EUROPEAN CHEMICALS AGENCY

~~Confidential~~

Helsinki, 2 June 2021

Addressees
Registrants of JS_111-92-2_Dibutylamine as listed in the last Appendix of this decision

Date of submission of the dossier subject to this decision
04 April 2014

Registered substance subject to this decision ("the Substance")
Substance name: Dibutylamine
EC number: 203-921-8
CAS number: 111-92-2

Decision number: Please refer to the REACH-IT message which delivered this communication (in format CCH-D-XXXXXXXXXX-XX-XX/F)

DECISION ON A COMPLIANCE CHECK

Under Article 41 of Regulation (EC) No 1907/2006 (REACH), you must submit the information listed below, by the deadline of ***9 March 2023***.

Requested information must be generated using the Substance unless otherwise specified.

A. Information required from all the Registrants subject to Annex VII of REACH

1. In vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.; test method: EU B.13/14. /OECD TG 471) using one of the following strains: E. coli WP2 uvrA, or E. coli WP2 uvrA (pKM101), or S. typhimurium TA102

2. Justification for an adaptation of short-term toxicity testing on aquatic invertebrates (Annex VII, Section 9.1.1.) based on the results of the Long-term toxicity testing on aquatic invertebrates requested below (Annex IX, Section 9.1.5.)

3. Growth inhibition study aquatic plants (Annex VII, Section 9.1.2.; test method: EU C.3./OECD TG 201)

4. Ready biodegrability (Annex VII, Section 9.2.1.1.; test method: OECD TG 301C/D/F or OECD TG 310)

B. Information required from all the Registrants subject to Annex VIII of REACH

1. Justification for an adaptation of a Short-term repeated dose toxicity (28 days) based on the results of the Sub-chronic toxicity study (90 days) requested below (Annex VIII, Section 8.6.1.)

2. Justification for an adaptation of short-term toxicity testing on fish (Annex VIII, Section 9.1.3.) based on the results of the Long-term toxicity testing on fish requested below (Annex IX, Section 9.1.6.)

3. Adsorption/desorption screening (Annex VIII, Section 9.3.1.; test method: OECD TG 106)

C. Information required from all the Registrants subject to Annex IX of REACH

1. Sub-chronic toxicity study (90-day) (Annex IX, Section 8.6.2.; test method: OECD TG 408) by oral route, in rats, with a neutralised form of the Substance.

2. Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5.; test method: EU C.20./OECD TG 211)

3. Long-term toxicity testing on fish (Annex IX, Section 9.1.6.; test method: OECD TG 210)

D. Information required from all the Registrants subject to Annex X of REACH

1. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.; test method: OECD TG 414) by oral route, in a second species (rabbit) with a neutralised form of the Substance.

Reasons for the request(s) are explained in the following appendices:

- Appendix entitled "Reasons common to several requests";
- Appendices entitled "Reasons to request information required under Annexes VII to X of REACH", respectively.

Information required depends on your tonnage band

You must provide the information listed above for all REACH Annexes applicable to you, and in accordance with Articles 10(a) and 12(1) of REACH:

- the information specified in Annex VII to REACH, for registration at 1-10 tonnes per year (tpa), or as a transported isolated intermediate in quantity above 1000 tpa;
- the information specified in Annexes VII and VIII to REACH, for registration at 10-100 tpa;
- the information specified in Annexes VII, VIII and IX to REACH, for registration at 100-1000 tpa;
- the information specified in Annexes VII to X to REACH, for registration at more than 1000 tpa.

You are only required to share the costs of information that you must submit to fulfil your information requirements.

How to comply with your information requirements

To comply with your information requirements you must submit the information requested by this decision in an updated registration dossier by the deadline indicated above. You must also update the chemical safety report, where relevant, including any changes to classification and labelling, based on the newly generated information.

You must follow the general testing and reporting requirements provided under the Appendix entitled "Requirements to fulfil when conducting and reporting new tests for REACH purposes". In addition, you should follow the general recommendations provided under the Appendix entitled "General recommendations when conducting and reporting new tests for REACH purposes". For references used in this decision, please consult the Appendix entitled "List of references".

Appeal

This decision, when adopted under Article 51 of REACH, may be appealed to the Board of Appeal of ECHA within three months of its notification to you. Please refer to http://echa.europa.eu/regulations/appeals for further information.

Failure to comply

If you do not comply with the information required by this decision by the deadline indicated above, ECHA will notify the enforcement authorities of your Member State.

Authorised[1] under the authority of Christel Schilliger-Musset, Director of Hazard Assessment

[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 on Reasons common to several requests

1. Assessment of your read-across approach under Annex XI, Section 1.5.

You seek to adapt the following standard information requirements by applying a read-across approach in accordance with Annex XI, Section 1.5:

- Pre-natal developmental toxicity study in a second species (Annex X, Section 8.7.2.)
- Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5.)

ECHA has considered the scientific and regulatory validity of your read-across approach(es) in general before assessing the specific standard information requirements in the following appendices.

Annex XI, Section 1.5. specifies two conditions which must be fulfilled whenever a read-across approach is used. Firstly, there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category. Secondly, it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (addressed under 'Assessment of prediction(s)').

Additional information on what is necessary when justifying a read-across approach can be found in the ECHA Guidance R.6. and related documents[2,3].

A. Predictions for toxicological properties

You have not provided any read-across justification document in your registration dossier.

You read-across between the following:

- Dibutylamine hydrochloride, EC No. 228-521-0,
- Dimethylamine hydrochloride, EC No. 208-046-5, and
- Tributylamine, EC No. 203-058-7

as source substances and the Substance as target substance.

You have not provided any reasoning for the prediction of toxicological properties.

ECHA assumes that you predict the properties of the Substance using a read-across hypothesis which assumes that different compounds have the same type of effects. The properties of your Substance are predicted to be quantitatively equal to those of the source substance.

ECHA notes the following shortcomings with regards to predictions of toxicological properties.

A. Absence of read-across documentation

Annex XI, Section 1.5 requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must provide a justification for the read-across including a hypothesis, explanation of

[2] Read-Across Assessment Framework (RAAF). 2017 (March) ECHA, Helsinki. 60 pp. Available online: Read-Across Assessment Framework (https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across)
[3] Read-across assessment framework (RAAF) - considerations on multi-constituent substances and UVCBs. 2017 (March) ECHA, Helsinki. 40 pp. Available online: https://doi.org/10.2823/794394

the rationale for the prediction of properties and robust study summary(ies) of the source study(ies) (ECHA Guidance R.6.2.6.1).

You have provided studies conducted with other substances than your Substance in order to comply with the REACH information requirements. You have not provided documentation as to why this information is relevant for your Substance.

In the absence of such documentation, ECHA cannot verify that the properties of your Substance can be predicted from the data on the source substances. The only exception is Dibutylamine hydrochloride with EC No. 228-521-0 because it is a neutralised salt of the Substance and the read-across is thus plausible as such.

B. Adequacy and reliability of source study

We have identified deficiencies with the source studies provided on (some) of the selected analogue substances. These deficiencies are addressed under the corresponding Appendix D.1.

B. Predictions for ecotoxicological properties

You have provided a read-across justification document in IUCLID Section 13.

You read-across between the structurally similar substances, Diethylamine, EC No. 203-716-3 (CAS No. 109-89-7) as source substance and the Substance as target substance.

You have provided the following reasoning for the prediction of aquatic toxicity:

- The target and source substances are structurally similar and "*they only differ in the chain length: the target substance has two butyl chains, the source chemical two ethyl groups*";
- "[...] *the two substances are of high purity and that it is not likely that they contain any impurities which might have an influence on* [the prediction]";
- "[The] *target and source chemical are supposed to behave similarly in the environment*";
- "*A common mode of action* [for the target and source chemical] *can be hypothesized for ecotoxicity endpoints*";
- "[...] *the most critical physico-chemical properties for this assessment are comparable for these substances*";you consider that available evidence support that the target and source substance show similar acute toxicity to aquatic invertebrates and fish and similar toxicity on algae;

In addition, in your comments on the draft decision, you provide an updated read-across justification for the analogue approach with Diethylamine, EC No. 203-716-3 (CAS No. 109-89-7). You provided the following additional justification for this analogue approach:

- the target and sources substances have no impuritites relevant for classification or the PBT/vPvB assessment;
- according to the QSAR Toolbox v4.3.1, the calculated structure similarity between the two substances is 42.8%;
- you justify the expected common mode of action based on similar mechanistic similarity as illustrated by several mechanistic profilers from the QSAR Toolbox v4.3.1 and, the USEPA New Chemical Categories and the Aquatic Toxicity Classification by ECOSAR and the Acute Aquatic Toxicity Mode of Action (MOA) of OASIS;
- the target and souce substance share similaritires in metabolites as one metabolite of the target substance was identified to be identical to those of the source substance

using the CATABOL simulator of microbial metabolism and as these metabolites mostly belong to the classes aldehydes (mono) and aliphatic amines . You also state that "*The microbial metabolism estimated 15 metabolites for the target substance and 10 metabolites for the source substance*":

- you consider that the target and sources substances show similar fate and ecotoxicological properties based on experimental data on the target and source substance, where available, or EPI Suite v4.11 and other (Q)SAR models predictions.

ECHA understands that you predict the properties of the Substance using a read-across hypothesis which assumes that different compounds have the same type of effects. The properties of your Substance are predicted to be quantitatively equal to those of the source substance.

ECHA notes the following shortcomings with regards to prediction of long-term toxicity to aquatic invertebrates:

A. *Missing supporting information on the selected analogue substances*

Annex XI, Section 1.5 requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must provide a justification for the read-across including a hypothesis, explanation of the rationale for the prediction of properties and robust study summary(ies) of the source study(ies) (ECHA Guidance Section R.6.2.6.1.).

As part of your read-across justification, you refer to studies on short-term toxicity on fish and aquatic invertebrates and on toxicity to algae conducted with the source substance. However, you have not provided robust study summaries for these sources of information.

In the absence of this information, you have not demonstrated that this information is reliable and that it can be used to support that the properties of the Substance can be predicted from the data on the selected source substance.

In your comments on the draft decision, you explain that you have updated the IUCLID dossier for the Substance including the requested robust study summaries on short-term toxicity to fish and aquatic invertebrates as well as on toxicity to algae. However, you have not provided this information as part of your comments on the draft decision.

Please note that this decision does not take into account updates of the registration dossiers after the date on which you were notified of the draft decision according to Article 50(1) of REACH (see section 5.4. of ECHA's Practical Guide "How to act in Dossier Evaluation).

B. *Absence of reliable bridging information*

Annex XI, Section 1.5 of the REACH Regulation states that "*physicochemical properties, human health effects and environmental effects or environmental fate may be predicted from data for reference substance(s)*". For this purpose "*it is important to provide supporting information to strengthen the rationale for the read-across*" (ECHA Guidance R.6.2.2.1.f). The set of supporting information should allow to verify the crucial aspects of the read-across hypothesis and establish that the properties of the Substance can be predicted from the data on the source substance.

As indicated above, your read-across hypothesis is based on the assumption that the structurally similar substances cause the same type of effect(s). In this context, relevant, reliable and adequate information allowing to compare the properties of the Substance and of the source substance is necessary to confirm that both substances cause the same type of effects. Such information can be obtained, for example, from reliable bridging studies of comparable design and duration for the Substance and of the source substance.

As explained under issue A. above, you have not provided adequate documentation for the studies on short-term toxicity on fish and aquatic invertebrates and on toxicity to algae conducted with the source substance. Furthermore, as explained under Appendices A.2. to A.3., B.2., your registration dossier does not include any reliable source of information for these information requirements.

In the absence of reliable bridging studies to compare the properties of the Substance and of the selected source substance, you have not established that they are likely to have similar properties. Therefore you have not provided sufficient supporting information to strengthen the rationale for the read-across.

C. *Read-across hypothesis*

According to Annex XI, Section 1.5., two conditions shall be necessarily fulfilled. Firstly, there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category. Secondly, it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (read-across approach).
A read-across hypothesis needs to be provided, establishing why a prediction for a toxicological or ecotoxicological property is reliable. This hypothesis should be based on recognition of the structural similarities and differences between the source substance(s) and your Substance (ECHA Guidance R.6). It should explain why the differences in the chemical structures should not influence the toxicological/ ecotoxicological properties or should do so in a regular pattern.

Your read-across hypothesis is that the similarity in chemical structure and in some of the physicochemical and ecotoxicological properties between the source substance(s) and your Substance is a sufficient basis for predicting the properties of your Substance for other endpoints.

Similarity in chemical structure and similarity of some of the physicochemical and ecotoxicological properties does not necessarily lead to predictable or similar ecotoxicological properties in other endpoints. As described above, a well-founded hypothesis is needed to establish a reliable prediction for a ecotoxicological property, based on recognition of the structural similarities and differences between the source substance(s) and your Substance.

D. *Adequacy and reliability of the source study on long-term toxicity on aquatic invertebrates*

We have identified issues with the reliability of the source of information provided on the selected analogue substance. These deficiencies are addressed under the corresponding Appendix C.2.

C. Conclusions on the read-across approach

As explained above, you have not established that relevant properties of the Substance can be predicted from data on the analogue substance. Therefore, your adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. and your grouping and read-across approach is rejected except for Dibutylamine hydrochloride with EC No. 203-058-7 as explained above.

Appendix A: Reasons to request information required under Annex VII of REACH

1. In vitro gene mutation study in bacteria

An *in vitro* gene mutation study in bacteria is an information requirement under Annex VII to REACH (Section 8.4.1.).

You have provided the following information:

i. a non-guideline *in vitro* gene mutation study in bacteria ([redacted], 1986) on the Substance and with the strains TA 98, TA 100, TA 1535 and TA 1537, which all gave negative results.

We have assessed this information and identified the following issue:

To fulfil the information requirement, a study must comply with OECD TG 471 (1997) (Article 13(3) of REACH). Therefore, the following specifications must be met:

- the test must be performed with 5 strains: four strains of *S. typhimurium* (TA98; TA100; TA1535; TA1537 or TA97a or TA97) and one strain which is either *S. typhimurium* TA102 or *E. coli* WP2 uvrA or *E. coli* WP2 uvrA (pKM101).

The reported data for the study you have provided did not include results from 5 appropriate strains. The required fifth strain, *S. typhimurium* TA102 or *E. coli* WP2 uvrA or *E. coli* WP2 uvrA (pKM101) is missing.

The information provided does not cover one of the key investigations required by OECD TG 471.

On this basis, the information requirement is not fulfilled.

Study design

To fulfil the information requirement for the Substance, the *in vitro* gene mutation study in bacteria (OECD TG 471) should be performed using one of the following strains: *E. coli* WP2 uvrA, or *E. coli* WP2 uvrA (pKM101), or *S. typhimurium* TA102.

In your comments on the draft decision, you agreed to perform the requested study.

2. Justification for an adaptation of Short-term toxicity testing on aquatic invertebrates based on the results of the Long-term toxicity study on aquatic invertebrates

Short-term toxicity testing on aquatic invertebrates is an information requirement under Annex VII to REACH (Section 9.1.1.). This information may take the form of a study record or a valid adaptation in accordance with either a specific adaptation rule under Annex VII, Section 9.1.1., Column 2 or a general adaptation rule under Annex XI.

You have provided the following information:

i. a key study according to a ASTM/US EPA guideline on the Substance ([redacted], 1994);
ii. a supporting study according to EU Method C.2. on the Substance ([redacted] 1988)

We have assessed this information and identified the following issue:

To fulfil the information requirement, a study must comply with OECD TG 202 and the

requirements of OECD GD 23 (ENV/JM/MONO(2000)6/REV1) if the substance is difficult to test (Article 13(3) of REACH). Therefore, the following specifications must be met:

Technical specifications impacting the sensitivity/reliability of the test
- test animals are not fed during the test;
- the test medium fulfils, among others, the following condition: total organic carbon (TOC) ≤ 2 mg/L;

Characterisation of exposure
- a reliable analytical method for the quantification of the test material in the test solutions with reported specificity, recovery efficiency, precision, limits of determination (i.e. detection and quantification) and working range must be available. The sample preparation must also be described;
- the concentrations of the test material are measured at least at the highest and lowest test concentration, at the beginning and end of the test;

Reporting of the methodology and results
- the methods used to prepare stock and test solutions is reported;
- the number of immobilised daphnids is determined at 24 and 48 hours. Data are summarised in tabular form, showing for each treatment group and control, the number of daphnids used, and immobilisation at each observation;

Your registration dossier provides a key study (study i. above) showing the following:
- the test animals were fed during the test as your report *"Feeding during test: combination of a unicellular green algae (Selenastrum capricornutum YCT mixture)"*;
- you have not reported the TOC content of the test medium;
- you have not reported how samples were prepared prior to the analytical verification of exposure concentrations;
- you report that a solvent was used without any further information or justification;
- you have not reported the number of immobilised daphnids at 24 and 48 hours for each treatment group and control.

Based on the above,
- as explained further under Appendix A.3. below, the Substance is difficult to test and there are critical methodological deficiencies resulting in the rejection of the study results. More specifically, the presence of the feed may have reduced exposure to the Substance through adsorption leading to an underestimation of the intrinsic toxicity of the test material;
- the reporting of the study is not sufficient to conduct an independent assessment of its reliability. More specifically:
  - as you have not reported the TOC content of the test medium, it is not possible to verify if the test medium composition complies with the specifications of OECD TG 202 and OECD GD 23;
  - as you have not specified how samples were prepared prior to the analytical verification of exposure concentrations, it is not possible to assess whether the extraction procedure was adequate to determine truly dissolved concentrations;
  - it is not possible to verify if the solvent identity and concentration in the test solutions was adequate;
  - in the absence of adequate reporting of the data on immobilised daphnids, it is not possible to verify that the validity criteria of OECD TG 202 were met and that the interpretation of the results is adequate.

In your comments on the draft decision, you agreed that study i. above does not fully comply with the requirements of OECD TG 202. You specify that you will now only list this information as supporting study.

Your registration dossier also provides a supporting study (study ii. above) showing the following:

- you have not reported the TOC content of the test medium;
- you report that no analytical determination of exposure concentrations was conducted.

Based on the above,

- the reporting of the study is not sufficient to conduct an independent assessment of its reliability. More specifically, as you have not reported the TOC content of the test medium, it is not possible to verify if the test medium composition complies with the specifications of OECD TG 202 and OECD GD 23.

  In your comments on the draft decision, you acknowledge that this study does not provide information on the TOC content of the medium.

- there are critical methodological deficiencies resulting in the rejection of the study results. More specifically, as no analytical monitoring of exposure concentrations was conducted, you have not demonstrated that exposure was satisfactorily maintained over the duration of the test.

  In your comments on the draft decision, you acknowledge this deficiency.

In your comments on the draft decision, you consider that, despite the above deficiencies, this study is sufficiently reliable to be regarded as supporting information on the acute toxicity to aquatic invertebrates. However, you do not provide any justification that the conditions set out under section 1.1.2 of Annex XI ('*Data from experiments not carried out according to the test methods referred to in Article 13(3)*') are met.

Therefore, none of the provided studies meet the specifications of OECD TG 202 in conjunction with OECD GD 23.

On this basis, the information requirement is not fulfilled.

ECHA takes note that, in your comments on the draft decision, you specify that you now intend to adapt this information requirement under Annex XI, Section 1.5 using information from the analogue substance diethylamine (EC No. 203-716-3).

However, if you intend to do so, you must take account of the deficiencies identified on the proposed read-across approach for predicting ecotoxicological properties specified under the Appendix on Reasons common to several requests (and in particular issues B. and C.). In the absence of appropriate justification, your adaptation is rejected .

The present decision requests the registrant(s) concerned to conduct and submit a long-term toxicity study on aquatic invertebrates (OECD TG 211; see Appendix C.2. for details). According Annex VII, Section 9.1.1., Column 2 and to prevent unnecessary animal testing, a short-term toxicity study on aquatic invertebrates does not need to be provided.

Because you still must comply with the information requirement in Annex VII, Section 9.1.1., you are requested to submit a justification for the adaptation provided in Annex VII, Section 9.1.1., Column 2, second indent.

3. Growth inhibition study aquatic plants

Growth inhibition study aquatic plants is an information requirement under Annex VII to REACH (Section 9.1.2.).

You have provided the following information:

i. a key study according to DIN 38412, part 9 on the Substance ([redacted], 1988; Report no. [redacted]);
ii. a supporting study according to DIN 38412, part 9 on the Substance ([redacted], 1988; Report no. [redacted]);
iii. a supporting study according to DIN 38412, part 9 on the Substance ([redacted], 1988; Report no. [redacted]);
iv. a supporting study according to an unspecified method by US EPA 1971 on the Substance ([redacted] 1980);

We have assessed this information and identified the following issues:

A. To comply with this information requirement, the test material in a study must be representative for the Substance (Article 10 and Recital 19 of REACH; ECHA Guidance R.4.1).

For study i. to iii. above, you have identified the test material as "*N-butylbutan-1-amine / 111-92-2 / 203-921-8*" (*i.e.* the Substance) without further information, including composition, impurity profile and presence of impurities.

In the absence of composition information on the test material, the identity of the test material and its impurities cannot be assessed and you have not demonstrated that the test material is representative for the Substance. Therefore, the information provided is rejected.

B. To fulfil the information requirement, a study must comply with OECD TG 201 and the requirements of OECD GD 23 (ENV/JM/MONO(2000)6/REV1) if the substance is difficult to test (Article 13(3) of REACH). Therefore, the following specifications must be met:

Key parameter to be measured

- the concentrations of the test material leading to a 50 % and 0% (or 10%) inhibition of growth at the end of the test are estimated. Growth must be expressed as the logarithmic increase in biomass (average specific growth rate) during the exposure period;

Characterisation of exposure

- a reliable analytical method for the quantification of the test material in the test solutions with reported specificity, recovery efficiency, precision, limits of determination (i.e. detection and quantification) and working range must be available;
- the test media prepared specifically for analysis of exposure concentrations during the test is treated identically to those used for testing (*i.e.* inoculated with algae and incubated under identical conditions);
- the concentrations of the test material are measured at least at the beginning and end of the test:
  1) at the highest, and
  2) at the lowest test concentration, and

3) at a concentration around the expected EC_{50}.
For volatile, unstable or strongly adsorbing test substances, additional samplings for analysis at 24 hour intervals is required.

- if the concentration of the test material has not been maintained within 20 % of the nominal or measured initial concentration throughout the test, results must be based on the geometric mean of measured concentrations during exposure or on a model describing the decline of the concentration of the test material.

Reporting of the methodology and results

- the test design is reported (*e.g.*, number of replicates);
- the test conditions are reported (*e.g.*, composition of the test medium, biomass density at the beginning of the test);
- the results of algal biomass determined in each flask at least daily during the test period are reported in a tabular form;

Other considerations

- Algal biomass is determined based on dry weight per volume, or alternatively as cell counts or biovolume using microscopy or an electric particle counter. If an alternative method is used (*e.g.* flow cytometry, *in vitro* or *in vivo* fluorescence, or optical density), a satisfactory correlation with biomass must be demonstrated over the range of biomass occurring in the test.

Your registration dossier provides three study records by [redacted], 1988 (studies i. to iii. above) showing the following:

- no analytical monitoring of exposure concentrations was conducted
- tabulated data on the algal biomass determined daily for each treatment group and control(s) are not reported;
- biomass was measured using *in vivo* fluorescence. You state that "*Since no calibration curve data were available, fluorescence data were equated with cell numbers*". No justification is provided that *in vivo* fluorescence was adequate for the determination of biomass (*e.g.* evidence of correlation between the measured parameter and dry weight for both control and treated groups).

Based on the above,

- the reporting of the studies is not sufficient to conduct an independent assessment of their reliability. More specifically:
  - as you have not reported algal biomass determined daily for each treatment group and control(s), it is not possible to verify if validity criteria consistent with the specifications of OECD TG 201 were met;
  - as you have not provided any supporting information to demonstrate that *in vivo* fluorescence provides an adequate determination of algal biomass, it is not possible to verify that the study is reliable. The physiological status of algal cells is known to impact the efficiency of the non-photochemical quenching (NPQ) of fluorescence and differences in physiological status between treatments may bias the relationship between re-emitted fluorescence and biomass. You have not addressed this uncertainty;
- there are critical methodological deficiencies resulting in the rejection of the results of these studies. More specifically:
  - no analytical monitoring of exposure concentrations was conducted and therefore you have not demonstrated that exposure was satisfactorily maintained over the duration of the tests.

In your comments on the draft decision, you agreed that study i. to iii. above does not fully comply with the requirements of OECD TG 201. You specify that you will now only list this information as supporting study.

Your registration dossier also provides a study by [redacted]. 1980 (studies iv. above) showing the following:

- results were expressed based on biomass and not based on average specific growth rate;
- you report that an analytical monitoring of exposure concentration was conducted using GC-FID. You have not reported the performance parameters of the analytical method nor the results obtained;
- you have not reported key elements of the study design and procedure, including the number of replicates, the algal biomass at the beginning of the test and the composition of the test medium;
- tabulated data on the algal biomass determined daily for each treatment group and control are not reported.
- biomass was measured using *in vivo* fluorescence. No justification is provided that the method was adequate.

Based on the above,

- there are critical methodological deficiencies resulting in the rejection of this study. More specifically, as results were expressed based on biomass, the study does not provide an adequate coverage of the key parameter of OECD TG 201, *i.e.* inhibition of growth expressed as the logarithmic increase in biomass (average specific growth rate) during the exposure period;
- the reporting of this study is not sufficient to conduct an independent assessment of its reliability. More specifically:
    - as you have not provided adequate information on the analytical method and the results of the analytical determination of exposure concentrations, you have not demonstrated that exposure was satisfactorily maintained over the duration of the test;
    - as you have not reported the key information on the study design and procedure listed above, you have not demonstrated that the test was conducted under conditions that are consistent with the specifications of OECD TG 201;
    - as you have not provided tabulated data on the algal biomass determined during the test, it is not possible to verify if validity criteria consistent with the specifications of OECD TG 201 were met;
    - as you have provided, no information on the relationship between measured *in vivo* fluorescence and biomass, for the reasons already explained above, you have not demonstrated that *in vivo* fluorescence was adequate for the determination of biomass.

In your comments on the draft decision, you confirm that study iv. does not contain the above missing information. You specify that you will now assign a reliability score of 4 for this study.

Therefore, none of the provided studies meet the specifications of OECD TG 201 and OECD GD 23.

On this basis, the information requirement is not fulfilled.

In your comments on the draft decision, you specify that you now intend to adapt this information requirement under Annex XI, Section 1.5 using information from the analogue substance diethylamine (EC No. 203-716-3).

However, if you intend to do so, you must take account of the deficiencies identified on the proposed read-across approach for predicting ecotoxicological properties specified under the Appendix on Reasons common to several requests (and in particular issues B. and C.). In the absence of appropriate justification, your adaptation is rejected .

Study design

The Substance is difficult to test due to its volatility (Henry's Law constant of c.a. 1 Pa.m3/mol as measured in a HPLC-study using a thermodynamic method) and high adsorption potential (as the Substance is ionisable). OECD TG 201 specifies that, for difficult to test substances, you must consider the approach described in OECD GD 23 or other approaches, if more appropriate for your substance. In all cases, the approach selected must be justified and documented. Due to the properties of Substance, it may be difficult to achieve and maintain the desired exposure concentrations. Therefore, you must monitor the test concentration(s) of the Substance throughout the exposure duration and report the results. If it is not possible to demonstrate the stability of exposure concentrations (i.e. measured concentration(s) not within 80-120% of the nominal concentration(s)), you must express the effect concentration based on measured values as described in OECD TG 201. In case a dose-response relationship cannot be established (no observed effects), you must demonstrate that the approach used to prepare test solutions was adequate to maximise the concentration of the Substance in the test solution.

In your comments on the draft decision, you indicate that the available information on Henry's Law constant for the Substance correspond to the uncharged molecule. You point out that the pH-corrected Henry's Law constant method was withdrawn in more recent versions of the guidance and that no HLC can be derived for the charged molecule in the environmentally relevant pH range. However, you consider that the HLC for the uncharged molecule is rather low and will be even lower for the charged molecule, reducing the potential of the loss of the substance during the test via loss from the water phase to the air.

ECHA agrees that there is currently no valid estimate of the HLC of the Substance in your registration dossier and that it remains unclear to what extent loss by volatilisation may be lead to significant losses from the water phase in the course of aquatic ecotoxicity tests. However, the Substance is also ionised under environmentally relevant pH and therefore may have a high potential to bind to test vessels and/or dissolved organic matter and suspended solids. Therefore, based on the information currently available ECHA maintains that the Substance is to be regarded as difficult to test.

4. Ready biodegradability

Ready biodegradability is an information requirement in Annex VII to REACH (Section 9.2.1.1.).

You have provided the following information:

i. a key study according to OECD TG 301C on the Substance ([redacted], 1992);
ii. a non-guideline study on the Substance using three different types of bacterial inoculum ([redacted] 1980).

We have assessed this information and identified the following issues:

A. To comply with this information requirement, the test material in a study must be representative for the Substance (Article 10 and Recital 19 of REACH; ECHA Guidance R.4.1).

For study i. and ii. above, you have identified the test material as "*N-butylbutan-1-amine / 111-92-2 / 203-921-8*" (*i.e.* the Substance) without further information, including composition, impurity profile and presence of impurities.

In the absence of composition information on the test material, the identity of the test material and its impurities cannot be assessed and you have not demonstrated that the test material is representative for the Substance. Therefore, the information provided is rejected.

In your comments on the draft decision, you explain that you had access to the full study report for a study referred to as [redacted] (1991) and that you will include further information on the test material identity in the robust study summary for that study.

However, we note that it is unclear if this study corresponds to study i. listed above.

B. To fulfil the information requirement, a study must comply with the OECD TG 301 or 310 (Article 13(3) of REACH). Therefore, for a study according to OECD TG 301, the following requirements must be met:

Technical specifications impacting the sensitivity/reliability of the test
- determination is carried out at least in duplicate;
- the inoculum is not be pre-adapted to the test material;
- for an OECD TG 301C study, the concentration of the inoculum is set to reach a bacterial cell density of 10^7 to 10^8 cells/L in the test vessel. The suspended solid concentration is 30 mg/L;
- for an OECD TG 301C study, biodegradation is followed by monitoring oxygen uptake by the inoculum. The percentage primary biodegradation must also be calculated from supplemental specific chemical analysis made at the beginning and end of incubation. DOC analysis is only an optional additional parameter;

Reporting of the methodology and results
- the concentration of the inoculum in the test and any pre-conditioning treatment are reported;
- the results of measurements at each sampling point in each replicate is reported in a tabular form;
- for an OECD TG 301C study, the determination of the biodegradation using a specific chemical analytical method is reported;

Your registration dossier provides an OECD TG 301C study (study i. above) showing the following:
- you report that the number of culture flasks per concentration is 1;
- you report that the concentration of the inoculum was "*30 mg/L*". You have not reported the concentration of the inoculum in cells/L;
- you have not reported the percentage of primary biodegradation calculated from supplemental specific chemical analysis;

Based on the above,

- there are critical methodological deficiencies resulting in the rejection of the results of this study. More specifically:
  - the test was not conducted in at least duplicates;
  - primary degradation was not monitored using specific chemical analysis, which is a mandatory element of OECD TG 301C;
- the reporting of the study is not sufficient to conduct an independent assessment of its reliability. More specifically:
  - as you have not reported information on inoculum density in cells/L, you have not demonstrated that inoculum density was consistent with the specifications of OECD TG 301C;
  - as you have not provided adequate reporting of the study results, it is not possible to verify if validity criteria consistent with the specifications of OECD TG 301C were met.

In your comments on the draft decision, you explain that you had access to the full study report for a study referred to as [redacted] (1991) and that you will include further information on "*the analytical method (including performance parameters), on the study design and the procedure as well as tabulated data on the algal biomass*".

However, we note that the study you are referring to seems to correspond to a growth inhibition study in alage and that it is unclear to what extent this information is relevant to this information requirement.

Your registration dossier also provides a non-guideline study conducted with three types of bacterial inoculum (study ii. above) showing the following:
- one of the inoculum used was pre-adapted to the test material;
- you have not reported key element of the study design (e.g. number of replicates), study procedure (*e.g.* composition of the test medium, initial inoculum concentration) and study results (*e.g.* results of measurements at each sampling point in each replicate in the test and control bottles).

Based on the above,
- there are critical methodological deficiencies resulting in the rejection of some of the results of these studies. More specifically, the results obtained using a a pre-adapted inoculum is rejected as the use of a non-pre-adapted inoculum is a mandatory element of any ready biodegradability study;
- the reporting of these studies is not sufficient to conduct an independent assessment of their reliability. More specifically, in the absence of an adequate reporting of the elements of the study design, study procedure and study results listed above, it is not possible to conduct an independent assessment of the relevance and reliability of this information.

Therefore, none of the provided studies meet the specifications of OECD TG 301 or 310.

In your comment on the draft decision, you state that study ii. still supports the conclusion that the Substance is readily biodegradable as high biodegradation was observed. However, you have not addressed any of the above deficiencies.

In your comments on the draft decision, you also state that "*the conclusion on the ready biodegradability of the Substance are supported by two QSAR calculations: - CATALOGIC v5.14.5 BOD 28 days MITI (OECD 301C) v11.16 - CATALOGIC v5.14.1.5, CATALOGIC Kinetic*

301F v14.17". As an annex to your comments on the draft decision, you have provided a QSAR Prediction Reporting Format (QPRF) for each of these two models.

We have assessed this additonal information from your comments on the draft decision and identified the following issue:

> Under Section 1.3., first paragraph, third indent of Annex XI to REACH, a study may be omitted if QSAR results are adequate for the purpose of classification and labelling and/or risk assessment, including PBT assessment. ECHA Guidance R.7.9.5.1. specifies that (Q)SARs for predicting ready biodegradation are not yet sufficiently accurate to predict rapid degradation. However, when no useful information on degradability is available (either experimentally derived or estimated), (Q)SAR predictions can be used as supporting evidence of that the substance is not rapidly degradable.
>
> You have provided the following results from the CATALOGIC v5.14.5. software:
> - OECD 301C model: 70 ± 5% biodegradation based on theoretical BOD removal after 28 days;
> - OECD 301F model: 80 ± 8% biodegradation based on theoretical BOD removal after 28 days but failing the 10d-window criteria.
>
> As explained above, you registration dossier currently does not include adequate experimental or estimated information on rapid biodegradation for the Substance. In addition, as explained in ECHA Guidance R.7.9.5.1. , (Q)SAR predictions are, on their own, not adequate to conclude on rapid biodegradation. Finally, we note that the results (Q)SAR predictions included in your comments on the draft decision provide limited support to conclude that the Substance is readily biodegradable, because the OECD 301C does not inform on the 10d-window criteria and the 10d-window criteria was not met according to the results of the OECD 301F model. Therefore, you have not demonstrated that the Substance is to be regarded as readily biodegradable.

On this basis, the information requirement is not fulfilled.

Appendix B: Reasons to request information required under Annex VIII of REACH

1. Justification for an adaptation of a Short-term repeated dose toxicity (28 days) based on the results of the Sub-chronic toxicity study (90 days)

A Short-term repeated dose toxicity study (28 days) is an information requirement under Annex VIII to REACH (Section 8.6.1.). This information may take the form of a study record or a valid adaptation in accordance with either a specific adaptation rule under Column 2 of Annex VIII or a general adaptation rule under Annex XI.

You have provided the following information:

i. a key study similar to OECD 413 via inhalation in rats with the Substance ([redacted], 2003).

We have assessed this information and identified the following issue:

To be considered compliant and enable concluding whether the Substance has dangerous properties and supports the determination of the No-Observed Adverse Effect Level (NOAEL), a study has to meet the requirements of OECD TG 413. The following key parameter(s) of this test guideline include, among others:

- Ophthalmological examination, haematology, clinical biochemistry, and pathology of sexual (male and female) organs, full detailed gross necropsy and subsequent histopathology

In the study i. you have provided, the following key parameters are missing: ophthalmological examination, haematology and clinical biochemistry. In addition, full detailed gross necropsy and histopathology is missing as the provided study focused only on the respiratory system and reproductive organs. Therefore, this study does not meet the specifications of OECD TG 413.

In your comments on the draft decision, you agreed that the key parameters set out in the OECD test guideline were not assessed in the 90-day study (i).

On this basis, the information requirement is not fulfilled.

Annex VIII, Section 8.6.1., Column 2 provides that an experimental study for this endpoint is not needed if a reliable sub-chronic (90 days) or chronic toxicity study is available.

The present decision requests the registrants concerned to generate and submit a reliable sub-chronic toxicity study (90 days) (see Appendix C.1.). According to Annex VIII, Section 8.6.1., Column 2 and to prevent unnecessary animal testing, a short term toxicity study (28 days) does not therefore need to be conducted.

Because you still must comply with the information requirement in Annex VIII, Section 8.6.1., you are requested to submit a justification for the adaptation provided in Column 2 of that provision.

2. Justification for an adaptation of Short-term toxicity testing on fish based on the results of the Long-term toxicity study on fish

Short-term toxicity testing on fish is an information requirement under Annex VIII to REACH (Section 9.1.3.). This information may take the form of a study record or a valid adaptation in accordance with either a specific adaptation rule under Annex VIII, Section 9.1.3, Column

2 or a general adaptation rule under Annex XI.

You have provided the following information:

i. a key study according to a unspecified methodology from IRSA on the Substance ([redacted] 1980)

We have assessed this information and identified the following issue:

To fulfil the information requirement, a study must comply with OECD TG 203 and the requirements of OECD GD 23 (ENV/JM/MONO(2000)6/REV1) if the substance is difficult to test (Article 13(3) of REACH). Therefore, the following specifications must be met:

Technical specifications impacting the sensitivity/reliability of the test

- the test is conducted on juveniles of similar age (or size);
- the test medium fulfils the following condition(s): particulate matter ≤ 5 mg/L, total organic carbon (TOC) ≤ 2 mg/L or carbon oxygen demand (COD) ≤ 5 mg/L;

Characterisation of exposure

- a reliable analytical method for the quantification of the test material in the test solutions with reported specificity, recovery efficiency, precision, limits of determination (i.e. detection and quantification) and working range must be available;
- in static tests, if the concentrations of the test material are not expected to remain within ± 20 % of the nominal, then the test substance concentration is determined (in one replicate) in all concentrations at the beginning, at 48 hours and at the end of the test;
- the determinations of exposure concentrations reflect the concentrations of the dissolved chemical;

Reporting of the methodology and results

- the test design is reported (*e.g.* number of test animals per concentration);
- the test procedure is reported (*e.g.* composition of the test medium, fish loading);
- in static tests, the results of at least daily measurements of dissolved oxygen, pH, salinity (if relevant) and temperature measured daily in each test vessel are reported. The results of hardness and TOC determinations at the beginning of the exposure in the dilution water are reported;
- mortalities and sub-lethal effects (*e.g.* with regard to equilibrium, appearance, ventilator and swimming behaviour) are reported. The frequency of observations includes at least 2 observations within the first 24 hours and at least two observations per day from day 2 to 4.

Your registration dossier provides a study by [redacted] 1980 (study i. above) showing the following:

- no information is provided on the life-stage of the test animals;
- you report that an analytical monitoring of exposure concentration was conducted suing GC-FID. However, you have not reported the performance parameters of the analytical method nor the results obtained;
- you have not reported key elements of the study design and procedure, including the tested concentrations, number of test animals per concentration and the composition of the test medium;
- mortalities, sub-lethal effects, oxygen concentration, pH and temperature, in each treatment group and control are not reported.

Based on the above, the reporting of this study is not sufficient to conduct an independent assessment of its reliability. More specifically:

- as you have provided no information on life-stage of the test animals, you have not demonstrated that the test was conducted on juveniles as required by the OECD TG 203;
- as you have not provided adequate information on the analytical method and the results of the analytical determination of exposure concentrations, you have not demonstrated that exposure was satisfactorily maintained over the duration of the test;
- as you have not reported the key information on the study design and procedure listed above, you have not demonstrated that the test was conducted under conditions that are consistent with the specifications of OECD TG 203;
- as you have not reported the key information on the study results listed above, it is not possible to verify if validity criteria consistent with the specifications of OECD TG 203 were met.

Therefore, this study does not meet the specifications of OECD TG 203 in conjunction with OECD GD 23.

In your comments on the draft decision, you acknowledge that the information listed above is not available for study i. and that it does not fully comply with the requirements of OECD TG 202. You specify that you will now only list this information as supporting study.

On this basis, the information requirement is not fulfilled.

In your comments on the draft decision, you also specify that you now intend to adapt this information requirement under Annex XI, Section 1.5 using information from the analogue substance diethylamine (EC No. 203-716-3).

However, if you intend to do so, you must take account of the deficiencies identified on the proposed read-across approach for predicting ecotoxicological properties specified under the Appendix on Reasons common to several requests (and in particular issues B. and C.). In the absence of appropriate justification, your adaptation is rejected .

The present decision requests the registrant(s) concerned to conduct and submit a long-term toxicity study on fish (OECD TG 210; see Appendix C.3. for details). According Annex VIII, Section 9.1.3., Column 2 and to prevent unnecessary animal testing, a short-term toxicity study on fish does not need to be provided.

Because you still must comply with the information requirement in Annex VIII, Section 9.1.3., you are requested to submit a justification for the adaptation provided in Annex VIII, Section 9.1.3., Column 2, second indent.

3. Adsorption/ desorption screening

Adsorption/desorption screening is an information requirement under Annex VIII to REACH (Section 9.3.1.).

You have adapted this information requirement under Annex XI, Section 1.3 ('(Q)SAR'). In support of your adaptation, you provided the following information:

i. a log Koc value estimated using the first-order Molecular Connectivity Index (MCI) method from SRC KOCWIN (v2.00) for the uncharged molecule;
ii. a correction of the log Koc value using a method described in a publication by Franco

& Trapp (2008)

We have assessed this information and identified the following issues:

A. Annex XI, Section 1.3. states that (Q)SAR results must be adequate for the purpose of risk assessment, including PBT assessment. ECHA Guidance R.7.1.15.4 specifies that a measured adsorption coefficient is usually needed for ionising substances, since it is important to have information on pH-dependence. The guidance further clarifies that, if estimation methods are not appropriate (e.g. because the substance is a surfactant or ionisable at environmentally-relevant pH), then a batch equilibrium test is essential under Annex VIII.

   The log Koc value predicted using the MCI index (see i. above) does not provide information on pH-dependence of the adsorption potential of the Substance. Therefore, this predicted value is not adequate for the purpose of risk assessment, including PBT assessment.

   In your comments on the draft decision, you agree with ECHA's assessment and state that this information will be removed for your registration dossier.

B. Under ECHA Guidance R.6.1.3.4 a prediction is adequate for the purpose of classification and labelling and/or risk assessment when the model is applicable to the chemical of interest with the necessary level of reliability. ECHA Guidance R.6.1.5.3. specifies that, among others, the following cumulative conditions must be met:
   - the model predicts well substances that are similar to the substance of interest,
   - reliable input parameters are used.

   As an attachment to your comments on the draft decision, you have provided a QMRF and QPRF for the QSAR prediction ii. listed above, including references to literature. The R2 obtained on the training set (available as supplement data in the paper of Franco & Trapp, 2008) is 0.76, while R2 obtained on the validation set (i.e. the overall predictivity of the model) was determined to be 0.55. The model predicts a log Koc of 3.12 at pH 5-8 for the Substance (as retrieved from your registration dossier).

   Supporting information on the original publication describing the model is publicly available (Franco & Trapp, 2008). This information indicates that:
   - the training set includes only one secondary aliphatic amine (i.e. dimethylamine). It also includes one tertiary aliphatic amine (i.e. trimethylamine);
   - the validation set does not include any secondary aliphatic amine and includes a single primary aliphatic amine (i.e. n-butyl amine);
   - for these substance, the input parameter (i.e. measured log Koc) were retrieved from a publication by van Oepen, Körder and Klein, 1991 (Chemosphere, vol 22(3-4):285-304). In this publication, log Koc was determined using a modified OECD TG 106 on podzol, alfisol and sediment. For dimethylamine, trimethylamine and n-butyilamine, log Koc values ranging from 0.60 to 2.70, 0.78 to 2.83 and 0.70 to 2.03, respectively, are reported. For the same substances, input values of 2.04, 1.99 and 1.74 were used.

   However, the prediction for the Substance used as input is not reliable because:
   - the training and validation sets include only a very limited number of substances with some structural similarity with the Substance. In particular, the training set includes only methylated aliphatic amines and no aliphatic amines with C-chain length similar to the Substance. Also the validated set does not include any

secondary aliphatic amine;
- it is unclear how the experimental values on aliphatic amines were selected in the paper of Franco & Trapp (2008) and why they differ from the experimental values retrieved from the source publication by van Oepen, Körder and Klein (1991);
- the publication van Oepen, Körder and Klein (1991) indicates significant variation in log Koc depending on the nature of the soil/sediment matrix. This information can only be obtained from testing the substance in different matrices (as required by the OECD TG 106) and is not reflected by the model prediction.

Therefore, you have not demonstrated that the prediction for the Substance is adequate for the purpose of classification and labelling and/or risk assessment.

On this basis, the information requirement is not fulfilled.

Appendix C: Reasons to request information required under Annex IX of REACH

1. Sub-chronic toxicity study (90-day)

A Sub-chronic toxicity study (90 day) is a standard information requirement under Annex IX to REACH (Section 8.6.2.).

You have provided the following information:

i. a key study study similar to OECD 413 via inhalation in rats with the Substance ([redacted], 2003).

We have assessed this information and identified the following issue:

To be considered compliant and enable concluding whether the Substance has dangerous properties and supports the determination of the No-Observed Adverse Effect Level (NOAEL), a study has to meet the requirements of OECD TG 413. The following key parameter(s) of this test guideline include, among others:

- Ophthalmological examination, haematology, clinical biochemistry, and pathology of sexual (male and female) organs, full detailed gross necropsy and subsequent histopathology.

In the study i. you have provided, the following key parameters are missing: ophthalmological examination, haematology and clinical biochemistry. In addition, full detailed gross necropsy and histopathology is missing as the provided study focused only on the respiratory system and reproductive organs. Therefore, this study does not meet the specifications of OECD TG 413.

In your comments on the draft decision, you agreed that the key parameters set out in the OECD test guideline were not assessed in the 90-day study (i).

On this basis, the information requirement is not fulfilled.

Study design

Referring to the criteria provided in Annex IX, Section 8.6.2, Column 2, the oral route is the most appropriate route of administration to investigate repeated dose toxicity, because the Substance is a corrosive liquid and you apply self-classification as Skin Corr. 1A (H314). ECHA Guidance R.7.6.2.3.2 specifies that corrosive or highly irritating substances must be tested preferably via the oral route. However, testing at concentration/dose levels causing corrosivity must be avoided. Testing of neutral salts of alkaline or acidic substances is therefore more appropriate as it allows the investigation of intrinsic properties at adequate dose levels. These specifications are valid also for testing of repeated dose toxicity.

In your comments, you proposed to perform a 90-day repeated dose study via inhalation according to OECD TG 413 with the Substance due to the following reasons:

- *"According to REACH Annex IX, 8.6.2, column 2, a substance shall be tested via inhalation, if exposure is likely, e.g., because of a high vapour pressure."*
- *"ECHA Guidance R.7 more explicitly defines that "Testing by the inhalation route is the [..] preferred route for liquids of high to very high vapour pressure at ambient temperature (>25 kPa or boiling point below 50°C) for which inhalation is usually the predominant route of human exposure."(R.7.5.6.3.4). In the same section [...] it is stated that testing via inhalation shall be performed, "if there is some concern for local*

effects in the respiratory tract for which a qualitative assessment might not be sufficiently robust to demonstrate safe handling and use of the substance (A concern for local effects in the respiratory tract might be assumed inter alia for substances that are corrosive or irritating for the skin and/or eye)."

- *"Dibutylamine has a vapour pressur of 2.2hPa, is corrosive, and local effects are expected to be the leading health hazard for workers. Dibutylamine is only used in industrial and professional applications; no exposure of consumers will occur."*
- *"Consequently, the assessment of the local effects after repeated inhalation and derivation of an appropriate DNEL will be an essential part of the exposure assessment. Testing of the neutral salt is considered inappropriate, since it masks the most important intrinsic property with regard to risk assessment."*
- *"In the draft decision, ECHA guidance 7.6.2.3.2 is quoted. This section deals with reproductive toxicity and differs from the section applicable here for repeated dose toxicity, where likely exposure routes for workers and associated risks play a major role."*
- *"Regarding the second reference to REACH Annex IX, 8.6.2, column 2 in the draft decision: "Referring to the criteria provided in Annex IX, Section 8.6.2, Column 2, the oral route is the most appropriate route of administration to investigate repeated dose toxicity, because the Substance is a corrosive liquid and you apply self-classification as Skin Corr. 1A (H314).", it should be noted, that section 8.6.2 does not mention corrosivity nor any explicit criteria for oral testing."*

ECHA notes that the vapor pressure of the Substance (2.2 hPa equals to 0.22 kPa) cannot be regarded as high and it does not warrant default testing via inhalation route. The sub-chronic 90-d study (i) focusses mainly on the local effects in respiratory tract. In your registration dossier, you have already calculated DNEL value of 29 mg/m^3 for local effects (acute and long-term). This is based on local irritating effects. You provided the same information in the attached CSR in your comments to the draft decision. ECHA considers that the available data sufficiently covers the local effects in respiratory tract. Therefore, it is essential to investigate also the potential systemic effects. ECHA agrees with your comment that ECHA guidance 7.6.2.3.2 refers to reproductive toxicity only. However, introductory sections to Annexes VII – X of REACH require that *in vivo* testing with corrosive substances at concentration/dose levels causing corrosivity must be avoided. Therefore, ECHA considers that testing of a neutralised form of the Substance would enable to investigate its intrinsic properties and systemic effects also in a repeated dose toxicity study at adequate dose levels as the known hazard for local effects due to corrosivity would not be the key determinant in the selection of appropriate dose levels. Based on these considerations, ECHA considers that oral route is the most appropriate route of administration in this case although Section 8.6.2, column 2, at Annex IX does not mention corrosivity nor any explicit criteria for oral testing.

Therefore the sub-chronic toxicity study must be performed according to the OECD TG 408, in rats and with oral administration of a neutralised form of the Substance.

2. Long-term toxicity testing on aquatic invertebrates

Long-term toxicity testing on aquatic invertebrates is an information requirement under Annex IX to REACH (Section 9.1.5.).

You have adapted this information requirement under Annex XI, Section 1.5 ('Grouping of substances and read-across approach'). In support of your adaptation, you provided the following information:

i. a key study according to OECD TG 211 with the analogue substance Diethylamine, EC No. 203-716-3 ([redacted]., 1999).

We have assessed this information and identified the following issues:

A. As explained in the Appendix on Reasons common to several requests your read-across adaptation under Annex XI, Section 1.5 is rejected. In addition, as further explained under issue B. below, deficiencies were identified on the studies included in your registration dossier.

B. According to Annex XI, Section 1.5., the results to be read across should have adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3), in this case OECD TG 211 and OECD GD 23 (ENV/JM/MONO(2000)6/REV1) if the substance is difficult to test. Therefore, the following specifications must be met:

Reporting of the methodology and results

- the test design is reported (*e.g.* semi-static or flow-through, number of replicates, number of parents per replicate);
- the test procedure is reported (*e.g.* loading in number of *Daphnia* per litre, test medium composition);
- the methods used to prepare stock and test solutions is reported;
- if an undefined organic material is included in the test medium, its composition, source, method of preparation, TOC/DOC of stock preparations, estimation of resulting TOC/DOC in the final test medium are provided;
- detailed information on feeding, including amount (in mgC/daphnia/day) and schedule is reported;
- results from any preliminary studies on the stability of the test substance is reported;
- the nominal test concentrations and the results of all analyses to determine the concentration of the test substance in the test vessels are reported;
- water quality monitoring within the test vessels (*i.e.* pH, temperature and dissolved oxygen concentration, and TOC and/or COD and hardness where applicable) is reported;
- the full record of the daily production of living offspring during the test by each parent animal/in each replicate is provided;
- the number of deaths among the parent animals (if any) and the day on which they occurred is reported;
- the coefficient of variation for control reproductive output is reported.

You have not reported any of the information listed above. In the absence of this information, it is not possible to conduct an independent assessment of the study and therefore this information is rejected.

In your comments on the draft decision, you explain that the robust study summary on the long-term toxicity on aquatic invertebrates has been updated and now includes the requested information. However, you have not provided this information as part of your comments on the draft decision.

Please note that this decision does not take into account updates of the registration dossiers after the date on which you were notified of the draft decision according to Article 50(1) of REACH (see section 5.4. of ECHA's Practical Guide "How to act in Dossier Evaluation).

On this basis, the information requirement is not fulfilled.

Study design

OECD TG 211 specifies that for difficult to test substances OECD GD 23 must be followed. As already explained above, the Substance is difficult to test. Therefore, you must fulfil the requirements described in 'Study design' under Appendix A.3.

3. Long-term toxicity testing on fish

Long-term toxicity testing on fish is an information requirement under Annex IX to REACH (Section 9.1.6.).

You have provided the following information:

i. a justification to omit the study which you consider to be based on Annex IX, Section 9.1., Column 2. In support of your adaptation, you did not provide a justification/provided the following justification: "The hazard assessment of *dibutylamine reveals neither a need to classify the substance as dangerous to the environment, nor is it a PBT or vPvB substance, nor are there any further indications that the substance may be hazardous to the environment. Therefore, and for reasons of animal welfare, a long-term toxicity testing in fish is not provided*".

We have assessed this information and identified the following issue:

Annex IX, Section 9.1., Column 2 does not allow omitting the need to submit information on long-term toxicity to fish under Column 1. It must be understood as a trigger for providing further information on long-term toxicity to fish if the chemical safety assessment according to Annex I indicates the need (Decision of the Board of Appeal in case A-011-2018).

Your adaptation is therefore rejected.

In your comments on the draft decision, while you recognize the rejection of the adaptation of the information requirement, you also specify that you intend to adapt this information requirement under Annex XI, Section 1.2. ('Weight of evidence'). You intend to provide the following justification:

i. The structure as well as the physico-chemical properties of the Substance are clearly identified. The Substance is readily biodegradable; therefore, relevant metabolites do not need to be considered;
ii. The substance does not produce an alert for protein binding in the schemes by OECD and OASIS (OECD QSAR Toolbox v4.3; see Chapter 2.5 of the updated Read-Across Justification). According to the modified classification scheme of Verhaar, the mode of action of the Substance is narcosis of baseline toxicity. Therefore, it can be concluded that the Substance has no specific mode of action and critical long-term effects are not to be expected;
iii. You specify that no information on long-term toxicity to fish is available for the analogue substance diethylamine (CAS 109-89-7) and that no reliable QSAR predictions or in-vitro results for long-term toxicity to fish are not available;
iv. Fish are not the most sensitive aquatic trophic level;
v. The Substance is neither acutely nor chronically hazardous to the aquatic environment according to the CLP-Regulation (EC) No 1272/2008. You based you reasoning on aquatic chronic classification on the result of the data currently available on short-term toxicity to fish and the concept of acute-to-chronic ratio;
vi. You further consider that this information is not needed for the PBT assessment of the Substance as it is concluded no P/vP based on ready biodegradability;
vii. You refer to Article 25 to REACH to specify that vertebrate animal testing should be

undertaken as a last resort.

We take note of your intention to submit an adaptation. However, we emphasize that the justification above does not rely on any source of information that could be used to conclude on long-term fish toxicity.

Relevant information that can be used to support weight of evidence adaptation for long-term toxicity to fish includes similar information that is produced by the OECD TG 210. The following aspects need to be covered: Parameters related to the survival and development of fish in early life stages from the stage of fertilized egg until the juvenile life-stage following exposure to the test substance are measured, including:

1) the stage of embryonic development at the start of the test, and
2) hatching of fertilized eggs and survival of embryos, larvae and juvenile fish, and
3) the appearance and behaviour of larvae and juvenile fish, and
4) the weight and length of fish at the end of the test.

As you did not submit such information, ECHA concludes that there is, in your justification, no weight of evidence to be assessed. Furthermore, your argument that no significant long-term toxicity on fish is expected based on available information on short-term toxicity to fish is not valid as, for the reasons explained under Appendix B.2, the information requirement for that endpoint is not fulfilled. Finally, the use of the acute-to chronic ratio concept on its own is not regarded as providing sufficient weight of evidence to conclude on chronic toxicity (ECHA Guidance R.7.8.5.).

On this basis, the information requirement is not fulfilled.

Study design

To fulfil the information requirement for the Substance, the Fish, Early-life Stage Toxicity Test (test method OECD TG 210) is the most appropriate (ECHA Guidance R.7.8.2.).

OECD TG 210 specifies that for difficult to test substances OECD GD 23 must be followed. As already explained above, the Substance is difficult to test. Therefore, you must fulfil the requirements described in 'Study design' under Appendix A.3.

Appendix D: Reasons to request information required under Annex X of REACH

1. Pre-natal developmental toxicity study in a second species

Pre-natal developmental toxicity (PNDT) studies (OECD TG 414) in two species is an information requirement under Annex X to REACH (Section 8.7.2.).

You have adapted this information requirement under Annex XI, Section 1.5 ('Grouping of substances and read-across approach'). In support of your adaptation, you provided the following information:

i. a key study according to OECD TG 414 via oral route (gavage) in rats with an analogue substance Dibutylamine hydrochloride with EC No. 228-521-0 ([redacted], 2010);
ii. a key study according to OECD TG 414 via oral route (gavage) in rats with an analogue substance Dimethylamine hydrochloride with EC No. 208-046-5 ([redacted], 2009);
iii. a supporting study according to OECD TG 414 via oral route (gavage) in rats with an analogue substance Tributylamine with EC No. 203-058-7 ([redacted], 1991).

We have assessed this information and identified the following issues:

A. As explained in the Appendix on Reasons common to several requests your read-across adaptations under Annex XI, Section 1.5. using information from Dimethylamine hydrochloride with EC No. 208-046-5 and from Tributylamine with EC No. 203-058-7 are rejected. In addition, as further explained under issue B. below, deficiencies were identified on one of the studies included in your registration dossier.

B. Under Annex XI, Section 1.5., the results to be read across must provide adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3), in this case OECD TG 414. Therefore, the following specification must be met:
   - dosing of the Substance from implantation until the day prior to scheduled caesarean section

   In the study iii., the animals were exposed on gestation days 6-15 and sacrificed on gestation day 20. The study does not have a required exposure duration because the exposure duration is not from implantation until the day prior to scheduled caesarean section as specified in OECD TG 414.

   Therefore, the study iii. does not have adequate and reliable coverage of the key parameters of the OECD TG 414.

C. You have not provided any reliable information on a second species. In order to be compliant and enable concluding if the Substance is a developmental toxicant, information provided has to meet the requirements of OECD TG 414 in two species.

On this basis, the information requirement is not fulfilled.

Study design

A PNDT study according to the OECD TG 414 study should be performed in the rabbit or rat as the preferred species. The test in the first species was carried out by using a rodent species (rat). Therefore, a PNDT study in a second species must be performed in the rabbit as preferred non-rodent species.

The Substance is a corrosive liquid and you apply self-classification as Skin Corr. 1A (H314). ECHA Guidance R.7.6.2.3.2 specifies that corrosive or highly irritating substances must be tested preferably via the oral route. However, testing at concentration/dose levels causing corrosivity must be avoided.

As already explained in Appendix C.1., testing of neutral salts of alkaline or acidic substances is more appropriate as it allows the investigation of intrinsic properties at adequate dose levels.

In your comments, you agreed to perform a study according to OECD TG 414 in the second species (rabbit) via the oral route. However, you did not agree to use the neutralised salt of the Substance due to the following reasons:

- *"Testing of the salt ignores intrinsic properties of the registered substance that might alter absorption and limit exposure under real-life conditions."*
- *"According to REACH Annex V and the corresponding guidance, attachment 1 (3), "deliberate neutralization of acids or bases to form the corresponding salts [...] is not covered by this exemption." Consequently, the "neutral salt" as stated in the draft decision would not be covered by the registration but would require a read across justification."*

ECHA disagrees with your argument that testing the neutralised salt would ignore the intrinsic properties of the Substance. Dissociation constant (pKa) of the Substance is 11. Therefore, the Substance will exist as a protonated form (NH_2^+) under physiological conditions as will the neutralised form of the Substance. Therefore, similar absorption and systemic effects are expected for the Substance and its neutralised form under physiological conditions. ECHA notes that you have provided an adequate PNDT study according to OECD TG 414 in one species (rat) via oral route with a neutralised salt of the Substance, i.e. Dibutylamine hydrochloride (EC No. 228-521-09) and read-across to the Substance is plausible as such as already explained in the Appendix on Reasons common to several requests. This will also apply for the request to use a neutralised form of the Substance in the PNDT study in rabbits.

Therefore, the study must be performed with oral administration (ECHA Guidance R.7.6.2.3.2.) of a neutralised form of the Substance.

Appendix E: Requirements to fulfil when conducting and reporting new tests for REACH purposes

A. Test methods, GLP requirements and reporting

1. Under Article 13(3) of REACH, all new data generated as a result of this decision must be conducted according to the test methods laid down in a European Commission Regulation or to international test methods recognised by the Commission or ECHA as being appropriate.
2. Under Article 13(4) of REACH, ecotoxicological and toxicological tests and analyses must be carried out according to the GLP principles (Directive 2004/10/EC) or other international standards recognised by the Commission or ECHA.
3. Under Article 10(a)(vi) and (vii) of REACH, all new data generated as a result of this decision must be reported as study summaries, or as robust study summaries, if required under Annex I of REACH. See ECHA Practical Guide on How to report robust study summaries[4].

B. Test material

Before generating new data, you must agree within the joint submission on the chemical composition of the material to be tested (Test Material) which must be relevant for all the registrants of the Substance.

1. Selection of the Test material(s)
   The Test Material used to generate the new data must be selected taking into account the following:
   - the variation in compositions reported by all members of the joint submission,
   - the boundary composition(s) of the Substance,
   - the impact of each constituent/ impurity on the test results for the endpoint to be assessed. For example, if a constituent/ impurity of the Substance is known to have an impact on (eco)toxicity, the selected Test Material must contain that constituent/ impurity,
   - as explained under Appendix C.1., the use of a neutral salts of the Substance (*e.g.* hydrochloride salt of the Substance) is more appropriate for conducting the tests requested under Appendix C.1. and D.1. as it allows the investigation of intrinsic properties at adequate dose levels. When selecting a neutral salt, the potential impact of the counterion must be considered. The counterion must have no known systemic toxicity.

2. Information on the Test Material needed in the updated dossier
   - You must report the composition of the Test Material selected for each study, under the "Test material information" section, for each respective endpoint study record in IUCLID.
   - The reported composition must include all constituents of each Test Material and their concentration values and other parameters relevant for the property to be tested.

This information is needed to assess whether the Test Material is relevant for the Substance and whether it is suitable for use by all members of the joint submission.
Technical instructions on how to report the above is available in the manual on How to prepare registration and PPORD dossiers[5].

[4] https://echa.europa.eu/practical-guides
[5] https://echa.europa.eu/manuals

Appendix F: Procedure

The information requirement for an Extended one-generation reproductive toxicity study (EOGRTS; Annex X, Section 8.7.3.) is not addressed in this decision. This may be addressed in a separate decision once the information from the Sub-chronic toxicity study (90-day) requested in the present decision is provided; due to the fact that the results from the 90-day study is needed for the design of the EOGRTS.

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

ECHA followed the procedure detailed in Articles 50 and 51 of REACH.

The compliance check was initiated on 24 March 2020.

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

ECHA took into account your comments and did not amend the requests.

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

As no amendments were proposed, ECHA adopted the decision under Article 51(3) of REACH.

Appendix G: List of references - ECHA Guidance[6] and other supporting documents

Evaluation of available information
Guidance on information requirements and chemical safety assessment, Chapter R.4 (version 1.1., December 2011), referred to as ECHA Guidance R.4 where relevant.

QSARs, read-across and grouping
Guidance on information requirements and chemical safety assessment, Chapter R.6 (version 1.0, May 2008), referred to as ECHA Guidance R.6 where relevant.

Read-across assessment framework (RAAF, March 2017)[7]

RAAF - considerations on multiconstituent substances and UVCBs (RAAF UVCB, March 2017)[7]

Physical-chemical properties
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Toxicology
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

Environmental toxicology and fate
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7b (version 4.0, June 2017), referred to as ECHA Guidance R.7b in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

PBT assessment
Guidance on information requirements and chemical safety assessment, Chapter R.11 (version 3.0, June 2017), referred to as ECHA Guidance R.11 in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.16 (version 3.0, February 2016), referred to as ECHA Guidance R.16 in this decision.

Data sharing
Guidance on data-sharing (version 3.1, January 2017), referred to as ECHA Guidance on data sharing in this decision.

[6] https://echa.europa.eu/guidance-documents/guidance-on-information-requirements-and-chemical-safety-assessment
[7] https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across

OECD Guidance documents[8]
Guidance Document on aqueous-phase aquatic toxicity testing of difficult test chemicals – No 23, referred to as OECD GD 23.

Guidance document on transformation/dissolution of metals and metal compounds in aqueous media – No 29, referred to as OECD GD 29.

Guidance Document on Standardised Test Guidelines for Evaluating Chemicals for Endocrine Disruption – No 150, referred to as OECD GD 150.

Guidance Document supporting OECD test guideline 443 on the extended one-generation reproductive toxicity test – No 151, referred to as OECD GD 151.

[8] http://www.oecd.org/chemicalsafety/testing/series-testing-assessment-publications-number.htm

Appendix H: Addressees of this decision and their corresponding information requirements

You must provide the information requested in this decision for all REACH Annexes applicable to you.

Registrant Name	Registration number	Highest REACH Annex applicable to you
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]

Where applicable, the name of a third party representative (TPR) may be displayed in the list of recipients whereas ECHA will send the decision to the actual registrant.