EUROPEAN CHEMICALS AGENCY

~~Confidential~~

Helsinki, 03 February 2021

Addressees
Registrant(s) of DimerFA_PEPA_PAA as listed in the last Appendix of this decision

Date of submission of the dossier subject to this decision
07/11/2018

Registered substance subject to this decision ("the Substance")
Substance name: Fatty acids, C18-unsatd., dimers, reaction products with polyethylenepolyamines
EC number: 614-452-7
CAS number: 68410-23-1

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 ***10 May 2024***.

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. Water solubility (Annex VII, Section 7.7.; test method: EU A.6./OECD TG 105/OECD GD 29)

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)

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

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

2. If negative results are obtained in test performed for the information requirement of Annex VIII, Section 8.4.2. then: In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.; test method: OECD TG 476 or TG 490)

3. and 4. Combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (OECD TG 422) (Annex VIII, Sections 8.6.1 and 8.7.1.; test method OECD 422) in rats, oral route;

5. Short-term toxicity testing on fish (Annex VIII, Section 9.1.3.; test method: OECD TG 203)

6. Simulation testing on ultimate degradation in surface water (triggered by Annex VIII, Section 9.2.; test method: EU C.25./OECD TG 309) at a temperature of 12 °C

7. Soil simulation testing (triggered by Annex VIII, Section 9.2.; test method: EU C.23./OECD TG 307) at a temperature of 12 °C

8. Sediment simulation testing (triggered by Annex VIII, Section 9.2.; test method: EU C.24./OECD TG 308) at a temperature of 12 °C

9. Identification of degradation products (triggered by Annex VIII, Section 9.2.; test method: using an appropriate test method)

10. Bioaccumulation in aquatic species (triggered by Annex I, Sections 0.6.1. and 4; Annex XIII, Section 2.1.; test method: OECD TG 305)

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

- Appendix entitled "Reasons common to several requests";
- Appendix/Appendices entitled "Reasons to request information required under Annexes VII to VIII 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 Annexes VII and VIII to REACH, for registration at 10-100 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".

The studies relating to biodegradation and bioaccumulation are necessary for the PBT assessment. However, to determine the testing needed to reach the conclusion on the persistency and bioaccumulation of the Substance you should consider the sequence in which these tests are performed and other conditions described in Appendix entitled "Requirements to fulfil when conducting and reporting new tests for REACH purposes".

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 information requirements for the following standard information requirements by grouping substances in the category and applying a read-across approach in accordance with Annex XI, Section 1.5:

- In vitro cytogenicity study in mammalian cells or in vitro micronucleus study (Annex VIII, Section 8.4.2.)
- In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.)
- Short-term repeated dose toxicity (28 day), (Annex VIII, Section 8.6.1.)
- Screening for reproductive/developmental toxicity (Annex VIII, Section 8.7.1.)
- Short-term toxicity testing on aquatic invertebrates (Annex VII, Section 9.1.1.)
- Short-term toxicity testing on fish (Annex VIII, Section 9.1.3.)

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

Grouping of substances and read-across approach

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 (addressed under 'Scope of the grouping'). 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.

A. Scope of the grouping

In your registration dossier you have formed a group (category) of 'Polyamidoamine substances'. You have provided a read-across justification document [redacted] in IUCLID Section 13. In this document you have addressed chemical and structural considerations, toxicokinetics and toxicological properties of the substances. You have also provided a data matrix on physico-chemical and (eco)toxicological properties of the substances.

In the read-across justification document you list the substances below as members of the polyamidoamine group:

[#8 Dimer trimer FA TETA PAA]	Fatty acids, C18-unsatd., dimers, oligomeric reaction products with tail-oil fatty acids and triethylenetriamine, CAS No. 68082-29-1 (EC No. 500-191-5), hereinafter the "source substance [1]";
[#9 Dimer trimer FA TETA PAA]	Reaction products of fatty acid dimers and trimers, C18 (unsaturated) alkyl and fatty acids, C18 (unsaturated)

	alkyl with amines, polyethylenepoly-, triethylenetetramine fraction, CAS No. 68154-62-1 (EC No. 701-120-2), hereinafter the "source substance [2]";
[#11 MonoFA TEPA PAA]	Reaction product of Fatty acids, C18 alkyl with amines, polyethylenepoly-tetraethylenepentamine fraction, CAS No. 103758-98-1 (EC No. 701-046-0) and
[#12 DimerFA PEPA PAA]	Fatty acids, C18-unsatd., dimers, reaction products with polethylenepolyamines, CAS No. 68410-23-1 (EC No. 614-452-7), hereinafter the "Substance"

You have provided the following reasoning for the grouping:

The four polyamidoamines substances are formed as a result of a chemical reaction between: an amine: [redacted] As a result *"The polyamidoamine substances are a mixture of constituents which include* [redacted]*. The substances therefore have common functional groups based on* [redacted] *moieties and are sufficiently similar in terms of chemical structure to support a category read-across approach".*

Further, in Section 1.1. of the justification document you state that as a result of the manufacturing process "[...] *part of the starting amine material does not react and is still present in the final reaction mixture. The unreacted amine is considered to be a constituent of the polyamidoamine substance (no attempt to remove the unreacted amine e.g. by distillation, preparative chromatography etc, is made). The concentration of free, unreacted amine in the polyamidoamine substances is in the range* [redacted] *(wt)".*

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

B. Predictions for properties

a. Prediction for toxicological properties

You have provided the following reasoning for the prediction of toxicological properties: "*The polyamidoamine substances are sufficiently structurally similar to support a category read-across approach, with common functional groups based on* [redacted] *moieties"* (Section 1.6, p. 7.). You consider that the substances have similar physico-chemical properties, toxicokinetic behaviour and toxicological profile. In Section 2.1. of your justification document, you state that *"According to Lipinski's Rule of Five (OECD QSAR Toolbox prediction using a representative structure), the polyamidoamine substances will not be bioavailable and oral absorption and systemic distribution are not predicted".* However, you further acknowledge that *"the polyamidoamine substances also contain some low molecular weight constituents; free or unreacted amines, which may be absorbed".* In order to assess the impact of the free, unreacted amines on the toxicity of the polyamidoamine substances, you assume that the "*toxicity/bioavailability decreases with increasing molecular weight and chain length*". In line with this argument, you state that TETA, being the amine with the shorter chain and the lower molecular weight "*is considered to represent the most conservative toxicity profile when compared to TEPA and PEPA*" and therefore you propose that "[...] *the use of the registered substance Fatty acids, C18-unsatd., dimers, polymers with*

tall-oil fatty acids and triethylenetetramine CAS 68082-29-1 (EC 500-191-5) as the source, represents the conservative profile within the group due to its free TETA content" (Section 2.1., p. 8 of the justification document).

You intend to predict the properties for the category members from information obtained from the substance Fatty acids, C18-unsatd., dimers, polymers with tall-oil fatty acids and triethylenetetramine EC No. 500-191-5 (CAS No. 68082-29-1) as source substance [1].

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 based on a worst-case approach.

Specifically, your read-across hypothesis is based on two arguments. Firstly you assume that the polyamidoamine constituents are not systemically available as a result of their physico-chemical properties and size while smaller constituents such as free unreacted amines may be systemically available. Secondly, you consider that the toxicity of these free, unreacted amines decreases with increasing molecular weight and chain length. On that basis you conclude that the category member containing the amine with the shortest chain and the lowest molecular weight, TETA, presents the most conservative toxicity profile.

Due to the different nature of each hazard property and consequent difference in scientific considerations (e.g. key parameters, biological targets), ECHA has analysed the information you have provided in light of the prediction of *in vitro* (genotoxicity) and *in vivo* (reproductive toxicity) properties of the Substance, using data from the source substance [1]. ECHA has identified the following issues:

(i) Assessment of the prediction for in vitro genotoxicity properties

Read-across hypothesis

According to Annex XI, Section 1.5., two conditions shall be necessarily fulfilled to apply grouping and read-across. 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).

An endpoint-specific 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 substances[2]. 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, as described above, and the supporting information you have provided, focus exclusively on considerations on exposure relating to testing in *in vivo* test systems, i.e. bioavailability in laboratory animals.

However, ensuring exposure of the test systems to the test item is a pre-requisite for deriving meaningful hazard information from experimental data. You have not provided considerations on the potential of the Substance to be absorbed in *in vitro* test systems, such as mammalian somatic cultured cell lines used in the OECD TG 473/487 and OECD TG 476/490.

[2] ECHA *Guidance*, Chapter R.6: QSARs and grouping of chemicals

Your read-across hypothesis does not address the specific test conditions in *in vitro* systems. Therefore ECHA considers that your read-across hypothesis does not constitute a reliable basis to predict the genotoxic properties of the Substance from information on the source substance [1].

(ii) Assessment of the prediction for in vivo properties (reproductive toxicity)

As indicated above your read-across hypothesis is based on 2 arguments and ECHA has identified the issues with each of them as follows.

a. Missing supporting information on the toxicokinetic properties of polyamidoamines

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*"[3]. 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(s).

As indicated above, you consider that the polyamidoamine substances will not be bioavailable after oral administration as a result of their molecular weight and low water solubility. In this context, relevant, reliable and adequate information characterising the toxicokinetic behaviour of the polyamidoamines is necessary to support your claim of no absorption.

You provided a statement based on OECD QSAR Toolbox prediction that based on their high molecular weight and low water solubility "*the polyamidoamine substances will not be bioavailable*".

You did not provide any experimental toxicokinetic data to support your claim that the polyamidoamine substances are not bioavailable under physiological conditions. The assumption that higher molecular weight results in lower bioavailability is an over simplification of the numerous factors which determine bioavailability of a substance. The gastrointestinal tract have active absorption mechanisms for certain types of substances; this includes amides of fatty acids. Given the lipophilic nature of the substances inclusion into micelles and subsequent absorption via the lymphatic system is not excluded.

Based on this information, ECHA considers that you have not established that the polyamidoamine substances are not absorbed.

b. Missing supporting information to substantiate worst-case consideration

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*"[4]. 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(s).

[3] ECHA Guidance R.6: Section R.6.2.2.1.f
[4] ECHA Guidance R.6: Section R.6.2.2.1.f

As indicated above, your read-across hypothesis is based on the assumption that the source substance [1] containing TETA constitutes a worst-case for the prediction of the property under consideration of the Substance. This consideration is established on your claim that the amine with the shortest chain and the lowest molecular weight, TETA, is considered to represent the most conservative toxicity profile.

Relevant, reliable and adequate information allowing to compare the properties of the amines is necessary to confirm a conservative prediction of the properties of the Substance from the data on the source substance(s). Such information can be obtained, for example, from studies of comparable design and duration for the different types of free or unreacted amines included in the composition of the substances.

In the section 2.1 of your justification document you claim that "*the polyamidoamine substance also contain some low molecular weight constituents; free or unreacted amines, which may be absorbed*". You address the impact of exposure to the different types of free, unreacted amines in the substances based on the "*data/read-across argumentation within the appropriate amines and fatty acid registrations*". You stress that "*repeated dose toxicity studies and a read-across approach are available for each of these amines and it is therefore possible to characterise their respective toxicological profile*". You consider that "*it is well known that toxicity/bioavailability decreases with increasing molecular weight and chain length, therefore data on the amine with the shortest chain length, TETA, is considered to represent the most conservative toxicity profile when compared to TEPA and PEPA*".

In order to justify your assumption on the relative toxicity of the free amines included in the composition of the members of this category, you refer to information and to a read-across adaptation argumentation included in registration dossiers of "*the appropriate amines and fatty acid*". However you do not provide information on:

- The identity of these amines and fatty acids;
- The existing repeated dose toxicity studies on these substances in the form of study summaries.

In the absence of such information, you have not established that the toxicity of the free amines decreases with increasing alkyl chain length and molecular weight and that the source substance [1] constitutes a worst-case for the prediction of the property under consideration of the Substance.

c. *Adequacy and reliability of source studies*

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

- be adequate for the purpose of classification and labelling and/or risk assessment;
- have adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3).

Test material identity

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*". Therefore, the unambiguous characterisation of the composition of the test material used to generate the source data is required to assess whether the test material is representative for the source substance as defined in the read-across justification document

and thus relevant to the Substance.

Your read-across justification document contains compositional information for the members of your category in Table 5. This table illustrates that the category members are UVCBs with composition varying in the residual amines (TETA, TEPA or PEPA) and in the amine-fatty acid adducts (low or high MW). However, the information on the composition of the test materials of the source studies provided in your dossier is limited in general to the numerical identifier and it does not contain information on the identity and quantitative occurrence of its constituents (groups of), in particular, no information on the amount of unreacted TETA, associated with your "worst case" hypothesis.

Due to the above deficiency, ECHA concludes that it is not possible to assess whether the test material is representative for the source substance [1] and thus relevant to the Substance. Therefore, the provided for the screening study for reproductive/developmental toxicity for source substance [1], as listed under the relevant information requirement in Appendix B, section 3. below, cannot be considered as adequate for the purpose of classification and labelling and/or risk assessment.

Conclusion for the prediction of toxicological properties
As explained above, you have not established that relevant properties of the Substance can be predicted from data on the source substance [1].

b. Prediction for ecotoxicological properties

You have provided the following reasoning for the prediction of ecotoxicological properties: *"The polyamidoamine substances are sufficiently structurally similar to support a category read-across approach, with common functional groups based on [redacted] moieties"* (Section 1.6, p. 7.). You consider that the category members have similar physico-chemical and ecotoxicological properties. In Section 3.4 of your justification document, you state that *"it is assumed that all four polyamidoamine substances have qualitatively similar toxic potential based on their structural similarity".*

You intend to predict the properties for the Substance from information obtained from the substance Reaction products of fatty acid dimers and trimers, C18 (unsaturated) alkyl and fatty acids, C18 (unsaturated) alkyl with amines, polyethylenepoly-, triethylenetetramine fraction, EC No. 701-120-2 (CAS No. 68154-62-1) as source substance [2].

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 that with regards to prediction(s) of ecotoxicological properties there are shortcoming(s) that are common to all aquatic information requirements under consideration and also shortcoming(s) that are specific for these information requirements individually. Altogether they result in a failure to meet the requirement of Annex XI, 1.5. The common shortcoming(s) are set out here, while the specific shortcomings are set out under the information requirement concerned in the Appendices below.

a. Read-across hypothesis

According to Annex XI, Section 1.5., two conditions shall be necessarily fulfilled to apply grouping and read-across. 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).

An endpoint-specific 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 substances[5]. 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.

According to the information provided in your dossier, you consider that the properties of the Substance can be predicted from information on other category members as a result of similarities in their chemical structures and in their physico-chemical properties.

While structural and physico-chemical similarity is a prerequisite for applying the grouping and read-across approach, it does not necessarily lead to predictable or similar ecotoxicological properties. You have not provided a well-founded hypothesis to establish a reliable prediction for an ecotoxicological property, based on recognition of the structural and compositional similarities and differences (e.g. differences in free amines and differences in the amine-fatty acids adducts with low molecular weight and with high molecular weight) between the category members.

b. Missing supporting 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 approach. Thus, in addition to the property/endpoint being read-across, it is also useful to show that additional properties, relevant to the endpoint, are also (qualitatively or quantitatively) similar between the source and target chemicals*". [6] 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(s).

Supporting information must include for example bridging studies of comparable design and duration for the Substance and the source substance(s)

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(s) is necessary to confirm that the category members cause the same type of effects.

In the technical dossier you have provided short-term toxicity studies to aquatic invertebrates and to fish for source substance [2], as listed under the relevant information requirements in Appendix A, Section 2; and Appendix B, Section 5. Furthermore, the data matrix included in the read-across justification document reports data for these two endpoints also for source substance [1].

[5] ECHA *Guidance*, Chapter R.6
[6] ECHA Guidance R.6, Section R.6.2.2.1.f

Furthermore, in order to support your claim that your Substance and the category members have similar properties for the endpoints under consideration in the read-across approach, you refer to their toxicity to algae and to microorganism properties.

However, the information you provided cannot be used to support your hypothesis, for the following reasons:

With respect to the data on short-term toxicity to aquatic invertebrates and to fish for source substance [2], the studies provided are considered as not adequate, for the reasons explained under the relevant information requirements in Appendix A, Section 2; and Appendix B, Section 5. The studies on short-term toxicity to aquatic invertebrates and to fish for source substance [1] are not provided in the dossier of the Substance. Furthermore, in the data matrix no data for these two endpoints is available for any other members of the category.

Consequently, since there are no reliable studies for aquatic toxicity across the category, no comparison of toxicity can be made.

With respect to the studies on toxicity to aquatic microorganisms, they do not inform on the aquatic toxicity properties of the target and source substances to fish and aquatic invertebrates. With respect to the studies on algae growth inhibition, while the data matrix reported in the read-across justification document suggests that the category members have similar toxicity to algae, you have not provided any evidence that this information is relevant for the prediction of toxicity to fish and aquatic invertebrates for the Substance (e.g. considering differences in uptake and toxicity among different trophic levels).

Accordingly, this information is not considered as relevant to support prediction of short-term toxicity testing on aquatic invertebrates and short-term toxicity testing on fish.

As explained above, the data set reported in the technical dossier does not include adequate information to support your read-across hypothesis.

In the absence of such information, you have not established that the Substance and the source substance(s) are likely to have similar properties.

c. Adequacy and reliability of source studies

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

- be adequate for the purpose of classification and labelling and/or risk assessment;
- have adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3).

Test material identity

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*". Therefore, the unambiguous characterisation of the composition of the test material used to generate the source data is required to assess whether the test material is representative for the source substance as defined in the read-across justification document and thus relevant to the Substance.

Your read-across justification document contains compositional information for the members of your category in Table 5. This table illustrates that the category members are UVCBs with composition varying in the residual amines (TETA, TEPA or PEPA) and in the amine-fatty acid adducts (low or high MW). However, the information on the composition of the test materials of the source studies provided in your dossier is limited in general to the numerical identifier and it does not contain information on the identity and quantitative occurrence of its constituents (groups of). More specifically, for the test materials you did not report any other information relevant to identify the tested material and concentration levels of the groups of constituents present in the composition of the test material, such as a specific and detailed description of the manufacturing process (including reactants, ratio of starting material, temperature etc).

Due to the above deficiency, ECHA concludes that it is not possible to assess whether the test material is representative for the source substance [2] and thus relevant to the Substance. Therefore, the provided short-term toxicity studies to aquatic invertebrates and to fish for source substance [2], as listed under the relevant information requirements in Appendix A, Section 2; and Appendix B, Section 5 below, cannot be considered as adequate for the purpose of classification and labelling and/or risk assessment.

Further deficiencies

The source studies with source substance [2] on short-term toxicity to aquatic invertebrates and to fish were not performed according to the testing specifications set out in the corresponding OECD TGs. Therefore, the source studies are not adequate for the purpose of classification and labelling and/or risk assessment. The specific reasons are explained further below under the relevant information requirements in Appendix A, Section 2; and Appendix B, Section 5.

For the reasons listed above, the predictions fail.

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 category members [1] and [2]. 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.

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

1. Water solubility

Water solubility is a standard information requirement in Annex VII to REACH.

You have adapted this information requirement by providing
1. a statement indicating that it is not possible to test your substance experimentally,
2. predicted water solubility values, and
3. a value for critical micelle concentration (CMC) conducted on an analogue substance TOFA_DimerFA_TEPA_PAA (EC 500-289-8).

We have assessed this information and identified the following issues:

You have not claimed concrete adaptations. However, from your reasoning ECHA understands that you invoke adaptations of Annex XI, Section 2, Annex XI, Section 1.3. and Annex XI, Section 1.5. of REACH.

According to Annex XI, Section 2, testing may be omitted, if it is technically not possible to conduct the study as a consequence of the properties of the substance, e.g. if the substance is volatile, highly reactive or unstable, reactive in water or radioactive.

You provided the following statement: "*The test substance is a UVCB substance. It is therefore not possible to experimentally determine the water solubility.*"

The mere fact that a substance is a UVCB substance is not a valid adaptation under Annex XI, Section 2. You have not substantiated why an experimental test is not possible for your Substance. Specifically, you have not provided evidence of attempts to determine the water solubility experimentally for your Substance.

In your comments to the draft decision you maintain that it is not technically feasible to determine the water solubility of the Substance due to its properties ("*UVCB, surface activity and hydrophobic properties*"). The UVCB issue has been already explained above. You further state that OECD 105 is only designed to determine the water solubility of pure substances and not of UVCBs, such as your Substance. However, as stated in Guidance R.7a Table R.7.1-5 the flask method, one of the methods available in the OECD 105, is suitable for complex substances, like your Substance. The additional arguments you provided in your comments, Substance being surface active and hydrophobic, are not in isolation valid adaptations under Annex XI, Section 2. You still have not substantiated why an experimental test is not possible for your Substance. Specifically, you have not provided evidence of attempts to determine the water solubility experimentally for your Substance.

Therefore, your adaptation is rejected.

Annex XI, Section 1.3. states that results obtained from valid QSAR models may be used instead of testing when the following cumulative conditions are met, in particular:

- the substance falls within the applicability domain of the QSAR model;
- adequate and reliable documentation of the applied method is provided; and
- the results are adequate for classification and labelling and/or risk assessment.

According to ECHA's Practical guide "How to use and report (Q)SARs", section 3.4, a QSAR Model Reporting Format (QMRF) and a QSAR Prediction Reporting Format (QPRF) are required to verify that the Substance falls within the applicability domain of the model, and to assess

the adequacy of the prediction for the purposes of classification and labelling.

To consider the water solubility information adequate for REACH purposes, among others, the information must cover all the relevant structures of the Substance, especially in the case of a UVCB substance.[7]. Further, to consider the substance to fall within the applicability domain of the model, among others, the model must predict well substances that are similar to the substance[8].

You provided calculated values from EPISuite software for the "representative component", namely linear and cyclic C18 derivative of Dimer FA PEPA PAA. You justify the selection of the representative components based on a) "*It is the main target substance that the reaction was intended to produce*"; and b) "*It was expected to be the major component of the mixture*". You report estimated water solubility values of 5.1 x 10^{-8} and 2.0 x 10-7 mg/l (WSKOW module) and 4.7 and 1.3 x 10-5 mg/l (WATERNT module) for your Substance based on the predictions, respectively, for the linear and cyclic "representative component".
Under "Any other information on results incl. tables", you state that "*Before the EPISuite program was used to estimate the water solubility of the test substance, the validity of the program's calculations was evaluated by comparing the calculated vapour pressure for several related substances with known measured values.*" We understand that in this sentence you mean "*water solubility*" and not "*vapour pressure*". You provide the comparison in Table 1 in the IUCLID dossier. You further state that "*The correlation between the calculated and measured values is considered good and acceptable for the purposes of this report.*"

We have assessed this information and identified the following deficiencies:

A. You have not provided any documentation for the QSAR prediction. In particular, you have not included a QMRF and/or a QPRF in your technical dossier. You have not reported the molecular structure (e.g. SMILES code) used for the prediction. However, based on the information in the technical dossier, ECHA can conclude on the applicability and adequacy of the prediction, as explained below.

B. Regarding the applicability domain of the model, Table 1 in the IUCLID dossier, Section 4.8, shows that the predicted values for related substances and fragments of the Substance are either significantly lower than the corresponding measured water solubility values (for [redacted] or show high solubility of the substances (for DETA and TEPA). You did not justify why you consider this correlation as good. In absence of such justification or further documentation, ECHA considers the structure you used for your prediction outside the applicability domain of the model.

C. You have predicted the water solubility value for a single structure. A single structure, even if fulfilling the requirements under Annex XI, Section 1.3. for a QSAR prediction, is not adequate to cover the water solubility endpoint of a UVCB substance.

In your comments to the draft decision you acknowledge that the QSAR prediction for water solibility provided in the dossier is not adequate. You propose to use the OECD QSAR toolbox to run a prediction based on multiple constituents that you deem most representative of the UVCB Substance. You further propose to conduct QSAR prediction on the chemical moieties with molecular weights below or equal to 600 DA.

It is unclear what models and methods you propose to use for predicting the water solubility.

[7] OECD GD 23, p. 11.
[8] ECHA Guidance R.6

You mention the QSAR Toolbox and "category reports", which indicates that you plan to use the Category approach in the Toolbox. Please note that category approach predictions with analogues are considered read-across, rather than QSAR, and would have to follow the rules specified under Annex XI, section 1.5.

You generically mention QSARs without specifying the models you intend to use. If you plan to use the Episuite WSKOW and WATERNT models please be aware that they are likely to be partly unsuitable for your Substance. The input structures are large, which means that the larger structures would be outside of the parametric and structural domain of both models.

Furthermore, as explained above, the information must cover all the relevant structures of the Substance, especially in the case of a UVCB substance. You propose to provide predictions only for those constituents that you deem are the most representative of the UVCB Substance, i.e. those constituents with molecular weight below or equal 600 DA. However, ECHA notes that the Substance contains several constituents above 600 DA (*"the Substance is a UVCB substance containing at least 16 components ranging in molecular weights from 146.15 to 1417.37 Da")*. Therefore, the selection of constituents you proposed for the QSAR predictions would not cover the whole UVCB Substance.

Since you have not provided in your comments any new predictions addressing the information requirement other than describing your intentions, the data gap remains.

Therefore, your adaptation is rejected.

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 (addressed under 'Scope of the grouping'). 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.

You have provided a CMC value based on a study conducted with another substance than your Substance. You justify the selection of this analogue substance based on slightly higher hydrophilicity of TEPA (tetraethylenepentamine) compared to TETA (triethylenetetramine, present in the Substance), and higher amount of monomeric fatty acids resulting in higher relative amount of adducts with lower MW. ECHA understands that you justify the selection of the source substance to be more water soluble than the Substance, i.e. apply a "worst case scenario".

We have assessed this information and identified the following issues:

Adequacy and reliability of source studies

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

- be adequate for the purpose of classification and labelling and/or risk assessment;
- have adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3)

Specifically, you have provided a CMC value of 60 mg/l (corrected for amine 54 mg/l)

conducted with an analogue substance "TOFA_DimerFA_TEPA_PAA" in a surface tension study. The provided surface tension study indicates surface tension values between ~64 and ~69 mN/m for the analogue substance test material concentrations between 150.7 mg/l and 30.14 mg/l, respectively.

You estimated a value of 40 mg/l for water solubility of your Substance based on the QSAR predictions above and the CMC of an analogue substance.

The experimentally measured surface tension values for the analogue substance are above the limit value of 60 mN/m indicated in the EU Test method A.5 for surface active substances. Therefore, you have not demonstrated that a CMC value is a reliable estimate of the water solubility value for such substances and for your Substance. For these reasons, the surface tension study conducted with the analogue substance is not adequate for predicting the water solubility of the Substance.

Moreover, you have not explained how you concluded on a water solubility value of 40 mg/l based on the CMC value of 54 mg/l and predicted water solubility values of 5.1 x 10-8 and 2.0 x 10-7 mg/l (WSKOW module) and 4.7 and 1.3 x 10-5 mg/l (WATERNT module).

Therefore, your adaptation is rejected.

2. Short-term toxicity testing on aquatic invertebrates

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

You have adapted this information requirement:

i. by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. You have provided in the dossier one short-term toxicity study on aquatic invertebrates (according to OECD TG 202, GLP) with the source substance [2] (CAS No, 68154-62-1).

Furthermore, in your comments to the draft decision you have provided the following information:

ii. an adaptation under Annex VII, Section 9.1.1., Column 2 with the following justification: "aquatic toxicity is unlikely to occur due to very low water solubility and unlikelihood to cross biological membranes".

We have assessed this information and identified the following issue(s):

i. Adaptation based on Annex XI, Section 1.5

A. Predictions

The study listed above was conducted with an analogue substance. As explained in the Appendix on Reasons common to several requests, your adaptation in accordance with Annex XI, Section 1.5. is rejected.

B. Source study is not adequate and reliable

To be adequate for the purpose of classification and labelling and/or risk assessment of the Substance, the source study must be conducted in accordance with the applicable OECD test guidelines or other internationally recognised test methods (Article 13(3) of REACH). For the purpose of classification and labelling, as set out in the CLP Regulation, the study

must provide information on intrinsic properties i.e. the basic properties of a substance or mixture as determined in standard tests or by other means designed to identify hazards. This is to be derived without consideration of exposure under realistic environmental conditions.[9] As a consequence of the above, studies performed with modification to standard tests procedures impacting exposure cannot be considered relevant to derive intrinsic properties.

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 requirements must be met:

- the test medium fulfils the following condition(s): total organic carbon (TOC) ≤ 2 mg/L;
- use of 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, when available; alternatively, a justification why the analytical monitoring of exposure concentrations is not technically feasible must be provided. For this purpose, a sufficiently sensitive analytical method must be used for the analysis of the test chemical in the test solutions or a statement from an analytical chemist must be provided to justify why lower detection limits (LOD) were not feasible (any preliminary analytical efforts should also be described in the report);
- the effect values can only be based on nominal or measured initial concentration if evidence is provided that the concentration of the test material has been satisfactorily maintained within 20 % of the nominal or measured initial concentration throughout the test (see also ECHA Guidance R.7b, Section R.7.8.4.1).

The Substance is UVCB, highly adsorptive (the estimated log Koc ranges from 4.9 to 11.6) and based on structural formula it indicates some surface active properties.

Your registration dossier provides an OECD TG 202 study showing the following:

- The test medium used was natural river water with the following characteristics: DOC of 2.98 mg/L. and suspended matter of 13.4 mg/L was used as the test media. You provide the following justification for the deviation from standard medium: *OLEIC_DimerFA_TETA_PAA has low water solubility and may sorb to organic and inorganic materials by different mechanisms. Due to these properties the test item was difficult to test in artificial water (e.g. sorption to the test organism and walls of the test vessel). Natural river water contains particulate as well as dissolved organic carbon to which the test item can sorb partially preventing that the test item settles onto surfaces. The sorbed fraction of the test item was difficult to extract from the test system which normally leads to low analytical recoveries. Nevertheless the test item was present in the test system and therefore available for exposure (dissolved in water and sorbed).*
- Analytical monitoring of exposure concentrations was not performed. You provide the following justification: *No determination of the test item concentrations was carried out. Preliminary investigations (non GLP) including the use of different analytical techniques showed that no specific method was available that meets the required sensitivity in the range of the test concentrations*. Moreover you summarised the methods you tried to use: the photometric method (limit of quantification of 1.6 mg/L, 230 nm wavelength), the evaporative light scattering detection (ELSD) (limit of detection above 10 mg/L) and the liquid chromatography with electrospray ionisation in positive mode (LC-ESI-MS).
- Effect values were based on nominal concentrations and you have not provided any evidence that the exposure concentrations have been maintained for the test substance during the study period.

[9] CLP Guidance, Section 1.1.3.

The Substance is difficult to test due to the adsorptive and potentially surface active properties as explained above.

Moreover, the study has the following deviations from the requirements of OECD TG 202 and OECD GD 23:

- The study was conducted with non-standard test media (natural water) with TOC above 2 mg/L, hence it does not meet the specifications given in OECD TG 202. The test substances is highly adsorptive and is therefore expected to bind to dissolved organic matter and particulate matter. Since river water differs from standard media with regards to the content of higher organic matter and particulate matter, the use of this modified test medium impacts the exposure to the test substance and decreases bioavailability of the substance. As a consequence the study does not inform on the intrinsic properties of the Substance hence the modification of the test media is not acceptable.

  In your comments to the draft decision you consider that the studies currently in the dossier conducted with modified test media "*show low potential for acute toxicity in a realistic environment and are scientifically acceptable*". You consider the study is sufficient to facilitate hazard identification and risk characterisation but at the same time you acknowledge that for the purpose of classification and labelling, studies must provide information on the intrinsic properties without consideration of exposure under realistic environmental conditions.

  As explained above, studies conducted in natural media cannot be accepted to fulfil this standard information requirement. You have not brought any new argumentation with this comments. Moreover, you do not explain why you consider it fine to submit data that cannot be used to fulfil all of the regulatory requirements identified above in this decision.

- Regarding your justification for not performing any determination of the test item concentrations, it is not acceptable for the following reasons:
  You summarise the three methods you tried to use.
  - Regarding the photometric method (limit of quantification of 1.6 mg/L, 230 nm wavelength), you indicate that it does not guarantee the required sensitivity. ECHA agrees that this method is not sufficiently sensitive, since it is well know that photometric measurements are not suitable to achieve low detection levels as it is required and in addition, it suffers of many interferences and a wavelength of 230 nm does not provide any selectivity.
  - Regarding the evaporative light scattering detection (ELSD) (limit of detection above 10 mg/L), you indicate that it does not guarantee the required sensitivity. ECHA agrees that this method is not sufficiently sensitive, and does not allow determination of the test item concentrations at adequatly low detection level.
  - Regarding the liquid chromatography with electrospray ionisation in positive mode (LC-ESI-MS), you explained that it was not suitable because only the minor fractions were detectable. ECHA notes that the detection method (ESI-MS) can provide sensitivity and selectivity superior to the other 2 other detection methods explained above (i.e. photometric method and ELSD). However, ECHA does not agree with your claim that LC-ESI-MS is unsuitable, since the number of applicable MS techniques with Liquid Chromatography are multiple and very suitable for higher molecular weight above 2000/3000 Daltons as the heavier fractions of your substance. It is also possible to apply different LC-MS strategies to detect and quantify fractions with lower molecular weight and with high molecular weight by multiple analyses of the same sample.

You have not explained nor justified why you have not attempted different strategies to quantify the constituents of your Substance. Therefore, you have not demonstrated that you have applied the most appropriate techniques to ensure that the system is adequately monitored and results are documented.

In your comments to the draft decision you further explain that developing a sufficiently sensitive method is not technically feasible due to the substance properties. You highlight that the Substance is a highly adsorptive UVCB, and based on structural formula it indicates some surface-active properties, making it difficult to analyse and that the attempts to develop the method to monitor the exposure concentrations during the test were not successful. You repeat the statement that "*Preliminary investigations (non GLP) including the use of different analytical techniques showed that no specific method was available that meets the required sensitivity in the range of the test concentrations.*" Finally you again discuss the detection limits of the three methods you tried to use (photometric and evaporative light scattering detection (ELSD) methods and the liquid chromatography with electrospray ionisation in positive mode (LC-ESI-MS)).

The justification provided in the comments is not acceptable, as explained in the following.

Regarding the discussed methods, you state that: "*ECHA has acknowledged that photometric and evaporative light scattering detection (ELSD) methods were not sensitive enough for the low detection levels necessary.*" ECHA notes that it has not excluded any detection technique "a priori" but only acknowledged recognised differences in the detection level among the different detectors: however, a method detection level shall be considered within the entire analytical protocol including extraction, purification, concentration, isolation, detection and quantification to fully judge the applicability and suitability of a method and not only by comparing values of the final detection phase.

Therefore, in your assessment of the suitable analytical methods you are lacking the evaluation covering of all the steps: the extraction, cleaning, detection and quantification protocols. Different final detection techniques could be used on different aliquots of the same sample to handle detection issues related to the molecular weight and structures of different fractions of the Subtance.

In addition, your comment that "*liquid chromatography with electrospray ionisation in positive mode (LC-ESI-MS) was not suitable because only the minor fractions were detectable*" is unclear as the number of applicable MS techniques with Liquid Chromatography are known to be suitable for molecular weight above 2000/3000 Daltons. It is also possible to apply different LC-MS techniques to detect and quantify fractions with lower and higher molecular weights by multiple analyses of the same sample.

In conclusion, the statements provided by you are not acceptable since you do not explain in detail the analytical methodologies investigated and the issues encountered in the various steps, and you do not include the results achieved with specificity, recovery efficiency, precision, limits of determination (i.e. detection and quantification) that would potentially enable you to justify why the analytical monitoring of the exposure concentrations is not technically feasible.

- Since the test substance is highly adsorptive, it is expected that considerable losses will occur during the exposure period. In the absence of analytical monitoring of exposure concentrations, you have not demonstrate that the test substance concentration during the test was maintained within the required 20% of nominal concentrations.

Hence, the source study provided does not meet the conditions listed above and therefore it is not adequate for the purpose of classification and labelling and/or risk assessment.

ii) Column 2 adaptation:

Under Section 9.1.1., Column 2, first indent, Annex VII to REACH, the study may be omitted if aquatic toxicity is unlikely, for instance if the Substance is highly insoluble in water. ECHA Guidance R.7.8.5 explains that there is no scientific basis to define a cut off limit for solubility below which toxicity is unlikely. Therefore, the justification must demonstrate very low water solubility and low likelihood to cross biological membranes. For the latter, the indicators used for low likelihood of a high bioaccumulation potential (ECHA Guidance R.11, Figure R.11-4) must be considered, including:

- physico-chemical indicators of hindered uptake, and
- supporting experimental evidence of hindered uptake (no chronic toxicity for mammals and birds, no chronic ecotoxicity, no uptake in mammalian toxicokinetic studies, very low uptake after chronic exposure).

To conclude, provided justification cannot rely solely on physico-chemical properties of the substance and must be further supported by lack of adsorption and effect in toxicological/ecotoxicological studies. Unless it can reliably be demonstrated that aquatic toxicity is unlikely to occur, the Substance must be considered as poorly water soluble.

In your comments to the draft decision you provide:

- a conclusion of low likelihood to cross biological membranes substantiated with the following physico-chemical indicators: "the predicted Koc values vary from 6.5 to 8.6"

Your registration dossier provides:

- an estimated value of 40 mg/l for water solubility of your Substance based on the QSAR predictions and the CMC of an analogue substance (addressed under Section A.1);
- short-term toxicity studies in aquatic invertebrates and fish and algae growth inhibition study.

However, there is currently no reliable information on water solubility of the Substance (Appendix A.1). Moreover, regarding unlikelihood to cross biological membranes as explained above physico-chemical parameters (log Koc) value on its own cannot be used to assess this property. Furthermore, even if the aquatic toxicity studies provided in the dossier are not reliable (as addressed under Sections A.2, A.3 and B.5), effects are observed indicating systemic exposure. Therefore, you have not demonstrated that toxicity is unlikely to occur and your adaptation is rejected.

In your comments to the draft decision, you agree to conduct a new study with the Substance if the information provided in the comments would not address the deficiencies of the current study.

As explained above, the information provided in the dossier and in the comments to the draft decision is not acceptable. Hence, the data gap remains.

You stated in your comments that if the test needs to be repeated, you 1) propose to conduct a further OECD 202 test using (i) synthetic test media and (ii) semi-static test design with 24h renewal of test media and without analytical determination and 2) you indicate that "the Registrants would need to know which substance should be selected by the Registrants and would possibly be accepted by ECHA as Test Material".

We have assessed your comment and identified the following issue(s):

1) 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. Therefore, the following requirements must be met (among others):
   - the test medium fulfils the following condition(s): particulate matter ≤ 20 mg/L, total organic carbon (TOC) ≤ 2 mg/L, hardness between 140 and 250 mg/L (as $CaCO_3$), pH between 6 and 9;
   - the selection of the exposure system to be used in a tests should be guided by the time course of the experiment, the study design, the test species used, the characteristics (e.g. physicochemical properties) of the test chemical, and/or the results of a preliminary stability study.
   - 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;
   - due to the properties of a Substance, it may be difficult to achieve and maintain 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.

You have not provided any detailed information on the test conditions listed above. E.g. you have not specified what is a composition of the syntetic test media and therefore it is not possible for ECHA to assess if a new test, as described by you in your comment, will provide information on intrinsic properties of the substance. In addition, it needs to be demonstrated that with the selected route/type of exposure the concentration of the test material is maintained through out the test.

Finally, your comment: "*Based on the registrant's extensive knowledge of the molecules we highlight that the sensitivity of the analytical cannot be improved and therefore if a repeat OECD 202 study is conducted, ECHA should be aware that analytical sensitivity will not be improved*" would need to be substantiated with data and scientifically valid justification.

2) 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). Therefore, the unambiguous characterization of the composition of the test material is required to assess whether the test material is representative for the Substance.

In the general comments you stated that: "*the Substance is a UVCB substance containing at least 18 components ranging in molecular weights from 189.19 to 1753.69 Da. (see detailed Report of Lead registrant). Each individual component varies in unknown concentrations within the registered UVCB substance*". Moreover you state that: "*This significant compositional variation is the reason why analytical confirmation during aquatic testing is not possible, no specific moiety can be used for analytical determination in any of the requested simulation or bioaccumulation tests*". Finally you stated: "*the Registrants would need to know which substance should be selected by the Registrants and would possibly be accepted by ECHA as Test Material*".

As explained above, unambiguous characterization of the composition of a test material is required to assess whether a test material is representative for the Substance.

Moreover, according to the ECHA Guidance R.11, the following assessment approaches could be considered for UVCB substances: "known constituents" approach, "fraction profiling" (or

"block profiling") approach, The whole substance approach, or any combination of these approaches.

If you decide to use the fraction profiling or known constituent approach, as described in ECHA Guidance R.11, in your robust study summary you need to sufficiently justify the reasons for selection of the relevant fraction/constituent and explain why testing of this selected fraction/constituent would be appropriate to fulfil the purposes of the chemical safety assessment (PBT assessment, or risk characterization, classification) of the registered substance as a whole.

According to ECHA Guidance R.11 for the purpose of the PBT/vPvB assessment "Known constituents" approach "*can be applied when a substance is 'a priori' known to contain specific constituents at relevant concentrations, these constituents are suspected based on available information to represent the worst case of the (v)P, (v)B and T properties of all constituents of the substance, and these specific constituents can be isolated or separately manufactured or otherwise acquired for the purpose of testing.*"

The worst-case constituent(s) are to be predicted in regards of the property under consideration (e.g. aquatic toxicity or degradability or bioaccumulation). Intrinsic property is not dependent on the quantity/concentration of the constituent in the fraction or substance. Instead, the selection of the most (suspected) constituent must consider, e.g. the carbon chain length, molecular weight, type of bonds present in the structure, presence of the amine groups, and possible impact of unreacted amines.

We acknowledge that you have recognized at least 18 representative structures. As their variability in concentrations within the Substance is high, it requires further assessment by applying a sound scientific approach. Moreover, in your comments you state that the variability refers to the substance as manufactured in different sites and with a variability of conditions for raw material and manufacturing process. ECHA is not in a position to indicate the representative structures and their concentrations to select for further testing as you, based on your knowledge of your Substance, shall select the representative structure(s) and concentration(s) that will cover the Substance. In addition as indicated in ECHA Guidance R.11, you must report any relevant information justifying your selection.

However once selected the test material should undergo sufficient characterization that would allow establishing the analytical protocol for further testing. Therefore, a thorough characterisation of the test material is a pre-requisite for most information requirements, as Article 13(4) of REACH stipulates "Ecotoxicological and toxicological tests and analyses shall be carried out in compliance with the principles of good laboratory practice".

Therefore, the information requirement is not fulfilled.

Study design

The Substance is difficult to test. OECD TG 202 specifies that for difficult to test substances, the OECD GD 23 is to be followed. To get reliable results, the substance properties need to be considered when performing the test, in particular with regard to the test design; including exposure system, test solution preparation, and sampling. OECD GD 23 (Table 1) describes testing difficulties related to a specific property of the substance. You may use the approaches described in OECD GD 23 or other approaches if more appropriate for your substance. The approach selected must be justified and documented.

Due to the substance properties it may be difficult to achieve and maintain the exposure concentrations. Therefore, you have to demonstrate that the concentration of the substance

is stable throughout the test (i.e. measured concentrations remains within 80-120% of the nominal concentration). If it is not possible to demonstrate the stability, you must express the effect concentration based on measured values as described in the applicable test guideline. In case a dose-response relationship cannot be established (no observed effects), you must demonstrate that the test solution preparation method applied was sufficient to maximise the concentration of the Substance in the test solution. Furthermore, exposure concentrations must be below the critical micelle concentration (CMC). This will ensure that test organisms are exposed to the freely dissolved chemical species and not the micelle which can alter the uptake of the test chemical.

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:
[redacted] 2013, key study, according to OECD TG 201 with the Substance;

We have assessed this information and identified the following issues:

A. To be adequate for the purpose of classification and labelling and/or risk assessment of the Substance, the study must be conducted in accordance with the applicable OECD test guidelines or other internationally recognised test methods (Article 13(3) of REACH). For the purpose of classification and labelling, as set out in the CLP Regulation, the study must provide information on intrinsic properties i.e. the basic properties of a substance or mixture as determined in standard tests or by other means designed to identify hazards. This is to be derived without consideration of exposure under realistic environmental conditions.[1] As a consequence of the above, studies performed with modification to standard tests procedures impacting exposure cannot be considered relevant to derive intrinsic properties.

   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 requirements must be met:

   - one of the two alternative growth media (*i.e.* the OECD or the AAP medium) is used. Any deviations from recommended test media must be described in details and justified in a way that ensures that the objective of the study is reached;
   - use of 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, when available. Alternatively, a justification why the analytical monitoring of exposure concentrations is not technically feasible must be provided; For this purpose, a sufficiently sensitive analytical method must be used for the analysis of the test chemical in the test solutions or a statement from an analytical chemist must be provided to justify why lower detection limits (LOD) were not feasible (any preliminary analytical efforts should also be described in the report);
   - the results can be based on nominal (or measured initial concentration) only if evidence is provided that the concentration of the test material has been maintained within 20 % of the nominal (or measured initial concentration) throughout the test.

The Substance is UVCB, highly adsorptive (the estimated log Koc ranges from 4.9 to 11.6) and based on structural formula it indicates some surface active properties.

[1] CLP Guidance, Section 1.1.3.

Your registration dossier provides an OECD TG 201 study showing the following:

- The test medium was prepared by using 50% of river water and 50% of the OECD medium. The natural river water had the following characteristics: DOC of 2.98 mg/L and suspended matter of 13.4 mg/L. You provide the following justification for the deviation from standard medium: *OLEIC_DimerFA_TETA_PAA has low water solubility and may sorb to organic and inorganic materials by different mechanisms. Due to these properties the test item was difficult to test in artificial water (e.g. sorption to the test organism and walls of the test vessel). Natural river water contains particulate as well as dissolved organic carbon to which the test item can sorb partially preventing that the test item settles onto surfaces. The sorbed fraction of the test item was difficult to extract from the test system which normally leads to low analytical recoveries. Nevertheless the test item was present in the test system and therefore available for exposure (dissolved in water and sorbed).*
- Analytical monitoring of exposure concentrations was not performed. You provide the following justification: *No determination of the test item concentrations was carried out. Preliminary investigations (non GLP) including the use of different analytical techniques showed that no specific method was available that meets the required sensitivity in the range of the test concentrations*. Moreover you summarised the methods you tried to use: the photometric method (limit of quantification of 1.6 mg/L, 230 nm wavelength), the evaporative light scattering (ELS) detection (limit of detection above 10 mg/L) and the liquid chromatography with electrospray ionisation in positive mode (LC-ESI-MS).
- Effect values were based on nominal concentrations and you have not provided any evidence that the exposure concentrations have been maintained for the test substance during the study period.

The Substance is difficult to test due to the adsorptive and potentially surface active properties as explained above.

Moreover, the study has the following deviations from the requirements of OECD TG 201 and OECD GD 23:

- The study was conducted with non-standard test media (natural water) and the provided justification is not acceptable for the reasons explained in detail in Appendix A, Section 2, hence the study does not meet the specifications given in OECD TG 201.

- No analytical monitoring was conducted and the given justification is not accepted. (For detail see explanation in Appendix A, Section 2);

- Since the test substance is highly adsorptive, it is expected that considerable losses will occur during the exposure period. In the absence of analytical monitoring of exposure concentrations, you have not demonstrate that the test substance concentration during the test was maintained within the required 20% of nominal concentrations.

Thus, the requirements listed above are not met and therefore the study is not adequate for the purpose of classification and labelling and/or risk assessment.

B. 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). Therefore, the unambiguous characterization of the composition of the test material is required to assess whether the test material is representative for the Substance.

   The information on the composition of the test materials, provided in your dossier is limited in general to the numerical identifier and it does not contain information on the identity

and quantitative occurrence of its constituents (groups of). More specifically for the test materials you did not report any other information relevant to identify the tested material and concentration levels of the groups of constituents present in the composition of the test material, such as a specific and detailed description of the manufacturing process (including reactants, ratio of starting material, temperature etc).

The test material composition is not sufficiently characterised, since identity and quantitative occurrence of the constituents is not provided. Therefore, this information is not sufficient to demonstrate that the test material is representative for the Substance.

All comments on the request for Growth inhibition study aquatic plants are identical as for request on Short-term toxicity testing on aquatic invertebrates. Therefore they were already addressed under Section A.2 to which ECHA refers to.

On this basis, the information requirement is not fulfilled.

Study design

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' in Appendix A, Section 2.

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

1. *In vitro* cytogenicity study in mammalian cells or *in vitro* micronucleus study

An *in vitro* cytogenicity study in mammalian cells or an *in vitro* micronucleus study is a standard information requirement in Annex VIII to REACH.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. You have provided one *in vitro* cytogenicity study (according to OECD TG 487, GLP) with the source substance, giving negative results.

As explained in the Appendix on Reasons common to several requests, your adaptation in accordance with Annex XI, Section 1.5. is rejected. Therefore, the information requirement is not fulfilled.

In your comments to the draft decision you accept "*that further clarification on the in vitro mutagenicity potential is necessary to fulfil Annex VIII section 8.4.2 requirements under REACH*". You propose to fill this information requirement using *in silico* screening methods, such as OECD TG QSAR Toolbox. Further you claim that "*if unfavourable results come to light through in silico testing, an OECD TG 487 will be commissioned*".

ECHA understands that you intend to explore ways to adapt the information requirement, among which you refer to the use of OECD QSAR Toolbox. As no information on the intended adaptation is available yet, no assessment or conclusions on the compliance of that future adaptation can currently be made. ECHA notes that the nature of the adaptation should be specified and justified accordingly to explain how it fulfils this information requirement.

Therefore, the information requirement is not fulfilled.

To fulfil the information requirement for the Substance, both *in vitro* cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2., test method OECD TG 473) and *in vitro* micronucleus study (Annex VIII, Section 8.4.2., test method OECD TG 487) are considered suitable.

2. In vitro gene mutation study in mammalian cells

An *in vitro* gene mutation study in mammalian cells is a standard information requirement in Annex VIII to REACH in case of a negative result in the *in vitro* gene mutation test in bacteria and the *in vitro* cytogenicity test.

For Annex VIII, 8.4.3., you have not provided any study with the Substance, However, you have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. You have provided one *in vitro* gene mutation in mammalian cells (according to OECD TG 476, GLP) with the source substance, giving negative results.

As explained in the Appendix on Reasons common to several requests, your adaptation in accordance with Annex XI, Section 1.5. is rejected. Therefore, the information requirement is not fulfilled.

Your dossier contains (i) a negative result for *in vitro* gene mutation study in bacteria with the Substance (Annex VII, Section 8.4.1.) and inadequate data for an *in vitro* cytogenicity study in mammalian cells with the source substance (Annex VII, Section 8.4.2.) which is rejected for the reasons provided in Appendix B, Section 1.

In your comments to the draft decision you propose "*to re-evaluate the need of OECD TG 476 testing* based on the results of the *in silico* and potential OECD TG 487 testing".

ECHA reiterates that an *in vitro* gene mutation study in mammalian cells is an information requirement under Annex VIII to REACH (Section 8.4.3.) in case of a negative result in the in vitro gene mutation test in bacteria and the in vitro cytogenicity test.

The result of the request for information in Appendix B, Section 1. will determine whether the present requirement for an *in vitro* mammalian cell gene mutation study in accordance with Annex VIII, Section 8.4.3 is triggered.

To fulfil the information requirement for the Substance, both the *in vitro* mammalian cell gene mutation tests using the hprt and xprt genes (OECD TG 476) and the thymidine kinase gene (OECD TG 490) are considered suitable.

3. Short-term repeated dose toxicity (28 days)

A Short-term repeated dose toxicity study (28 days) is a standard information requirement in Annex VIII to REACH.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. You have provided one screening for reproductive/developmental toxicity study (according to OECD TG 422, GLP) with the source substance.

As explained in the Appendix on Reasons common to several requests, your adaptation in accordance with Annex XI, Section 1.5. is rejected. Therefore, the information requirement is not fulfilled.

When there is no information available neither for the 28-day repeated dose toxicity endpoint (EU B.7, OECD TG 407), nor for the screening study for reproductive/ developmental toxicity (OECD TG 421 or TG 422), the conduct of a combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (OECD TG 422) is preferred to ensure that unnecessary animal testing is avoided. Such an approach offers the possibility to avoid carrying out a 28-day study according to OECD TG 407, because the OECD TG 422 can at the same time fulfil the information requirement of REACH Annex VIII, 8.6.1 and that of REACH Annex VIII, 8.7.1.[10]

Information on the design of the study to be performed (species and route)

ECHA has evaluated the most appropriate route of administration for the study. Based on the information provided in the technical dossier and in the Chemical safety Report, ECHA considers that the oral route is the most appropriate route of administration to investigate repeated dose toxicity, as the Substance is a liquid of very low vapour pressure (0.002 Pa at 25°C) and no uses with spray application are reported that could potentially lead to aerosols of inhalable size. Hence, the test shall be performed by the oral route.

According to test method OECD TG 422, the test is designed for use with rats. On the basis of this default assumption ECHA considers that testing should be performed with rats.

4. Screening for reproductive/developmental toxicity

[10] ECHA Guidance R.7a, Section R.7.6.2.3.2.

A Screening for reproductive/developmental toxicity study (test method: EU B.63/OECD TG 421 or EU B.64/OECD TG 422) is a standard information requirement under Annex VIII to REACH, if there is no evidence from analogue substances, QSAR or *in vitro* methods that the Substance may be a developmental toxicant. There is no information available in your dossier indicating that your Substance may be a developmental toxicant.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. You have provided one screening for reproductive/developmental toxicity study (according to OECD TG 422, GLP) with the source substance.

As explained in the Appendix on Reasons common to several requests**,** your adaptation in accordance with Annex XI, Section 1.5. is rejected. Therefore, the information requirement is not fulfilled.

In your comments to the draft decision you state that you "*will not proceed with this testing until the results of the OECD 408 and OECD 414 studies become available. These tests were previously requested by ECHA via decision TPE-D-2114466078-42-01/F*".

ECHA notes that you are referring to a testing proposal examination for a different substance (EC: 701-046-0) which is not related to this compliance check decision. As far as the testing proposal examinations for the Substance are concerned, ECHA notes that the testing proposal examination procedure has been terminated. The termination process has been communicated to you with a Termination Letter issued on 18 December 2018. Therefore, the tests you are referring to (OECD TG 408 and OECD TG 414) have not been requested by ECHA and no decision is pending.

For the reasons explained above under request 3., the conduct of a combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (OECD TG 422) is preferred to ensure that unnecessary animal testing is avoided.

Information on study design

Similarly as explained above under request 3., a study according to the test method EU B.64/OECD TG 422 must be performed in rats with oral administration of the Substance.

5. Short-term toxicity testing on fish

Short-term toxicity testing on fish is an information requirement under Annex VIII to REACH (Section 9.1.3.).

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. You have provided one short-term toxicity study on fish (according to OECD TG 203, GLP) with the source substance [2] (CAS No., 68154-62-1).

We have assessed this information and identified the following issue(s):

A. Predictions

The study listed above was conducted with an analogue substance. As explained in the Appendix on Reasons common to several requests, your adaptation in accordance with Annex XI, Section 1.5. is rejected.

B. Source study is not adequate and reliable

To be adequate for the purpose of classification and labelling and/or risk assessment of the Substance, the study must be conducted in accordance with the applicable OECD test guidelines or other internationally recognised test methods (Article 13(3) of REACH). For the purpose of classification and labelling, as set out in the CLP Regulation, the study must provide information on intrinsic properties i.e. the basic properties of a substance or mixture as determined in standard tests or by other means designed to identify hazards. This is to be derived without consideration of exposure under realistic environmental conditions.[11] As a consequence of the above, studies performed with modification to standard tests procedures impacting exposure cannot be considered relevant to derive intrinsic properties.

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 requirements must be met:

- the test medium fulfils the following condition(s): particulate matter ≤ 5 mg/L and total organic carbon (TOC) ≤ 2 mg/L;
- use of 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, when available. Alternatively, a justification why the analytical monitoring of exposure concentrations is not technically feasible must be provided; For this purpose, a sufficiently sensitive analytical method must be used for the analysis of the test chemical in the test solutions or a statement from an analytical chemist must be provided to justify why lower detection limits (LOD) were not feasible (any preliminary analytical efforts should also be described in the report);
- the results can be based on nominal (or measured initial concentration) only if evidence is provided that the concentration of the test material has been maintained within 20 % of the nominal (or measured initial concentration) throughout the test.

The Substance is UVCB, highly adsorptive (the estimated log Koc ranges from 4.9 to 11.6) and based on structural formula it indicates some surface active properties.

Your registration dossier provides an OECD TG 203 study showing the following:

- The test medium used was natural river water with the following characteristics: DOC of 2.98 mg/L and suspended matter of 13.4 mg/L. You provide the following justification for the deviation from standard medium: *OLEIC_DimerFA_TETA_PAA has low water solubility and may sorb to organic and inorganic materials by different mechanisms. Due to these properties the test item was difficult to test in artificial water (e.g. sorption to the test organism and walls of the test vessel). Natural river water contains particulate as well as dissolved organic carbon to which the test item can sorb partially preventing that the test item settles onto surfaces. The sorbed fraction of the test item was difficult to extract from the test system which normally leads to low analytical recoveries. Nevertheless the test item was present in the test system and therefore available for exposure (dissolved in water and sorbed).*
- Analytical monitoring of exposure concentrations was not performed. You provide the following justification: *No determination of the test item concentrations was carried out. Preliminary investigations (non GLP) including the use of different analytical techniques showed that no specific method was available that meets the required sensitivity in the range of the test concentrations.* Moreover you summarised the methods you tried to use: the photometric method (limit of quantification of 1.6 mg/L, 230 nm wavelength), the evaporative light scattering (ELS) detection (limit of detection above 10 mg/L) and

[11] CLP Guidance, Section 1.1.3.

the liquid chromatography with electrospray ionisation in positive mode (LC-ESI-MS).
- Effect values were based on nominal concentrations and you have not provided any evidence that the exposure concentrations have been maintained for the test substance during the study period.

The Substance is difficult to test due to the adsorptive and potentially surface active properties as explained above.

Moreover, the study has the following deviations from the requirements of OECD TG 203 and OECD GD 23:
- The study was conducted with non-standard test media (natural water) with TOC above 2 mg/L and particulate matter above 5 mg/L, hence it does not meet the specifications given in OECD TG 203. As explained in detail in Appendix A, Section 2. for highly adsorptive substances, the use of natural water as the test medium impacts the exposure to the test substance, decreases bioavailability of the substance and in consequence does not inform on the intrinsic properties of the substance.

- No analytical monitoring was conducted and the given justification is not accepted. (For detail see explanation in Appendix A, Section 2);

- Since the test substance is highly adsorptive, it is expected that considerable losses will occur during the exposure period. In the absence of analytical monitoring of exposure concentrations, you have not demonstrate that the test substance concentration during the test was maintained within the required 20% of nominal concentrations.

Thus, the requirements listed above are not met and therefore the study is not adequate for the purpose of classification and labelling and/or risk assessment.

In your comments to the draft decision you do not agree to perform the requested study. You argue that algae is the most sensitive trophic level and therefore you believe that repeating the vertebrate test is not in the interest of animal welfare.

We have assessed your comment and identified the following issues.

As specified in the ECHA Guidance R.7b, for PNEC derivation, "if there is compelling evidence, to suggest that the fish value is likely to be at least a factor of about 10 less sensitive than invertebrates or algae there are no further requirements for acute fish testing".

All of the short-term studies reported in your registration dossiers are considered incompliant with the respective REACH standard information requirements as explained under requests A2, A3 and B5 of this decision. There is therefore no reliable evidence to show that a trophic level would be less sensitive than another. Furthermore, your claim is not even supported by the data referred to by you and submitted in your registration dossier since you have reported for algae the 72-h ErC50 value of 4.34 mg/L, for fish and Daphnia you reported a value of 7.07 mg/L (for the 96-h LC50 and 48-h EC50 respectively). These effect values do not differ by a factor of ten.

Your general comments on selection of the test material has been addressed under Section A.2 to which ECHA refers to.

On this basis, the information requirement is not fulfilled.

Study design

OECD TG 203 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' in Appendix A, Section 2.

6.-9. Simulation testing on ultimate degradation in surface water, soil simulation testing, sediment simulation testing and identification of degradation products (triggered by Annex VIII, Section 9.2., column 2)

Further degradation testing must be considered if the chemical safety assessment (CSA) according to Annex I indicates the need to investigate further the degradation of the substance (Annex VIII, Section 9.2., column 2).

You have not provided any studies but a claim in your dossier that further degradation studies are not considered necessary *as this substance is considered to be inherently biodegradable and it is likely to degrade quickly in the environment.* '

This information requirement is triggered in case the chemical safety assessment (CSA) indicates the need for further degradation investigation (Annex I, Section 4; Annex XIII, Section 2.1), such as if the substance is a potential PBT/vPvB substance (ECHA Guidance R.11.4). This is the case if the Substance itself or any of its constituent or impurity present in concentration ≥ 0.1% (w/w) or relevant transformation/degradation product meets the following criteria:

- The Substance is potentially persistent or very persistent (P/vP) as it is not readily biodegradable (*i.e.* <60% degradation in OECD TG 301 B/D)
- The Substance is potentially bioaccumulative or very bioaccumulative (B/vB) (e.g. log Kow > 4.5).

Screening information provided in your dossier indicates that the Substance may have PBT/vPvB properties:

- The Substance is potentially P or vP since it is not readily biodegradable (15% degradation in 28 days and 19% in 60 days in the OECD TG 301D test and 0-27% in 28 days and 0-70% in 74 days in the OECD TG 301B test).
- It is not possible to conclude on the B or vB potential because as specified in request in Appendix B, Section 10 below, there is no compliant information on Bioaccumulation and further testing is therefore requested.

Although you claim that the Substance is inherently biodegradable, you have not provided any evidence for that assumption. Based on the screening information, the Substance is a potential PBT/vPvB substance and therefore, the chemical safety assessment (CSA) indicates the need for further investigation on degradation.

The Substance has also a high adsorption coefficient (log Koc >5) and therefore has high potential for adsorption to soil and to sediment. Based on this, soil and sediment represent relevant environmental compartments.

In your comments, to the draft decision you question the applicability of the ready biodegradability tests for complex UVCB substances. You claim among others that "*OECD 301B and 301D reported in your dossier are not appropriate for assessing the biodegradation of multiple molecules at once and historically were designed for assessment of single pure substances.*" You quote also from the ECHA guidance on Application of the CLP that "*the results of biodegradability tests on complex or multi-constituent substances should be carefully evaluated before use for classification purposes is considered*".

We have assessed your comments however note that as the ready biodegradability study was not requested in this decision, the comments do not directly address the request for simulation studies and identification of degradation products included in this decision. Based on evidence provided for the ready biodegradation endpoint, we consider the information in the dossier as sufficient to confirm that the substance is not readily biodegradable and hence, the substance is potentially persistent and further testing is required to assess its persistence based on Annex I, Section 4; Annex XIII, Section 2.1.

In your comments to the draft decision, you do not agree to conduct the requested studies. due to the following reasons:

1) Instead of conducting the studies you "*propose to invoke an adaptation and use a QSAR approach, similar to the Water solubility approach described and conduct a QSAR assessment on identified components with molecular weights ≤600Da.*"
2) You consider that conducting these studies "*would not produce information of scientific merit or appropriate new information for hazard or risk assessment*" due to A) analytical challenges and B) the variable nature of the UVCB Substance.
3) You consider that conducting simulation studies without having reliable information on water solubility (requested under A.1) is unjustified because "*knowledge of the water solubility is a prerequisite for setting up test conditions for a range of fate (e.g. biodegradation, bioaccumulation) and effects studies.*"

We have assessed this information and identified the following issues.

1) Regarding your proposal to provide QSAR predictions for these endpoints

Under ECHA Guidance R.6.1.7.3. a prediction is adequate for the purpose of classification and labelling and/or risk assessment if the following cumulative conditions is/are met:
- the composition of the substance is clearly defined, and
- representative structure(s) for the assessment are selected.

In addition the following cumulative conditions is/are to be met:
- the composition of the substance is clearly defined, and
- different constituents of the same substance are predicted individually.

As indicated in ECHA Guidance R.11. the use of QSAR predictions for identifying substances for persistence (P and vP) might be used at the screening level. However, QSAR results alone are in most cases not sufficient to conclude on non-persistence but should be supported by additional information. In every case, it should be verified that the QSAR model and predictions are reliable and applicable to the Substance. QSAR predictions can be used as part of a Weight-of-Evidence approach.
From your comments it cannot be concluded that the selection of constituents proposed for the QSAR prediction for persistence would be representative of the whole UVCB, as it is described in the paragraph above.

Moreover, it is not clear which QSAR model(s) you intend to use and if you also intend to provide predictions for identification of degradation products.

Currently, we do not know of any sufficiently reliable models to predict persistence in surface water, soil or sediment. QSAR models for ready biodegradation, such as Biowin, can be used at the screening stage of the PBT assessment and CATALOGIC can also be used for ready biodegradation and mapping of degradation products, given that the predictions are reliable (within the applicability domain of the models) and adequate for the purpose.

In consequence your proposal of using QSAR approach for simulation studies and identification of degradation products is not considered as valid.

2) Regarding your statement that performing the requested studies "would not produce information of scientific merit or appropriate new information for hazard or risk assessment", ECHA notes the following.

2A) Regarding your comments on analytical challenges

You state in your comments that "*the identification of degradation products would be impossible due to a lack of single representative chemical structure and no common structures to facilitate the radiolabelling of all structure within the UVCB*". You explain why NMR and mass spectrometry would not be suitable methods for the identification of the degradation products of the Substance.

In the comments you also state that "*the variable nature of the components in a UVCB also mean it would be difficult to identify if individual components had undergone degradation, or if they are simply a part of the UVCB itself*" and that "*The results of a simulation study would have no scientific value as the test would only be relevant to the batch of UVCB tested at the point in time when it was tested. Due to the variable nature a second study on a different batch could yield very different results. The information which could be gained about the degredates from these studies will be limited by the nature of the test material, in particular it's complex constituents, lack of UV chromophore and low molecular weight constituents would require a non-labelled method of analysis which will have the same complications of analytical sensitivity as those identified in previous aquatic testing*".

Finally, you submit a statement from a contract research laboratory explaining the analytical challenges of complex UVCBs, such as the Substance.

We have assessed your comments and identified the following issues.

To fulfil the information requirement, a study must comply with the OECD TGs: 309, 308, 307 (Article 13(3) of REACH). Therefore, the following requirements must be met (among others):

- the analytical method used for the quantification of the test material and its transformation/degradation products needs to be described. The recovery efficiency, precision, limits of determination (i.e. detection and quantification) and working range need to be reported;

The contract research laboratory in their statement indicates that "*given the complexity of the UVCB, radiolabelling the commercial product is not a feasible option and there is no obvious representative candidate molecule for radiolabelling.*" Such situation is quite common for UVCB substances and a selected test material shall be identified and prepared prior to the testing. Please note, the laboratory statement is often applicable also for well-defined substances as the tested material shall be appropriately selected, well known, prepared and controlled.

The contract research laboratory also indicates that the selection of the representative candidate molecules for radiolabelling must be carefully performed. You are expected to perform this selection by the use of you knowledge of the Substance and of its molecular structures.

We acknowledge that studies on the rate of degradation conducted with non-radiolabelled test material present more limitations on the information that can be obtained, since with

radiolabelled test material it is possible to accurately measure multiple parameters such as mineralisation and the amount of unextractable material (therefore to calculate carefully the mass balance). With the use of radiolabelled test material it is possible also to quantify and identify degradation products.

Despite the relevance of the use of radiolabelling and the more efficient monitoring of degradation product it appears that you have decided to only consider analytical methods applied to not radiolabelled test material and you concluded that "*due to the complex nature of the UVCBs and lack of test design appropriate for measuring the degradation of UVCB/mixtures. In addition, the identification of degradation products would be impossible due to a lack of single representative chemical structure and no common structures to facilitate the radiolabelling of all structure within the UVCB.*" Please be aware that on the contrary the identification of degradation products can be performed for more than one structures as those structures, within a sound analytical protocol, are known (i.e. the simplicity of the molecular structures within this UVCB substance the components will be ultimately biodegradable and many of the structural moieties are expected to biodegrade quickly), and the degradation are better identified when derived by radiolabelled material.

Regarding your statement related to the presence of a UV chromophore, we would like to highlight that the presence of a UV chromophore is only relevant if a photometric method is used. This is not the case and therefore the lack a UV chromophore is not an issue. It is also important to stress that the comparison with a reference standard shall be always performed with the same analytical detection method as used for the test sample.

In regards to your comment on NMR method, the applicability of this method shall be assessed on the basis of a full method taking into account all phases of the analytical protocol. Moreover there are various available extraction and concentration methods that result in an increase of the detection limit of the substance. Also the statement on mass spectrometry cannot be accepted as the potency of the mass spectroscopy techniques and the number of their varieties make your statement unjustified.

Finally, the information on the degradation products is necessary to better assess the substance properties. Moreover, the lack of a UV chromophore has no impact on the selection of the monitoring analytical methods and it is not clearly explained and justified why the nature of the test material would require a non-labelled method of analyses.

2B) Regarding your comments on the variable nature of the UVCB Substance

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). Therefore, unambiguous characterization of the composition of the test material chosen is required to assess whether the test material is representative for the Substance.

You declare that "*the results of a simulation study would have no scientific value as the test would only be relevant to the batch of UVCB tested at the point in time when it was tested. Due to the variable nature a second study on a different batch could yield very different results*". With this statement you confirm that a thorough characterisation of the test material is a pre-requisite for most information requirements. As indicated above, the unambiguous characterization of the composition of the test material is required to assess whether the test material is representative for the Substance.

Your general comments on selection of the test material has been addressed under Section A.2 to which ECHA refers to.

3) In your comments you claim that "*ECHA have requested further investigation of water solubility and therefore committal of the registrants to higher tier simulation studies at this stage is considered unjustified*".

The timeline (39 months) given to you to perform the tests allows for sequential testing of water solubility and simulation studies (including identification of degradation products) and designing an appropriate testing strategy. As you indicate in your comments, you may start your testing from deriving the water solubility values and then based on that to design an appropriate testing strategy with a possibility of performing simulation studies in a sequence.

Study design
OECD TG 309 is an appropriate method for studying degradation in surface water and OECD TG 307 and 308 are the appropriate methods for studying degradation in soil and sediment. Under Annex XIII, the information must be based on data obtained under conditions relevant for the PBT/vPvB assessment. Therefore:

- You must perform the OECD TG 309 test, by following the pelagic test option with natural surface water containing approximately 15 mg dw/L of suspended solids (acceptable concentration between 10 and 20 mg dw/L) (ECHA Guidance R.11).

- You must perform the OECD TG 307 test using five soils representing a range of relevant soils (*i.e.* varying in their organic content, pH, clay content and microbial biomass).

- You must perform the OECD TG 308 test using two sediments. One sediment should have a high organic carbon content (2.5-7.5%) and a fine texture, the other sediment should have a low organic carbon content (0.5-2.5%) and a coarse texture.

- You must perform the tests at the temperature of 12°C, the average environmental temperature for the EU (ECHA Guidance R.16, Table R.16-8). Performing the tests at this temperature is in line with the applicable test conditions of the OECD TG 307.

Non-extractable residues (NER) must be quantified in all simulation studies. The reporting of results must include a scientific justification of the used extraction procedures and solvents. By default, total NER is regarded as non-degraded Substance. However, if reasonably justified and analytically demonstrated a certain part of NER may be differentiated and quantified as irreversibly bound or as degraded to biogenic NER. Such fractions could be regarded as removed when calculating the degradation half-life(s) (ECHA Guidance R.11).

For the information on degradation products you must obtain this information while performing the simulation studies. You must provide a scientifically valid justification for any other method you have used for identification of the transformation/degradation products. Identity, stability, behaviour, and molar quantity of the degradation/ transformation products relative to the Substance must be evaluated and reported, when analytically possible. In addition, degradation half-life, potential for bioaccumulation and toxicity of the transformation/degradation product must be investigated.

10. Bioaccumulation in aquatic species

Bioaccumulation in aquatic species is required for the purpose of PBT/vPvB assessment (Annex I, Sections 0.6.1 and 4 to REACH).

You have sought to adapt this information requirement by using data from Qualitative or quantitative structure-activity relationship (QSAR) in accordance with Annex XI, Section 1.3.

We have assessed this information and identified the following issue:

Triggering for the test
This information requirement is triggered in case the chemical safety assessment (CSA) indicates the need for further investigation on bioaccumulation in aquatic species (Annex I, Section 4; Annex XIII, Section 2.1), such as if the substance is a potential PBT/vPvB substance (ECHA Guidance R.11.4). This is the case if the Substance itself or any of its constituent or impurity present in concentration ≥ 0.1% (w/w) or relevant transformation/degradation product meets the following criteria:

- The Substance is potentially persistent or very persistent (P/vP) as it is not readily biodegradable
- The Substance is potentially bioaccumulative or very bioaccumulative (B/vB) (e.g. log Kow > 4.5, calculated BCF >2000 etc);

As indicated under the requests in Appendix B, sections 6-9 the Substance is a potential PBT/vPvB substance and therefore, the chemical safety assessment (CSA) indicates the need for further investigation on bioaccumulation in aquatic species.

Annex XI, Section 1.3. states that results obtained from valid QSAR models may be used instead of testing when the following cumulative conditions are met, in particular:

- the substance falls within the applicability domain of the QSAR model;
- the results are adequate for classification and labelling and/or risk assessment.

To consider the prediction adequate for REACH purposes, among others, predictions must cover all the relevant structures of the Substance. Specifically for UVCB substances, a set of representative structures for each constituent should be identified and subject to prediction (ECHA Guidance R.6, Section R.6.1.7.3). Further, to consider the substance to fall within the applicability domain of the model, among others, the substance must fall within the applicability domain as defined by the model developer.

Moreover for surface active substances, log Kow is not a valid descriptor to predict bioaccumulation potential of the substance (ECHA Guidance R.7c, Appendix R.7.10-3)

Your Substance is a UVCB and based on structural formula it indicates some surface active properties.

In your registration dossier bioaccumulation was calculated as BCF from BCFBAF v 3.01 model of EpiSuite. The BCF calculation was derived:

- based on the constituent's predicted Log Kow value (12.31).

1. The BCFBAF calculations are based on log Kow. As the Substance indicates some surface active properties, the provided QSAR prediction based on log Kow is considered not adequate to reliably predict the bioaccumulation of the Substance.

2. The model developer indicates a maximum log Kow of 11.26 for Estimation Domain (ref BCFBAF help file). The value that you used (log Kow = 12.31) exceeds the maximum value indicated in the "estimation domain" by the model developer. The substance is therefore considered to be outside the applicability domain of the model.

Due to the above, the prediction is considered as not adequate for the purpose of classification and labelling and/or risk assessment.

As a consequence, the adaptation you provided does not fulfil the criteria specified in Annex XI, Section 1.3. and it is therefore rejected.

In your comments to the draft decision you acknowledge that the QSAR prediction for bioaccumulation provided in the dossier is not adequate. However, you do not agree to conduct the requested study due to the following reasons:

1) You propose to provide in an updated dossier new QSAR predictions for bioaccumulation using more suitable tool for predictions due to *"advancements in in silico QSPR technology."* In addition you inform that *"further justification will be provided and the predictions based on multiple constituents of the UVCB that are deemed representative of the substance"*.

2) You state that you *"understand the request is based on uncertainty in the current log Kow, biodegradability and water solubility measurements. However, this uncertainty is not suitable to justify the need for the bioaccumulation study and the committal of significant numbers of vertebrate animals (>400)."* Therefore, you propose *"to invoke further adaptations and create a weight of evidence, by refinement of the log Kow and water solubility values before committing to bioaccumulation testing"*

Moreover you consider that conducting this study will *"not produce information of scientific merit or appropriate new information for hazard or risk assessment"* due to A) analytical challenges and B) the variable nature of the UVCB Substance, since you claim that *"The information which could be gained about bioaccumulation from a study will be limited by the nature of the test material and it's complex constituents, lack of UV chromophore and low molecular weight constituents would require a non-labelled method of analysis which will have the same complications of analytical sensitivity as those identified in previous aquatic testing"*.

3) You consider that conducting bioaccumulation study without having reliable information on water solubility (requested under A.1) is unjustified because *"knowledge of the water solubility is a prerequisite for setting up test conditions for a range of fate (e.g. biodegradation, bioaccumulation) and effects studies."*
We have assessed this information and identified the following issues.

1) Regarding your proposal to provide new QSAR predictions for bioaccumulation

In addition to the cumulative conditions for obtaining valid results from QSAR models mentioned above, the following cumulative conditions are to be also met:

- the composition of the substance is clearly defined, and
- different constituents of the same substance are predicted individually.

From your comments, it seems that you intend to apply the same BCFBAF model for the updated predictions, although this is not fully clear. Please note that the use of BCFBAF calculations and the use QSAR predictions based on logKow are already assessed and addressed above.

Moreover the BCF predictions (and the WSKOW predictions) both in BCFBAF (Episuite) model and Catalogic BCF models are based on logKow. As long as there are reliability issues with the logKOW predictions, there will be issues with the BCF predictions, in both models. Moreover there is a risk that even though you would identify a representative number of constituents, some of them (e.g. [redacted]) would be outside of the applicability domain of at least the logKOW model (but probably also related models).

2) Regarding your comment on the complexity of the test material and the selection of analytical monitoring method, as already explained under sections (B6-B9) the lack of a UV chromophore cannot impact the selection of the monitoring analytical methods as you have decided to exclude pothometric methods. It is also unclear and not justified why the nature of the test material would require a non-labelled method of analyses as such affirmation only derive from a wrong interpretation of the statement of the laboratory.

3) Finally, we consider that the timeline (39 months) given to you to perform the tests allows for sequential testing. As you indicate in your comments, you may start your testing from deriving the water solubility values and then based on that to design an appropriate testing strategy taking into account the study design on the OECD TG 305. Moreover please note that the bioaccumulation of each relevant constituent and degradation products shall be assessed to reach a conclusion of PBT properties (ECHA Guidance R.11). Therefore conducting first the studies requested under B6-B9 and identifying the relevant constituents and/or degradation products would be necessary to decide on appropriate test material for the bioaccumulation study.

Study design

Bioaccumulation in fish: aqueous and dietary exposure (Method EU C.13 / OECD TG 305) is the preferred test to investigate bioaccumulation (ECHA Guidance R.7.10.3.1.). Exposure via the aqueous route (OECD TG 305-I) must be conducted unless it can be demonstrated that:

- a stable and fully dissolved concentration of the test substance in water cannot be maintained within ± 20% of the mean measured value, and/or
- the highest achievable concentration is less than an order of magnitude above the limit of quantification (LoQ) of a sensitive analytical method.

This test set-up is preferred as it allows for a direct comparison with the B and vB criteria of Annex XIII of REACH.

You may only conduct the study using the dietary exposure route (OECD 305-III) if you justify and document that testing through aquatic exposure is not technically possible as indicated above. You must then estimate the corresponding BCF value from the dietary test data according to Annex 8 of the OECD 305 TG and OECD Guidance Document on Aspects of OECD TG 305 on Fish Bioaccumulation (ENV/JM/MONO(2017)16).

Appendix C: 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[12].

B. Test material

1. *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 boundary composition(s) of the Substance,
   b) 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.

2. *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 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,

With that detailed information, ECHA can confirm whether the Test Material is relevant for the Substance.

Technical instructions on how to report the above is available in the manual on How to prepare registration and PPORD dossiers[13].

[12] https://echa.europa.eu/practical-guides
[13] https://echa.europa.eu/manuals

Appendix D: General recommendations when conducting and reporting new tests for REACH purposes

A. Strategy for the PBT/vPvB assessment

You are advised to consult ECHA Guidance R.7b (Section R.7.9.), R.7c (Section R.7.10) and R.11 on PBT assessment to determine the sequence of the tests needed to reach the conclusion on PBT/vPvB. The guidance provides advice on 1) integrated testing strategies (ITS) for the P, B and T assessments and 2) the interpretation of results in concluding whether the Substance fulfils the PBT/vPvB criteria of Annex XIII.

In particular, you are advised to first conclude whether the Substance fulfils the Annex XIII criteria for P and vP, and then continue with the assessment for bioaccumulation. When determining the sequence of simulation degradation testing you are advised to consider the intrinsic properties of the Substance, its identified uses and release patterns as these could significantly influence the environmental fate of the Substance. You must revise your PBT assessment when the new information is available.

B. Environmental testing for substances containing multiple constituents

Your Substance contains multiple constituens and, as indicated in ECHA Guidance R.11 (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 synthetize its relevant constituents and/or fractions.

Appendix E: 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 13/08/2019.

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

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 F: List of references - ECHA Guidance[14] 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)[15]

RAAF - considerations on multiconstituent substances and UVCBs (RAAF UVCB, March 2017)[15]

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.

OECD Guidance documents[16]
Guidance Document on aqueous–phase aquatic toxicity testing of difficult test chemicals – No 23, referred to as OECD GD 23.

[14] https://echa.europa.eu/guidance-documents/guidance-on-information-requirements-and-chemical-safety-assessment
[15] https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across
[16] http://www.oecd.org/chemicalsafety/testing/series-testing-assessment-publications-number.htm

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.

Appendix G: Addressees of this decision and the corresponding information requirements applicable to them

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]

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.