium salt shows a better absorption and bioavailability. Therefore, with respect to this decision, testing with the analogue substance (magnesium salt) is requested.

You have specified the design of the proposed extended one-generation reproductive toxicity study as follows:

- Extension of Cohort 1B will most likely be included;
- Cohorts 2A and 2B will be included;
- Two-week premating exposure;
- Dose levels of 30, 100 and 300 mg/kg bw/day;

- Study will be perfomed in the same test house where the OECD TG 407, 421, 408 and TG 414 studies were conducted;
- Study will be perfomed similarly to the OECD TG 407, 421, 408 and TG 414 studies:
  - Rat species: Sprague-Dawley;
  - Exposure route: Oral (feed);
  - Vehicle: Mixture of mineral oil and corn oil.

ECHA considers that the proposed study design requires modification to fulfil the information requirement of Annex X, Section 8.7.3. of the REACH Regulation. You proposed that extension of Cohort 1B is conditional. However, the extension of Cohort 1B to produce the F2 generation is already triggered and needs to be conducted. The developmental neurotoxicity triggers are not met and therefore the Cohorts 2A/2B are not requested. Ten-week premating exposure duration is needed as the registered substance is lipophilic and absorption is expected to be slow.

Thus, an extended one-generation reproductive toxicity study according to columns 1 and 2 of 8.7.3., Annex X is required. The following refers to the specifications of this required study.

Premating exposure duration and dose-level setting

To ensure that the study design adequately addresses the fertility endpoint, the duration of the premating exposure period and the selection of the highest dose level are key aspects to be considered. According to the ECHA Guidance[2], the starting point for deciding on the length of premating exposure period should be ten weeks to cover the full spermatogenesis and folliculogenesis before the mating, allowing meaningful assessment of the effects on fertility.
You proposed a 2-week premating period because "*it is most likely an F2 generation will be included, so the F1 generation would dose from weaning for a minimum of 10 weeks prior to mating which would cover a full cycle of spermatogenesis or folliculogenesis. This will satisfy any concerns arising from the theoretical high tonnage/exposure and properties of the substance which might in theory have suggested a need for longer exposure.*"

ECHA does not agree with your proposal because a 2-week premating exposure duration is not sufficiently long to cover the full spermatogenesis and folliculogenesis before the mating, allowing meaningful assessment of the effects on fertility. In this specific case, a ten-week exposure duration is supported by the lipophilicity of the registered substance ($\log K_{ow}$ = 5.32 at 40 °C) and its expected slow absorption from the gastrointestinal tract after oral administration to ensure that the steady state in parental animals has been reached before mating. Therefore, the requested premating exposure duration is 10 weeks.

You proposed a dose level setting of 30, 100 and 300 mg/kg bw/day based on the experiences gained in the conducted OECD TG 407 and 421 studies on the registered substance and OECD TG 408 and 414 studies on the analogue substance, Magnesium, bis(2-hydroxybenzoato-o1,o2)-, ar,ar'-di-c14-18-alkyl derivs. (List No 931-371-5). Furthermore, you point out that the OECD TG 443 study should be conducted similarly to these available studies, *i.e.* by oral route.

However, ECHA notes that the four studies (OECD TG 407, 421, 408 and 414) were all carried out by gavage dosing whereas you propose an OECD TG 443 feeding study. In this respect, ECHA emphasises that change of dosing from gavage to feed might have significant effects on toxicokinetic parameters (*e.g.* bioavailability, AUC, C_{max}) and, hence, on the

[2] ECHA *Guidance on information requirements and chemical safety assessment*, Chapter R.7a, Section R.7.6 (version 6.0, July 2017)

toxicological dynamics of the registered substance. Frequently, gavage dosing results in bolus effects not observable in feeding studies. Furthermore, palatability problems resulting from the analogue substance as well as the proposed vehicle (mixture of mineral oil and corn oil) must be considered when changing the administration route from gavage to feed. Therefore, the dose levels and formulation used in the already conducted gavage studies might not be directly applicable to the proposed OECD TG 443 feeding study.

Therefore, ECHA emphasises that the highest dose level shall aim to induce systemic toxicity, but not death or severe suffering of the animals, to allow comparison of reproductive toxicity and systemic toxicity. The dose level selection should be based upon the fertility effects with the other cohorts being tested at the same dose levels.

If there are no relevant data to be used for the dose-level setting, it is recommended that a range-finding study (or range finding studies) is performed and that its results are reported with the main study. This will support the justifications of the dose level selections and interpretation of the results.

Species and route selection

You proposed testing by the oral route in rats. According to the test method OECD TG 443, the rat is the preferred species. ECHA agrees with your proposal, since the substance to be tested is a solid, and taking into account the discussion above, regarding the choice between gavage versus feed administration.

Extension of Cohort 1B

If the column 2 conditions of 8.7.3., Annex X are met, Cohort 1B must be extended, which means that the F2 generation is produced by mating the Cohort 1B animals. This extension provides information also on the sexual function and fertility of the F1 animals.

You proposed to extend Cohort 1B "*if there are triggers seen in the first breeding and F1 generation which require further confirmation, i.e. equivocal findings.*"

However, ECHA concludes that the extension of Cohort 1B is already triggered by the available information because:

- The use of the registered substance is leading to significant exposure of consumers and professionals because the registered substance is used by consumer and professionals as lubricants and greases in vehicles or machinery which include filling and draining of containers and enclosed machinery (including engines);

- There are indications that the internal dose for the registered substance will reach a steady state in the test animals only after an extended exposure because its $\log K_{ow}$ is ≥ 4.5 ($\log K_{ow}$ = 5.32 at 40 °C) and you also stated that "*absorption is expected to be slow/inefficient via the gastrointestinal tract*" ; and

- There are indications for endocrine-disrupting modes of action because in the OECD TG 408 study conducted with the accepted read-across substance, statistically significant higher relative adrenal gland weights (56% and 35% in males and females, respectively) were noted in the high dose groups (300 mg/kg bw/day). Test substance-related microscopic findings (adrenal cortical cell hypertrophy) were also noted in the adrenal cortex of high dose females at necropsy.

Therefore, Cohort 1B must be extended to include mating of the animals and production of the F2 generation because (i) the uses of the registered substance is leading to significant

exposure of professionals and consumers, (ii) the internal dose for the registered substance will reach a steady state in the test animals only after an extended exposure and (iii) the OECD TG 408 study indicates one or more relevant modes of action related to endocrine disruption.

Cohorts 2A and 2B

The developmental neurotoxicity Cohorts 2A and 2B need to be conducted in case of a particular concern on (developmental) neurotoxicity as described in column 2 of 8.7.3., Annex X. When there are triggers for developmental neurotoxicity, both the Cohorts 2A and 2B are to be conducted as they provide complementary information.

You proposed to include Cohorts 2A and 2B based on the following reasoning: "*In terms of other parameters/concerns, it is quite likely based on all the data generated so far that the thyroid effects seen may be due to hepatic enzyme induction and hence increased Thyroid hormone (TH) degradation. This is most probably adaptive and not toxicologically significant.*" Furthermore, "*ECHA require investigation of these parameters to confirm this, and so inclusion of a DNT cohort will allow determination of whether there is any effect on pup neurodevelopment, regardless of the process by which that may occur. Correlation of dose levels and responses between this study and preceding work identifying these effects will allow better understanding of the processes which may be occurring, and their significance.*"

ECHA only requires investigation of developmental neurotoxicity in case of a particular concern as decribed in ECHA Guidance[2]. ECHA notes that in the OECD TG 408 study provided, an increase in relative (but not absolute) thyroid/parathyroid weight of 16% was observed in high dose females only in the presence of general toxicity (increased liver weight, significant increase of serum liver enzymes of up to 579%, and test substance-related microscopic findings in the liver) without reaching statistical significance.

ECHA concludes that the developmental neurotoxicity Cohorts 2A and 2B need not to be conducted because there is no particular concern on (developmental) neurotoxicity based on the available information.

b) Consideration of the information received during third party consultation

ECHA received third party information concerning the testing proposal during the third party consultation.

The third party considered that conducting the study with the analogue Magnesium, bis(2-hydroxybenzoato-o1,o2)-, ar,ar'-di-c14-18-alkyl derivs. (List No 931-371-5) might be acceptable. As outlined above, ECHA agrees that the read-across is plausible and requests testing with this analogue magnesium salt.

Furthermore, regarding the study design, the third party stated that the basic study design (Cohorts 1A and 1B without extension) "*is considered to be appropriate in the absence of any triggers or conditions necessitating the inclusion of additional cohorts or a further generation*". However, the third party did not provide any scientific data which would support this statement and subsequently fulfil this information requirement.

c) Outcome

Therefore, pursuant to Article 40(3)(d)(c) of the REACH Regulation, you are requested to carry out the study with the analogue substance analogue substance Magnesium, bis(2-

hydroxybenzoato-o1,o2)-, ar,ar'-di-c14-18-alkyl derivs. (List No 931-371-5), as specified above.

Notes for your consideration

No triggers for the inclusion of Cohorts 2A and 2B (developmental neurotoxicity) and Cohort 3 (developmental immunotoxicity) were identified. However, you may expand the study by including Cohorts 2A and 2B and/or Cohort 3 if new information becomes available after this decision is issued to justify such an inclusion. Inclusion is justified if the available information, together with the new information, shows triggers which are described in column 2 of Section 8.7.3., Annex X and further elaborated in ECHA Guidance[2]. You may also expand the study to address a concern identified during the conduct of the extended one-generation reproduction toxicity study and also due to other scientific reasons in order to avoid a conduct of a new study. The justification for the expansion must be documented.

Appendix 2: Procedural history

ECHA received your registration containing the testing proposals for examination in accordance with Article 40(1) on 6 February 2018.

ECHA held a third party consultation for the testing proposals from 23 April 2018 until 7 June 2018. ECHA received information from third parties (see Appendix 1).

This decision does not take into account any updates after **12 September 2018**, 30 calendar days after the end of the commenting period.

The decision making followed the procedure of Articles 50 and 51 of the REACH Regulation, as described below:

ECHA notified you of the draft decision and invited you to provide comments.

ECHA did not receive any comments by the end of the commenting period.

ECHA notified the draft decision to the competent authorities of the Member States for proposals for amendment.

As no amendments were proposed, ECHA took the decision according to Article 51(3) of the REACH Regulation.

Appendix 3: Further information, observations and technical guidance

1. This decision does not imply that the information provided in your registration dossier is in compliance with the REACH requirements. The decision does not prevent ECHA from initiating a compliance check on the registration at a later stage.

2. Failure to comply with the requests in this decision, or to otherwise fulfil the information requirements with a valid and documented adaptation, will result in a notification to the enforcement authorities of the Member States.

3. In relation to the information required by the present decision, the sample of the substance used for the new tests must be suitable for use by all the joint registrants. Hence, the sample should have a composition that is suitable to fulfil the information requirement for the range of substance compositions manufactured or imported by the joint registrants.

   It is the responsibility of all joint registrants who manufacture or import the same substance to agree on the appropriate composition of the test material and to document the necessary information on their substance composition. In addition, it is important to ensure that the particular sample of the substance tested in the new tests is appropriate to assess the properties of the registered substance, taking into account any variation in the composition of the technical grade of the substance as actually manufactured or imported by each registrant.

   If the registration of the substance by any registrant covers different grades, the sample used for the new tests must be suitable to assess these grades. Finally there must be adequate information on substance identity for the sample tested and the grades registered to enable the relevance of the tests to be assessed.

4. If the required tests are conducted with an analogue substance in the context of a read-across approach, the identity of the test material used to perform the test should be specified in line with the ECHA's Practical Guide on "How to use alternatives to animal testing to fulfil your information requirements" (chapter 4.4). This is required to show that the test material is representative of the analogue substance identified in the read-across approach and used to predict the properties of the registered substance.