ECHA
EUROPEAN CHEMICALS AGENCY

Helsinki, 23 July 2019

Addressee: [redacted]

Decision number: CCH-D-2114476324-47-01/F
Substance name: Quaternary ammonium compounds, di-C12-18-alkyldimethyl, chlorides
EC number: 269-924-1
CAS number: 68391-05-9
Registration number: [redacted]
Submission number: [redacted]
Submission date: 27/11/2012
Registered tonnage band: Over 1000

DECISION ON A COMPLIANCE CHECK

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

1. **Robust study summary (RSS) for [redacted] 2010) (Annex VII, Section 9.1.2. in conjunction with Annex I, Section 3.1.5)**

   OR

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

2. **Robust study summary (RSS) for [redacted] 2010), Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5. in conjunction with Annex I, Section 3.1.5)**

   OR

   Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5.; test method: Daphnia magna reproduction test, EU C.20./OECD TG 211) with the registered substance;

3. **Long-term toxicity testing on fish (Annex IX, Section 9.1.6.1.; test method: Fish, early-life stage (FELS) toxicity test, OECD TG 210) with the registered substance;**

4. **Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: CO2 evolution test, OECD TG 301B) or**

   Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: MITI test (I), OECD TG 301C) or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: Closed bottle test, OECD TG 301D) or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: Manometric respirometry test, OECD TG 301F) or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: Ready biodegradability – CO2 in sealed vessels (headspace test), OECD TG 310) with the registered substance.

You have to submit the requested information in an updated registration dossier by ***30 July 2020***. You shall also update the chemical safety report, where relevant.

The reasons of this decision are set out in Appendix 1. The procedural history is described in Appendix 2 and advice and further observations are provided in Appendix 3.

Appeal

This decision can be appealed to the Board of Appeal of ECHA within three months of its notification. An appeal, together with the grounds thereof, has to be submitted to ECHA in writing. An appeal has suspensive effect and is subject to a fee. Further details are described under: http://echa.europa.eu/regulations/appeals.

Authorised[1] by **Claudio Carlon**, Head of Unit, 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 1: Reasons

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

ECOTOXICOLOGICAL PROPERTIES

Notes for your consideration:

Once adequate data on the ecotoxicological properties of the registered substance to aquatic organisms are available, you shall revise (if necessary) the chemical safety assessment as necessary according to Annex I of the REACH Regulation.

In case you need to conduct new studies, considering the properties of the substance (low solubility (CMC = 2.3 mg/L) and adsorptive properties (log Kow > 4.5)) you should consult OECD Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures, ENV/JM/MONO (2000)6 and ECHA *Guidance on information requirements and chemical safety assessment* (version 4.0, June 2017), Chapter R7b, Table R.7.8-3 summarising aquatic toxicity testing of difficult substances for choosing the design of the requested ecotoxicity tests) and for calculation and expression of the result of the tests.

In addition, regarding the use of the Water Accommodated Fraction (WAF) approach, please note that the WAF approach is problematic when used with a test substance containing several constituents, as in the case of the registered substance. In such cases the toxicity cannot be allocated to specific constituents directly and interpretation of the results in the risk assessment requires careful consideration taking into account differences in fate of the constituents in the environment. When constituents of varying solubility are present there can be partitioning effects which limit dissolution in the water. These effects should be minimised and appropriate loadings selected accordingly to allow an appropriate determination of the toxicity of the different constituents. In that respect, it is critical that a robust chemical analysis is carried out to identify those constituents present in the water to which the test organisms are exposed. Additionally, chemical analysis to demonstrate attainment of equilibrium in WAF preparation and stability during the conduct of the test is required. Methods capable of identifying gross changes in the composition of WAFs with time are required. Methods such as ultra-violet spectroscopy or total peak area have been used successfully for this purpose. The method used to prepare the WAF should be fully described in the test report and evidence of its compositional stability over time should be provided.

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

"Growth inhibition study in aquatic plants" is a standard information requirement as laid down in Annex VII, Section 9.1.2. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement. Furthermore, pursuant to Article 10(a)(vii) and Annex I, Section 3.1.5. where there is more than one study addressing the same effect, then the study or studies giving rise to the highest concern shall be used to draw a conclusion and a robust study summary shall be prepared for that study or studies and included as part of the technical dossier. Robust study summaries will be required of all key data used in the hazard assessment.

Pursuant to Articles 10(a)(vii) of the REACH Regulation, the information set out in Annexes VII to XI must be provided in the form of a robust study summary, if required under Annex I. Article 3(28) of the REACH Regulation defines a robust study summary as a detailed summary of the objectives, methods, results and conclusions of a full study report providing sufficient information to make an independent assessment of the study minimising the need to consult the full study report. Guidance on the preparation of the robust study summaries is provided in the ECHA Practical Guide 3: 'How to report robust study summaries'.

In the technical dossier you have provided the following study records to fulfil the standard information requirement of Annex VII, Section 9.1.2.:

- Study no. 1: Key study, reliability 1, [redacted] 2010), GLP compliance: yes, test method: equivalent or similar to OECD Guideline 201 (OECD TG 201) with the registered substance;
- Study no. 2: Key study, reliability 1, [redacted] 1998), GLP compliance: yes, test method: equivalent or similar to ISO 10253 with the registered substance.

Regarding Study no. 1 ([redacted], 2010), ECHA notes that you have not provided sufficient information in the technical dossier to allow an independent assessment of the relevance and reliability of the study. In particular:

a. ECHA notes that the test was conducted with river water. OECD TG 201 specifies that two alternative mineral growth media are recommended (i.e. the OECD and AAP medium) and that the use of a modified test medium should be described in details and justified. However, you did not describe how the test medium was prepared (e.g. filtration, removal of the native phytoplankton community, addition of a nutrient stock solution etc.) and you did not provide any justification that this modified test is adequate to fulfil the requirements of the REACH Regulation, including Classification and Labelling.
b. You state that "*At the end of the test 5 random samples were microscopically checked for purity of the algal culture. The 5 random samples checked at the end of the test pure and were not significantly contaminated with bacteria, although due to natural river water being used some bacteria were present*". However, you did not describe in sufficient details how the purity of the phytoplankton cultures was determined. More specifically, depending on how the river water was prepared, the test medium may have been contaminated by picophytoplankton which cannot be detected using a light microscope (epifluorescence should be used instead). If algal biomass was determined using fluorescence, there could potentially have been interference with the quantification of algal biomass during the test;
c. You did not describe how the algal biomass was determined;
d. You note that "*the effect assessment can be based on nominal concentrations*". However, in the 'effect concentrations' table, you specify that effect values were derived based on initial measured test material concentrations. You should thus clarify in your robust study summary how effect values were determined and if nominal concentrations were used you should correct the reported values. ECHA acknowledges that in some cases it may be adequate to derive effect values based on measured concentrations at test initiation. OECD TG 201 states that "*actual exposure concentrations may be difficult to define, especially for adsorbing substances tested at low concentrations. In such cases, disappearance of the test substance from solution by adsorption to the increasing algal biomass does not mean*

that it is lost from the test system. When the result of the test is analysed, it should be checked whether a decrease in concentration of the test substance in the course of the test is accompanied by a decrease in growth inhibition. If this is the case, application of a suitable model describing the decline of the concentration of the test substance may be considered. If not, it may be appropriate to base the analysis of the results on the initial (nominal or measured) concentrations". ECHA notes that based on the raw biomass data reported in your study record, it appears that at 0.19 mg/L (nominal) a marked growth rate inhibition was observed after 24h. However, from 24h onwards, the growth rate reached values similar to control. You provided (limited) analytical monitoring data after test initiation showing that over 75% of nominal concentration is lost in the 0.19 mg/L (nominal) test solution after 72h. Accordingly, it could be assumed that the decrease in concentration of the test substance was accompanied by a decrease of growth inhibition. ECHA notes that contrary to OECD TG 201 recommendations, you did not report using a suitable model describing the decline of the test substance concentration to account for this effect.

e. Finally, you did not provide sufficient information on how test samples were prepared for the quantification of the test substance. Based on the information reported ECHA cannot determine if dissolved test substance concentrations were measured or if the measures also account for test substance bound to dissolved organic matter.

Firstly, in your comments on the draft decision, you provide additional methodological information on the above study. However, this information still does not address fully ECHA's concerns, in particular:

- Regarding point a. above on test medium preparation, you state that the natural water used to prepare the growth medium was not filtered but was frozen until use to preserve the organic content and reduce microbial activity. However, the fact that the water was frozen may not be sufficient to kill all native phytoplankton from the natural river water (see also point b. below). You clarify that the river water was supplemented with standard OECD recommended nutrients (150% the recommended concentrations of NaHCO3 and 50% for all other nutrients). Based on the information provided, ECHA notes that the final test medium had a hardness of c.a. 129 mg/L $CaCO_3$ while the standard OECD medium has a hardness of c.a. 21 mg/L $CaCO_3$. You have not justified that the difference in water hardness did not impact the determination of effect values.
- Regarding point b., you state that the purity of the algal culture was determined microscopically but no specific analysis was carried out to determine the occurrence of picophytoplankton. You consider that picophytoplankton biomass was likely low compared to the inoculum density as the sampled natural water was oligotrophic. However, you do not provide any phytoplankton count data in support of your statement. In addition, in oligotrophic waters phytoplankton communities tend to be dominated by picophytoplankton (i.e. higher relative abundance) and an increase in optical density due to picophytoplankton growth cannot be ruled out unless an appropriate control is added to the study design (e.g. medium containing natural water but no inoculum). However, there is no information that such a control was included in your study.
- Regarding point c., you clarify that phytoplankton growth was monitored using a UV/VIS Spectrophotometer and that absorbance data were then converted to cell numbers using a calibration curve. However, you do not specify what test medium was used to prepare the calibration curve (i.e. fully mineral medium or modified medium containing natural river water) as it may have an impact on the parameters of the equation used to convert optical density into cell numbers. Therefore, your

comments do not fully demonstrate the reliability of the algal density quantification method.

- Regarding point d., you indicate that effect values were not corrected for adsorption to glassware. You intend to correct the calculation in a dossier update. As explained above, the decrease in the concentration of the test substance was accompanied by a decrease of growth inhibition. You must provide a clear description of how this effect was accounted for in the calculation of effect values.
- Regarding point e., you state that "*the samples were diluted with leaching solution 2 and filtered over 0.45 µm Pall GHP acrodisc syringe filter to remove the suspended solids and or algae. At the end of the test after 72h the sorption to glassware in the 0.19 mg/L was analysed to evaluate the sorption to glassware. The concentrations of test substance present in these samples were quantified using LC-MS/MS*". However, you still do not provide sufficient information to demonstrate that the method is adequate to provide a reliable determination of truly dissolved concentrations. First, the substance is a UVCB and you must specify what component(s) of the registered substance was(were) quantified and how gross changes in the composition of the substance in the test water were monitored. You need to provide this information along with a detailed description of the analytical results. Then, you indicate that filtration of suspended matter followed the addition of the leaching solution. Therefore, this implies that the fraction of the substance adsorbed to DOC and/or particulate matter may have been desorbed prior to the filtration. Therefore you must demonstrate that the method provide a reliable determination of truly dissolved concentrations.

Secondly, you claim that the method/study used is adequate to fulfil the information requirement of REACH. You explain that the properties of cationic surfactant have significant impact on exposure under realistic environmental conditions. You first acknowledge that "*the environmental fate of cationic surfactants deviates from that of standard chemicals*" and therefore "*the Koc does not adequately represent the partitioning of cationic surfactants*". You consider that effect values obtained using natural (river) water and expressed as 'bulk concentrations' are relevant for PNEC derivation and you state that "*the bulk approach is* [...] *both conservative and more environmentally realistic than the standard method*". You further state that "[it] *has been accepted by the Technical Meetings (TM's) for the EU risk assessments of* [...] *DODMAC* [...] *and the primary alkyl amines category*". You also consider that these data are adequate for classification and labelling. Finally, you specify that "*for classification and labelling as well as for PBT assessment, the available (bulk) effect data are divided by a factor of 10 (worst case)*"

ECHA disagrees with your conclusion that in general the data generated using the 'bulk approach' are adequate for classification and labelling. In this context ECHA notes the following:

- The CLP Guidance, Section 1.1.3. clarifies that classification must be based on intrinsic hazards, i.e. the basic properties of a substance or mixture as determined in standard tests or by other means designed to identify hazards. As the CLP Regulation is hazard-based, the data on intrinsic properties must not take exposure into consideration. Therefore, the bulk approach which aims at mimicking exposure under "*more environmentally realistic*" conditions must not be used for classification and labelling.
- Similar considerations apply for the PBT assessment. As per Annex XIII of REACH, the PBT assessment should be based on data generated under '*relevant conditions*', i.e. those conditions that allow for an objective assessment of the PBT/vPvB properties of a substance and not the PBT/vPvB properties of a substance in

particular environmental conditions. This has been also confirmed by theBoard of Appeal in its Decision of 7 December 2016 in case A-013-2014;

- The EU RAR reports cited by you did not conclude that classification and labelling can be based on effect values based on 'bulk' concentrations (i) the EU RAR on DODMAC (EC Number 203-508-2) does not discuss the classification of the substance and (ii) the Draft EU RAR on Primary Alkyl Amines has not been endorsed by the European Commission (as clearly specified in the foreword section of the document). On the latter, ECHA points out that adopted RAC Opinions are available on the individual substances originally included in the Draft EU RAR on Primary Alkyl Amines (i.e. EC No. 204-015-5, EC No. 204-695-3, EC No. 262-977-1, EC No. 263-125-1, EC No. 262-976-6). RAC concluded that, for studies conducted with a dilution water containing a high level of suspended matter and humic acid, nominal concentrations do not represent truly dissolved concentrations and that such study has limited usefulness for the purposes of classification.
- You applied a correction factor of 10 to the available (bulk) effect data for classification and labelling and the PBT assessment. You state that this correction factor is conservative as it assumes "*that 90% was sorbed where EUSES calculates that only 7.2% is sorbed using the Koc of C12-18 DAQ*". However, in your comments you also state that "*the Koc does not adequately represent the partitioning of cationic surfactants*". ECHA concludes that there is currently no scientific justification that the proposed correction factor of 10 may be considered as a realistic worst case to correct effect values based on 'bulk' concentrations.

Therefore ECHA concludes that, to be considered valid for classification and labelling and the PBT assessment, you must derive relevant effect values based on reliable estimates of truly dissolved concentrations of the test substance in order to allow an objective assessment of the properties of the substance. In addition, you must clarify the remaining uncertainties related to the study design and methodology detailed above.

Regarding study no. 2, ECHA notes that you have not provided sufficient information in the technical dossier to allow an independent assessment of the relevance and reliability of the study. In particular:

a. In the absence of adequate information on biomass in the control cultures during the test, it is not possible to verify if the validity criteria described in OECD TG 201 have been fulfilled: (i) the mean coefficient of variation for section-by-section specific growth rates (days 0-1, 1-2 and 2-3, for 72-hour tests) in the control cultures must not exceed 35% and (ii) the coefficient of variation of average specific growth rates during the whole test period in replicate control cultures must not exceed 10%;
b. You specified that an analytical monitoring of test substances concentration was conducted. However, you did not report any results on the determination of test concentrations throughout the test. Accordingly, ECHA cannot verify that the exposure was adequately maintained throughout the experiment;
c. You derived the effect concentrations based on nominal test concentrations. However, you report that the test substance contains [redacted] dicocodimethyl ammonium chloride. Accordingly, you did not demonstrate that the test substance falls within the substance identity profile of the registered substance as you did not provide any information on the composition of the remaining [redacted] If it can be demonstrated that the remaining [redacted] does not interfere with the determination of the ecotoxicological properties of the main component, you should derive effect concentrations based on active ingredient concentrations;
d. The test significantly deviates from an OECD TG 201 study as a marine phytoplankton species was used. Accordingly a test medium with high salinity was

used. ECHA notes that the solubility of the test substance might be influenced by salinity. In the marine environment the salinity is so high that the solubility of most substances decreases and precipitation may occur by a process known as salting out. Accordingly, you should provide adequate justification that this study provides relevant information for the purpose of the hazard assessment of the registered substance.

In your comments on the draft decision, you provided further information which leads to the conclusion that this study is of insufficient reliability to be considered as key information:

- Regarding point a., you clarify that the mean coefficient of variation for a section-by-section specific growth rate is not available for this study. Therefore ECHA concludes that the available information on this study do not allow verifying that all validity criteria of OECT TG 201 were fulfilled (i.e., mean coefficient of variation for section-by-section specific growth rates in the control should be < 35%).
- Regarding point b., you clarify that no analytical monitoring was conducted or reported in the study report. Therefore ECHA concludes that exposure cannot be verified.
- Regarding point c., you provide a certificate of analysis for the test material used in this study and you report that the test material composition was [redacted] dicocodimethyl ammonium chloride, [redacted] propan-2-ol and [redacted] water. The observed ErC50 and NOEC values obtained in this study were much lower than in the study by [redacted] (2010) and you consider that the presence of propan-2-ol may explain to some extent the higher toxicity of this test material. You also specify that the effect concentration should be corrected for active ingredient by multiplying the effect values with a factor 0.75.
- Regarding point d., you consider that using *Phaeodactylum tricornutum* as test species is acceptable as it "*is a recommended species in the 1993 PARCOM protocol for algae growth inhibition (ENV/MC/CHEM(98)19/PART2) and in OECD review paper on Aquatic Testing Methods for Pesticides and Industrial Chemicals (ENV/MC/CHEM(98)19/PART1)*". However, you must justify that using a marine growth medium had no impact on exposure to the test substance.

You have acknowledged the uncertainties with this study and you specify that you intend to reduce the reliability score of the study to 2 (reliable with restrictions) and that the study will no longer be used as key information.

In view of the above, ECHA has removed the request for a RSS to be provided for this study.

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

In order to allow an independent assessment of the key study ([redacted], 2010) submitted, pursuant to Article 41(1) and (3) of the REACH Regulation you are requested to provide a complete robust study summary with the above missing elements and justifications described above. To summarise you must provide:

- a justification that the difference in water hardness between the modified test water and the standard OECD medium did not impact the determination of effect values;
- further information to justify that the algal biomass quantification method is reliable;
- further information to justify that the analytical monitoring method provided reliable

determination of truly dissolved concentration. Considering that the substance is a UVCB, the analytical determination of exposure level should also allow the identification of any gross changes in the composition of the substance in the test water;
- effect values based on truly dissolved concentrations. The calculation of effect values must also be corrected to account for the reduction of growth inhibition with the decrease in the concentration of the test substance, as explained in paragraph 40 of OECD TG 201.

Alternatively, if you cannot submit this information or if available information indicate that the study is not reliable as per the criteria indicated above and/or not adequate to fulfil the information requirement, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Growth inhibition study aquatic plants (EU C.3./OECD TG 201) with the registered substance.

2. Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5.)

"Long-term toxicity testing on aquatic invertebrates" is a standard information requirement as laid down in Annex IX, Section 9.1.5. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement. Furthermore, pursuant to Article 10(a)(vii) and Annex I, Section 3.1.5. where there is more than one study addressing the same effect, then the study or studies giving rise to the highest concern shall be used to draw a conclusion and a robust study summary shall be prepared for that study or studies and included as part of the technical dossier. Robust study summaries will be required of all key data used in the hazard assessment.

Pursuant to Articles 10(a)(vii) of the REACH Regulation, the information set out in Annexes VII to XI must be provided in the form of a robust study summary, if required under Annex I. Article 3(28) of the REACH Regulation defines a robust study summary as a detailed summary of the objectives, methods, results and conclusions of a full study report providing sufficient information to make an independent assessment of the study minimising the need to consult the full study report. Guidance on the preparation of the robust study summaries is provided in the ECHA Practical Guide 3: 'How to report robust study summaries'.

In the technical dossier you have provided the following study record to fulfil the standard information requirement of Annex IX, Section 9.1.X.: Key study, reliability 1, [redacted] 2010), GLP compliance: yes, test method: equivalent or similar to OECD Guideline 211 (OECD TG 211) with the registered substance.

ECHA notes that you have not provided sufficient information in the technical dossier to allow an independent assessment of the reliability of the study. In particular:

a. The study was conducted using river water. You specified that the test water originated from the [redacted] river and that the NPOC and TSS was 2.348 mg/L and 18.2 mg/L, respectively. OECD TG 211 recommends that "*a fully defined medium be used in this test. [...] It is further recommended that TOC levels in the medium (i.e. before addition of the algae) be below 2 mg/l*". Based on the information reported ECHA notes that the TOC content of the test water is above 2

mg/L and you should justify why you consider that the test medium is appropriate for this test;

b. Based on the information you reported, it is unclear if the analytical monitoring method allows measuring truly dissolved test item concentrations. You indicate that test samples were diluted in the leaching solution used for the LC-MS/MS (but you did not clearly specify the composition of this solution) and then filtered through 0.45 µm filter. ECHA notes that depending on the affinity to leaching solution, the test item may be desorbed from dissolved and/or particulate organic water, which would lead to an overestimation of true exposure concentrations. Accordingly, you should provide a clear description of the method and a justification of its adequacy to reliably estimate exposure levels.

In your comments on the draft decision, you state the following:

- Regarding point a., you state that "*river water typically has a higher organic content and total suspended solids (TSS) and cannot be compared to standard medium as these studies have been performed according to the bulk approach ECETOC TR 88 (2003) where the presence of DOC and suspended matter is considered acceptable*". ECHA notes that you justify the use of a modified test medium because the study was conducted based on the bulk approach. Therefore, as already explained in request 1, such a test is not considered as adequate for classification and labelling unless a reliable analytical determination of truly dissolved test substance concentrations is available.
- Regarding point b., you specify that "*the main component, C12 DAQ (representing ±50%), was analysed during the experiment [using HPLC with mass spectrometry (LC-MS/MS)] and the results of this component were treated as representative for the test substance".* You consider that it is "*justified to use the bulk test concentrations which are in this case equal to the nominal concentration to derive the effect values*". As already explained under request 1, you need to provide sufficient information to demonstrate that the method is adequate to provide a reliable determination of truly dissolved concentrations for this particular substance. First, the substance is a UVCB and you did not specify how gross changes in the composition of the substance in the test water were monitored. You must provide this information along with a detailed description of the results. As already explained, filtration of suspended matter followed the addition of the leaching solution. Therefore, this implies that the fraction of the substance adsorbed to DOC and/or particulate matter may have been desorbed prior to the filtration and that hence the analysed concentrations may not represent truly dissolved concentrations.

Hence, the information currently provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently, there is an information gap and it is necessary to provide information for this endpoint. In order to allow an independent assessment of the study submitted, pursuant to Article 41(1) and (3) of the REACH Regulation you are requested to provide complete robust study summary with the above described missing elements for the study. To summarise, you must provide:

- further information to justify that the analytical monitoring method provided reliable determination of truly dissolved concentration. Considering that the substance is a UVCB, the analytical determination of exposure level should also allow the identification of any gross changes in the composition of the substance in the test water;

effect values based on truly dissolved concentrations.

Alternatively, if you cannot submit this information or if available information indicate that the study is not reliable as per the criteria indicated above and/or not adequate to fulfil the information requirement, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: *Daphnia magna* reproduction test (test method: EU C.20./OECD TG 211).

3. Long-term toxicity testing on fish (Annex IX, Section 9.1.6.1.)

"Long-term toxicity testing on fish" is a standard information requirement as laid down in Annex IX, Section 9.1.6. of the REACH Regulation. Adequate information on Fish, early-life stage (FELS) toxicity test (Annex IX, 9.1.6.1.), or Fish, short-term toxicity test on embryo and sac-fry stages (Annex IX, 9.1.6.2.), or Fish, juvenile growth test (Annex IX, 9.1.6.3.) needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have sought to adapt this information requirement according to Annex IX, Section 9.1.6., column 2 and you provided the following statement: *"For this type of substance algae are the most sensitive species, then Daphnia magna and lowest toxicity was found in fish, when looking to acute toxicit results. Therefore the reproduction toxicity is performed in Daphnia magna. - The substance has a low bioaccumulation potential since an estimated BCF value of 70.79 L/kg wet weight derived. - In accordance with Annex IX column 2 adaptation, testing for long term toxicity to fish of quaternary ammonium compounds, di-C12-18-alkyldimethyl, chlorides is not deemed necessary as it is moderately water soluble".*

ECHA understands that your justification is based on the fact:

a. that you consider fish to be less sensitive than invertebrates based on the reported acute toxicity data;
b. that you consider that the Chemical Safety Assessment does not show the need to generate long-term toxicity data on fish.

However, ECHA notes that the registered substance is surface active UVCB with a critical micelle concentration of [redacted] mg/L and log Kow > 4.5. You did not report specific information on the physicochemical properties of the constituents of the registered substance, however considering the data generated on the whole substance and the wide C-chain length distribution (i.e. from C12 to C18) it is likely that at least some consituents of the registered substance are poorly water soluble[2]. Accordingly, as specified in Annex VIII, Section 9.1.3., column 2, the long-term toxicity test on fish shall be considered. Indeed, poorly soluble and adsorptive substances require longer time to be significantly taken up by the test organisms and so steady state conditions are likely not to be reached within the duration of a short-term toxicity test. For this reason, short-term tests may not give a true measure of toxicity for poorly soluble substances. Therefore, the results of acute toxicity studies cannot be used to demonstrate a species sensitivity difference between aquatic invertebrates and fish.

[2] The concept of "poorly water soluble" is associated with the need to consider long-term tests instead of short-term tests. The ECHA *Guidance on Information Requirements and Chemical Safety Assessment* section R.7.8.5 (Endpoint Specific Guidance R.7.b) suggests that water solubility below 1mg/L or below the detection limit of the analytical method of the tested substance should be used for considering the substance as poorly water soluble and performing the long-term tests instead of the short-term tests.

As specified in ECHA Guidance on information requirements and chemical safety assessment, Chapter R.7b, Section R.7.8.5.3., a risk is indicated by the Chemical Safety Assessment (CSA) for substances with log Kow > 3 and a PEC_{local} or $PEC_{regional}$ > 1/100th of the water solubility. As you did not demonstrate that PEC_{local} or $PEC_{regional}$ is < 1/100th of the water solubility, ECHA concludes that the CSA cannot be used to demonstrate that the risks to the aquatic environment are adequately controlled.

In your comments on the draft decision, you state the following:

- Regarding point a. above, you agree that the results of short-term toxicity studies might not be sufficient to support a sensitivity difference between fish and aquatic invertebrates. However, you consider this hypothesis valid and you provided the following information:
  - a higher NOEC was observed for fish for the read-across substance Dihydrogenated tallow Dimethyl Ammonium Chloride (C16-18 DAQ, EC number 263-090-2);
  - data on a similar substance, dimethyl didecyl ammonium chloride (DDAC, CAS number 7173-51-5) showing a *"Fish chronic 34d NOEC = 0.0322 mg/L, an Algae 96h NOEC = 0.011 mg/L and a Daphnia chronic 21d NOEC = 0.014 mg/L";*
  - a statement that algae is the most sensitive trophic level to the toxicity of quaternary ammonium compounds.

  ECHA Guidance on Information Requirement and Chemical Safety Assessment, Chapter R.7b, Section R.7.8.5.3. (Version 4.0, June 2017) specifies that testing on fish may be omitted only if there is compelling evidence to suggest that the fish value is likely to be at least a factor of 10 less sensitive than invertebrates or algae. You state that the higher NOEC was observed for fish for Dihydrogenated tallow Dimethyl Ammonium Chloride (C16-18 DAQ, EC number 263-090-2). However you did not provide any evidence that it can be concluded that the difference in sensitivity is above a factor of 10. Then, based on the data reported by you on dimethyl didecyl ammonium chloride (DDAC, CAS number 7173-51-5), the difference in sensitivity between fish and algae or invertebrates is less than a factor of 3. Therefore you did not provide compelling evidence to support that the fish value is likely to be at least a factor of 10 less sensitive than invertebrates or algae.
- You also refer to experimental BCF data on DHDTMAC (EC number 263-090-2) and DDAC (CAS number 7173-51-5) and to the biodegradability of quaternary ammonium compounds. You claim that these data support a low risk to the aquatic environment. ECHA notes that this is not a valid adaptation of the information requirement for long-term toxicity on fish.
- Finally regarding point b. above, you state that the exposure scenarios currently included in the Chemical Safety Report (CSR) *"demonstrate that PEClocal and regional are way below <1/100th of the water solubility".*
  However, ECHA notes that PEC estimations are not provided for all exposure scenarios. For instance, no PEC is available for the uses of the substance in plant protection products. For other scenarios, the PEC is estimated based on environmental monitoring data. However, either the sampling methodology is not described or the data were generated based on a single or very few samples. Additionally, the description of the analytical monitoring method is either omitted or lacks critical information (e.g. recovery rate) to assess the reliability of the reported values. Accordingly, ECHA concludes that the CSR currently does not support with sufficient confidence that PEC_{local} or $PEC_{regional}$ is < 1/100th of the water solubility for all identified uses.

ECHA emphasizes that for the reasons explained above the Chemical Safety Assessment cannot be used to adapt the information requirement for long-term toxicity on fish. Therefore your adaptation of the information requirement is rejected.

Additionally, you sought to adapt the information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing (a) study record for a long-term toxicity study to fish (based on US EPA: Methods for acute toxicity tests with fish, macroinvertebrates and amphibians. Ecological Research Service EPA-66013-75-00) with the analogue substance Dihydrogenatedtallow Dimethyl Ammonium Chloride (CAS number 61789-80-8).

ECHA considers that the read-across between the source and target substance is plausible.

However, ECHA notes that, contrary to Article 3(28) of the REACH Regulation the documentation of the study is insufficient and does not allow an independent assessment of the adequacy of the study, their results and use for hazard assessment.

Pursuant to Articles 10(a)(vii) of the REACH Regulation, the information set out in Annexes VII to XI must be provided in the form of a robust study summary, if required under Annex I. Article 3(28) of the REACH Regulation defines a robust study summary as a detailed summary of the objectives, methods, results and conclusions of a full study report providing sufficient information to make an independent assessment of the study minimising the need to consult the full study report. Guidance on the preparation of the robust study summaries is provided in the ECHA Practical Guide 3: 'How to report robust study summaries'.

In particular, the following elements are not reported:

a. detailed description of the test substance (purity, nature and quantification of impurities etc.);
b. test procedure used (e.g. semi-static or flow-through);
c. test design (e.g. number of test chambers and replicates, number of eggs per replicate, material and size of the test chamber (height, width, volume), water volume per test chamber);
d. method of preparation of stock solutions and frequency of renewal (the solubilising agent and its concentration should be given);
e. detailed description of the preparation of test sample for analytical quantification;
f. the recovery efficiency of the method and the nominal test concentrations, the limit of quantification, the means of the measured values and their standard deviations in the test vessels and the method by which these were attained and evidence that the measurements refer to the concentrations of the test chemical in true solution;
g. evidence that controls survival was acceptable;
h. data on mortality at each stage (embryo, larval and juvenile) and cumulative mortality;
i. days to hatch, numbers of larvae hatched each day, and end of hatching;
j. number of healthy fish at end of test;
k. incidence, description and number of morphological abnormalities, if any;
l. incidence, description and number of behavioural effects, if any;
m. approach for the statistical analysis (regression analysis or analysis of the variance) and treatment of data (statistical test or model used).

In your comments on the draft decision, you provided further information which leads ECHA

to the conclusion that this study is not appropriate to fulfil information requirements, for the reasons explained below:

- Regarding point a., you indicate that the substance is reported as "*71.4% active, 8% monoalkyl impurities*". ECHA notes that the test material used to conduct the study is insufficiently defined;
- Regarding point b., you clarified that a flow-through setup was used;
- Regarding point c. and d., you specify that "*In the river water study, the five test concentrations were: 0.040, 0.061, 0.130, 0.230 and 0.45 mg a.i./L (measured conc); In the well water study, the five test concentrations were: 0.006, 0.013, 0.024, 0.053 and 0.090 mg a.i./L (measured conc)*" and that "*For the river water study, 0.59 mg/L methylene blue active substance (MBAS) and triethyleneglycol was used as carrier solvent. For well water study, isopropanol was used as carrier solvent*".
  ECHA notes that the use of a carrier solvent and its impact on the bioavailability of the active substance is not justified. In particular, the use of an anionic surfactant (0.59 mg/L methylene blue active substance) as a co-solvent in the test with River water at a concentration higher than the highest concentration of the active substance itself would likely impact the assessment of the properties of the test material. Furthermore, there is no scientific justification for using different solvents in the study conducted with River and Well water.
- Regarding point e., you specify that no details are reported. You did not provide further information on point f. Without this information ECHA concludes that it cannot be verified if reliable estimates of truly dissolved concentrations were determined.
- Regarding point g., you provided a summary table of the test results supporting that fish survival in the control was acceptable.
- Regarding point h. to m., you specified that no information is reported. ECHA concludes that this study does not fulfil the requirement of Annex XI, Section 1.1.2. as it does not provide an adequate coverage of the key parameters foreseen to be investigated in the corresponding test methods referred to in Article 13(3).

Finally you specify that this study reviewed and cited as part of the EU RAR on DODMAC (EC Number 203-508-2). However, as already explained under request 1., this report does not discuss the adequacy of this study for the purpose of classification and labelling.

To conclude, the additional information provided as part of your comments indicate that key parameters of OECD TG 210 have not been documented in this study. In addition, the information available on this study does not allow verifying the reliability of the determination of dissolved concentrations. Therefore this study does not fulfil the information requirement and ECHA has removed the request for a robust study summary

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Fish, early-life stage (FELS) toxicity test (OECD TG 210).

ENVIRONMENTAL FATE PROPERTIES

4. Ready biodegradability (Annex VII, Section 9.2.1.1.)

"Ready biodegradability" is a standard information requirement as laid down in Annex VII, section 9.2.1.1. of the REACH Regulation. Adequate information on this endpoint needs to

be present in the technical dossier for the registered substance to meet this information requirement.

In the technical dossier you have provided the following study record to fulfil the standard information requirement of Annex VII, Section 9.2.1.1.: Key study, reliability 1, [redacted] (2006), GLP compliance: yes, test method: according to OECD Guideline 301B (OECD TG 301B) with the registered substance. However, this study does not provide the information required by Annex VII, Section 9.2.1.1., because it is not reliable.

In particular, you specified that "*to reduce the toxicity of the test material 20 mg/L humic acid was added to the test item and toxicity control replicates*". As specified in *ECHA Guidance on information requirement and chemical safety assessment*, Chapter R.7b, Section 7.9.4.1. (version 4.0., June 2017), ECHA considers that this deviation from the standard ready biodegradability test design may be considered acceptable to test poorly water soluble substances. However, as specified in Appendix R.7.9-3. of ECHA Guidance R7.b, to be considered acceptable such tests require additional information to be reported in the robust study summary, appropriate additional flask series and the use of a poorly soluble positive control.

More specifically, the study did not include the following flask series:

a. Poorly soluble positive control (either diisooctylphthalate or anthraquinone introduced by direct addition);
b. Poorly soluble positive control using the same choice of introduction as the test substance;
c. Direct addition control;
d. Test substance (introduced by direct addition for conservative assessment).

ECHA further notes that you did not report the method of test substance introduction, the description of pre-treatments (if any), the nominal versus measured carbon concentrations of the stock solution containing the test substance and humic acid including the degree of recovery (if applicable to your test design).

Considering that the design of this study did not include appropriate test controls, ECHA considers that its results are not reliable. Hence, the information provided by this study does not meet the information requirement.

In your comments on the draft decision, you provide the following comments:

- You clarify that "*the reason for using humic acid in the study was to reduce the toxicity of the substance to the inoculum*". You also specify that you do not consider it necessary to include additional control flasks in the test design. You state that cationic surfactants form complexes with humic acid and thus behave differently than the standard control substances diisooctylphthalate or anthraquinone.
  ECHA points out that Annex II of OECD TG 301 specifies that "*if inhibition due to toxicity is to be avoided, it is suggested that the test substance concentrations used in ready biodegradability testing should be less than 1/10 of the EC50 values* [e.g. based on OECD TG 209]" and recommends "*the use of the stringent and sensitive Closed Bottle test or the use of C14-labelled material*". ECHA points out that, in a recent OECD TG 209 study included in your technical dossier, an EC50 of 68 mg/L was determined for the registered substance. ECHA concludes that a ready biodegradability test conducted with OECD TG 301D is technically feasible without

further modification to mitigate inoculum toxicity and that you did not follow the recommendations of OECD TG 301.

- You specify that "*under environmental conditions, cationic surfactants are unlikely to pose a toxicity risk to microorganisms because these compounds will be present in the environment in the µg/L range. The use of linear anionic surfactants, humic acid and silica gel does not lead to a false negative result and thus provide a fair interpretation of the biodegradation potential of the test substance*".
  As already explained under request 1, ECHA emphasizes that the information provided in the technical dossier should be appropriate to determine the classification of the substance in accordance with the criteria in Regulation (EC) No 1272/2008 including any M-factor resulting from the application of Annex 10 of this Regulation. The data should also be adequate for the purpose of the PBT assessment as per Annex XIII of the REACH Regulation. In this context it should allow an objective assessment of the properties of the substance and should not relate to particular environmental conditions.
- You provided additional methodological information on the preparation of the test solutions. You state in your comments that "*A test concentration of 10 mg/L, corresponding to a carbon content of 7.3 mg C/L in the test vessels was selected*".

ECHA notes that the test substance concentration is below the minimum test substance concentration set out in OECD 301B (i.e. 10-20 mg DOC/L). As specified in ECHA Guidance on Information Requirement and Chemical Safety Assessment, Chapter R.7b, section R.7.9.4.1. (Version 4.0 – June 2017) there is already some flexibility with the inoculum and test substance concentrations given in Ready Biodegradability Tests and going beyond the limits defined will change the ratio of substance to inoculum in a way that is deemed to be too favourable.

Based on the above, ECHA maintains that this study does not provide adequate information to fulfil the information requirement for this endpoint.

In addition, you have sought to adapt this information requirement according to Annex XI, Section 1.2., weight of evidence. Hence, ECHA has evaluated your adaptation with respect to this provision. You have not provided an explanation or justification on how the sources of information/studies, which you have provided enable an assumption or conclusion that the registered substance is readily biodegradable.

To support your weight of evidence adaptation you have provided the following sources of information:

1. Weight of evidence: [redacted] 1999), reliability 2, equivalent or similar to OECD 301D with the registered substance, GLP compliance: not specified.
2. Weight of evidence: [redacted] 1990a), reliability 4, equivalent or similar to OECD 301D with the registered substance, GLP compliance: yes.
3. Weight of evidence: [redacted] 1990b), reliability 4, equivalent or similar to OECD 301D with the registered substance, GLP compliance: yes.

You also refer to a report from [redacted] (1974) which you disregarded due to major methodological deficiencies.

An adaptation pursuant to Annex XI, Section 1.2. requires sufficient weight of evidence from several independent sources of information leading to the assumption/conclusion that a substance has or has not a particular dangerous property with respect to the information requirement in question including an adequate and reliable documentation while the information from each single source alone is regarded insufficient to support this notion.

With regard to quality and relevance, ECHA notes that the above studies were all conducted with a test substance containing 14-15% of 2-propanol. ECHA considers that 2-propanol may be subject to biodegradation and thus interfere with the evaluation of the biodegradability of the registered substance. Therefore, those studies specified above do not provide scientific evidence, which can contribute to a weight of evidence adaptation with respect to the information requirement in question.

In your comments on the draft decision, you disagree that the presence of a co-solvent may have impacted the reliability of the results of this study because "*it is* [...] *very unlikely that ethanol and alkylbenzyldimethylammonium salts will be degraded by the same species*".

ECHA notes that the addition of a co-substrate may cause additional uncertainty due to unspecific sum parameters measured in ready biodegradability tests. Therefore, it is not possible to determine the O_2 consumption that can be attributed to the test substance or to the co-solvent in studies based on OECD TG 301D. ECHA concludes that the active substance must be the sole source of carbon added to the medium.

In addition, you have concluded that the registered substance is readily biodegradable. However, ECHA notes that under the conditions of these tests, none of the test substances tested were found to fulfil the criteria for ready biodegradability. Hence those studies specified above do not provide additional support to your conclusion with respect to the information requirement in question.

As explained above, the sources of information you provided (i) do not allow to assume/conclude on the dangerous (hazardous) property of the registered substance with respect to the information requirement for Annex VII, Section 9.2.1.1 and (ii) do not provide additional support to your conclusion that the registered substance should be regarded as readily biodegradable.

Therefore, the general rules for adaptation laid down in Annex XI, Section 1.2. of the REACH Regulation are not met and your adaptation of the information requirement is rejected.

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

Regarding the test method, depending on the substance profile, you may conclude on ready biodegradability, by applying the most appropriate and suitable test guideline among those listed in the ECHA *Guidance on information requirements and chemical safety assessment, Chapter R.7b* (version 4.0, June 2017) and in the paragraph below. The test guidelines include the description of their applicability domain.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to perform one of the following tests with the registered substance subject to the present decision:

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: CO2 evolution test, OECD TG 301B)

or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: MITI test (I), OECD TG 301C)

or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: Closed bottle test, OECD TG 301D)

or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: Manometric respirometry test, OECD TG 301F)

or

Ready biodegradability (Annex VII, Section 9.2.1.1.; test method: Ready biodegradability – CO2 in sealed vessels (headspace test), OECD TG 310).

Appendix 2: Procedural history

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

The compliance check was initiated on 16 October 2017.

The decision making followed the procedure of Articles 50 and 51 of the REACH Regulation, as described below:

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

ECHA took into account your comments and amended the requests.

ECHA notified the draft decision to the competent authorities of the Member States for proposal(s) for amendment.

As no amendments were proposed, ECHA took the decision according to Article 51(3) of the REACH Regulation.

Appendix 3: Further information, observations and technical guidance

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

2. Failure to comply with the requests in this decision, or to otherwise fulfil the information requirements with a valid and documented adaptation, will result in a notification to the enforcement authorities of your Member State.

3. In relation to the information required by the present decision, the sample of the substance used for the new tests must be suitable for use by all the joint registrants. Hence, the sample should have a composition that is suitable to fulfil the information requirement for the range of substance compositions manufactured or imported by the joint registrants.

   It is the responsibility of all joint registrants who manufacture or import the same substance to agree on the appropriate composition of the test material and to document the necessary information on their substance composition. In addition, it is important to ensure that the particular sample of the substance tested in the new tests is appropriate to assess the properties of the registered substance, taking into account any variation in the composition of the technical grade of the substance as actually manufactured or imported by each registrant.

   If the registration of the substance by any registrant covers different grades, the sample used for the new tests must be suitable to assess these grades. Finally there must be adequate information on substance identity for the sample tested and the grades registered to enable the relevance of the tests to be assessed.