~~Confidential~~

Helsinki, 15 March 2023

Addressees
Registrant(s) of Tallow DPG triamine AC - (T) as listed in Appendix 3 of this decision

Date of submission of the dossier subject to this decision
29/06/2012

Registered substance subject to this decision ("the Substance")
Substance name: N-(3-ammoniopropyl)-N'-(C16-C18, C18 unsatured)alkylpropane-1,3-diaminium triacetate
EC/List number: 700-693-6

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 ***20 June 2024***.

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

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: OECD TG 471, 2020)

2. Short-term toxicity testing on aquatic invertebrates (Annex VII, Section 9.1.1.; test method: EU C.2./OECD TG 202)

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

4. Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: EU C.4. C/D/E/F/OECD TG 301B/C/D/F or EU C.29./OECD TG 310) on relevant constituent(s)/fraction(s) of the Substance, as described under the corresponding appendix on reasons for the request.

The reasons for the decision(s) are explained in Appendix 1.

Information required depends on your tonnage band

You must provide the information listed above for all REACH Annexes applicable to you in accordance with Articles 10(a) and 12(1) of REACH. The addressees of the decision and their corresponding information requirements based on registered tonnage band are listed in Appendix 3.

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 requirements for testing and reporting new tests under REACH, see Appendix 4.

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 Mike Rasenberg, Director of Hazard Assessment

Appendix 1: Reasons for the request(s)
Appendix 2: Procedure
Appendix 3: Addressees of the decision and their individual information requirements
Appendix 4: Conducting and reporting new tests under REACH

[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 for the request(s)

Contents

0. Reasons common to several requests

0.1. Read-across adaptation rejected

1 You have adapted the following standard information requirements by using grouping and read-across approach under Annex XI, Section 1.5:

- In vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.)
- Long-term toxicity testing on aquatic invertebrates (Annex VII, Section 9.1.1., column 2)
- Growth inhibition study aquatic plants (Annex VII, Section 9.1.2.)
- Ready biodegradability (Annex VII, Section 9.2.1.1.)

2 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 sections.

3 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.

4 Additional information on what is necessary when justifying a read-across approach can be found in the Guidance on IRs and CSA, Chapter R.6. and related documents (RAAF, 2017; RAAF UVCB, 2017).

0.1.1. Scope of the grouping of substances (category)

5 You provide a read-across justification document in IUCLID Section 13.

6 For the purpose of this decision, the following abbreviations are used for the category members:

7 Linear Polyamines:

- Triamine C: Coco dipropylene triamine (CAS RN 91771-18-5)
- Triamine OV: Oleyl (vegetable oil) dipropylene triamine (CAS RN 28872-01-7)
- Triamine T: Tallow dipropylene triamine (CAS RN 61791-57-9) also referred to as N-(3-aminopropyl)-N'-C16-18 (evennumbered), C18 unsaturated alkyl -propane-1,3-diamine (CAS RN 1219458-14-6)
- Tetramine T: N-tallow alkyltripropylene tetramine (CAS RN 68911-79-5) also referred to as N-(3-aminopropyl)-N'-[3-(C16-18 (evennumbered), C18 unsaturated alkyl amino)propyl]propane-1,3- diamine (CAS RN 1219458-11-3)
- Tetramine OV: Oleyl(vegetable oil) tripropylene tetramine (CAS RN 67228-83-5)

8 Branched polyamines:

- Triamine Y12: Dodecyl dipropylene triamine, branched (CAS RN 2372-82-9)
- Triamine YT: Tallow dipropylene triamine, branched (CAS RN 85632-63-9) also referred to as N-(3-aminopropyl)-N-N-(C16-18 evennumbered, 18 unsaturated)-alkylpropane-1,3-diamine (CAS RN 1219826-66-0),

9 Polyamine acetates:

- Triamine OV acetate: (Z)-N-(3-aminopropyl)-N'-[3-(9-octadecenylamino)propyl] propane-1,3-diamine acetate (No CAS RN or EC No. provided)
- Triamine OV acetate : (Z)-N-(3-aminopropyl)-N'-9-octadecenylpropane-1,3-diamine acetate (No CAS RN or EC No. provided)
- Triamine T acetate: (N-(3-aminopropyl)-N'-[3-(C16-18(evennumbered), C18 unsaturated alkyl amino)propyl]propane-1,3- diamine acetate (No CAS RN or EC No. provided)

10 Branched polyamine acetate

- Triamine YT acetate: N-(3-aminopropyl)-N-N-(C16-18 evennumbered, 18 unsaturated)-alkylpropane-1,3- diamine acetate (No CAS RN or EC No. provided)

11 You justify the grouping of the substances as:

- "*Structurally, the linear di-, tri- and tetramines are very similar: a linear alkyl chain and a primary amine at the end, with 1, 2 or 3 secondary amines in between. Consequently, they share the same chemical reactivity and their physico-chemical properties are very similar from which a comparable toxicological profile can be expected"*.
- "*The variability of the alkyl chain length* […] *is suspected to influence aspects related to bioavailability, but not aspects of chemical reactivity, route of metabolism, and specific mechanisms of toxicity e.g. sensitization and genotoxicity. For these reasons, many of the toxicological studies can best be performed on the substance with the shortest chain length within the sub-category, as this is considered to result to the lowest NOAEL or most likely able to show specific effects where for ecotoxicology and fate studies can best be focussed on the extremes of the category*".
- "*Addition of an acetate group to the polyamines increases their water solubility. Due to dissociation in the gastrointestinal tract, a principal difference in their toxicological profile compared to the polyamines without the acetate for systemic toxicity cannot be expected. However as the acetate group is added to increase water solubility of the compound, and influences the pH of the substance, small differences can be expected in the toxicity profile for acute toxicity and local effects*".
- "*General profiling in OECD Toolbox are the same for all the linear triamine and tetramine structures: Aliphatic amines, no protein or DNA binding, and no specific alerts. Lipinski rule indicates that the longer chain lengths are likely less bioavailable compared to the shorter chain lengths*".

12 You define the applicability domain as: "*substances that contain 1 or more repeating 1,3-diamino propane (DP) groups linked to a fatty amine. These can be linearly linked based on one DP and fatty amine (diamine), two DP and fatty amine (triamine structure: alkyl dipropylene triamine) or 3 DP with a fatty amine (tetramine structure: alkyl tripropylene tetramine), or in a branched or Y-amine form of two DP that are both linked to the nitrogen of a fatty amine (The annotation 'branched' in this case does not refer to the alkyl chain)* […] [The] *alkyl chain rang[e] from 12 (Dodecane) to 18 (Octadecane), depending on the source of the fatty amine. For alkyl chain lengths, largely the following ranges are implied in this group: Tallow C16 (25-40%) and C18 (50-75%); Oleyl C18 (>70 %); Coco C12 (45-62%) and C14 (15-25%)*". You specify that "*tetraamines also contain for a large part triamines and some diamines, and the triamines can contain a considerable*

amount of diamines and some tetraamines". Finally, you state that "[the] *previously formed category of polyamines is extended to include polyamine acetates*".

13 ECHA understands that this is the applicability domain of the grouping and your predictions are assessed on this basis.

14 We have identified the following issue with the proposed scope of the grouping:.

0.1.1.1. Incomplete characterisation of the group members

15 Annex XI, Section 1.5 of the REACH Regulation provides that "*substances whose physicochemical, toxicological and ecotoxicological properties are likely to be similar or follow a regular pattern as a result of structural similarity may be considered as a group*."

16 According to the Guidance on IRs and CSA, Section R.6, "*in identifying a category, it is important that all potential category members are described as comprehensively as possible*", because the purity profile and composition can influence the overall toxicity/properties of the potential category members (Guidance on IRs and CSA, Section R.6.2.4.1.). Therefore, qualitative and quantitative information on the compositions of the category members must be provided to confirm the category membership.

17 Furthermore, the provided information for categories consisting of UVCB (Unknown or Variable composition, Complex reaction products or of Biological materials) substances needs to include qualitative compositional information of the individual constituents of the category members; as well as quantitative characterisation in the form of information on the concentration of the individual constituents of these substances; to the extent that this is measurable (Guidance on IRs and CSA, Section R.6.2.5.5.).

18 Your definition of the applicability domain of the category can be summarised as linear and branched triamine and tetraamine substance with C-chain length ranging mostly from C12 to C18. The alkyl chain can be saturated or unsaturated. The category is extended to include polyamine acetates.

19 Your read-across justification document contains compositional information for some polyamine acetates but no information on polyamines from the category.

20 In the absence of comprehensive qualitative and quantitative information on the compositions of the category members, the category membership of these substances cannot be confirmed.

21 Despite of the above issue, ECHA understands that this is the applicability domain of the grouping and your predictions are assessed on this basis.

0.1.2. Predictions for (eco)toxicological and fate properties

22 You provide a read-across justification document in IUCLID Section 13.

23 <u>Toxicological properties</u>

24 You predict the toxicological properties of the Substance from information obtained from the following source substance:

Triamine Y12 Dodecyl dipropylene triamine, branched (CAS RN 2372-82-9)

25 You provide the following reasoning for the prediction of toxicological properties:

- "*Due to dissociation in the gastrointestinal tract, a principal difference in their toxicological profile compared to the polyamines without the acetate for systemic toxicity cannot be expected.*"
- "*However as the acetate group is added to increase water solubility of the compound, and influences the pH of the substance, small differences can be expected in the*

toxicity profile for acute toxicity and local effects."

- "*As the acetate will dissociate from the substance during the process of absorption, there is no principal difference between systemic exposure to the polyamines acetate and the polyamine without the acetate. Consequently, cross-reading is applied for this endpoint from the corresponding polyamine substance without acetate."*

26 ECHA understands that your read-across hypothesis assumes that different compounds have the same type of effects. You predict the properties of your Substance to be quantitatively equal to those of the source substance.

27 Ecotoxicological properties

28 You predict the ecotoxicological properties of the Substance from information obtained from the following source substance(s):

Triamine OV Oleyl (vegetable oil) dipropylene triamine (CAS RN 28872-01-7)

29 You have not provided any specific reasoning for the predictions of ecotoxicity.

30 ECHA understands that your read-across hypothesis assumes that different compounds have the same type of effects. You predict the properties of your Substance based on a worst-case approach claiming that the addition of acetates increases the molecular weight of the substance and reduces the toxicity.

31 Fate properties

32 You predict the fate properties of the Substance from information obtained from the following source substance(s):

Triamine C Coco dipropylene triamine (CAS RN 91771-18-5)

33 You provide the following reasoning for the prediction of fate properties: "*Based on the broad substrate specificity of micro-organisms degrading fatty amine derivatives with respect to the alkyl chain length it is unlikely that the biodegradability of these surfactants differs significantly with varying alkyl chain lengths. Biocidal effects explain negative results obtained in ready biodegradability tests*".

34 ECHA understands that your read-across hypothesis assumes that different compounds have the same type of effects. You predict the properties of your Substance to be quantitatively equal to those of the source substance.

35 We have identified the following issues which are common to the predictions of toxicological, ecotoxicological and fate properties:

0.1.2.1. Insufficient data density

36 Annex XI, Section 1.5. provides that "*substances whose physicochemical, toxicological and eco-toxicological properties are likely to be similar or follow a regular pattern as result of structural similarity may be considered as a group or 'category' of substances*".

37 According to the Guidance on IRs and CSA, Section R.6.2.1.5., one of the factors in determining the robustness of a category is the density and distribution of the available data across the category. To identify a regular pattern and/or to derive reliable prediction of the properties of the members of the category, adequate and reliable information covering the range of structural variations identified among the category members needs to be available.

38 Furthermore, in larger categories there may be breaks in trends which could affect the reliability of interpolation (Guidance on IRs and CSA, Section R.6.2.2.2.). To confirm that there are no such breakpoints, adequate and reliable information needs to cover also substances within a range of homologous series.

39 You have provided information on

- a single category member (*i.e.*, Triamine Y12) for *in vitro* gene mutation study in bacteria
- a single category member (*i.e.*, Triamine YT) for long-term toxicity on aquatic invertebrates and growth inhibition on aquatic plants
- a single category member (*i.e.*, Triamine C) for ready biodegradability

40 The selected analogues Triamine C and Triamine Y12 are on the lower border of the sub-category triamines.

41 Information for one category member is not sufficient to establish a trend across the category consisting of 11 substances. In particular, you have not provided data for the upper and lower borders of the category for the corresponding endpoints. In addition, in the absence of data for substances between the upper and lower borders of the category, it cannot be confirmed that there is no breakpoint in toxicity trend within the given range of chain length and that (i) the relative abundance of mono, di, tri and tetramine, (ii) the presence of amines in a branched or Y-amine form (iii) the presence of unsaturation of the alkyl chain and (iv) the presence of acetate as counterion will not impact the predictions. Therefore, the information provided is not sufficient to conclude that (eco)toxicological and properties are likely to follow a regular pattern.

0.1.2.2. Inadequate or unreliable source study

42 According to Annex XI, Section 1.5., if the grouping concept is applied then in all cases the results to be read across must:

(1) be adequate for the purpose of classification and labelling and/or risk assessment;

(2) have adequate and reliable coverage of the key parameters addressed in the corresponding study that shall normally be performed for a particular information requirement.

43 Specific reasons why the studies on the source substance do not meet these criteria are explained further below under the applicable information requirement sections 1, 2, 3, 4.

44 Therefore, no reliable predictions can be made for these information requirements

0.1.3. Conclusion on the read-across approach

45 For the reasons above, you have not established that relevant properties of the Substance can be predicted from data on the source substance(s). Your read-across approach under Annex XI, Section 1.5. is rejected.

Reasons related to the information under Annex VII of REACH

1. In vitro gene mutation study in bacteria

46 An in vitro gene mutation study in bacteria is an information requirement under Annex VII, Section 8.4.1.

1.1. Information provided

47 You have adapted this information requirement by using Annex XI, Section 1.5. (Grouping of substances and read-across approach) based on experimental data from the following substances:

(i) *in vitro* gene mutation study in bacteria (2002) with an analogue substance N-(3-aminopropyl)-N-dodecylpropane-1,3-diamine (CAS RN 2372-82-9).

1.2. Assessment of the information provided

1.2.1. Read-across adaptation rejected

48 As explained in Section 0.1., your adaptation based on grouping of substances and read-across approach under Annex XI, Section 1.5 is rejected. In addition, ECHA identified endpoint specific issues addressed below.

1.2.2. Insufficient information provided to confirm test material identity for study (i)

49 To comply with this information requirement, the test material in a study must be representative for the substance to be tested; Article 10 and Recital 19 of REACH; Guidance on IRs and CSA, Section R.4.1). The Test Methods Regulation (EU) 440/2008, as amended by Regulation (EU) 2016/266, requires that "*if the test method is used for the testing of a* [...] *UVCB* [...] *sufficient information on its composition should be made available, as far as possible, e.g. by the chemical identity of its constituents, their quantitative occurrence, and relevant properties of the constituents*". Such information includes purity, composition, carbon chain length, saturation, branching, depending on the type of UVCB substance.

50 The study (i) has been conducted with the UVCB substance listed above. You claim that the purity of the test material is [redacted]%. However, you have not provided any information on composition for the test material.

51 In the absence of detailed information on the UVCB 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 that was intended to be tested.

1.2.3. Inadequate or unreliable study (i) on the source substance

52 As explained in Section 0.1., the results to be read across must have an adequate and reliable coverage of the key parameters addressed in the test guideline for the corresponding study that shall normally be performed for a particular information requirement, in this case OECD TG 471. Therefore, the following specifications must be met:

a) one positive control is included in the study and the positive control substance produces a statistically significant increase in the number of revertant colonies per

plate compared with the concurrent negative control;
b) the number of revertant colonies per plate for the concurrent negative control is inside the historical control range of the laboratory;
c) the mean number of revertant colonies per plate is reported for the treated doses and the controls.

53 In study (i) described as an in vitro gene mutation study on bacteria:

a) you have not shown that the positive control substance produced a statistically significant increase in the number of revertant colonies per plate compared with the concurrent negative control;
b) you have not shown that the number of revertant colonies per plate for the concurrent negative control was inside the historical control range of the laboratory;
c) the mean number of revertant colonies per plate for the treated doses and the controls was not reported.

54 The information provided does not cover the specification(s) required by the OECD TG 471.

55 Therefore, the information requirement is not fulfilled.

1.3. Specification of the study design

56 To fulfil the information requirement for the Substance, the in vitro gene mutation study in bacteria (OECD TG 471, 2020) is considered suitable.

2. Short-term toxicity testing on aquatic invertebrates

57 Short-term toxicity testing on aquatic invertebrates is an information requirement under Annex VII to REACH (Section 9.1.1.).

2.1. Information provided

58 You have adapted this information requirement by using Column 2 of Annex VII, Section 9.1.2. To support the adaptation, you have provided following information:

(i) a study on long-term toxicity to aquatic invertebrates (2010) according to OECD TG 211 with the analogue substance Oleyl (vegetable oil) dipropylene triamine (CAS RN 28872-01-7)

2.2. Assessment of the information provided

59 Under Annex VII, Section 9.1.2., Column 2, second indent, the study may be omitted if a long-term aquatic toxicity study on invertebrates is available.

2.2.1. Read-across adaptation rejected

60 As explained in Section 0.1., your adaptation based on grouping of substances and read-across approach under Annex XI, Section 1.5. is rejected. In addition, ECHA identified endpoint specific issue(s) addressed below.

2.2.2. Insufficient information provided to confirm test material identity

61 To comply with this information requirement, the test material in a study must be representative for the substance to be tested; Article 10 and Recital 19 of REACH; Guidance

on IRs and CSA, Section R.4.1). The Test Methods Regulation (EU) 440/2008, as amended by Regulation (EU) 2016/266, requires that "*if the test method is used for the testing of a* [...] *UVCB* [...] *sufficient information on its composition should be made available, as far as possible, e.g. by the chemical identity of its constituents, their quantitative occurrence, and relevant properties of the constituents*". Such information includes purity, composition, carbon chain length, saturation and branching.

62 The study (i) has been conducted with a test material considered by you as representative to the Substance. You provide the following information on the test material: "*Primary fatty amine:* [redacted] *(area %), Di amine:* [redacted] *(area %), Tri amine:* [redacted] *(area %), Tetra amine:* [redacted] *(area %)*". You have not provided information on the distribution of the C-chain length, on the presence of unsaturated constituents and on branching.

63 In the absence of detailed information on the UVCB 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 that was intended to be tested.

2.2.3. Inadequate or unreliable study on the source substance

64 Under Annex XI, Section 1.5., the results to be read across must have an adequate and reliable coverage of the key parameters addressed in the study that must normally be performed for a particular information requirement, in this case for long-term toxicity to aquatic invertebrates the OECD TG 211. If the analogue substance is difficult to test, the requirements of OECD GD 23 must be followed. Therefore, the following specifications must be met:

65 Technical specifications impacting the sensitivity/reliability of the test

a) the test medium fulfils the following condition(s): total organic carbon (TOC) ≤ 2 mg/L;

66 Characterisation of exposure

b) if the concentrations of the test material in a semi-static test are not expected to remain within ± 20 % of the nominal concentration, then all test concentrations must be determined when freshly prepared and at the time of renewal on one occasion during each week of the test;

67 Additional requirements applicable to difficult to test substances

c) a continuous flow through exposure system is used if exposure concentrations cannot be maintained within 80-120% of nominal in a semi-static exposure system with a renewal frequency of 24 hours.

68 In study (i) described as a long-term toxicity study on daphnids according to OECD TG 211:

69 Technical specifications impacting the sensitivity/reliability of the test

a) you specify that Natural river water of the river [redacted] was used as test medium. You report that the TOC concentration was [redacted] mg/L;

70 Characterisation of exposure

b) you report measured concentrations that are below ± 20 % of the nominal concentration in both fresh and old media throughout the test at 30, 90 and 270 µg/L. However, you have not provided the results of the analytical monitoring for the test at 10 µg/L and you have provided a single measurement at 90 µg/L;

71 Additional requirements applicable to difficult to test substances

c) test was conducted under semi-static conditions with a renewal rate of test solutions (frequency) of 3 times per week. Measured test concentrations were:

- 270 µg/L: recovery in fresh media ranged from 52 to 87% and in old media from 0% (i.e. measured value was below LOQ) to 44%
- 90 µg/L (measured only on one occasion): recovery in fresh media was 9% and in old media recovery was 0% (i.e. measured value was below LOQ);
- 30 µg/L: recovery in fresh media ranged from 0% (i.e. measured value was below LOQ) to 167%% and in old media recovery was always 0% (i.e. measured value was below LOQ).

72 Based on the above, there are critical methodological deficiencies resulting in the rejection of the study results. More, specifically

- the TOC content of the test medium was above the mandatory value of 2 mg/L which is not adequate to investigate the intrinsic hazards of the Substance. You justify the use of natural water with high TOC by referring to the "bulk approach" (ECETOC, 2001). However, ECHA notes that information on intrinsic properties of a substance must be generated independently from exposure considerations (*e.g.*, decision of the Board of Appeal of 11 December 2018 in case A-006-2017, para. 133-135). The Guidance on Application of CLP Criteria, Section 1.1.3., specifies that classification must be based on intrinsic hazards, *i.e.* the basic properties of a substance as determined in standard tests or by other means designed to identify hazards. Therefore, the bulk approach which aims at mimicking exposure under "more environmentally realistic" conditions must not be used for classification and labelling. Similar considerations apply for the PBT assessment. As per Annex XIII of REACH, the PBT assessment should be based on data generated under 'relevant conditions', i.e. those conditions that allow for an objective assessment of the PBT/vPvB properties of a substance and not the PBT/vPvB properties of a substance under particular environmental conditions. This has been also confirmed by the Board of Appeal in its Decision of 7 December 2016 in case A-013-2014.
- the monitoring of exposure concentrations did not cover all required concentrations and, for some concentrations did not have an appropriate frequency over the exposure phase.
- the test design for the study was not adequate to maximize the exposure to the test material. The reported results on the analytical monitoring of exposure shows that concentrations were not maintained below ± 20 % of the nominal concentration. However, you have not attempted to increase the frequency of test medium renewal to 24 hours or used a flow-through test set-up as required by the OECD GD 23.

73 Therefore, the study submitted in long-term toxicity study present in your registration dossier does not provide an adequate and reliable coverage of the key parameter(s) of the corresponding OECD TG 211.

74 On this basis, you have not provided adequate information for long-term aquatic toxicity study on invertebrates. Therefore, your adaptation is rejected and the information requirement is not fulfilled.

2.3. Study design and test specifications

75 The Substance is difficult to test due to its low water solubility (CMC of 46 mg/L), surface activity (surface tension of 36 mN/m at 1 g/L), ionisable properties (pKa for the first amine of > 9) and adsorptive properties (sorption to suspended matter and DOC is reported ecotox studies). OECD TG 202 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 202. 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.

3. Growth inhibition study aquatic plants

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

3.1. Information provided

77 You have also adapted this information requirement by using Annex XI, Section 1.5. (Grouping of substances and read-across approach) based on experimental data from the following substances:

(i) a toxicity study to aquatic algae and cyanobacteria (2009) according to OECD TG 201 with the analogue substance Oleyl (vegetable oil) dipropylene triamine (CAS RN 28872-01-7)

3.2. Assessment of the information provided

3.2.1. Read-across adaptation for growth inhibition study with aquatic plants is rejected

78 As explained in Section 0.1., your adaptation based on grouping of substances and read-across approach under Annex XI, Section 1.5. is rejected. In addition, ECHA identified endpoint-specific issue(s) addressed below.

3.2.1. Insufficient information provided to confirm test material identity

79 To comply with this information requirement, the test material in a study must be representative for the substance to be tested; Article 10 and Recital 19 of REACH; Guidance on IRs and CSA, Section R.4.1). The Test Methods Regulation (EU) 440/2008, as amended by Regulation (EU) 2016/266, requires that "*if the test method is used for the testing of a [...] UVCB [...] sufficient information on its composition should be made available, as far as possible, e.g. by the chemical identity of its constituents, their quantitative occurrence, and relevant properties of the constituents*". Such information includes purity, composition, carbon chain length, saturation and branching.

80 The study (i) has been conducted with the UVCB listed above. You claimed that the test material was representative of the boundary composition of the Substance. However, you did not provide any information on purity and composition (including carbon chain length, saturation, branching) to support your claim.

81 In the absence of detailed information on the UVCB 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 that was intended to be tested.

3.2.2. Inadequate or unreliable study on the source substance

82 Under Annex XI, Section 1.5., the results to be read across must have an adequate and reliable coverage of the key parameters addressed in the study that must normally be performed for a particular information requirement, in this case the OECD TG 201. If the analogue substance is difficult to test, the requirements of OECD GD 23 must be followed. Therefore, the following specifications must be met:

83 Technical specifications impacting the sensitivity/reliability of the test

a) one of the two alternative growth medium (*i.e.* the OECD or the AAP medium) is used. Any deviations from recommended test media must be described and justified;

84 Characterisation of exposure

b) 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);
c) the concentrations of the test material are measured at least at the beginning and end of the test:
   - at the highest, and
   - at the lowest test concentration, and
   - at a concentration around the expected EC_{50}.
d) for volatile, unstable or strongly adsorbing test substances, additional samplings for analysis at 24 hour intervals is required;

85 Additional requirements applicable to difficult to test substances

e) for adsorbing test chemical, dissolved total organic carbon concentrations (other than that due to the test chemical) must be maintained in all test solutions at or below 2 mg/L.

86 In study (i) described as growth inhibition study on aquatic plants/algae:

87 Technical specifications impacting the sensitivity/reliability of the test

a) the test medium is described as natural water from the river Innerste. You have provided the following justification for not using one of the two alternative growth medium of OECD TG 201: "*The aquatic ecotoxicity tests with polyamines were therefore performed in river water to allow a PECaquatic, bulk/PNECaquatic, bulk approach and is considered to be conservative but more environmentally realistic than the standard method";*

88 Characterisation of exposure

b) the test media prepared specifically for analysis of exposure concentrations was not inoculated with algae;
c) the concentration of the test material was only verified at beginning and end of the test, at the second lowest and the second highest test concentration ([redacted] and [redacted] mg/L);
d) as explained under request 2, the substance is considered to be adsorptive. You have observed significant loss from the test medium at t=72h and no additional sampling for analysis at 24 h interval was conducted;

89 Additional requirements applicable to difficult to test substances

e) as already explained under request 2, the substance is considered to be adsorptive. You report that the test was conducted with natural freshwater with a TOC content of 2.9 mg/L.

90 Based on the above, there are critical methodological deficiencies resulting in the rejection of the study results. More specifically,

- you have not used one of the two alternative growth medium and the TOC content of the test medium was above the mandatory value of 2 mg/L. You justify this deviation by referring to the "bulk approach" (ECETOC, 2001). As already explained under section 2.2.3.3., the bulk approach is not adequate for the purpose of classification and labeling and the PBT assessment.
- you have not demonstrated that exposure was satisfactorily maintained and that effect concentrations can be expressed based on nominal concentrations as (i) not all required test concentrations were analytically monitored, (ii) the samples were not inoculated with algae, and (iii) the sampling frequency was not adequate.

91 Therefore, the requirements of OECD TG 201 are not met.

92 On this basis, the information requirement is not fulfilled.

3.3. Study design and test specifications

93 OECD TG 201 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 and test specifications' under Request 2.

4. Ready biodegradability

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

4.1. Information provided

95 You have adapted this information requirement by using Annex XI, Section 1.5. (Grouping of substances and read-across approach) based on experimental data from the following substance:

(i) a ready biodegradability study (2009) according to OECD TG 301D with the analogue Substance Coco dipropylene triamine (CAS RN 91771-18-5)

4.2. Assessment of information provided

4.2.1. Read-across adaptation for ready biodegradation is rejected

96 As explained in Section 0.1., your adaptation based on grouping of substances and read-across approach under Annex XI, Section 1.5. is rejected. In addition, ECHA identified endpoint-specific issue(s) addressed below.

4.2.2. Insufficient information provided to confirm test material identity

97 To comply with this information requirement, the test material in a study must be representative for the substance to be tested; Article 10 and Recital 19 of REACH; Guidance on IRs and CSA, Section R.4.1). The Test Methods Regulation (EU) 440/2008, as amended by Regulation (EU) 2016/266, requires that "*if the test method is used for the testing of a* [...] *UVCB* [...] *sufficient information on its composition should be made available, as far as possible, e.g. by the chemical identity of its constituents, their quantitative occurrence, and relevant properties of the constituents*". Such information includes purity, composition, carbon chain length, saturation and branching.

98 The study (i) has been conducted with the UVCB substances listed above. You provide the following information on the test material: "*coco dipropylene triamine ([redacted]%); coco propylene diamine ([redacted]%); coco propylene tetramine ([redacted]%); coco amine ([redacted]%)*". You have not provided information on the distribution of the C-chain length, on the presence of unsaturated constituents and on branching.

99 In the absence of detailed information on the UVCB 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 that was intended to be tested.

4.2.3. Inadequate or unreliable study on the source substance

100 Under Annex XI, Section 1.5., the results to be read across must have an adequate and reliable coverage of the key parameters addressed in the study that must normally be performed for a particular information requirement, in this case the OECD TG 301. Therefore, for a study according to OECD TG 301D, the following specifications must be met:

101 Technical specifications impacting the sensitivity/reliability of the test

a) test solutions are prepared using an appropriate nutrient medium, which includes ammonium chloride;
b) dilute inoculum without sludge flocs is used. The inoculum is normally derived from the secondary effluent of a treatment plant or laboratory-scale unit receiving predominantly domestic sewage;

102 Reporting of the methodology and results

c) the test conditions are reported, e.g.: adaptation of inoculum (if any), density of the inoculum in cells/mL and in mg/L suspended solid, test medium composition including organic carbon content which should not be higher than 10% of the organic carbon content introduced by the test material;
d) the results of measurements at each sampling point in each replicate is reported in a tabular form;
e) the calculation of the ThOD is described and justified;
f) for nitrogen-containing test materials, correction for nitrification is applied on the theoretical oxygen demand (i.e. ThODNO3) unless it can be demonstrated that nitrification did not occur (e.g. by monitoring changes in concentrations in nitrite and nitrate);

103 Your registration dossier provides an OECD TG 301D study showing the following:

104 Technical specifications impacting the sensitivity/reliability of the test

a) you report that "*ammonium chloride was not added to prevent oxygen consumption due to nitrification*". You justify the deviation by stating that "omission does not result in nitrogen limitation as shown by the biodegradation of the reference compound";
b) for study (i), you described the inoculum as "*River water* […] *from the Rhine near Heveadorp*". You state that particles were removed by sedimentation.

105 Reporting of the methodology and results

c) you have not specified the volume of inoculum added to the test bottles and you have not reported inoculum density in cells/mL;
d) the results of measurements at each sampling point in each replicate are not reported;
e) you provide a generic description of the equation used to derive the $ThOD_{NH3}$.

However, you do not describe how this equation was used considering the UVCB nature of the tested substance.

f) you have not reported whether a correction for nitrification was applied on the theoretical oxygen demand.

106 Based on the above,

- there are critical methodological deficiencies resulting in the rejection of the study results. More, specifically:
  - you have not used a standard test medium as you report that Ammonium chloride was omitted from the test medium. This deviation is no considered acceptable as it may artificially reduce oxygen consumption and lead to underestimating respiration in the inoculum blank (i.e. one of the validity criteria of OECD TG 301D). The lack of nitrogen limitation in the positive control does not address the above issue as it does not provide additional information with regard respiration in the inoculum blank.
- the reporting of the study is not sufficient to fully assess its reliability. More specifically:
  - as you have not reported inoculum concentration in the test vessel in cells/L, it is not possible to verify if the inoculum density was low enough to be consistent with the specifications of OECD TG 301D.
  - as you have not provided an adequate reporting of the study results, it is not possible to verify if validity criteria consistent with the specifications of OECD TG 301D were met;
  - you have not specified how ThOD was estimated and, as the test material is a nitrogen-containing substance, that the calculated ThOD takes into account oxygen consumption through nitrification (or alternatively supporting information that nitrification did not occur).

107 Therefore, the requirements of OECD 301 D are not met.

108 On this basis, the information requirement is not fulfilled.

4.3. Study design and test specification

109 The revised introduction to the OECD Guidelines For Testing Of Chemicals, Section 3 Part I states that ready biodegradability tests are intended for pure substances but may also be relevant, on a case-by-case basis, to mixtures of structurally similar chemicals (i.e. which are composed of constituents expected to show similar degradation kinetics). However, such tests are not generally applicable for complex mixtures or substances (i.e. UVCB or multi-constituent substances) containing different types of constituents. For complex substances, a single ready biodegradability test does not allow to conclude on the ready biodegradability of all constituents and therefore, does not fulfil the information requirement. The Substance is a UVCB that includes amines, diamines, triamines and tetramines with varying C-chain length. Some constituents are also unsaturated.

110 The Substance is a complex substance and contains constituents with significant structural differences described above.

111 For the reasons provided above, testing on the Substance as a whole does not fulfil the information requirement. For the generation of information on ready biodegradability, you must consider the level of information required for the purposes of classification and labelling and, if applicable to your registration, the PBT/vPvB assessment and the exposure assessment/risk characterisation. In order to conclude on which of constituents of the Substance are and which are not readily biodegradable, you may have to consider conducting more than one study using selected individual constituents and/or fractions. If you choose to test one (or more) fraction(s) of the Substance, you must provide a

justification that their constituents within chosen fraction(s) are similar enough so that similar degradation kinetics can be assumed. If you decide to conduct a single study in order to prove that all constituents of the Substance are readily biodegradable, you must provide a justification that the selected constituent/fraction can be considered a reasonable worst-case for the Substance as a whole in terms of degradation kinetics.

112 Justification for selection of relevant constituent and/or fractions for the testing, must consider degradation kinetics of constituents of the Substance based, as minimum, on the similarity/differences of the chemical structures and the physico-chemical properties of constituents of the Substance. For that purpose, tools and approaches mentioned in Guidance on IRs and CSA, Sections R.7b and R.11 should be considered.

References

The following documents may have been cited in the decision.

Guidance on information requirements and chemical safety assessment (Guidance on IRs & CSA)

Chapter R.4 Evaluation of available information; ECHA (2011).
Chapter R.6 QSARs, read-across and grouping; ECHA (2008).
Appendix to Chapter R.6 for nanoforms; ECHA (2019).
Chapter R.7a Endpoint specific guidance, Sections R.7.1 – R.7.7; ECHA (2017).
Appendix to Chapter R.7a for nanomaterials; ECHA (2017).
Chapter R.7b Endpoint specific guidance, Sections R.7.8 – R.7.9; ECHA (2017).
Appendix to Chapter R.7b for nanomaterials; ECHA (2017).
Chapter R.7c Endpoint specific guidance, Sections R.7.10 – R.7.13; ECHA (2017).
Appendix to Chapter R.7a for nanomaterials; ECHA (2017).
Appendix R.7.13-2 Environmental risk assessment for metals and metal compounds; ECHA (2008).
Chapter R.11 PBT/vPvB assessment; ECHA (2017).
Chapter R.16 Environmental exposure assessment; ECHA (2016).

Guidance on data-sharing; ECHA (2017).
Guidance for monomers and polymers; ECHA (2012).
Guidance on intermediates; ECHA (2010).
All guidance documents are available online: https://echa.europa.eu/guidance-documents/guidance-on-reach

Read-across assessment framework (RAAF)

RAAF, 2017 Read-across assessment framework (RAAF); ECHA (2017).
RAAF UVCB, 2017 Read-across assessment framework (RAAF) – considerations on multi- constituent substances and UVCBs; ECHA (2017).

The RAAF and related documents are available online:
https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across

***OECD Guidance documents* (OECD GDs)**

OECD GD 23 Guidance document on aquatic toxicity testing of difficult substances and mixtures; No. 23 in the OECD series on testing and assessment, OECD (2019).
OECD GD 29 Guidance document on transformation/dissolution of metals and metal compounds in aqueous media; No. 29 in the OECD series on testing and assessment, OECD (2002).
OECD GD 150 Revised guidance document 150 on standardised test guidelines for evaluating chemicals for endocrine disruption; No. 150 in the OECD series on testing and assessment, OECD (2018).
OECD GD 151 Guidance document supporting OECD test guideline 443 on the extended one-generation reproductive toxicity test; No. 151 in the OECD series on testing and assessment, OECD (2013).

Appendix 2: Procedure

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 07 December 2021.

The deadline of the decision is set based on standard practices for carrying out OECD TG tests. It has been exceptionally extended by 6 months from the standard deadline granted by ECHA to take into account currently longer lead times in contract research organisations.

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

ECHA did not receive any comments within the commenting period.

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

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

Appendix 3: Addressees of this decision and their corresponding information requirements

In accordance with Articles 10(a) and 12(1) of REACH, the information requirements for individual registrations are defined as follows:

- 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;

Registrant Name	Registration number	Highest REACH Annex applicable to you
[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.

Appendix 4: Conducting and reporting new tests for REACH purposes

1. Requirements when conducting and reporting new tests for REACH purposes

1.1. 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[2].

(4) Under the introductory part of Annexes VII/VIII/IX/X to REACH, where a test method offers flexibility in the study design, for example in relation to the choice of dose levels or concentrations, the chosen study design must ensure that the data generated are adequate for hazard identification and risk assessment.

1.2. 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.

Selection of the Test material(s)

The Test Material used to generate the new data must be selected taking into account the following:

a) the variation in compositions reported by all members of the joint submission,
b) the boundary composition(s) of the Substance,
c) 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.

Information on the Test Material needed in the updated dossier

a) 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.
b) The reported composition must include the careful identification and description of

[2] https://echa.europa.eu/practical-guides

the characteristics of the Tests Materials in accordance with OECD GLP (ENV/MC/CHEM(98)16) and EU Test Methods Regulation (EU) 440/2008 (Note, Annex), namely all the constituents must be identified as far as possible as well as their concentration. Also any constituents that have harmonised classification and labelling according to the CLP Regulation must be identified and quantified using the appropriate analytical methods.

c) The reported composition must also include other parameters relevant for the property to be tested, in this case the relative abundance of monoamine, diamine and triamine, the distribution of C-chain length, the degree of unsaturation within each of fractions and the relative abundance of branched versus linear polyamines.

With that detailed information, ECHA can confirm 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 (https://echa.europa.eu/manuals).

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.

2. General recommendations for conducting and reporting new tests

2.1. Environmental testing for substances containing multiple constituents

Your Substance contains multiple constituents and, as indicated in Guidance on IRs & CSA, Section R.11.4.2.2, you are advised to consider the following approaches for persistency, bioaccumulation and aquatic toxicity testing:

- the "known constituents approach" (by assessing specific constituents), or
- the "fraction/block approach, (performed on the basis of fractions/blocks of constituents), or
- the "whole substance approach", or
- various combinations of the approaches described above

Selection of the appropriate approach must take into account the possibility to characterise the Substance (i.e. knowledge of its constituents and/or fractions and any differences in their properties) and the possibility to isolate or synthesize its relevant constituents and/or fractions.

References to Guidance on REACH and other supporting documents can be found in Appendix 1.