EUROPEAN CHEMICALS AGENCY

Helsinki, 13 December 2018

Addressee: [redacted]

Decision number: CCH-D-2114453541-54-01/F
Substance name: Anisaldehyde
EC number: 204-602-6
CAS number: 123-11-5
Registration number: [redacted]
Submission number: [redacted]
Submission date: 03/05/2018
Registered tonnage band: 100-1000

DECISION ON A COMPLIANCE CHECK

Based on Article 41 of Regulation (EC) No 1907/2006 (the REACH Regulation), ECHA requests you to submit information on:

Extended one-generation reproductive toxicity study (Annex IX, Section 8.7.3.; test method OECD TG 443) in rats, oral route with the registered specified as follows:

- **At least two 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); and**
- **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 ***20 December 2021***. You shall also update the chemical safety report, where relevant.

The scope of this compliance check decision is limited to the standard information requirements of Annex IX, Section 8.7.3. to the REACH Regulation.

The reasons of 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 **Ofelia Bercaru**, Head of Unit, Evaluation E3

[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

Consideration on uses of the substance in relation to the tests requested in the decision

In your registration dossier you indicated that the substance has cosmetic uses.

ECHA notes that your substance, in addition to the use in cosmetic products, is also used in cleaning agents and polishes, air care products and as fragrance material by consumers. Furthermore, ECHA notes that the substance has professional end-use of washing and cleaning products and polishes and wax blends.

ECHA therefore concludes that your registration dossier reports other uses beyond cosmetic uses. Consequently you cannot exclude that there is potential consumer and worker exposure to the substance without demonstrating strictly controlled conditions, as you have reported the following PROCs: 1, 2, 4, 8a, 8b, 10, 11 and 13. ECHA's factsheet[2] on the interface between REACH and Cosmetics Regulations, developed jointly with the European Commission, provides that registrants of substances that use the substance also for non-cosmetic uses (i.e. mixed-use substances) are permitted to perform animal testing, as a last resort, for all endpoints requiring vertebrate testing.

The requested vertebrate tests are therefore justified for the purposes of assessing hazards for workers and consumers. Such testing would not trigger the testing and marketing bans under the Cosmetics Regulation as the testing is to be performed for the purposes of meeting the requirements of the REACH Regulation.[3]

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

In accordance with Articles 10(a) and 12(1) of the REACH Regulation, a technical dossier registered at 100 to 1000 tonnes per year must contain, as a minimum, the information specified in Annexes VII to IX to the REACH Regulation. The information to be generated for the dossier must fulfil the criteria in Article 13(4) of the same regulation.

The basic test design of an extended one-generation reproductive toxicity study (test method OECD TG 443 with 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 IX of the REACH Regulation, if the available repeated dose toxicity studies (e.g. 28-day or 90-day studies, OECD TGs 421 or 422 screening studies) indicate adverse effects on reproductive organs or tissues or reveal other concerns in relation with reproductive toxicity. Furthermore column 2 defines when the study design needs to be expanded. Further detailed guidance on study design and triggers is provided in the ECHA Guidance[4].

In your comments, you agreed to perform the test.

a) The information requirement

[2] https://echa.europa.eu/documents/10162/13628/reach_cosmetics_factsheet_en.pdf
[3] http://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:52013DC0135&from=EN
[4] ECHA *Guidance on information requirements and chemical safety assessment* Chapter R.7a, Section R.7.6 (version 6.0, July 2017)

In the Decision CCH-D-2114359254-48-01/F, ECHA concluded, after evaluating the relevant information in your registration dossier, that an extended one-generation reproductive toxicity study according Annex IX, Section 8.7.3. is required. Indeed, the decision indicated that the information provided for the registered substance in the technical dossier did not meet the information requirement. Consequently it is necessary to provide information for this endpoint.

In the same decision ECHA required that you provide a sub-chronic toxicity study (90-day, OECD TG 408) which shall be conducted before the extended one generation reproductive toxicity study. Furthermore the results from the 90-day study shall be used, among other relevant information, to decide on the study design of the extended one generation reproductive toxicity study. In accordance with that decision you have provided the results of a sub-chronic toxicity study.

b) The specifications for the required study

Based on the experimental results submitted for the sub-chronic toxicity study (90-day), ECHA has re-evaluated the design of the EOGRT study and concluded that a new decision needs to be taken following the procedure under Articles 50 and 51 addressing the design of that study. The reasoning for extension of Cohort 1B is given below.

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[4], the starting point for deciding on the length of the 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.

Ten weeks premating exposure duration is required if there is no substance specific information in the dossier supporting shorter premating exposure duration. In this specific case, animals of Cohort 1B are mated to produce the F2 generation and, thus, the premating exposure duration will be ten weeks for these Cohort 1B animals and the fertility parameters will be covered allowing an evaluation of the full spectrum of effects on fertility in these animals. Thus, shorter premating exposure duration for parental (P) animals may be considered. However, the premating period shall not be shorter than two weeks and must be sufficiently long to reach a steady-state in reproductive organs as advised in the ECHA Guidance[4]. The consideration should take into account whether the findings from P animals after a longer premating exposure duration would provide important information for interpretation of the findings in F1 animals, e.g. when considering the potential developmental origin of such findings as explained in ECHA Guidance[4].

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 is no relevant data to be used for 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.

In your comments, you disagree with ECHA's request that the highest dose level shall aim to induce systemic toxicity. Instead, you consider that toxicokinetic data should be used to justify the dose level selection.

ECHA notes that for REACH purposes, the study should be adequate for risk assessment as well as classification and labelling. To this end, the dose level selection should be based on toxicity. According to ECHA Guidance[4], "*The highest dose for an extended one-generation reproductive toxicity study should be selected with the aim to induce some toxicity (or to use the limit dose of 1000 mg/kg bw/day if humans are not exposed to higher dose levels), in order to allow a conclusion on whether effects on reproduction are considered to be secondary, non-specific consequence of other toxic effects seen [...]. Only in this way is it possible to assess if the substance is a reproductive toxicant and/or if the effects on reproduction are potentially associated with systemic toxicity and to what extent.*

The possibility to select the highest dose level, based on the toxicokinetic data as mentioned in EU B.56 (OECD TG 443) and in the OECD GD 151, may not allow comparison of adverse effects on fertility with systemic toxicity and, thus, does not support production of data for classification and labelling purposes, including categorisation. Regarding the highest dose level, it is important to ensure that toxicity in both female and male animals is considered to ensure that reproductive toxicity in either gender is not overlooked."

ECHA Guidance[4] highlights the use of dose range-finding studies and where information on toxicokinetics may be helpful: "*Dose level selection is assisted by the information from existing studies as well as from specific dose range-finding studies that may need to be conducted. Toxicokinetic information may provide reasons to adjust for example, the dosing route and regime. In addition, it should be considered that toxicity and toxicokinetics in pregnant animals may differ to that in non-pregnant animals. This may cause challenges in selecting the highest dose level for the study as at various phases of the study the sensitivity of the animals may differ.*"

Dosing information from previous studies (OECD TG 408 and 422) shows that the highest dose of 500 mg/kg bw/day did not cause excessive systemic toxicity: apart from slight local irritation (squamous cell hyperplasia of the stomach), the livers showed centrilobular hypertrophy ("Grade 1" in OECD TG 408 and "very slight-slight" in OECD TG 422). At this dose level, the OECD TG 408 study reported decreased urine pH values (no further details given in the dossier) as well as the changes in weight and histopathology of epididymides. The OECD TG 422 study also showed a significant decrease in epididymal weights as well as a significant decrease in number of pregnant dams (6 vs. 12 in control group) and respective fertility index (46.2% vs. 92.3%).

As the OECD TG 422 indicated adverse effects on fertility, the extended one-generation reproductive toxicity study should be designed to follow up on these findings. As explained in ECHA Guidance[4], a comparison between the severity of the effects on fertility/development and the severity of other toxicological findings must then be performed. Therefore, the highest dose level should be intended to replicate the findings above in order to provide adequate information on reproductive toxicity for the purpose of both classification (including categorisation within the Reproductive toxicity hazard class) and risk assessment.

A dose-range finding study may help in justifying dose level selection and the results from a range-finding study should be reported with the main study to support the justifications of the dose level selections and interpretation of the study results.

Extension of Cohort 1B

If the column 2 conditions of Section 8.7.3., Annex IX 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.

The use of the registered substance in the joint submission is leading to significant exposure of consumers and professionals because the registered substance is used by professionals as cleaning agents and polishes (PROCs 1, 2, 4, 8a, 8b, 10, 11 and 13) and consumers as cleaning agents and polishes, air care, cosmetics as well as fragrance material.

Furthermore, there are indications for endocrine-disrupting modes of action observed in the available OECD TG 408 and 422 studies: the sub-chronic toxicity study (OECD TG 408) showed significantly decreased weight of cauda epididymis (71% of control) and epididymides (83% of control) at the high dose (500 mg/kg bw/day). At this dose level there was only a slight decrease in male mean body weights (-5.3%). The organ weight changes also correlated with histopathological changes: ductal atrophy (grades 1-3), associated with oligospermia, were observed in all animals. The OECD TG 422 study, with a shorter exposure duration compared to OECD TG 408, had also shown a significant decrease in the epididymis weight (91% of control) at 500 mg/kg bw/day, although without histopathological changes.

In your comments, you disagree with the requirement to extend the Cohort 1B to produce F2 animals and consider that the underlying mode of action for the observed effects is currently not known. To support this statement, you refer to negative results in EPA's Endocrine Disruptor Screening Program for the 21 century (EDSP21) and an experimental study (Zondek *et al.* 1938)[5].

Although the precise underlying modes of action are uncertain, ECHA considers that indications of one or more mode of action related to endocrine disruption to justify the inclusion of Cohort 1B are involved, as effects in reproductive organs are observed. According to the ECHA Guidance[4], changes in reproductive organs (e.g. epididymides), observed in repeated dose studies, are an indication of endocrine (disrupting) mode of action.

The EPA screening program presents *in vitro* data as well as ToxCast/CERAPP QSAR model predictions for androgen, oestrogen and thyroid hormone receptors; all are negative for the registered substance. ECHA notes that negative *in vitro* data does not invalidate a concern based on *in vivo* experiments (OECD TG 408 and 422 studies conducted with the registered substance).

Zondek *et al.* (1938), with very limited documentation, did not demonstrate oestrogenic activity in rats or mice for the registered substance. ECHA notes that the experiments were not repeated dose studies but animals were exposed within 24 hours and hence this does not invalidate the abovementioned concern. ECHA further notes that even if the substance does not appear to show oestrogenic activity, it does not exclude other endocrine (disrupting) modes of action.

In conclusion, ECHA considers that your arguments do not invalidate the observed trigger(s) indicating one or more mode of action related to endocrine disruption.

[5] Zondek, B. and Bergmann, E. 1938. LXXXIV. Phenol methyl ethers as estrogenic agents. Biochemical Journal, 32(Part 1), 641-645.

Furthermore, you consider that the F1 generation is fully sufficient for risk assessment and classification purposes, with the F2 generation having limited added scientific and regulatory value. To support this statement, you refer to five publications[6].

ECHA notes that these publications do not inform on the properties of the registered substance, nor do they inform why and how the information from these publications should be used to address the information requirement for production of the second filial generation (extension of Cohort 1B) which is triggered because the regulatory scientific criteria are met for the registered substance.

Therefore, ECHA concludes that Cohort 1B must be extended to include mating of the animals and production of the F2 generation because the uses of the registered substance is leading to significant exposure of professionals and consumers and there are indications of modes of action related to endocrine disruption from the two available studies (OECD TG 408 and OECD TG 422) indicating endocrine-disruption modes of action for the registered substance.

Species and route selection

As described in the Decision CCH-D-2114359254-48-01/F, ECHA considers that testing should be performed in rats, by the oral route.

c) Outcome

Based on the available information, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Extended one-generation reproductive toxicity study (test method OECD TG 443), in rats, oral route, 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 relevant 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 IX and further elaborated in ECHA Guidance[4]. 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.

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[6] Piersma AH *et al.* Combined retrospective analysis of 498 rat multi-generation reproductive toxicity studies: on the impact of parameters related to F1 mating and F2 offspring. Reprod Toxicol. 2011 May;31(4):392-401.

Martin MT *et al.* Profiling the reproductive toxicity of chemicals from multigeneration studies in the toxicity reference

Janer G *et al.* A retrospective analysis of the twogeneration study: what is the added value of the second generation? Reprod Toxicol. 2007 Jul;24(1):97-102.

Beekhuijzen M *et al.* Implementing the extended one-generation reproductive toxicity study (EOGRTS): important points to consider. Crit. Rev. Toxicol. 2016; 46:4, 332-347.

Rorije E *et al.* On the impact of second generation mating and offspring in multi-generation reproductive toxicity studies on classification and labelling of substances in Europe. Regul Toxicol Pharmacol. 2011 Nov;61(2):251-60.

Deadline to submit the requested information in this decision

In the draft decision communicated to you the time indicated to provide the requested information was 24 months from the date of adoption of the decision. In your comments on the draft decision, you requested an extension of the timeline to 45 months, due to the development of microencapsulated formulation, palatability and dose-range studies, as well as limited capacity in testing laboratories. You also submitted a statement from the company's own test laboratory. ECHA acknowledges that an extension is justified because of the need of the above-mentioned additional experiments. Hence, ECHA has only partially granted the request and set the deadline to 36 months.

Appendix 2: Procedural history

For the purpose of the decision-making, this decision does not take into account any updates of your registration after the date when the draft decision was notified to you under Article 50(1) of the REACH Regulation.

On 26 April 2017 ECHA issued the Decision CCH-D-2114359254-48-01/F.

On 3 May 2018 you submitted an update of your registration dossier, including the results of a 90-day sub-chronic toxicity study.

On 5 June 2018 ECHA informed you that the request for an EOGRT study was withdrawn and would be addressed in a separate, the current, decision.

The compliance check of the information requirement for an extended one-generation reproductive toxicity study was initiated on 14 May 2018.

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 but amended the deadline.

ECHA notified the draft decision to the competent authorities of the Member States for proposal(s) 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 compliance check decision does not prevent ECHA from initiating further compliance checks on the present 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 your Member State.

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.