ECHA
EUROPEAN CHEMICALS AGENCY

Helsinki, 13 June 2016

Addressee [redacted]

Decision number: CCH-D-2114332899-33-01/F
Substance name: benzaldehyde
EC number: 202-860-4
CAS number: 100-52-7
Registration number: [redacted]
Submission number: [redacted]
Submission date: 03.09.2015

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

1. **In vitro cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2, test method: OECD TG 473) or in vitro micronucleus study (Annex VIII, Section 8.4.2, test method: OECD TG 487) with the registered substance;**

2. **In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3; test method: OECD TG 476 or OECD TG 490) with the registered substance; provided that the study requested under 1. has negative results;**

3. **Pre-natal developmental toxicity study (Annex IX, Section 8.7.2; test method: EU B.31/OECD TG 414) in a first species (rats or rabbits), oral route with the registered substance;**

4. **Pre-natal developmental toxicity study (Annex X, Section 8.7.2; test method: EU B.31/OECD TG 414) in a second species (rats if first species was rabbits or rabbits if first species was rats), oral route with the registered substance;**

5. **Extended one-generation reproductive toxicity study (Annex IX/X, Section 8.7.3; test method: 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); and**
   - **Cohort 1B (Reproductive toxicity) without extension to mate the Cohort 1B animals to produce the F2 generation;**

6. **Short-term toxicity testing on aquatic invertebrates (Annex VII, Section 9.1.1; test method: Daphnia sp. Acute immobilisation test, EU C.2/OECD TG 202) with the registered substance;**

7. **Growth inhibition study aquatic plants (Annex VII, Section 9.1.2; test method: Alga, growth inhibition test, EU C.3/OECD TG 201) with the registered substance;**

8. **Identification of PNEC and risk characterisation (Annex I, Section 3.3.1. and 6.): revise PNECs for freshwater, marine water, intermittent releases, freshwater sediment, marine sediment and soil using the assessment factors recommended by ECHA Guidance R.10 for PNEC derivation and revise the risk characterisation accordingly *or* provide a detailed justification for not using the recommendations of ECHA guidance in PNEC derivation.**

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 of the REACH Regulation. In order 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 are required to submit the requested information in an updated registration dossier by **20 June 2019**. You shall also update the chemical safety report, where relevant. The timeline has been set to allow for sequential testing.

The reasons of this decision are set out in Appendix 1. The procedural history is described in Appendix 2. 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, shall 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

0. Grouping of substances and read-across approach

In the registration, you have adapted the standard information requirements for

- Pre-natal developmental toxicity (Annex IX/X, Section 8.7.2) and
- Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3)

by applying a read-across adaptation following REACH Annex XI, Section 1.5. The read-across approach is described in the following section.

Article 13(1) of the REACH Regulation provides that information on intrinsic properties of substances may be generated by means other than tests. Such other means include the use of information from structurally related substances (grouping of substances and read-across), "provided that the conditions set out in Annex XI are met".

Annex XI, 1.5. requires a structural similarity among the substances within a group or category such that relevant properties of a substance within the group can be predicted from the data on reference substance(s) within the group by interpolation. The following analysis presents your justification for the proposed grouping approach and read-across hypothesis, together with ECHA's analysis concerning the justification in both a generic and a property-specific context.

0.1. Introduction of the grouping and read-across approach proposed by the Registrant

Your proposed read-across is based on the source substances benzoic acid and its salt sodium benzoate (both referred to as "benzoate" here below). To justify the read-across, you stated that no reproductive and developmental studies with the registered substance are available. Benzoate is a metabolite of the test substance formed by the enzyme aldehyde dehydrogenase, that is present in the liver, but also in the stomach. Based on this, you expected that the outcome of studies with the metabolite benzoate is representative for the toxicity of the registered substance and can be used in a read-across approach.

ECHA understands that the basis for prediction of the proposed read-across is the hypothesis that benzaldehyde is metabolised to the common substance benzoate and that this common substance is representative for the toxicological properties of the registered substance benzaldehyde; *i.e.* the parent substance benzaldehyde and possible other (bio)transformation products do not significantly influence the observed toxicity profile.

0.2. Information submitted by the Registrant to support the grouping and read-across approach

You have provided no further read-across justification than that outlined under 0.1. above.

0.3. ECHA analysis of the grouping and read-across approach in light of the requirements of Annex XI, Section 1.5.

With regard to the proposed predictions, ECHA has the following observations:

(i) The substance characterisation of the source studies needs to be sufficiently detailed in order to assess whether the attempted prediction is not compromised by the composition and/or impurities. In the ECHA practical guide "*How to report on Read-Across*" it is recommended to follow the Guidance on identification and naming of substances und REACH (version 1.3, February 2014) also for the source substances. This ensures that the identity of the source substance and its impurity profile allows an assessment of the suitability of the substances for read-across purposes.

ECHA notes that the source substances are identified by their chemical names and CAS numbers. However, the impurity profiles of the source substances cannot be assessed using the information provided in the registration dossier and, hence, ECHA cannot verify the suitability of the substances for read-across. As the structural similarity between the source substances and the target substance cannot be established, prediction of toxicological properties is not possible.

(ii) In order to meet the provisions in Annex XI, Section 1.5 to predict human health effects from data for a source substance, ECHA considers that structural similarity alone is not sufficient. It has to be justified why such prediction is possible in view of the identified structural differences and the provided evidence has to support such explanation. In particular, the structural similarities must be linked to a scientific explanation of how and why a prediction is possible.

You explained that benzoate "*is a metabolite of the test substance formed by the enzyme aldehyde dehydrogenase, that is present in the liver, but also in the stomach*" with references to [redacted] and [redacted]. Although the referenced articles have not been provided in the registration dossier, ECHA concludes that it is likely that benzoate is formed from benzaldehyde via a metabolic oxidation pathway. However, you have not provided any evidence that this *in vivo* oxidation is the only (bio)transformation pathway acting on benzaldehyde; *i.e.* you have not addressed the question whether other (bio)transformation products are formed which might influence the prediction. You have also not considered whether benzoate undergoes further (bio)transformation to other (bio)transformation products which might influence the prediction.

Furthermore, you have not shown that (bio)transformation of benzaldehyde to benzoate is sufficiently rapid and complete to exclude systemic bioavailability and internal exposure to benzaldehyde itself. When considering that bioavailability of benzaldehyde is likely, it should be explained why systemic exposure to benzaldehyde would not significantly influence the toxicological properties under consideration. However, you have not included any such explanation. ECHA concludes that you did not address important aspects such as the toxicokinetics of the parent substances and their metabolic fate and the resulting possible differences in their metabolite profiles. Therefore, it is not possible to verify that the source and target substances have the same, common mechanism of action which would allow predicting toxicological properties as a result of structural similarity in accordance with Annex XI, Section 1.5. In this respect, ECHA further notes that the structural differences between benzaldehyde and benzoate are significant:

Whereas benzaldehyde contains an aldehyde function, benzoate bears a carboxylate group instead. It is emphasised that an aldehyde function exerts a significantly different reactivity compared to a carboxylate group. An aldehyde exerts high reactivity towards nucleophiles; for example, the reaction of amino groups of peptides/proteins with aldehydes by nucleophilic addition to yield Schiff base. Such reactivity is not observed for carboxylate groups. You have not explained why these structural differences and their inherent different reactivity result in similar toxicological properties with respect to reproductive and pre-natal developmental toxicity. Therefore, the provided explanation provides no basis for predicting the properties of benzaldehyde from benzoate which does not rely upon conversion to benzoate.

You have provided study records for repeated dose toxicity studies which show significant differences with respect to toxicological effects (e.g. NOAELs based on kidney effects in comparison to NOAELs based on effects on body weight). Such differences may indeed stem from qualitative differences (i.e. exposure to different substances) and/or quantitative differences (e.g. exposure of target tissues to different concentrations of a substance). The observed differences may actually indicate that benzaldehyde is systemically available and exerts different effects than benzoate. Therefore, ECHA concludes that the presented evidence contradicts your hypothesis that target and source substance have the same properties, and on this basis also, it is not possible to predict the toxicological properties under consideration.

(iii) Annex XI, Section 1.1.2 (2) and Annex XI, Section 1.5 require for non-GLP studies and studies used for read-across purposes that "*adequate and reliable coverage of the key parameters foreseen to be investigated in the corresponding test method referred to in Article 13(3)*". Annex XI, Section 1.1.2 (2) and (3) and Annex XI, Section 1.5 require for non-GLP studies and studies used for read-across purposes "reliable coverage" and "reliable documentation".

With respect to the information requirement for extended one-generation reproductive toxicity (Annex X, Section 8.7.3.), ECHA notes that you have provided a non-GLP, non-guideline four-generation study with the source substance benzoic acid in rats in which only some reproduction parameters were assessed: "*Percentage of infertility, delayed sexual maturation, litter size, total pups, surviving pups.*" However, according to the OECD testing guideline, comprehensive examinations on functional fertility such as mating and gestation indices and male and female reproductive systems is required including, in particular, examination of the oestrous cycle; sperm parameters; organ weights of uterus, ovaries, testes, epididymides, prostate, seminal vesicles with coagulating glands and their fluids and prostate, thyroid, pituitary, adrenal glands; histopathology of vagina, uterus with cervix, ovaries, testis, epididymis, seminal vesicles, prostate, and coagulating gland. ECHA notes that these key parameters have not been examined. Furthermore, ECHA notes that the dose level setting, which is a key parameter, does not comply with the requirements of OECD TG 416 because only two doses were used (0.5% and 1%), no rationale for dose selection has been provided and no toxicity was observed at the highest dose.

With respect to the information requirement for pre-natal developmental toxicity according to Annex IX/X, Section 8.7.2., ECHA notes that you have provided four non-GLP, pre-natal developmental toxicity studies with the source substance sodium benzoate in mice, rats, hamsters and rabbits which are designated as equivalent or similar to OECD TG 414. ECHA notes that the exposure duration deviates from that prescribed in the current test guideline because it was from day 6 through day 15 of gestation in rats and mice, from day 6 through day 10 of gestation in hamsters, and from day 6 to day 18 of gestation in rabbits. OECD TG 414 states that the study "*is not intended to examine solely the period of organogenesis, (e.g. days 5-15 in the rodent, and days 6-18 in the rabbit) but also effects from preimplantation [...] through the entire period of gestation to the day before caesarean section.*" Hence, the effects from preimplantation through the entire period of gestation, which is a key parameter foreseen to be investigated, is not covered. According to ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.0, July 2015), R.7.6.4.2.2 "*If a study is conducted according to an old test method and thus uses a shorter administration period than current test methods, it is important that there is no indication challenging the exposure period used. Thus, if there is a concern suggesting that a longer exposure period would have revealed developmental toxicity or more profound findings affecting also lower dose levels that were not observed using shorter exposure duration, this should be addressed*". However, ECHA notes that you have not explained whether a longer administration period is not deemed necessary for the registered substance. Furthermore, according to OECD TG 414, "*unless limited by the physical/chemical nature or biological properties of the test substance, the highest dose should be chosen with the aim to induce some developmental and/or maternal toxicity (clinical signs or a decrease in body weight) but not death or severe suffering.*" However, you have not reported any rationale for dose selection and according to the study records no effects have been observed for any species at the highest dose administered. In this respect ECHA notes that the highest dose tested was 175 mg/kg bw/day in rats and mice, 300 mg/kg bw/day in hamsters, and 250 mg/kg bw/day in rabbits only, and that a limit test according to OECD TG 414, paragraph 16, was not performed. Therefore, ECHA concludes that the dose level setting was inadequate to investigate pre-natal developmental toxicity for the registered substance. Moreover, due to limited reporting in all four study records, ECHA cannot assess which examinations have been actually performed on maternal animals and offspring. It is also noted that you have stated that the performed studies deviate in "*foetal examinations*" but you did not specify in detail which examinations were and were not conducted.

ECHA concludes that the source studies do not provide the information required by Annex IX/X, Section 8.7.2. (pre-natal developmental toxicity) and Annex X, Section 8.7.3. (extended one-generation reproductive toxicity), because they do not meet the requirements of Annex XI, Section 1.1.2 nor Annex XI, Section 1.5.

0.4. Conclusion

The adaptation of the standard information requirements for pre-natal developmental toxicity (Annex IX/X, Section 8.7.2.) and extended one-generation reproductive toxicity (Annex X, Section 8.7.3.) in the technical dossier is based on the proposed read-across approach examined above. ECHA does not consider the read-across justification to be a reliable basis to predict the properties of the registered substance for the reasons set out above. Thus, the adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. Therefore, ECHA rejects the read-across adaptations in the technical dossier for pre-natal developmental toxicity and extended one-generation reproductive toxicity.

1. In vitro cytogenicity study in mammalian cells or in vitro micronucleus study (Annex VIII, Section 8.4.2.)

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.

An "*In vitro* cytogenicity study in mammalian cells or an *in vitro* micronucleus study" is a standard information requirement as laid down in Annex VIII, Section 8.4.2. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

In the technical dossier you have provided two study records for in vitro mammalian chromosome aberration tests. The test from 1986 is designated as key study, whereas the test from 1982 is designated as supporting study. However, neither the key study nor the supporting study provide the information required by Annex VIII, Section 8.4.2., because they were not performed according OECD TG 473. In the 1986 study only 100 cells and in the 1982 study only 162 cells were scored. Furthermore, for both tests no data on mitotic index nor rationale for top concentration were provided, and no tabulated data is presented. The scoring of only 100 and 162 cells represents a significant deviation from the testing guideline which prescribes scoring of "at least 300 cells". Such deviation in the number of scored cells significantly impacts the statistical analysis which renders the results of the tests questionable. Moreover, ECHA cannot conclude whether the study design with respect to concentration selection was appropriate as no data on mitotic index nor rationale for top concentration was provided. ECHA also notes that the 1982 study was performed in the presence of metabolic activation only and it does not provide any information in absence of metabolic activation. In view of the significant deviations as described here above, ECHA concludes that the tests themselves and their results cannot be accepted to fulfil the standard information requirement according to Annex VIII, 8.4.2.

In addition to the two in vitro chromosome aberration tests, you have also provided two study records for sister chromatid exchange assays in mammalian cells, one study record for a single cell gel/comet assay in mammalian cells for detection of DNA damage and one study record for a DNA damage and repair assay, unscheduled DNA synthesis in mammalian cells in vitro. Furthermore, you have also provided three study records in the IUCLID section "*Genetic toxicity in vivo*", namely for a Drosophila SLRL test, a somatic mutation assay in Drosophila and an Ames test of metabolites of benzaldehyde extracted from rat urine. However, none of these in vitro and in vivo assays provides the information required by Annex VIII, Section 8.4.2., because these assays neither identify agents that cause structural chromosome aberrations nor detect micronuclei in the cytoplasm of interphase cells.

As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.

Therefore, 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: *In vitro* cytogenicity study in mammalian cells (test method: OECD TG 473) or *in vitro* mammalian cell micronucleus study (test method: OECD TG 487).

2. In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.)

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.

An "*In vitro* gene mutation study in mammalian cells" is an information requirement as laid down in Annex VIII, Section 8.4.3. of the REACH Regulation, "if a negative result in Annex VII, Section 8.4.1. and Annex VIII, Section 8.4.2." is obtained. ECHA notes that the registration dossier contains negative results for one of these information requirements; i.e. in vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.). However, ECHA notes that the registration dossier does not contain appropriate study records for the information requirement in vitro cytogenicity study in mammalian cells or in vitro micronucleus study according to Annex VIII, Section 8.4.2. (see 1. above). Therefore, adequate information *on in vitro* gene mutation in mammalian cells needs to be present in the technical dossier for the registered substance to meet this information requirement provided that the study requested under 1. has negative results.

In the technical dossier you have provided a study record for a non-guideline mammalian cell gene mutation assay, designated as key study. However, this study does not provide the information required by Annex VIII, Section 8.4.3., because no data on the relative total growth (RTG) nor rationale for top concentration has been provided. Therefore, ECHA cannot conclude whether the study design with respect to concentration selection was appropriate. ECHA also notes that the study was performed in the absence of metabolic activation only and it does not provide any information in the presence of metabolic activation. Furthermore, no tabulated data is presented and, therefore, it is not possible to verify that the increase in the mutant frequency is higher than the threshold 126 global evaluation factor (GEF value); i.e. your overall conclusion "positive at slightly toxic doses" cannot be verified. Based on the information provided, ECHA concludes that this study deviates significantly from OECD TG 476. In view of the significant deviations as described here above, ECHA concludes that the test itself and its results cannot be accepted to fulfil the information requirement of Annex VIII, Section 8.4.3.

In addition to the key study above, you have provided a study record for a non-GLP mammalian cell gene mutation assay, designated as supporting study. You classified this study as reliability 4 (not assignable). ECHA concludes that also this study does not provide the information required by Annex VIII, Section 8.4.2. because no tabular results, no reporting on cytogenicity, no rational for selection of concentrations has been provided; furthermore, the study is unreliable and was performed without positive control.

You have also provided three study records in the IUCLID section "*Genetic toxicity in vivo*", namely for a Drosophila SLRL test, a somatic mutation assay in Drosophila and an Ames test of metabolites of benzaldehyde extracted from rat urine. However, all these in vivo assays do not provide the information required by Annex VIII, Section 8.4.3., because these assays do not detect gene mutations in mammalian cells.

As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.

Therefore, 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: *In vitro* mammalian cell gene mutation test (test method: OECD TG 476 or OECD TG 490) provided that the study requested under 1. has negative results.

3. Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.) in a first species

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.

A "pre-natal developmental toxicity study" for a first species is a standard information requirement as laid down in Annex IX, Section 8.7.2. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. However, as explained above in Appendix 1, section 0 of this decision, your adaptation of the information requirement cannot be accepted.

You have also provided two study records with reliability 4 for the publications by Watanuki and Sakaguchi K (1981) and Abramovici and Rachmuth-Roizman (1983). Abramovici and Rachmuth-Roizman (1983) investigated the embryotoxic effect of a single dose of the test substance on young chick embryos. In the endpoint study record of Watanuki and Sakaguchi K (1981), the reporting is very limited; e.g. the principal of the study used is not reported and seems limited to investigate effects on rat embryo fibroblasts. These two study records do not inform on induction of developmental toxicity including effects on growth, survival, external, skeletal and visceral malformations and variations due to exposure during the whole prenatal period and the potential relationship of effects to maternal toxicity. Therefore, these study records do not fulfil the standard information required by Annex IX, Section 8.7.2., because they are not reliable and do not cover key parameters and exposure duration of a pre-natal developmental toxicity study (see Annex XI, Section 1.1.2.).

Hence, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.
According to the test method EU B.31/OECD TG 414, the rat is the preferred rodent species and the rabbit the preferred non-rodent species. On the basis of this default assumption ECHA considers testing should be performed with rats or rabbits as a first species.

ECHA considers 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 4.0, July 2015) R.7a, chapter 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.

Therefore, 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: Pre-natal developmental toxicity study (test method: EU B.31./OECD TG 414) in a first species (rats or rabbits) by the oral route.

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

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.

Pre-natal developmental toxicity studies (test method EU B.31./OECD TG 414) on two species are part of the standard information requirements for a 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).

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. However, as explained above in Appendix 1, section 0 of this decision, your adaptation of the information requirement cannot be accepted.
You have also provided two study records with reliability 4 for the publications by Watanuki and Sakaguchi K (1981) and Abramovici and Rachmuth-Roizman (1983). Abramovici and Rachmuth-Roizman (1983) investigated the embryotoxic effect of a single dose of the test substance on young chick embryos. In the endpoint study record of Watanuki and Sakaguchi K (1981), the reporting is very limited; e.g. the principal of the study used is not reported and seems limited to investigate effects on rat embryo fibroblasts. These two study records do not inform on induction of developmental toxicity including effects on growth, survival, external, skeletal and visceral malformations and variations due to exposure during the whole prenatal period and the potential relationship of effects to maternal toxicity. Therefore, these study records do not fulfil the standard information required by Annex X, Section 8.7.2. , column 2, because they are not reliable and do not cover key parameters and exposure duration of a pre-natal developmental toxicity study (see Annex XI, Section 1.1.2.).

Hence, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.
According to the test method EU B.31/OECD TG 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 rabbits or rats as a second species, depending on the species tested in the first pre-natal developmental toxicity study (see requested study under 3. above).

ECHA considers 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 4.0, July 2015) R.7a, chapter 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.

Therefore, 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: Pre-natal developmental toxicity study (test method: EU B.31./OECD TG 414) in a second species (rats if first species was rabbits or rabbits if first species was rats) by the oral route.

Notes for your consideration

You are reminded that before performing a pre-natal developmental toxicity study in a second species you must consider the specific adaptation possibilities of Annex X, Section 8.7.2., column 2 and general adaptation possibilities of Annex XI. If the results of the test in the first species enable such adaptation, testing in the second species should be omitted and the registration dossier should be updated containing the corresponding adaptation statement.

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

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.

The basic test design of an extended one-generation reproductive toxicity study (test method B.56./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 X. 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. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

a) The information requirement

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. However, as explained above in Appendix 1, section 0 of this decision, your adaptation of the information requirement cannot be accepted.

In the technical dossier you have provided a study record for a publication of a non-GLP and non-guideline, 32-week one-generation study, in rats, oral (gavage), with reliability 4, using ten rats and one dose of 5 mg/kg bw/day only, with very limited reporting and concluding that *"treatment did not affect reproduction"* although *"fewer treated females became pregnant"*; however, significance of this finding is unknown (Sporn et al. 1967). This study record does not provide the information required by Annex X, Section 8.7.3., because it is not reliable, reporting is very limited, and the study does not cover key parameters of an extended one-generation reproductive toxicity study, as required by Annex XI, Section 1.1.2; e.g. in the provided study only 10 animals were exposed to 1 dose.

Hence, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint. Thus, an extended one-generation reproductive toxicity study according Annex X, Section 8.7.3. is required. The following refers to the specifications of this required study.

b) The specifications for the study design

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.

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* R.7a, chapter R.7.6 (version 4.0, July 2015).

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.

It is recommended that results from a range-finding study (or range finding studies) for the extended one-generation reproductive toxicity study 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

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

ECHA considers 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 4.0, July 2015) R.7a, chapter 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.

c) Outcome

Therefore, 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 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.

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) and Cohort 3 (developmental immunotoxicity) were identified. However, you may expand the study by including the extension of Cohort 1B, Cohorts 2A and 2B and Cohort 3 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* R.7a, chapter R.7.6 (version 4.0, July 2015). 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.

6. Short-term toxicity testing on aquatic invertebrates (Annex VII, Section 9.1.1.)

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.

"Short-term toxicity testing on aquatic invertebrates" is a standard information requirement as laid down in Annex VII, Section 9.1.1. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have provided a non-GLP study from a publication (Bringmann and Kuhn 1977), where no guideline was followed and the duration of the test was of 24 hours.
Even while pointing the shortcomings of the study ECHA is of the opinion that you cannot use this study as a key study. The robust study summary does not provide sufficient information to establish the reliability of the results used and this impacts also on the PNEC derivation. In addition, the OECD Test Guideline 202 is performed at least for 48 hours; thus, between 24 hours and 48 hours the toxicity effects could have increased and the EC50 increased extensively. In the absence of 48-hour coverage and reliable documentation, the study does not meet the requirement set forth under Annex XI, Sections 1.1.2 (3) and (4) of the REACH Regulation.

As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.

In view of the biodegradation properties of the substance and no bioaccumulation potential, a short term study, as requested here, seems to be the most appropriate instead of a long term test.

Therefore, 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: *Daphnia* sp. Acute immobilisation test, EU C.2./OECD TG 202).

7. Growth inhibition study aquatic plants (Annex VII, Section 9.1.2.)

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(e) and 13(4) of the REACH Regulation, a technical dossier registered at more than 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to X of the REACH Regulation.
"Growth inhibition study aquatic plants" is a standard information requirement as laid down in Annex VII, Section 9.1.2. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have provided several non-GLP studies from publications, all assigned as reliability 4 according to the Klimisch score, two of which have been combined in a weight of evidence approach.

The robust study summaries do not provide sufficient information to establish the reliability of the results used and this impacts also on the PNEC derivation. As a consequence, also the weight of evidence approach proposed based on Annex XI, Section 1.2 cannot be accepted.

As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.

Therefore, 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: Freshwater Alga and Cyanobacteria, Growth Inhibition Test, EU C.3./OECD TG 201).

8. Identification of PNEC and risk characterisation (Annex I, Section 3.3.1. and 6.): revise PNECs for freshwater, marine water, intermittent releases, freshwater sediment, marine sediment and soil using the assessment factors recommended by ECHA Guidance R.10 for PNEC derivation and revise the risk characterisation accordingly *or* provide a detailed justification for not using the recommendations of ECHA guidance in PNEC derivation

Pursuant to Articles 10(b) and 14(1) of the REACH Regulation, the registration shall contain a chemical safety report which shall document the chemical safety assessment conducted in accordance with Article 14(2) to (7) and with Annex I of the REACH Regulation.

Annex I, Section 3.3.1. of the REACH Regulation requires to establish a PNEC for each environmental sphere based on the available information and to use an appropriate assessment factor to the effect values.

The ECHA *Guidance on information requirements and chemical safety assessment,* Chapter R.10 provides further details and specifically provides default assessment factors that should be applied to derive PNECs.

Further, pursuant to Annex I, Section 3.3.2. if it is not possible to derive the PNEC, then this shall be clearly stated and fully justified.

You have used two aquatic toxicity results from studies which you consider long-term and an assessment factor of 50 for the calculation of PNEC aquatic for freshwater.

ECHA notes that according ECHA *Guidance on information requirements and chemical safety assessment,* Chapter R.10 (May 2008), an assessment factor of 50 can be applied when two long-term aquatic toxicity results (e.g. EC10 or NOEC) from freshwater or saltwater species representing two trophic levels (algae and/or crustaceans and/or fish) are available. In your registration dossier you have provided information on long-term toxicity to fish; however, ECHA does not consider this test as fulfilling the requirements for a long-term study according to OECD 210, due to the considerably shorter duration of the test (7 days). The second long-term study you provided is on long-term toxicity to algae, the issues of which are outlined under point 7 above. Even if the information for this endpoint would have been adequate, ECHA notes that in view of the information provided in the registration dossier, it would not be possible to determine that algae are the most sensitive species in short term studies. Hence, you have selected the incorrect assessment factor.

As explained above, the information provided on PNEC for the registered substance in the chemical safety report does not meet the general provisions for preparing a chemical safety report as described in Annex I, 3.3.1.

Consequently, you are given two options:

(i) You shall revise the PNECs derived for freshwater by applying the assessment factors recommended by the ECHA Guidance that are appropriate in this case. Furthermore, you are requested to revise other relevant PNECs according to the points considered above, specifically marine water, intermittent releases, freshwater sediment, marine sediment and soil. Subsequently, you shall re-assess related risks.

(ii) Alternatively, in accordance with Annex I, 3.3.1. and ECHA *Guidance on information requirements and chemical safety assessment,* Chapter R.10. (May 2008), you shall provide a full justification for the PNECs derived for freshwater, marine water, intermittent releases, freshwater sediment, marine sediment and soil provided in the chemical safety report by specifying how the following has been taken into account:

   a. Intra- and inter-laboratory variation of toxicity data;
   b. Intra- and inter-species variations (biological variance);
   c. Short-term to long-term toxicity extrapolation;
   d. Laboratory data to field impact extrapolation.

A justification for varying the assessment factor could include one or more of the following:

- evidence from structurally similar compounds which may demonstrate that a higher or lower factor may be appropriate.
- knowledge of the mode of action as some substances by virtue of their structure may be known to act in a non-specific manner. A lower factor may therefore be considered. Equally a known specific mode of action may lead to a higher factor.
- the availability of data from a variety of species covering the taxonomic groups of species across at least three trophic levels. In such a case the assessment factors may only be lowered if multiple data points are available for the most sensitive taxonomic group (i.e. the group showing acute toxicity more than 10 times lower than for the other groups).

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to revise PNECs for freshwater, marine water, intermittent releases, freshwater sediment, marine sediment, soil using the default assessment factors and other recommendations of ECHA Guidance R.10 for PNEC derivation and to revise the risk characterisation accordingly *or* provide a detailed justification for not using the recommendations of ECHA Guidance R.10 for PNEC derivation.

Notes for your consideration

The results of the studies requested with this decision shall be taken into account when revising the PNECs.

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.

The compliance check was initiated on 16 October 2015.

The decision making followed the procedure of Articles 50 and 51 of the REACH Regulation:

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.

ECHA received proposals for amendment and did not modify the draft decision.

ECHA invited you to comment on the proposed amendments.

ECHA referred the draft decision to the Member State Committee.

Your comments on the proposed amendments were taken into account by the Member State Committee.

In addition, you provided comments on the draft decision. These comments were not taken into account by the Member State Committee as they were considered to be outside of the scope of Article 51(5).

The Member State Committee reached a unanimous agreement on the draft decision during its MSC-47 meeting and ECHA took the decision according to Article 51(6) of the REACH Regulation.

Appendix 3: Further information, observations and technical guidance

1. The substance subject to the present decision is provisionally listed in the Community rolling action plan (CoRAP) for start of substance evaluation in 2018.

2. This compliance check decision does not prevent ECHA from initiating further compliance checks on the present registration at a later stage.

3. Failure to comply with the request(s) in this decision, or to fulfil otherwise the information requirement(s) with a valid and documented adaptation, will result in a notification to the enforcement authorities of your Member State.

4. In relation to the information required by the present decision, the sample of substance used for the new test(s) 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 composition 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 substance tested in the new test(s) 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 test(s) must be suitable to assess these grades. Finally there must be adequate information on substance identity for the sample tested and the grade(s) registered to enable the relevance of the test(s) to be assessed.