EUROPEAN CHEMICALS AGENCY

Helsinki, 21 March 2018

Addressee: [redacted]

Decision number: TPE-D-2114394439-33-01/F
Substance name: tert-dodecanethiol
EC number: 246-619-1
CAS number: 25103-58-6
Registration number: [redacted]
Submission number: [redacted]
Submission date: 19/04/2017
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 is accepted and you are requested to carry out:

1. **Pre-natal developmental toxicity study (Annex X, Section 8.7.2.; test method: EU B.31./OECD TG 414) in a second species (rabbit), oral route using the registered substance.**

Your testing proposal is modified and you are requested to carry out:

2. **Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.; test method: EU B.56./OECD TG 443) in rats, oral route with the registered substance, specified as follows:**
   - **Ten weeks premating exposure duration for the parental (P0) generation;**
   - **Dose level setting shall aim to induce some toxicity at the highest dose level;**
   - **Cohort 1A (Reproductive toxicity);**
   - **Cohort 1B (Reproductive toxicity) without extension to mate the Cohort 1B animals to produce the F2 generation; and**
   - **Cohort 3 (Developmental immunotoxicity).**

You may adapt the testing requested above according to the specific rules outlined in Annexes VI to X and/or according to the general rules contained in Annex XI to the REACH Regulation.

To ensure compliance with the respective information requirement, any such adaptation will need to have a scientific justification, referring and conforming to the appropriate rules in the respective annex, and an adequate and reliable documentation.

You have to submit the requested information in an updated registration dossier by **28 July 2021**. You also have to update the chemical safety report, where relevant. The timeline has been set to allow for sequential testing.

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.

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 Claudio Carlon, Head of Unit, Evaluation E2

[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 submitted by you.

1. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.) in a second species

Pursuant to Article 40(3)(a) of the REACH Regulation, ECHA may require the Registrant to carry out the proposed test.

Pre-natal developmental toxicity studies on two species are part of the standard information requirements for substance registered for 1000 tonnes or more per year (Annex IX, Section 8.7.2., column 1, Annex X, Section 8.7.2., column 1, and sentence 2 of introductory paragraph 2 of Annex X of the REACH Regulation).

The dossier contains the following study reports for the developmental toxicity endpoint:

i. Key study ([redacted] 1983): Pre-natal developmental toxicity study in rats via the inhalation route (GLP compliant and reliability score of 2);
ii. Key study ([redacted] 1983): Pre-natal developmental toxicity study in mice, via the inhalation route (GLP compliant and reliability score of 2); and
iii. Supporting study ([redacted] 1983): Two-week range finding inhalation toxicity study in rats (GLP compliant and reliability score of 2).

Both studies i. and ii. follow the OECD TG 414. However they deviate from the test guideline as only two dose levels were tested.

ECHA notes in the rat study (i.) that the LOAEC for the maternal animals was established at the low dose (22.7 ppm) due to a decrease in the mean body weight gain, while for the foetuses, the NOAEC was determined to be the high dose (88.6 ppm) due to the absence of adverse toxic effects. ECHA considers that the rat study (i.) with its supporting study (iii.) is adequate to cover the information required on the first species in spite of some deficiences.

With reference to the mice study (ii.) the LOAEC for the maternal animals was determined to be the low dose (22.7 ppm) due to the increase in post-implantation loss, while for the foetuses the NOAEL was established at the high dose (88.6 ppm) due to the absence of adverse toxic effects. In the study report it was concluded that that the substance at "*an actual exposure level of 88.6 ppm or less did not produce a teratogenic effect in mice*".

ECHA considers that the results from the mouse study rise a concern on the adequacy of the mouse study, as well as potential pre-natal developmental toxicity. The number of dams with viable foetuses was reduced by 23-25% in exposure groups, but also 3 dams out of 22 in the control group (13.6%) did not produce viable foetuses. The dams with no viable foetuses (total resorptions) was reflected in the post-implantation loss parameter. This also lead to low number of live litters in the exposed groups (16 and 15 in the mid- and high-dose groups, respectively). Furthermore, there is no explanation or other clinical signs in dams to support the interpretation that increase in post-implantation loss is a sign of maternal toxicity.

In fact, the incidence of high post-implantation loss (caused by total resorptions) in the control group jeopardises the interpretation of the results of the study and may indicate some problems in the study set up.

In addition, an increase in post-implantation loss may also reflect pre-natal developmental toxicity. There was a statistically significant reduced mean foetal body weight in the low dose group, although not followed by a similar observation in the high-dose group, which may be due to a lower statistical power as there were less dams with viable foetuses in that group.

As explained above, there are uncertainties in the mice study that create difficulties to interpret the results obtained. ECHA considers that this study is not adequate to provide information on the second species. Hence, ECHA cannot conclude on the developmental toxicity endpoint. Consequently, further information on the second species is required to allow a comprehensive evaluation of pre-natal developmental toxicity.

You have submitted a testing proposal for a pre-natal developmental toxicity study in a second species rabbits according to EU B.31./OECD TG 414 by the oral route with the registered substance. For the reasons explained above, ECHA agrees that there is a need to further investigate pre-natal developmental toxicity.

ECHA requested your considerations for alternative methods to fulfil the information requirement for Reproductive toxicity (pre-natal developmental toxicity). 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. ECHA has taken these considerations into account.

ECHA considers that the proposed study performed with the registered substance is appropriate to fulfil the information requirement of Annex X, Section 8.7.2. of the REACH Regulation.

You proposed testing with the rabbit. The test in the first species was carried out with rats. As explained above, in the technical dossier there is only one adequate study that provides information on developmental toxicity, which is the study in rats. According to the test method EU B.31./OECD 414, the rat is the preferred rodent species and the rabbit the preferred non-rodent species. On the basis of this default consideration, ECHA considers testing should be performed with the rabbit as a second species.

You proposed testing by the oral route. ECHA agrees that the oral route is the most appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.6.2.3.2. Since the substance to be tested is a liquid, with a low vapour pressure and a very high boiling point (0.2 kPa / 237.85°C), ECHA concludes that testing should be performed by the oral route.

Outcome

Therefore, pursuant to Article 40(3)(a) of the REACH Regulation, you are thus requested to carry out the proposed study with the registered substance subject to the present decision: Pre-natal developmental toxicity study in a second species (rabbit, oral route (test method: EU B.31./OECD TG 414).

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

Pursuant to Article 40(3)(b) of the REACH Regulation, ECHA may require the Registrant to carry out the proposed test under modified conditions.

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. If the conditions described in column 2 of Annex X are met, the study design needs to be expanded to include the extension of Cohort 1B, Cohorts 2A/2B, and/or Cohort 3. Further detailed guidance on study design and triggers is provided in in ECHA *Guidance on information requirements and chemical safety assessment* ECHA Guidance on information requiChapter R.7a, Section R.7.6 (version 6.0, July 2017).

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.

ECHA requested your considerations for alternative methods to fulfil the information requirement for Reproductive toxicity (extended one-generation reproductive toxicity study). 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. ECHA has taken these considerations into account.

You have submitted a testing proposal for an extended one-generation reproductive toxicity study - basic test design (Cohorts 1A, and 1B without extension), according to EU B.56./OECD TG 443 by the oral route to be performed in rats with the registered substance, with the following justification: "*Based on the results of the OECD 413 study, there is no specific concern for neurotoxicity and immunotoxicity*". Consequently, you do "*not propose the inclusion of the developmental immunotoxicity (DIT) and developmental neurotoxicity (DNT) cohorts.*"

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. Specifically, ECHA is of the opinion that on the basis of the available information on the substance subject to this decision, Cohort 3 should be included in the study design. Further justification for this modification of the proposed study design is provided below.

Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement. 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 ECHA Guidance, 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 propose 4 weeks for the premating exposure duration. However ECHA notes that ten weeks premating exposure duration is required because there is no substance specific information in the dossier supporting shorter premating exposure duration as advised in the ECHA *Guidance on information requirements and chemical safety assessment* ECHA Guidance on information requiChapter R.7a, Section R.7.6 (version 6.0, July 2017). Ten weeks exposure duration is supported also by the lipophilicity of the substance (log Kow of 7.43) to ensure that the steady state in parental animals has been reached before mating.

The highest dose level shall aim to induce some toxicity to allow comparison of effect levels and effects of reproductive toxicity with those of 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 is no relevant data to be used for dose level setting, it is recommended that results from a range-finding study (or range finding studies) are reported with the main study. This will support the justifications of the dose level selections and interpretation of the results.

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 not to include an extension of Cohort 1B and provided justifications following the criteria described in column 2 of Section 8.7.3 of Annex X and detailed in ECHA *Guidance on information requirements and chemical safety assessment* ECHA Guidance on information requiChapter R.7a, Section R.7.6 (version 6.0, July 2017).

Therefore, ECHA agrees that the criteria to extend the Cohort 1B are not met and concludes that Cohort 1B must not be extended to include mating of the animals and production of the F2 generation.

The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

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 not to include Cohorts 2A and 2B and provided justifications following the criteria described in column 2 of Section 8.7.3 of Annex X and detailed in ECHA *Guidance on information requirements and chemical safety assessment* ECHA Guidance on information requiChapter R.7a, Section R.7.6 (version 6.0, July 2017).

Therefore, ECHA agrees that the criteria to include Cohorts 2A and 2B are not met and concludes that the developmental neurotoxicity Cohorts 2A and 2B need not to be conducted.

The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Cohort 3

The developmental immunotoxicity Cohort 3 needs to be conducted in case of a particular concern on (developmental) immunotoxicity as described in column 2 of 8.7.3., Annex X.

You proposed not to include Cohort 3 and provided the following justification: *"Based on the results of the OECD 413 study, there is no specific concern for...immunotoxicity".*

ECHA notes that existing information on the registered substance itself derived from the available *in vivo* study, that is the sub-chronic toxicity (90-day) inhalation study ([redacted] 2017) with the registered substance, shows evidence of toxicity in thymus and spleen. Specifically, a significant decrease in the thymus weight (-35%) in females, was reported for the high-dose group (100 ppm). ECHA notes that in the females of the high-dose group there was no exposure-related body weight changes. Hence, the 35% reduction in the thymus weight indicates concern for developmental immunotoxicity.

In your comments, you claimed that *"the study did show that high-dose animals experienced some exposure-related discomfort [exposure-related stress]"* including *"decreased growth and food intake".* However, ECHA notes that only the males of the high-dose group had a reduced growth throughout the exposure period. There was no exposure-related body weight changes observed in the females of the high-dose group. As regards food consumption, there was only an isolated statistically significant difference in females of the high-dose group wth a decreased food intake on days 0 to 7.

ECHA agrees that, in general, the thymus effects may indicate stress. However, it is not clear from the findings of this study whether it can be concluded that the effects seen in the thymus in females (-35% reduction) are due to *"exposure-related stress"*. As mentioned above, typical responses to stress, such as body weight or body weight gain, food consumption and circulating blood cells were unchanged in females of the high-dose gorup. Hence, the thymus effects seen in females, without any other signs of particular stress, suggest primary effects of the test article on the immune system. Consequently, in the absence of strong evidence of stress, additional investigations for developmental immunotoxicity are required. As a consequence, addition of Cohort 3 should be maintained as part of the EOGRTS design.

ECHA concludes that the developmental immunotoxicity Cohort 3 needs to be conducted because there is a particular concern on (developmental) immunotoxicity based on the results from the above-identified *in vivo* study on the registered substance itself.

The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Species and route selection

You proposed testing in rats. According to the test method EU B.56./ OECD TG 443, the rat is the preferred species. On the basis of this default consideration, ECHA considers that testing should be performed in rats.

You proposed testing by the oral route. ECHA agrees that the oral route is the most

appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.6.2.3.2. Since the substance to be tested is a liquid, ECHA concludes that testing should be performed by the oral route.

Outcome

Therefore, pursuant to Article 40(3)(b) of the REACH Regulation, you are requested to carry out the modified study with the registered substance subject to the present decision: Extended one-generation reproductive toxicity study (test method EU B.56./ OECD TG 443), in rats, oral route, according to the following study-design specifications:

- Ten weeks premating exposure duration for the parental (P0) generation;
- Dose level setting shall aim to induce some toxicity at the highest dose level;
- Cohort 1A (Reproductive toxicity);
- Cohort 1B (Reproductive toxicity) without extension to mate the Cohort 1B animals to produce the F2 generation; and
- Cohort 3 (Developmental immunotoxicity).

Notes for your consideration

The conditions to include the extension of Cohort 1B are currently not met. Furthermore, no triggers for the inclusion of Cohorts 2A and 2B (developmental neurotoxicity) were identified. However, you may expand the study by including the extension of Cohort 1B and Cohorts 2A and 2B if new information becomes available after this decision is issued to justify such an inclusion. Inclusion is justified if the new information shows triggers which are described in column 2 of Section 8.7.3., Annex X and further elaborated in ECHA *Guidance on information requirements and chemical safety assessment* ECHA Guidance on information requiChapter R.7a, Section R.7.6 (version 6.0, July 2017). 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. The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Deadline to submit the requested Information

In the registration dossier, in IUCLID Section 7.8.1 [Toxicity to reproduction] under the EOGRTS testing proposal you requested a 48 months' timeline to perform the pre-natal developmental toxicity study and the extended one-generation reproductive toxicity study. Following ECHA's request for further justification, you provided a proposition of a 40 months' time schedule, as evidence as to why the standard testing proposal draft decision deadline of 30 months would need to be extended to 40 months. Therefore, ECHA has granted the request and set the deadline to 40 months.

Appendix 2: Procedural history

ECHA received your registration containing the testing proposals for examination in accordance with Article 40(1) on 20 April 2017.

ECHA held a third party consultation for the testing proposals from 22 June 2017 until 7 August 2017. ECHA did not receive information from third parties.

This decision does not take into account any updates after **13 December 2017**, 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 took into account your comments and did not amend the request(s).

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.