ECHA
EUROPEAN CHEMICALS AGENCY

Helsinki, 9 January 2017

Addressee: [redacted]

Decision number: CCH-D-2114349164-51-01/F
Substance name: Turpentine oil
EC number: 932-349-8
CAS number: 8006-64-2
Registration number: [redacted]
Submission number: [redacted]
Submission date: 22.02.2013
Registered tonnage band: More than 1000 tonnes per year

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. **Composition of the substance (Annex VI, Section 2.3.);**

2. **Name(s) in the IUPAC nomenclature or other international chemical name(s) (Annex VI, Section 2.1.1.) of the registered substance;**

3. **In vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.; test method: Bacterial reverse mutation test, EU B.13/14 /OECD TG 471) with the registered substance;**

4. **In vitro cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2, test method: EU B.10/OECD TG 473) or in vitro micronucleus study (Annex VIII, Section 8.4.2, test method: OECD TG 487) with the registered substance;**

5. **In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3; test method: EU B.17/OECD TG 476) with the registered substance provided that the studies requested under 3 and 4 have negative results;**

6. **Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2; test method: EU B.26/OECD TG 408) in rats with the registered substance, modified to include urinalysis and a full histopathological examination which is to include immuno-histochemical investigation of renal pathology to determine if the pathology is mediated by alpha-2u globulin nephropathy;**

7. **Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3; test method: OECD TG 443) in rats, oral route with the registered substance with the following study design:**

   - **Ten weeks premating exposure duration for the parental (P0) generation;**
   - **Dose level setting shall aim to induce some toxicity at the highest dose level;**
   - **Cohort 1A (Reproductive toxicity);**

- **Cohort 1B (Reproductive toxicity) with extension to mate the Cohort 1B animals to produce the F2 generation;**

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

9. **Pre-natal developmental toxicity study (Annex X, Section 8.7.2; test method: EU B.31/OECD TG 414) in a second species (rats or rabbits, depending on the choice of species for the study requested under 8), oral route with the registered substance;**

10. **Ready biodegradability (Annex VII, Section 9.2.1.1; test method: MITI test (I), OECD TG 301C) with the registered substance**

    or

    Ready biodegradability (Annex VII, Section 9.2.1.1; test method: Closed bottle test, OECD TG 301D) with the registered substance

    or

    Ready biodegradability (Annex VII, Section 9.2.1.1; test method: Manometric respirometry test, OECD TG 301F) with the registered substance

11. **Simulation testing on ultimate degradation in surface water (Annex IX, Section 9.2.1.2; test method: Aerobic mineralisation in surface water – simulation biodegradation test, EU C.25/OECD TG 309) at a temperature of 12 °C with the registered substance. The biodegradation of each relevant constituent present in concentration at or above 0.1% (w/w) or, if not technically feasible, in concentrations as low as technically detectable shall be assessed. This can be done simultaneously during the same study;**

12. **Sediment simulation testing (Annex IX, Section 9.2.1.4; test method: Aerobic and anaerobic transformation in aquatic sediment systems, EU C.24 / OECD TG 308) at a temperature of 12 °C with the registered substance The biodegradation of each relevant constituent present in concentration at or above 0.1% (w/w) or, if not technically feasible, in concentrations as low as technically detectable shall be assessed. This can be done simultaneously during the same study;**

13. **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;**

14. **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.**

You are required to submit the requested information in an updated registration dossier by **18 January 2021** except for the information requested under point 6 for a sub-chronic toxicity study (90-day) which shall be submitted in an updated registration dossier by **16 January 2018**. You may only commence the extended one-generation reproductive toxicity study as requested under point 7 after **16 April 2018**, unless an indication to the contrary is communicated to you by ECHA before that date. You shall also update the chemical safety report, where relevant. The timeline has been set to allow for sequential testing.

You may adapt the testing requested above according to the specific rules outlined in Annexes VI to X and/or according to the general rules contained in Annex XI of the REACH Regulation. In order to ensure compliance with the respective information requirement, any such adaptation will need to have a scientific justification, referring and conforming to the appropriate rules in the respective Annex, and an adequate and reliable documentation.

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

Appeal

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

Authorised[1] by Hannu Braunschweiler, Acting Head of Unit, Evaluation E1

[2] 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

IDENTIFICATION OF THE SUBSTANCE

In order to ensure that potential hazardous properties of the substance are not underestimated, the substance identification deficiencies must be resolved before identifying the test sample to be used for the testing requested in the present decision.

1. Composition of the substance (Annex VI, Section 2.3.)

Pursuant to Article 10(a)(ii) of the REACH Regulation, the technical dossier shall contain information on the identity of the substance as specified in Annex VI, Section 2 of the REACH Regulation. In accordance with Annex VI, Section 2 the information provided shall be sufficient to enable the identification of the registered substance.

"Composition of the substance" is an information requirement as laid down in Annex VI, Section 2.3. of the REACH Regulation. The substance composition corresponds to the chemical representation of what the substance consists of and is therefore an essential part of substance identification and the cornerstone of all the REACH obligations. Adequate information needs to be present in the technical dossier for the registered substance to meet this information requirement.

In that respect, according to chapter 4.3 of the Guidance for identification and naming of substances under REACH and CLP (Version: 1.3, February 2014) – referred to as "the Guidance" thereinafter, you should note that for UVCB substances (substances of Unknown, or Variable Composition, or of Biological origin) presenting a large number of constituents, such as the registered substance, the following applies:

- All constituents present in the substance with a concentration of ≥ 10 % shall be identified and reported individually;
- All constituents relevant for the classification and/or PBT assessment of the registered substance shall be identified and reported individually; and
- Other constituents shall be identified by a generic description of their chemical nature.
- The composition shall represent the substance as it is manufactured.

You reported the constituents with their chemical name and numerical identifiers, and concentration ranges. However, ECHA has observed the following deficiencies in the description of the composition of the substance:

(i) The typical concentration of the constituents

The typical concentrations of the constituents were not reported.

You are accordingly requested, pursuant to Article 41(1) and (3) of the REACH Regulation, to revise the information on the composition of the registered substance in order to establish a precise chemical representation of what the substance consists of. In particular you shall include the typical concentration of the constituents reported.

(ii) The concentration ranges

Some of the constituents are reported with a very broad concentration range (i.e. [redacted] % for "[redacted]", [redacted] % for "[redacted]"), which is not justified by the manufacturing process description due to its lack of details. As a consequence, you shall provide an explanation for the high variability in the composition. A detailed description of the manufacturing process may help to clarify the broad concentration ranges reported for the registered substance. You should also revise the concentration ranges, if those provided in the present dossier are not representative for the registered substance as manufactured. In addition, you shall also clarify how the minimum and maximum values for each constituent or group of constituents were obtained. Without this information ECHA is not able to conclude on the representativeness of these values.

In your comments on the draft decision you highlighted that the registered substance may be considered as a multi-constituent substance. ECHA acknowledges the comment and remarks that the status of any constituent as main constituent or impurity shall be considered when revising the composition. However, any update submitted on the composition of the substance will be evaluated by ECHA during the follow-up process.

ECHA notes in this regard that according to chapter 4.2 of the Guidance, for well-defined substances, the following applies:

- Each main constituent (i.e. the constituent present at ≥80% for mono-constituent substance or each constituent present at ≥10% and <80% for multi-constituent substance) shall be identified and reported individually; and
- Each impurity present at ≥1% or relevant for the classification and/or PBT assessment of the registered substance shall be identified and reported individually.

In the event that the present registration dossier covers different compositions of the registered substance you shall report separately the compositional information.

Further technical details on how to report the composition of UVCB substances in IUCLID are available in paragraphs 2.1 and 2.2.2 of the Data Submission Manual – Part 18: How to report the substance identity in IUCLID 5 for registration under REACH (version: 2.0, July 2012) on the ECHA website. Information on how to report several compositions in IUCLID is specified in paragraph 2.3, Q&A8 of that manual.

2. Name(s) in the IUPAC nomenclature or other international chemical name(s) (Annex VI, Section 2.1.1.)

"Name or other identifier of the substance" is an information requirement as laid down in Annex VI, Section 2.1. of the REACH Regulation. The name and other identifiers are used to identify the substance in an unambiguous manner and are therefore fundamental for substance identification. Adequate information needs to be present in the technical dossier for the registered substance to meet this information requirement.

ECHA notes that you identified the registered substance as of Unknown or Variable composition, Complex reaction products or Biological materials (UVCB). The naming of UVCB substances such as the registered substance shall consist of two parts: (i) the chemical name and (ii) a more detailed description of the manufacturing process, as indicated in chapter 4.3 of the Guidance. ECHA observes that you did not provide sufficient information on the manufacturing process, as explained thereinafter.

ECHA notes that the composition of the registered substance reported in section 1.2 of the IUCLID dossier and as addressed above in Section A.1, indicates that it contains two constituents with a significant variation (i.e. [redacted] % and [redacted]%). While in some instances it may be possible for a constituent to have a broad concentration range as an inherent result of the manufacturing process of the substance, the dossier itself does not contain sufficient information about the manufacturing process to conclude whether this is true or whether multiple distinct compositions are obtained. Indeed in the IUCLID dossier you state that: *Exact composition varies with the age, location and species of the softwood* source. A detailed description of the manufacturing process, including the identity of the source and information on the most relevant steps and parameters of the manufacturing process, is therefore required.

In your comments on the draft decision you highlighted that the substance may be considered as a multi-constituent substance. ECHA acknowledges the comment. However any update submitted on the substance identity will be evaluated by ECHA during the follow-up process.

- In case, in line with your comments, you consider your substance as a multi-constituent substance, you are accordingly requested to revise the chemical name assigned to the registered substance. You shall ensure that the chemical name is representative of the specific substance which is the subject of this registration. Based on the information currently contained in the dossier (section 1.4) and on your comments, ECHA invites you to consider if a chemical name such as "[redacted]" would be appropriate for the identification of the registered substance.

  You shall also delete the CAS information currently assigned to the substance and provide instead any available CAS information specifically corresponding to the substance.

  As for the reporting of the information in IUCLID, you shall include the revised information in the reference substance assigned in IUCLID section 1.1.

  You shall note that the registration is currently linked to the list number 932-349-8 which refers to the chemical name "gum turpentine oil". You can however not remove or modify at this stage the EC number for technical reasons, because the registration is linked to that number in REACH-IT. To ensure unambiguous identification of the registered substance and in case the name provided in the registration dossier is not appropriate, you shall indicate, in the "Remarks" field of the reference substance in IUCLID section 1.1, the following: "The list number 932-349-8 currently assigned does not specifically correspond to the registered substance. This identifier cannot be modified or deleted at this stage in the present registration update for technical reasons". You shall also specify, in the same "Remarks" field, any available and appropriate EC or List number for the substance.

  You should note that ECHA has established a process, subject to certain conditions, enabling registrants to adapt the EC identifier of an existing registration, while maintaining the regulatory rights already conferred to the substance concerned.

However, pending the resolution of all the incompliances highlighted in the present decision, the adaptation of the identifier can only be effective once ECHA is at least in a position to establish unambiguously the identity of the substance intended to be covered by the Registrant with this registration. Should the information submitted by the Registrant as a result of the present decision enable ECHA to identify the substance unambiguously, the process of adapting the identifier will be considered as relevant. In that case, ECHA will inform you in due time as to when the identifier adaptation process shall be initiated.

In any case, you should note that the application of the process of adapting the identifier does not affect the obligation to fulfil the requirements specified in this decision.

- In case you still consider your substance as a UVCB, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to provide a more detailed information on the process used for the manufacturing of the registered substance. The manufacturing process description shall include:
    - The identity of the source in terms of composition or alternatively in terms of family, species and other relevant parameters;
    - The parameters of the distillation of oleoresin– pressure and temperature range (boiling point range) shall be specified.

  If the substance covered by the registration is manufactured according to different manufacturing processes, including the use of different sources, steps and/or processing parameters, then the detailed description of the manufacturing process required hereinabove shall be reported separately for each manufacturing process. You shall note that substances manufactured according to different manufacturing processes may indicate multiple substances and consequently the requirement for multiple registrations.

  Regarding how to report the chemical name and description of the UVCB substance, this information shall be included in the in the "IUPAC name" and Description field in IUCLID section 1.1, respectively.

You shall ensure that the information is consistent throughout the dossier. The information shall be sufficient to account for the variability in the composition of the substance (as noted in Section A.1. above).

Further technical details on how to report the identity substances in IUCLID are available in the Manual "How to prepare registration and PPORD dossiers" on the ECHA website.

PROPERTIES OF THE SUBSTANCE

Grouping of substances and read-across approach

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

- In vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.);
- In vitro cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2) 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);
- Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2);
- Extended one-generation reproductive toxicity study (Annex IX/X, Section 8.7.3) (previously two-generation reproductive toxicity study);

- Pre-natal developmental toxicity study (Annex IX, Section 8.7.2) in one species substance;
- Pre-natal developmental toxicity study (Annex X, Section 8.7.2) in a second species;
- Ready biodegradability (Annex VII, Section 9.2.1.1);
- Acute aquatic toxicity (Annex VII, Section 9.1.1 and 9.1.2 and Annex VIII, Section 9.1.3)

by applying a read-across adaptation following REACH Annex XI, Section 1.5.

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

Annex XI, Section 1.5 requires a structural similarity among the substances within a group or category so that relevant properties of a substance within the group can be predicted from the data on reference substance(s) within the group by interpolation. Such prediction for properties need to be based on a similar or regular pattern of these properties as a result of the structural similarity. The following analysis presents your justification for the proposed grouping approach and read-across hypothesis, together with ECHA's analysis concerning the justification in both a generic and an endpoint-specific context.

A. Description of the grouping and read-across approach proposed by the Registrant

Your read-across approach is based on the claimed structural similarity of alpha-pinene, beta-pinene, delta-3-carene and the UVCB substance gum turpentine oil, the registered substance. These substances are claimed to belong to the same chemical-structure category of bicyclic terpene hydrocarbons. Your strategy is to use results obtained in studies with the main constituents of the registered UVCB substance to predict its properties.

You claim that *"following extensive review of the existing data on alpha pinene, beta pinene, (-)-beta pinene, delta 3 carene, camphene and components of gum turpentine oil, a great extent of similar patterns was demonstrated for physico-chemical properties, environmental fate and ecotoxicological and toxicological properties."*

You state: *"...read-across strategy was used to compile the available data on the main components of gum turpentine oil, in order to cover as far as possible the variations in contents of individual components. Read-across strategy was also used to extrapolate results from one compound to another, or to increase the robustness of a weight-of-evidence approach, when it was judged as relevant, according to the endpoint required. When data was lacking, new testing was performed in order to cover the endpoint requirement, when it was both scientifically justified and technically feasible, keeping in mind that animal testing on vertebrate animals for the purposes of REACh shall be undertaken only as a last resort."*

And you continue: *"The use of read-across approach was then primarily based on the similarity of structure of these substances and components."*

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

You have provided a read-across document in the registration file. In this read-across document you provided the following arguments to justify the category read-across approach.

The category consists of:

i. Alpha-pinene (CAS 80-56-8) purity >/= 80 %.
ii. Beta-pinene (CAS 127-91-3) purity >/= 80%
iii. (-)-beta-pinene (CAS 18172-67-3) (specific stereoisomer of . beta pinene purity >/= 80%
iv. Delta-3-carene (CAS 13466-78-9) purity >/= 70%
v. Gum turpentine oil (CAS 8006-64-2) UVCB, registered substance

For the substances number i. – iv., further information on other constituents or impurities is not available in the read-across justification document. The substance identity of the registered substance (number v.) is described in section 1.1 and 1.2 of IUCLID.

In addition, you named two more substances for read-across purposes, but they are not members of the proposed category: Camphene (CAS 79-92-5) and crude sulphate turpentine (CAS 8006-64-2). For crude sulphate turpentine, differing compositions are provided in the different IUCLID endpoint summary sections.

"*The chemical category designated bicyclic terpene hydrocarbons includes four simple bicyclic terpene hydrocarbons and one UVCB composed primarily of alpha and beta pinene and smaller amounts of other chemically identified terpene hydrocarbons. Alpha pinene and beta pinene are bicyclic terpene hydrocarbons and are positional isomers of each other. Delta-3-carene is a bicyclic terpene hydrocarbon with a [4.1.0] skeleton. Camphene is a bicyclic terpene hydrocarbon, with a [2.2.1] carbon skeleton. Camphene is structurally related to beta-pinene in that both are bicyclic C10 hydrocarbons that contain an exocyclic methylene function. Gum turpentine oil in this chemical category is a UVCB composed primarily of alpha and beta pinene. The remaining fraction is accounted for mainly by other terpene bicyclic (camphene and delta 3 carene), monocyclic (limonene, beta phellandrene) and different terpene hydrocarbons.*"

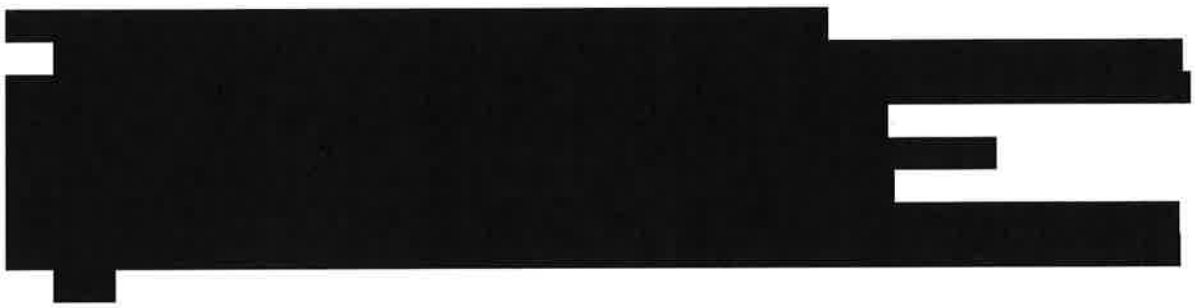
Tables (= data matrix) representing the available properties for the category members are presented for selected endpoints, to allow comparison of values across the category members. Ten physico-chemical properties are compared. Similarity of environmental behaviour is also claimed. However, no measured data are presented for environmental properties. For human health properties acute oral and dermal, skin and eye irritation, as well as sensitisation and Ames test results are in the data matrix.

A chapter on the similarity of the metabolism of bicyclic terpenes is presented to support the claim that the substances upon uptake are rapidly metabolised and excreted and that bioaccumulation does not occur. This chapter is concerned with alpha-pinene, beta-pinene and delta-3-carene.

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

As indicated in the section 1.2 of the IUCLID file the registered substance has the following composition (only constituents with a potential concentration of [redacted] % are listed here); there are other possible constituents in concentrations lower than [redacted] %.

[redacted]



ECHA understands your strategy is to predict properties of the registered substance by reading across the results obtained in studies with (some of) the constituents present in the registered substance. In this regard ECHA has the following observations:

(1) Substance characterisation of source and target substances

The characterisation of the source substances needs to be sufficiently detailed in order to assess what impact the composition and/or impurities may have on the attempted prediction. In the ECHA's practical guide 6 "How to report on Read-Across" it is recommended to follow the ECHA Guidance for identification and naming of substances under REACH and CLP (version 1.3, February 2014) also for the source substances. This ensures that the identity of the source substance and its impurity profile allows an assessment of the suitability of the substances for read-across purposes.

In the present case, the characterisation of the substances identified as category members and proposed to act as source substances for the registered substance is not sufficiently clear; in the category description the purity of the members are indicated as 80 % or higher. No information on the possible impurities of the source substances are provided in the justification document. In the endpoint study summaries sometimes there is more information, which is conflicting to the information provided in the justification document.

ECHA's conclusion is that currently the identity of the source substances and their impurity profiles cannot be assessed using the information provided in the registration dossier and the suitability of the substances for read-across purposes cannot be verified.

(2) Structural similarity and dissimilarity of the individual substances within the category and the scientific explanation on why and how the structural features allow predictions

In order to meet the provisions in Annex XI, Section 1.5. to predict human health and environmental effects from data from a reference substance within the group by interpolation to other substances in the group, ECHA considers that structural similarity alone is not sufficient. It has to be justified why such prediction is possible in view of the identified structural differences and the provided evidence has to support such explanation. In particular, the structural similarities must be linked to a scientific explanation of how and why a prediction is possible and of how the structural dissimilarities impact the prediction.

For a category used for a read-across approach it should be explained which structural moieties the category members have in common, and which structural differences are allowed for category members according to the category definition. ECHA notes that there are some explanations on the structural similarities for the mono-constituent members of the category provided (i-iv above), but the structural dissimilarities have not been explained nor analysed. For the UVCB member of the category (v., namely the registered substance) it has not been explained scientifically why the additional constituents apparently not present in other members of the category still allow the substance to be a member of the category.

In the present case, you propose to use results obtained in studies with the main constituents of the registered UVCB substance to predict its properties. In addition, camphene and crude sulphate turpentine are used for read-across purposes but they are not identified as members of the category. ECHA understands that they are used as additional analogue substances. However, ECHA notes that no further explanations are offered why and how these substances may be used to predict human health and environmental properties for the registered substance. Also no comparisons of inherent properties between crude sulphate turpentine or camphene with the registered substance are provided which would allow verifying the suitability of crude sulphate turpentine or camphene as analogue substance.

From the composition of the registered substance provided in the registration dossier it is obvious that a large portion of the composition of the registered substance is not covered by the main approach chosen (i.e. to predict the properties of the registered substance using results obtained with the members of the category, which are at simultaneously constituents of the registered substance). The monocyclic terpene hydrocarbons and other terpenes hydrocarbons (d. to i. above) potentially may make up to [redacted] % of the total composition, if the maximum concentrations in the provided concentration ranges of these constituents are added to each other. No explanation is offered in which way these substances influence the toxicity profile of the registered substance and thereby influence the reliability of predictions which do not take into account the presence of these constituents in the registered substance.

Moreover, in the composition of the registered substance, there are constituents listed in concentrations below [redacted]%. These constituents and their possible impact on the read-across have not been addressed. They also might have an impact on the toxicity profile of the registered substance and thereby influence the reliability of predictions which do not take into account the presence of these constituents in the registered substance.

As explained in more detail in the property specific point 5 below, the available data for studies investigating toxicity after repeated administration (repeated dose toxicity, pre-natal developmental toxicity and the reproductive toxicity study) is only from one member of the category or there is no information at all. In contrast to the principle purpose of a category, i.e. to provide more reliable data and come to a prediction associated with less uncertainty in comparison with an analogue approach, the proposed category provides one data point for one source substance, if one at all, for the properties under consideration.

ECHA concludes that you have not addressed the obvious structural differences between the source substances and the target substance and did not explain why those differences would not lead to differences in the mode of action and in the toxicity profile of target and source substances. The provided explanation is not considered as valid to establish the link between the structural similarity and the prediction. Consequently it is not possible to predict human adverse health effects based on the proposed approach nor to predict environmental effects.

(3) Information in the data matrix to support a similar or regular pattern as a result of structural similarity

Annex XI, Section 1.5 provides that "substances whose physicochemical, toxicological and eco-toxicological properties are likely to be similar or follow a regular pattern as result of structural similarity may be considered as a group or 'category' of substances. One prerequisite for a prediction based on read-across therefore is that the substances involved are structural similar and are likely to have similar properties. One important aspect in this regard is the data matrix comparing properties of source and target substances.

The data matrix provided by you has no information on toxicity studies using repeated administration nor any measured environmental concentrations for the different category members. Therefore, the data matrix does not allow verifying that the members of the category have a consistent toxicity profile which would support the claim of a similar pattern of toxicity for these properties.

Regarding the toxicological endpoints, the available data in the data matrix are concerned with acute effects, local effects or gene mutations and therefore do not cover the broad range of possible interactions with mammalian biological systems as investigated in repeated dose toxicity studies, pre-natal developmental toxicity studies, or reproductive toxicity studies. The physico-chemical properties or the environmental effects described in the two other tables also do not provide decisive information regarding studies with repeated administration to mammalian organisms and their consistent toxicity profile.

In the present case, additional complications for the attempted predictions arise by the fact that exposure to the registered substance results in a combined exposure of the experimental system to all constituents simultaneously. In contrast, exposure to a mono-constituent substance results in exposure to only this substance (plus its impurities) in the same study type. If the toxicity is to be predicted on the basis of the toxicity of the individual constituents it is mandatory to know about the individual toxicity profiles of the mixture constituents, including their dose-response relationships. Only then a sound assessment can be made on the possible action modes of the mixture constituents (joint or combined, dissimilar, or interaction) and the consequences for the prediction. None of such considerations have been addressed in the justification document and the toxicity profiles are not even known for the main constituents. The proposed predictions, therefore, appear to be not reliable.

Furthermore, there are large possible concentration variations of the constituents in the registered substance. This leads to additional uncertainty when predictions are attempted on the basis of results with several mono-constituents as source substances. The predictions need to cover the possible concentration ranges in a representative way. No attempt has been made to explain how such variations potentially influence the predictions.

The main constituents furthermore exist in different enantiomeric forms. It is not addressed by you which enantiomeric forms are present in the registered substance and which are present in the mono-constituent category members. Consequently, the possible impact of such enantiomeric structures on the attempted prediction is not addressed.

However, the data matrix allows ECHA to draw some conclusions. The registered substance is classified as skin sensitising. The same is true for (−)-beta-pinene and delta-3-carene, but not for beta-pinene nor for alpha-pinene. These substances, therefore, appear to show different reactivity towards biological macromolecules, which is the prerequisite to trigger a response of the immunological system. Such reactivity differences and whether they have impact on other properties of the substances in the category are not considered in making predicitions.

In addition there is information in the registration dossier for repeated dose toxicity for one constituent of the registered substance. The source substance alpha-pinene was investigated for repeated dose toxicity in rats and mice via inhalation. The studies demonstrate that the substance is causing systemic toxicity. For rats, nephrotoxicity has been observed in males only (LOAEL 50 ppm) which has been claimed to be due to alpha 2u-globulin induced nephropathy. Female rats showed mortality and reduced body weight gain at higher doses. Mice showed hyperplasia in the urinary bladder epithelium at concentrations > 50 ppm. The NOAEC observed in mice for alpha-pinene is read-across to the registered substance without further considerations on other constituents of the registered substance. ECHA interprets this to mean that the same target organ and the same level of toxicity is assumed for the registered substance and, therefore, that the other constituents present in this substance are claimed to have no independent toxicity of their own or do not influence the toxicity of alpha-pinene. Such claim is not substantiated by other data. The prediction on the basis of the studies with alpha-pinene alone may therefore underestimate the toxic potential of the registered substance.

ECHA concludes that the information in the data matrix does not confirm the statement that: "*...read-across strategy was used to compile the available data on the main components of gum turpentine oil, in order to cover as far as possible the variations in contents of individual components*". The data matrix for human health and environmental properties therefore does not allow verifying the claim of a similar pattern of toxicity between the constituents and the registered UVCB substance. In fact, for the properties discussed here, there is no information at all to assess variations in the toxicity profiles of the individual substances. Consequently the information in the data matrix does not support the possibility to predict adverse human health and environmental effects within the proposed category by interpolation to the group members.

(4) Exposure of the test organism to source and target substances and to their metabolites

Annex XI, Section 1.5. provides that "*substances whose physicochemical, toxicological and eco-toxicological properties are likely to be similar or follow a regular pattern as result of structural similarity may be considered as a group or 'category' of substances*". One prerequisite for a prediction based on read-across therefore is that the substances involved are structurally similar and are likely to have similar properties. One important aspect in this regard is the comparison of absorption, distribution, metabolism and elimination of source and target substances to allow assessing the qualitative and quantitative internal systemic exposure of the test organism when exposed to source and target, respectively.

You presented in the justification document a chapter on Absorption, Distribution, Metabolism and Excretion (ADME) of some main constituents to support the claim of similar properties of the category members. On the basis of the information provided the following conclusions are drawn by you:

- The constituents investigated in human volunteers when administered via inhalation (alpha-pinene, beta-pinene, delta-3-carene) are taken up rapidly and to a large degree.
- The investigated substances are metabolised via cytochrome P-450 catalysed oxidation at different positions of the molecules.
- The oxidation step leads to oxygenated metabolites which are subsequently conjugated and excreted in the urine.
- It is concluded that bioaccumulation does not occur.

ECHA notes that

- In the IUCLID file under 7.1.1 ([redacted]) absorption and excretion rates of alpha-pinene, beta-pinene and 3-carene after inhalation exposure in human volunteers to turpentine vapour (not clear which turpentine, but due to the study design ECHA assumes that it consists at least of alpha-pinene, beta-pinene and 3-carene as main constituents) and after exposure to alpha-pinene and 3-carene alone have been determined. The mean half-lives ($t_{1/2}$) of the last phase of alpha-pinene, beta-pinene, and 3-carene averaged 32, 25, and 42 h, respectively. This indicates that there are tissues in the body (most likely lipid rich tissues), in which the substances accumulate and from which the elimination of the substances are delayed by only slowly making them available to metabolism. Such distribution behaviour is consistent with the logKow of 4.49 which is provided by you in the dossier as representative for the registered substance. Therefore, it seems not to be supported to conclude a general rapid metabolism and excretion. Consequently, your conclusion on bioaccumulation appears to be doubtful. The average $t_{1/2}$ of the last phases of alpha-pinene and 3-carene tended to be higher after exposure to turpentine containing the monoterpenes simultaneously than after exposure to the monoterpenes alone. This appears to indicate that the simultaneous exposure to the turpentine mixture influences the clearance rate of the individual substance in this study. The impact of such changes in the clearance rates by combined exposures on the predictions based only on one constituent is not addressed in the justification document. You have, therefore, not established that the attempted predictions are possible despite the combination effects occurring in the toxicokinetic studies in human volunteers.

- The substances under consideration have quite complex chemical structures. Therefore, the metabolic fate after hydroxylation via P450 enzymes is expected to provide a complex picture as well, since different parts of the molecules are accessible to oxidation. Indeed, the studies by Ishida (1981) in rabbits present in the IUCLID file demonstrate that hydroxylation takes place at different positions of the molecules for the individual constituents which are then further converted to subsequence metabolic products. These products are different for each constituent of the registered substance and no information is presented on their potential toxicity. It is obvious that, when exposed to the registered substance, the test organism is exposed to a multitude of metabolites at the same time which are not present at exposures to one of the constituents alone. The impact of this situation on the toxicity profile of the registered substance and the reliability of a prediction only relying on one constituent is not addressed in the justification document.

ECHA concludes that you did not establish to which chemical substances the test organism is systemically exposed when external exposure to the registered substance occurs. The complex metabolic profile expected at exposure to the registered substance is not taken into account in the read-across approach. Therefore, it is not possible to verify that the source substances and the registered substance have the same, common mechanism of action which would lead to a similar or regular pattern of toxicity as a result of structural similarity. As a consequence it is not possible to predict properties within the proposed category by interpolation to the group members.

(5) Further observations

- The prediction for studies on mutagenic effects, ready biodegradability and acute aquatic toxicity relies on key studies conducted with crude sulphate turpentine. Comparing the test substance for the ready biodegradation studies provided by you in the technical dossier with the test substance for the acute aquatic toxicity, ECHA observes that they all refer to crude sulphate turpentine as test material, but with compositional variations between the different tests. Furthermore, for the provided ready biodegradability studies, the total concentration of the different compositional constituents adds up to [redacted]%.

- ECHA notes that crude sulphate turpentine is not a member of the proposed category and no justification for its use as analogue substance has been provided. In particular, reasoning is missing why and how this substance can be used to predict results in the Ames test, the chromosomal aberration study in vitro, the mammalian gene mutation study, ready biodegradability testing or the acute toxicity in fish for the registered substance, which has another composition and additional constituents. ECHA considers therefore that the predictions based on crude sulphate turpentine are not justified and the predictions cannot be accepted.

- The read-across approach for the prenatal developmental toxicity provides a study with camphene. Camphene is not a member of the proposed category and amounts to 0 - 2 % in the registered substance. It is not explained why and how the results obtained with camphene can be used for predicting the results which may be obtained with the registered substance. Therefore, the read-across approach does not take into account the structural differences between the source substance and the target substance, does not explain how and why a similar pattern of toxicity supports such predictions, and does not address the other constituents of the registered substance. Furthermore a study on Rowachol is provided. Rowachol is not mentioned in the justification document, no further explanations are provided and therefore the contribution of this study to the prediction for the registered substance cannot be assessed. ECHA considers, therefore, that the prediction of "no prenatal developmental effect" for the registered substance in a study conducted according to OECD 414 is not justified and cannot be accepted.

- There is no valid source study compliant with Annex X, 8.7.3 available for read-across of reproductive toxicity. Therefore, no predictions based on a read-across approach are possible.

D. Conclusion on the read-across approach

ECHA considers that structural similarity alone is not sufficient for predicting (eco)toxicological properties. It has to be justified why and how such prediction is possible in view of the identified structural differences and the provided evidence has to support such explanation. ECHA notes that in view of the issues listed above, it has not been demonstrated that the source and target substances have the same properties or follow a similar pattern with regard to studies on mutagenicity, repeated dose toxicity, pre-natal developmental toxicity, reproductive toxicity, ready biodegradability, and acute toxicity to fish. Besides the reference to the structural similarity, there is no valid mechanistic explanation provided by you why predictions can be made using the results from the source substances. ECHA concludes that you have failed to meet the requirement of Annex XI, Section 1.5. that human health and environmental effects may be predicted from data for reference substance(s) within the group by interpolation to other substances in the group (read-across approach).

In your comments to the draft decision you have indicated that "*you understand why the read-across was considered to be insufficient to characterise the properties of the registered substance and did not question the scientific rationale behind it*".

Pursuant to Article 41(1) of the REACH Regulation, ECHA concludes that the adaptation of the standard information requirements for the properties investigated by the Ames test, the chromosomal aberration study in vitro, the mammalian gene mutation study in vitro, the repeated dose toxicity study (90-day) in rodents, the pre-natal toxicity studies in rats and rabbits, the study on reproductive toxicity (EOGRTS), the testing on ready biodegradability, and on acute toxicity to fish in the technical dossier based on the proposed read-across approach does not comply with the general rules of adaptation as set out in Annex XI, 1.5. Therefore, ECHA rejects those adaptations in the technical dossier that are based on Annex XI, 1.5.

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

3. In vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.)

An "*In vitro* gene mutation study in bacteria" is a standard information requirement as laid down in Annex VII, Section 8.4.1. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have not provided any study record of an in vitro gene mutation study in bacteria in the dossier that would meet the information requirement of Annex VII, Section 8.4.1, for the registered substance. Instead, you have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. You have provided:

(i) RA 13466-78-9, Ames test, [redacted] 2010, supporting study, reliability 1, according to GLP and OECD TG 471 (2010), 5 strains, conducted with delta-3-carene, negative with and without metabolic activation.

(ii) RA 18172-67-3, Ames test, [redacted] 2006, supporting study, reliability 2 (no data on number of bacterial cells per culture, individual plate count and positive controls), no GLP, according to equivalent OECD TG 471 (2010), TA 1535, TA 1537, TA 98 and TA 100, TA 1538 strains, conducted with (-)-beta-pinene, negative with and without metabolic activation.

(iii) RA 80-56-8, Ames test, [redacted] 1989, supporting study, reliability 4 (no information on negative or positive controls, no information on cytotoxicity), TA 1535, TA 1537, TA 98, TA 100, TA 1538 strains, conducted with alpha-pinene, no GLP, unclear guideline, negative with and without metabolic activation

(iv) RA 127-91-3, Ames test, [redacted] 1989, supporting study, reliability 4 (no information on negative or positive controls, no information on cytotoxicity), TA 1535, TA 1537, TA 98, TA 100, TA 1538 strains, conducted with beta-pinene, no GLP, unclear guideline, negative with and without metabolic activation

(v) RA 80-56-8, Ames test, NTP 2005, supporting study, reliability 2 (no data on test material purity, number of cells per culture and number of replicates used; only 3 strains tested; no individual plate counts), TA 98, TA 100, TA 102, conducted with alpha-pinene, no GLP, according to equivalent OECD TG471, negative with and without metabolic activation

(vi) 8006-64-2, Ames test, NTP 2006, supporting study, reliability 2 (lack of details on test material, number of cells per culture and number of replicates used; only 3 strains tested; no individual plate counts) TA 98, TA 100, E. coli WP2 strains, conducted with turpentine (no constituents specified, but claimed to be the same test material as specified in section 1.1 and 1.2 of IUCLID) , no GLP, according to equivalent OECD TG471, negative with and without metabolic activation

(vii) RA 79-92-5 Ames test, [redacted] 1985, supporting study, reliability 4 (no details on test substance), TA 98, TA100, bacteria UTH8414 and UTH8413, conducted with camphene, no GLP, no guideline, negative with and without metabolic activation

(viii) RA 80-56-8, Ames test, [redacted] no publication year, supporting study, reliability 4, test with urinary extracts from rats treated with alpha-pinene. No GLP, no guideline, negative with and without metabolic activation

(ix) RA 8006-64-2, Ames test, [redacted] 2010, key study, reliability 1, according to GLP and OECD 471, 5 strains, conducted with Crude sulphate turpentine, percentage of components: typical Composition: Alpha Pinene ([redacted]%), Beta Pinene ([redacted]%), Delta carene ([redacted]%), Dipentene ([redacted]%), Sulphur ([redacted]%), negative with and without metabolic activation

ECHA does not consider that studies (iii), (iv), (vii), (viii) contribute to the assessment of the information requirement "*In vitro* gene mutation study in bacteria" for the registered substance. These studies have major deficiencies (see above), which do not allow a meaningful interpretation of the results. (i) investigated delta-3-carene, (v) investigated alpha-pinene, (ii) investigated (-)-beta-pinene, (vi) investigated turpentine, but no details on the substance tested are provided. (ix) investigated crude sulfate turpentine: alpha-pinene ([redacted]%), beta-pinene ([redacted]%), delta-carene ([redacted]%), dipentene ([redacted]%), sulfur ([redacted]%).

ECHA notes that the main constituents alpha-pinene, (-)-beta-pinene, and delta-3-carene have been tested in the Ames test and were found consistently to not result in a mutagenic response in this assay. However, the composition of the registered substance also show the presence of another isomer of beta-pinene (CAS 127-91-3) and the possible presence of monocycliclic and other terpene hydrocarbons which may reach concentrations up to [redacted] % (adding the maximum concentrations of the concentration ranges). You did not discuss whether such compounds may have an influence on the mutagenic response in this test system. There is also no valid information presented in the dossier on the results of such substances in the Ames test.

Crude sulphate turpentine is not a member of the proposed category and has a different composition compared to the registered substance, in particular it does only contain dipentene at a concentration of [redacted] % as additional substance whereas the registered substance contains this and other terpene hydrocarbons in potentially much higher concentrations. Furthermore it contains sulphur (not specified which type), which is not present in the registered substance. It is not explained why and how the results obtained with crude sulphate turpentine may be used to predict the results on an Ames test conducted with the registered substance.

In conclusion, as explained above in the section '*Grouping of substances and read-across approach'* of this decision, your adaptation of the information requirement is rejected. In the specific case of the Ames test information requirement, ECHA considers that you demonstrated for the source substances investigated with valid studies that they are likely to be similar for the property under consideration, i.e. mutagenicity in the Ames test. However, the prediction of a negative outcome in the Ames test for the registered substance on this basis is regarded as not acceptable, since a relevant part of the composition of the registered substance is not covered by the investigated source substances and no explanation has been provided why this would not be needed. Therefore, this adaptation does not meet the general rules for adaptation of Annex XI, 1.5. and the information provided on this property 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 information requirement.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

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: Bacterial reverse mutation test (test method: EU B.13/14. / OECD TG 471).

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

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

You have not provided any study record of an *In vitro* cytogenicity study in mammalian cells or *in vitro* micronucleus study in the dossier that would meet the information requirement of Annex VIII, Section 8.4.2, for the registered substance. Instead, you have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. You have provided:

(i) RA 8006-64-2, chromosome aberration test, [redacted] 2010, key study, reliability 1, according to GLP and OECD TG 473, conducted with crude sulfate turpentine, negative with and without metabolic activation.

(ii) RA 127-91-3, in Vitro Sister Chromatid Exchange Assay in Mammalian Cells, Sasaki 1090, supporting study, reliability 4 (individual results were not reported; no information on metabolic activation and on validity of controls), no GLP, equivalent to OECD Guideline 479, conducted with beta-pinene, no effect on cell cycle and induction of sister-chromatid exchanges in cultured Chinese hamster ovary cells

ECHA does not consider that study (ii) contributes to the assessment of the information requirement "*In vitro* cytogenicity study in mammalian cells or an *in vitro* micronucleus study" for the registered substance. The study has major deficiencies (see above), which do not allow a meaningful interpretation of the results. (i) investigated crude sulfate turpentine.

Crude sulphate turpentine is not a member of the proposed category and has a different composition compared to the registered substance, in particular it does only contain dipentene at a concentration of [redacted] % as additional substance whereas the registered substance contains this and other terpene hydrocarbons in potentially much higher concentrations. Furthermore it contains sulphur (not specified which type), which is not present in the registered substance. It is not explained why and how the results obtained with crude sulphate turpentine may be used to predict the results on an *in vitro* cytogenicity study in mammalian cells or an in vitro micronucleus study conducted with the registered substance.

In conclusion, as explained above in the section '*Grouping of substances and read-across approach'* of this decision, your adaptation of the information requirement is rejected. The prediction of a negative outcome in the *in vitro* cytogenicity study in mammalian cells or *in vitro* micronucleus study for the registered substance on this basis is regarded as not acceptable, since a) a relevant part of the composition of the registered substance is not covered by the investigated source substances and no explanation has been provided why this would not be needed and b) the same or a similar pattern for the property under consideration of the source substances in the category have not been established. Therefore this adaptation does not meet the general rules for adaptation of Annex XI, 1.5. and the information provided on this property 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 information requirement.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: *In vitro* cytogenicity study in mammalian cells (test method: EU B.10./OECD TG 473) or *in vitro* mammalian cell micronucleus study (test method: OECD TG 487).

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

An "*in vitro* gene mutation study in mammalian cells" is an information requirement as laid down in Annex VIII, Section 8.4.3. of the REACH Regulation, "if a negative result in Annex VII, Section 8.4.1. and Annex VIII, Section 8.4.2." is obtained. ECHA notes that the registration dossier currently does not contain information which allows concluding on the results for both of these information requirements.

Therefore, adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement only, if the results of the two other in vitro mutagenicity test requested in this decision are negative.

You have not provided any study record of an *in vitro* gene mutation study in mammalian cells in the dossier that would meet the information requirement of Annex VIII, Section 8.4.3, for the registered substance. Instead, you have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. You have provided:

(i) RA 18172-67-3, mammalian cell gene mutation (mouse lymphoma assay), [redacted] 2006, supporting study, reliability 2 (no information on negative and positive control data), no GLP and equivalent to OECD TG 476, conducted with (-)-beta-pinene, positive without metabolic activation (old evaluation) negative without metabolic activation (new evaluation), negative with metabolic activation (old evaluation), ambiguous with metabolic activation (new evaluation)

(ii) RA 80-56-8, UDS – test, [redacted] 1989, supporting study, reliability 4 (no data on test material purity, source and concentration units; conditions; details of metabolic activation systems; procedures used to block entry of cells into S-phase; negative/positive controls; grain count in individual culture), no GLP, no guideline, conducted with alpha-pinene, negative

(iii) RA 8006-64-2, mammalian cell gene mutation assay (mouse lymphoma assay), [redacted] 2010, key study, reliability 1, according to GLP and OECD 476, conducted on crude sulfate turpentine, negative with and without metabolic activation

ECHA does not consider that study (ii) contributes to the assessment of the information requirement "*in vitro* gene mutation study in mammalian cells" for the registered substance. The study has major deficiencies (see above), which do not allow a meaningful interpretation of the results. (i) investigated (-)-beta-pinene and was assessed as inconclusive by you and (iii) investigated crude sulfate turpentine.

Crude sulphate turpentine is not a member of the proposed category and has a different composition compared to the registered substance, in particular it does only contain dipentene at a concentration of [redacted] % as additional substance whereas the registered substance contains this and other terpene hydrocarbons in potentially much higher concentrations. Furthermore it contains sulphur (not specified which type), which is not present in the registered substance. It is not explained why and how the results obtained with crude sulphate turpentine may be used to predict the results on an *in vitro* gene mutation study in mammalian cells conducted with the registered substance.

In conclusion, as explained above in the section '*Grouping of substances and read-across approach*' of this decision, your adaptation of the information requirement is rejected. The prediction of a negative outcome in the *in vitro* gene mutation study in mammalian cells for the registered substance on this basis is regarded as not acceptable, since a) a relevant part of the composition of the registered substance is not covered by the investigated source substances and no explanation has been provided why this would not be needed and b) the same or a similar pattern for the property under consideration of the source substances in the category have not been established. Therefore this adaptation does not meet the general rules for adaptation of Annex XI, 1.5. and the information provided on this property 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 information requirement, if a negative result in Annex VII, Section 8.4.1. and Annex VIII, Section 8.4.2." is obtained.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: *In vitro* mammalian cell gene mutation test (test method: EU B.17./OECD TG 476) provided that both studies requested under 3. and 4. have negative results.

6. Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2.)

A "sub-chronic toxicity study (90 day)" is a standard information requirement as laid down in Annex IX, Section 8.6.2. 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. Pursuant to the last paragraph of column 2 of that Section, further studies may be required by the Agency in case of, for instance, indications of an effect for which the available evidence is inadequate for toxicological and/or risk characterisation.

You have not provided any study record of a sub-chronic toxicity study (90-day) in the dossier that would meet the information requirement of Annex IX, Section 8.6.2, for the registered substance. Instead you have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. You have provided:

(i) RA 80-56-8, repeated dose toxicity (90-days) in rats via inhalation, NTP 2006, key study, reliability 2 (food consumption, hematology, urinalysis, ophthalmological examination, some organ weights were not recorded, no functional observation battery), with alpha-pinene according to a study design equivalent or similar to OECD 413 (0, 50, 100, 200, 400 ppm), no GLP. The identified LOAEL in male rats was 25 ppm based on observed nephropathy. In female rats nephropathy was not observed and the NOAEL was determined as 200 ppm based on mortality and lower body weight gain at 400 ppm. You claim that the nephropathy is caused by accumulation of alpha2u-globulin in the form of hyaline droplets and that this is a male rat specific mechanism leading to kidney cell necrosis and tumors. ECHA concludes that the information provided is consistent with a male rat specific hyaline droplet accumulation due to alpha-2-microglobulin accumulation but that you did not prove for the tested substance that this mechanism is indeed causal for the observed kidney effects. ECHA also notes that other constituents of the registered substance also have been associated with such kidney effects.

(ii) RA 80-56-8, repeated dose toxicity (90-days) in mice via inhalation, NTP 2006, key study, reliability 2 (food consumption, hematology, urinalysis, ophthalmological examination, some organ weights were not recorded, no functional observation battery), with alpha-pinene according to a study design equivalent or similar to OECD 413 (0, 50, 100, 200, 400 ppm), no GLP. The identified NOAEL is at 50 ppm on the basis of cell hyperplasia of the urinary bladder epithelium observed in both sexes.

However, as explained above in the section *'Grouping of substances and read-across approach'* of this decision, your adaptation of the information requirement is rejected.

Therefore, 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. However since alpha-pinene is part of the composition of the registered substance, the results of the studies with alpha-pinene are considered by ECHA as relevant for the determination of the appropriate study design for the registered substance.

ECHA has evaluated the most appropriate route of administration for the study under consideration according to REACH Annex IX, Section 8.6.2. Based on the information provided in the technical dossier and the chemical safety report the testing by the dermal route is not appropriate. The properties of the registered substance and its uses indicate that human exposure by the inhalation route is likely. However, no repeated dose toxicity study by the oral route is provided in the technical dossier for the registered substance and according to ECHA Guidance R.7.a, chapter R.7.5.4.3, the oral route is the preferred one concerning repeated dose toxicity testing. Furthermore, the available information for the inhalation route for one of the constituents (alpha–pinene) indicate that systemic toxicity may be caused, if this constituent is part of the tested substance. Systemic toxicity may be extrapolated from the oral route to the inhalation route. Hence, ECHA considers that in this specific case the default oral route is the most appropriate route of administration.

According to the test method EU B.26/OECD TG 408 the rat is the preferred species. ECHA considers this species as being appropriate and testing should be performed with the rat.

A study in the rat will also provide the opportunity to prove or disprove the claimed alpha-2-microglobulin mediated nephropathy by one or more of the constituents. Since humans do not excrete alpha-2u-globulin, this mode of action is not relevant to humans. For this reason, ECHA decided to include in the request for a sub-chronic toxicity study urinalysis (which is optional in paragraph 30 of OECD 408, and the relevant part of Section 1.5.2.2. of EU Method B.26) to investigate kidney function in the request for a sub-chronic toxicity study. Furthermore, a full histopathological examination (paragraph 36 of OECD 408, Section 1.5.2.4. of EU Method B.26) including immunohisto-chemical investigation of renal pathology is to be conducted to determine if the pathology is indeed mediated by alpha-2-microglobulin.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

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:
Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2.; test method: EU B.26./OECD TG 408) in rats, with the registered substance modified to include urinalysis and a full histopathological examination which is to include immunohistochemical investigation of renal pathology to determine if the pathology is mediated by alpha-2u globulin nephropathy.

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

The basic test design of an extended one-generation reproductive toxicity study (Cohorts 1A and 1B, without extension of Cohort 1B to include a F2 generation, and without Cohorts 2A, 2B and 3) is a standard information requirement as laid down in column 1 of 8.7.3., Annex X. If the conditions described in column 2 of Annex X are met, the study design needs to be expanded to include the extension of Cohort 1B, Cohorts 2A/2B, and/or Cohort 3. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have not provided any study record of an extended one-generation reproductive toxicity study with the registered substance in the dossier that would meet the information requirement of Annex X, Section 8.7.3. You did also not provide as study record of a two-generation reproductive toxicity study (EU B.35, OECD TG 416) with the registered substance which was initiated before 13 March 2015 and which would be considerd appropriate to address this standard information requirement. Instead you have sought to adapt this information requirement by providing the following justification:

"*In a GLP teratogenicity study conducted according to OECD guideline 414 with camphene and in a teratogenicity/postnatal development study using rowachol (terpene mixture of alpha/beta-pinene (■%)), no teratogenic/postnatal development effects were identified. Moreover, in a 90-day repeated toxicity study conducted with alpha-pinene, no effects were observed on reproductive organs (tissues examined microscopically: epididymidis, preputial gland, prostate, seminal vesicle and testes for males, clitoral gland, ovary and uterus for females). Thus, considering the read-across approach, a reproductive toxicity study is not deemed necessary based on the results of these studies.*"

ECHA notes that there are no adequate studies meeting the information requirement in Section 8.7.3. conducted with any member of the proposed category. ECHA understands that you refer to Annex XI 1.2 in particular that sufficient weight of evidence is available to conclude on the presence or absence of a particular dangerous property, i.e. in this case reproductive toxicity as investigated in an extended one-generation reproductive toxicity study. To support this you provided:

(i) The results from the pre-natal developmental toxicity study conducted with camphene as described under 8 (i).
(ii) The results from a pre-natal developmental toxicity study conducted with rowachol as described under 8 (ii).
(iii) The results from a 90-day repeated dose toxicity study conducted with alpha-pinene as described under 6 (i) and 6 (ii).

ECHA notes that the information provided under (i) to (iii) was not obtained with the registered substance but with analogue substances. As explained above in the section '*Grouping of substances and read-across approach*' of this decision, your current read-across approach is rejected and these studies cannot be used for adapting other information requirements. Furthermore, a teratogenicity study and 90-day studies in combination, do not address the key information elements of Annex X, 8.7.3, in particular functional fertility, sperm parameter analysis, oestrous cyclicity, postnatal development, endocrine modes of action, and histopathology of gonads and accessor sex organs of F1 animals. Thus, it is not possible to conclude if the registered substance has or has not a hazardous property on sexual function and fertility and developmental toxicity according to Annex XI, 1.2.

ECHA concludes that the information available on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint. Thus, an extended one-generation reproductive toxicity study according to columns 1 and 2 of 8.7.3., Annex X is required. The following refers to the specifications of this required study.

Information from studies to be conducted before the extended one-generation reproductive toxicity study

The sub-chronic toxicity study shall be conducted before the extended one-generation reproductive toxicity study and the results from that study shall be used, among other relevant information, to decide on the study design of the extended one-generation reproductive toxicity study following ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.1, October 2015).The sub-chronic toxicity study may provide information on effects that is relevant for triggers (e.g. weight changes and histopathological observations of organs as indication(s) of one or more modes of action related to endocrine disruption which may meet the toxicity-trigger for extension of Cohort 1B or as evidence of specific mechanism/modes of action and/or neurotoxicity and/or immunotoxicity which may meet the particular concern criteria for developmental neurotoxicity and/or developmental immunotoxicity cohorts).

Premating exposure duration and dose-level setting

To ensure that the study design adequately addresses the fertility endpoint, the duration of the premating exposure period and the selection of the highest dose level are key aspects to be considered. According to ECHA Guidance, the starting point for deciding on the length of premating exposure period should be ten weeks to cover the full spermatogenesis and folliculogenesis before the mating, allowing meaningful assessment of the effects on fertility. Ten weeks premating exposure duration is required because there is substance specific information in the dossier indicating accumulation in lipid rich tissues (see 0.3 (12)). (Chapter R.7a: Endpoint specific guidance, R.7.6, Version 4.0 – July 2015).

The highest dose level shall aim to induce some toxicity to allow comparison of effect levels and effects of reproductive toxicity with those of systemic toxicity. The dose level selection should be based upon the fertility effects with the other cohorts being tested at the same dose levels.

It is recommended that results from a range-finding study (or range finding studies) for the extended one-generation reproductive toxicity study are reported with the main study. This will support the justifications of the dose level selections and interpretation of the results.

Extension of Cohort 1B

If the column 2 conditions of 8.7.3., Annex X are met, Cohort 1B must be extended, which means that the F2 generation is produced by mating the Cohort 1B animals. This extension provides information also on the sexual function and fertility of the F1 animals.

The use of the registered substance is leading to significant exposure of consumers because the registered substance is used by consumers in a wide range of application, e.g. adhesives, sealants, air care products, biocidal products (e.g. disinfectants, pest control), coatings and paints, thinners, paint removes, fillers, putties, plasters, modelling clay, finger paints, textile dyes, finishing and impregnating products; including bleaches and other processing aids, washing and cleaning products (including solvent based products), perfumes, fragrances, cosmetics, personal care products, finger paints, scented clothes, scented toys, etc. Also a significant exposure of professionals is noted e.g., use of fragrance products, use of adhesives and sealants, use of coatings and inks, etc.

Furthermore there are indications that the internal dose for the constituents of the registered substance will reach a steady state in the test animals only after an extended exposure. In human volunteers, the mean half-lives (t1/2) of the last phase of alpha-pinene, beta-pinene, and 3-carene averaged 32, 25, and 42 h, respectively. The average t1/2 of the last phases of alpha-pinene and 3-carene tended to be higher after exposure to turpentine than after exposure to monoterpenes alone (see also section *'Grouping of substances and read-across approach'* 0.3 (4)).

Therefore, ECHA concludes that Cohort 1B must be extended to include mating of the animals and production of the F2 generation because the uses of the registered substance is leading to significant exposure of consumers and there are indications that the internal dose for constituents of the registered substance will reach a steady state in the test animals only after an extended exposure.

Species and route selection

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

According to the test method EU B.56/ OECD TG 443, the test substance is usually administered orally. On the basis of this default consideration, ECHA considers testing should be performed by the oral route.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Extended one-generation reproductive toxicity study (test method EU B.56/ OECD TG 443), in rats, oral route, according to the following study-design specifications:

- Ten weeks premating exposure duration for the parental (P0) generation;
- Dose level setting shall aim to induce some toxicity at the highest dose level;
- Cohort 1A (Reproductive toxicity);
- Cohort 1B (Reproductive toxicity) with extension to mate the Cohort 1B animals to produce the F2 generation;

Currently, the inclusion of Cohorts 2A and 2B (developmental neurotoxicity) and Cohort 3 (developmental immunotoxicity) is not requested. However, the sub-chronic toxicity study (90-day) requested in this decision (request 6) and/or any other relevant information may trigger changes in the study design. Therefore, the sub-chronic toxicity study (90-day) is to be conducted first and the study results submitted to ECHA in a dossier update by **16 January 2018**. If, on the basis of this update and/or other relevant information, a need for changes to the study design is identified, ECHA will inform you by **16 April 2018** (i.e. within three months after expiry of the 12-month deadline to provide the sub-chronic toxicity study (90-day)) of its intention to initiate a new decision making procedure under Articles 41, 50 and 51 of the REACH Regulation to address the design of the extended one-generation reproductive toxicity study. If you do not receive a communication from ECHA by **16 April 2018**, the request of the present decision for the extended one-generation reproductive toxicity study remains effective and you may commence the conduct of the study and the results will need to be submitted by the deadline given in this decision **18 January 2021.**

Notes for your consideration

When submitting the study results of the sub-chronic toxicity study (90-day) you are invited to also include in the registration update your considerations whether changes in the study design are needed (see also *ECHA Guidance on information requirements and chemical safety assessment R.7a, chapter R.7.6 (version 4.1, October 2015)*).

Furthermore, after having commenced the extended one-generation reproduction toxicity study in accordance with the ECHA decision, you may also expand this study to address a concern identified during the conduct of it and also due to other scientific reasons in order to avoid a conduct of a new study. The justification for the changes in the study design must be documented. The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/ triggers must be documented.

8. Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.)

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

You have not provided any study record of a pre-natal developmental toxicity study in the dossier that would meet the information requirement of Annex IX, Section 8.7.2, for the registered substance. Instead you have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation.You have provided:

(i) RA 79-92-5, developmental toxicity/teratogenicity, [redacted] 1992, key study, reliability 2 (only 2 doses were tested up to the limit dose of 1000 mg/kg/day), according to GLP and OECD 414, conducted with camphene, the NOAEL for maternal toxicity and fetotoxicity was considered to be 250 mg/kg bw/day. At 1000 mg/kg bw/day maternal toxicity and fetotoxicity was observed. No teratogenic effects have been reported

(ii) RA 65546-76-9, developmental toxicity/teratogenicity, [redacted] 1978, supporting study, reliability 4, with rowachol (l-menthol [redacted]%; menthone [redacted]%; alpha, beta-pinene [redacted]%; borneol [redacted]%; cineol [redacted]%; d-camphene [redacted]%; rheochrysin [redacted]% and olive oil [redacted]%), no effects reported.

ECHA does not consider that study (ii) contributes to the assessment of the information requirement "pre-natal developmental toxicity" for the registered substance. The study does not allow a meaningful interpretation of the results. Furthermore it investigated a substance for which no relation to the registered substance has been established. Study (i) investigated camphene. Camphene is present as constituent in the registered substance in concentrations up to [redacted]%. However, the registered substance has other main constituents and other terpene hydrocarbons as constituents as well. Camphene is not a member of the proposed category and no explanation has been provided how and why this substance may be used to predict the pre-natal developmental toxicity for the registered substance.

In conclusion, as explained above in the section *'Grouping of substances and read-across approach'* of this decision, your adaptation of the information requirement is rejected. The prediction that in a pre-natal developmental toxicity study with the registered substance no adverse effects would be detected is regarded as not acceptable, since a) the major part of the composition of the registered substance is not covered by the investigated source substance and no explanation has been provided why this would not be needed and b) the same or a similar pattern for the property under consideration of the source substance in the category has not been established and c) the source study is not conducted with a category member, no data matrix has been provided, and the substance is therefore regarded as outside of the category definition. Therefore, this adaptation does not meet the general rules for adaptation of Annex XI, 1.5. and the information provided on this property 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 information requirement.

According to the test method EU B.31/OECD TG 414, the rat is the preferred rodent species and the rabbit the preferred non-rodent species. On the basis of this default consideration, ECHA considers testing should be performed with rats or rabbits as a first species.

According to the test method EU B.31/OECD TG 414, the test substance is usually administered orally. On the basis of this default consideration, ECHA considers testing should be performed by the oral route.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Pre-natal developmental toxicity study (test method: EU B.31./OECD TG 414) in a first species (rats or rabbits) by the oral route.

9. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.)

Pre-natal developmental toxicity studies on two species are part of the standard information requirements for a substance registered for 1000 tonnes or more per year (Annex IX, Section 8.7.2., column 1, Annex X, Section 8.7.2., column 1, and sentence 2 of introductory paragraph 2 of Annex X of the REACH Regulation).

As explained above, the technical dossier does not contain information on a pre-natal developmental toxicity study on a first species with the registered substance and the adaptation provided is rejected.

The technical dossier does not contain an adaptation for the second species in accordance with column 2 of Annex X, Section 8.7. or with the general rules of Annex XI for this standard information requirement.

According to the test method EU B.31/OECD TG 414, the rat is the preferred rodent species and the rabbit the preferred non-rodent species. On the basis of this default consideration, ECHA considers testing should be performed with rats or rabbits as a second species, depending on the choice of the species for the first pre-natal developmental toxicity study.

According to the test method EU B.31/OECD TG 414, the test substance is usually administered orally. On the basis of this default consideration, ECHA considers testing should be performed by the oral route.

In your comments to the draft decision you have agreed to the requested studies on toxicological properties.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Pre-natal developmental toxicity study (test method: EU B.31./OECD TG 414) in a second species (rabbits or rats, depending on the choice of the species for the first pre-natal developmental toxicity study) by the oral route.

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

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

'Ready biodegradability' is a standard information requirement for a substance registered in quantities of 1 tonnes or more per year (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.

You have not provided any study record for ready biogradability testing that would meet the information requirement of Annex VII, Section 9.2.1.1, for the registered substance. Instead, you have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation. You have provided:

(i) Results obtained in a study (STZ 2010) designated as key study with reliability 1 according to OECD Guideline 301 F (Ready Biodegradability: Manometric Respirometry Test) and GLP and performed with the analogue substance Crude Sulfate Turpentine (S: [redacted] %) which is a UVCB substance itself. The composition of the test material is described as alpha pinene ([redacted]%), beta pinene ([redacted]%), delta carene ([redacted]%), dipentene ([redacted]%). The test revealed 71.7% degradation in 28 days. It is noted that the concentrations of the constituents of the test substance adds up to 93%. 7 % are not accounted for.

(ii) Results obtained in a study ([redacted] 2001) designated as supporting study with reliability 2 and according to OECD Guideline 301 B (Ready Biodegradability: CO2 Evolution Test) and GLP that showed 52% degradation in 28 days, thus, missing the ready biodegradability threshold values. The technical dossier indicated that the test material is identical with the registered substance, but the details of the test material specify: *"Composition of test material, percentage of components: α-pinene ([redacted]%) and β-pinene ([redacted]%)"*, without further clarifications. The tested substance therefore appears not to be the registered substance and ECHA assumes that this result is intended to be read-across to the registered substance. It is noted that concentrations of the constituents of the test substance adds up to about [redacted]%. [redacted] % are not accounted for.

ECHA notes that the results of the two studies differ with regard to the ready biodegradability. Explanations on how these results are used to predict the ready biodegradability for the registered substance are missing.

As explained above in the section *'Grouping of substances and read-across approach'* of this decision, your adaptation of the information requirement is rejected.

Therefore, in the absence of other valid adaptation argumentation based on REACH Annex VII, Section 9.2.1.1., 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, Volume 5 Chapter R7b (May 2008) and in the paragraph below. The test guidelines include the description of their applicability domain.

In your comments on the draft decision you stated that due to substance properties and experimental experience with the method, the OECD 301D would be the most preferred guideline for the ready biodegradation study. ECHA agrees that the OECD 301D is a suitable guideline for the registered substance and notes that as discussed in the paragraph above, you may choose the most suitable guideline of the options given below. Furthermore, ECHA also agrees with your comments that due to high volatility of the registered substance the OECD 310 is not the most suitable guideline and has removed OECD 310 as an option from this draft decision.

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:

Ready biodegradability (Annex VII, 9.2.1.1.; test method: MITI test (I), OECD 301C).
or
Ready biodegradability (Annex VII, 9.2.1.1.; test method: Closed bottle test, OECD 301D).
or
Ready biodegradability (Annex VII, 9.2.1.1.; test method: Manometric respirometry test, OECD 301F).

11. Simulation testing on ultimate degradation in surface water (Annex IX, Section 9.2.1.2.) and

12. Sediment simulation testing (Annex IX, Section 9.2.1.4.)

"Simulation testing on ultimate degradation in water" and "sediment simulation testing" are standard information requirements for a substance registered in quantities of 100 tonnes or more as laid down in Annex IX, sections 9.2.1.2. and 9.2.1.4 of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have not provided any study record for simulation testing on ultimate degradation in water that would meet the information requirement of Annex IX, Sections 9.2.1.2 and 9.2.1.4, for the registered substance. Instead you have sought to adapt this information requirement on the basis of ready biodegradability: *"Gum turpentine oil has been shown to be readily biodegradable. According to Column 2 of Annex IX on the standard information requirements, simulation testing on ultimate degradation in water and sediment does not need to be conducted if the substance is readily biodegradable"*.

However, as described in the previous section of the draft decision, your proposed prediction based on read-across for ready biodegradability (REACH Annex VII, 9.2.1.1) has been evaluated not to meet Annex VII and REACH Annex XI, 1.5 adaptation criteria. Therefore, information compliant with Annex VII Section 9.2.1.1 on ready biodegradability is not present in the dossier and consequently cannot be used for adaptation purposes.

Furthermore, the substance is not highly insoluble in water (water solubility = 25.5 mg/L) whilst substance uses (fragranced products, coatings and inks, solvents) do not exclude direct and/or indirect exposure of the aquatic compartment.

Given the physicochemical (LogKoc = 3.4, LogKow = 4.49), use and exposure (as above) properties of the substance, affinity of the substance and indirect exposure to the sediment compartment are also likely.

ECHA notes that due to lack of information on the degradation of the substance you have not in your Chemical Safety Assessment (CSA) or the technical dossier justified that there is no need to investigate further the degradation of the substance or its degradation products.

Thus, the justification for the adaptation provided by you does not meet the criteria of either the specific adaptation rules of Column 2 of Annex IX, section 9.2, or the general adaptation rules of Annex XI. Therefore, the adaptations cannot be accepted.

As the registered substance is a UVCB, ECHA notes that the simulation test on degradation needs to be performed for each relevant group of homologous constituents, the constituents tested being the ones deemed to be relevant for the PBT/vPvB assessment.

In your comments on the draft decision you highlighted that the Annex IX requests for simulation testing may be adapted according to the specific rules outlined in Annexes VI to X and/or according to the general rules contained in Annex XI of the REACH Regulation and that the testing requirement may depend on the outcome of the ready biodegradation study also requested in the present decision. ECHA refers to the body of the decision above and notes further that any adaptation submitted will be evaluated by ECHA during the follow-up process.

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

One of the purposes of the simulation test is to provide information relevant for assessing the persistence (P) and very persistent (vP) properties of a substance in accordance with Annex XIII of the REACH Regulation. ECHA observes that Annex XIII also indicates that "*the information used for the purposes of assessment of the PBT/vPvB properties shall be based on data obtained under relevant conditions*". The Guidance on information requirements and chemical safety assessment R.7b (version 2.0, November 2014) defines the following with respect to simulation tests: "*these tests attempt to simulate degradation in a specific environment by use of indigenous biomass, media, relevant solids [...], and a typical temperature that represents the particular environment*". The Guidance on information requirements and chemical safety assessment Chapter R.16 (version: 2.1, October 2012), Table R.16-9 indicates 12°C (285K) as the average environmental temperature to be used at EU level to conduct a chemical safety assessment. Performing the test at the temperature of 12°C is within the applicable test conditions of the Test Guideline. Therefore, the test should be performed at the temperature of 12°C.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, the Registrant is requested to submit the following information derived with the registered substance subject to the present decision:

Simulation testing on ultimate degradation in surface water (Annex IX, 9.2.1.2.; test method: Aerobic mineralisation in surface water – simulation biodegradation test, EU C.25./OECD 309), and

Sediment simulation testing (test method: Aerobic and anaerobic transformation in aquatic sediment systems, EU C.24. / OECD 308).

In both studies requested the biodegradation of each relevant constituent present in concentration at or above 0.1% (w/w) or, if not technically feasible, in concentrations as low as technically detectable shall be assessed. This can be done simultaneously during the same study.

The identification of degradation products also needs to be performed, according to REACH Annex IX, 9.2.3 specifications. You may obtain this information from the simulation study also requested in this decision, or by some other measure. You will need to provide a scientifically valid justification for the chosen method.

13. 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 for a substance registered at 100 tonnes or more 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. Moreover, Annex VII Section 9.1.1. Column 2 of the REACH Regulation states that long-term aquatic toxicity on Daphnia shall be considered if the substance is poorly water soluble.

You have not provided any study record for long-term toxicity testing on aquatic invertebrates that would meet the information requirement of Annex IX, Section 9.1.5, for the registered substance. Instead you have sought to adapt this information requirement using the following justification: "*As there is no acute toxicity above the solubility limit and as the substance is rapidly biodegradable, the substance is not classified. It seems unnecessary to perform long-term test on invertebrates*".

ECHA notes that the adaptation argumentation cannot be verified in the absence of valid acute toxicity data for the registered substance (see Appendix 1, section '*Grouping of substances and read-across approach*'). Furthermore, the provided argumentation is not in line with REACH Annex IX, 9.1 specific rules for adaptation as absence of acute toxicity does not exclude manifestation of toxicity during aquatic toxicity tests of longer exposure duration. Additionally, the parameters that are the basis for effects in the short and long-term aquatic invertebrate tests are different. Requesting long-term aquatic toxicity testing is also scientifically justified based on the registered substance's exposure potential and water solubility profile, especially in the absence of reliable information on ready biodegradation.

In your comments on the draft decision you stated that "*contrary to what was concluded in the dossier, it may be anticipated that DRT's substance exhibits acute toxic effects below its water solubility limit*" and that "*reliable experimental acute toxicity data were generated and are available for several terpenes*". However, no actual biological toxicity data is provided and no explanation is provided how the data generated relates to the registered substance. ECHA hence considers there is still no valid acute toxicity data on the registered substance, in the current dossier.

You also explain that alongside the acute *Daphnia* studies "*the physical and chemical properties of (-)-alpha-pinene and (-)-beta-pinene were studied*", and that the acute *Daphnia* studies were carried out under specific test conditions to minimise the losses of the substance during testing due to high volatility and low water solubility. You have provided analytical data to show that the substances were not maintained within 20 % of initial concentration. ECHA notes that even if in the data provided the % loss seem to have been miscalculated and are consequently overestimated, more than 20 % seem to have been lost. However, in the OECD TG 211 the following is indicated: "*If, from preliminary stability tests (see paragraph 7), the test substance concentration is not stable (i.e. outside the range 80 - 120% of nominal or falling below 80% of the measured initial concentration) over the maximum renewal period (i.e. 3 days), consideration should be given to more frequent medium renewal, or to the use of a flow -through test*". Further, the guideline provides guidance on how to express results if the substance concentrations are not satisfactorily maintained within +/- 20 % of nominal or measured initial concentration. Thus currently ECHA does not consider it justified that it is technically not possible to carry out the study. Furthermore, the stability data provided is not for the registered substance but for (-)-alpha-pinene and (-)-beta-pinene, however, no read-across justification has been provided to show how this data relates to the properties of the registered substance.

In addition, you noted that "*the fact that the substance contains more than one constituent may enhance the difficulties of analytical monitoring in water phase*". ECHA notes that this is not a valid argument to adapt the information requirement as multiconstituent/UVCB substances are not exempt from aquatic toxicity testing.

However, as the substance may be difficult to test, ECHA refers the Registrant to consult the 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 3.0, February 2016), Chapter R7b, Table R.7.8-3 summarising aquatic toxicity testing of difficult substances for choosing the design of the requested ecotoxicity test(s) and for calculation and expression of the result of the test(s).

Lastly, you have provided the following proposal for a stepwise adaptation to cover the request for long-term toxicity data to aquatic invertebrates. Firstly, you have provided QSAR data derived from Kreatis model for (-)-alpha-pinene (CAS No 7785-26-4) and beta-pinene (CAS 127-91-3) with chronic aquatic toxicity values. First, ECHA has compared the information provided with the requirements set for acceptance of QSAR models in Annex XI section 1.3 as follows:

- Adequate and reliable documentation of the applied method is provided: You have submitted a QMRF and a QPRF.

- Results are derived from a (Q)SAR model whose scientific validity has been established: You have submitted a detailed and clear QMRF. You consider that the model is based on regression, however, the precise equation has not been provided. For ECHA to decide on the scientific validity of the model also the equation would need to be submitted and assessed. It is therefore not possible to conclude on scientific validity of the model submitted.

- The substance falls within the applicability domain of the (Q)SAR model: you have explained in the QMRF the stepwise approach used to check whether a chemical falls into the applicability domain of the model and concluded in the QPRF that the registered substance is within the domain. However, as no training set has been submitted it is not possible for ECHA to conclude whether this is true.

Due to the above short-comings it is not possible to evaluate whether the results are reliable and adequate for the purpose of classification and labeling and risk assessment. In conclusion, the QSAR information submitted is currently not sufficient to fulfill the requirements of Annex XI section 1.3.

Secondly, you have indicated that you plan to carry out the OECD 211 study requested using (-)-alpha-pinene (CAS No 7785-26-4) as the source substance. You state that this substance "*shows higher toxicity*" and that "*when testing mono-constituent substance rather than multi-constituent substance, the chance to obtain a scientifically valid long-term toxicity study would be higher*". However, no data is yet available on the proposed source substance. Thirdly, you have indicated that you plan to discuss the impact of impurities on the long-term toxicity of the registered substance. However, no such discussion is provided in the comments and, therefore, it cannot be taken into account.

ECHA notes that no read-across justification has been provided for using (-)-alpha-pinene (CAS No 7785-26-4) as a source substance to the registered substance and no new information on using beta-pinene as a source substance in general and for long-term aquatic toxicity testing specifically has been provided. Hence your proposed adaptations using (-)-alpha-pinene (CAS No 7785-26-4) and beta-pinene (CAS 127-91-3) as described above do not fulfil the rules of Annex XI, section 1.5. Testing on the registered substance is hence indicated.

ECHA considers that the justification for waiving provided by the Registrant does not meet the criteria of either the specific adaptation rules of Column 2 of Annex IX, section 9.1, or the general adaptation rules of Annex XI. Therefore, the adaptations cannot be accepted and there is an information gap for the long-term toxicity testing on aquatic invertebrates (Annex IX, section 9.1.5. of the REACH Regulation). Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: *Daphnia magna* reproduction test (test method: EU C.20./OECD 211).

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

"Long-term toxicity testing on fish" is a standard information requirement for a substance registered at 100 tonnes or more 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.) needs to be present in the technical dossier for the registered substance to meet this information requirement. Moreover, Annex VIII Section 9.1.3. Column 2 of the REACH Regulation states that long-term aquatic toxicity on fish shall be considered if the substance is poorly water soluble.

You have not provided any study record of for long-term toxicity testing on fish that would meet the information requirement of Annex IX, Section 9.1.6.1, for the registered substance. Instead you have sought to adapt this information requirement using the following justification: *"As there is no acute toxicity above the solubility limit and as the substance is rapidly biodegradable, the substance is not classified. It seems unnecessary to perform long-term test on fish"*.

ECHA notes that the adaptation argumentation cannot be verified in the absence of valid acute toxicity data for the registered substance (see Appendix 1, section *'Grouping of substances and read-across approach'*). Furthermore, the provided argumentation is not in line with REACH Annex IX, 9.1 specific rules for adaptation as absence of acute toxicity does not exclude manifestation of toxicity during aquatic toxicity tests of longer exposure duration. Long-term aquatic toxicity testing is also scientifically justified based on the registered substance's exposure potential and water solubility profile, especially in the absence of reliable information on ready biodegradation.

ECHA considers that the justification for the adaptation provided by the Registrant does not meet the criteria of either the specific adaptation rules of Column 2 of Annex IX, section 9.1, or the general adaptation rules of Annex XI. Therefore, the adaptations cannot be accepted and there is an information gap for this endpoint.

Regarding the long-term toxicity testing on fish pursuant to Annex IX, section 9.1.6.1, ECHA considers that the FELS toxicity test according to OECD 210 is the most sensitive of the standard fish test available as it covers several life stages of the fish from the newly fertilised egg, through hatch to early stages of growth and should therefore be used (see ECHA Guidance on information requirements and chemical safety assessment (version 2.0, November 2014), Chapter R7b, Figure R.7.8-4). The test method OECD 210 is also the only suitable test currently available for examining the potential toxic effects of bioaccumulation (ECHA Guidance R7b, version 2.0, November 2014). For these reasons, ECHA considers the FELS toxicity test using the test method OECD 210 as appropriate and suitable.

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 (test method: OECD 210).

Notes for your consideration

According to ECHA Guidance on information requirements and chemical safety assessment (version 2.0, November 2014), Chapter R7b (Section R.7.8.5., including Figure R.7.8-4), if based on the results of the long-term Daphnia study and the application of a relevant assessment factor, no risks are observed (PEC/PNEC<1), no long-term fish testing may need to be conducted. However, if a risk is indicated, the long-term fish study needs to be conducted.

If you consider your substance as difficult to test 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 3.0, February 2016), Chapter R7b, Table R.7.8-3 summarising aquatic toxicity testing of difficult substances for choosing the design of the requested ecotoxicity test(s) and for calculation and expression of the result of the test(s).

Appendix 2: Procedural history

The compliance check was initiated on 20 October 2015.

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

ECHA took into account your comments and amended a request and the deadline.

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

ECHA received proposal(s) for amendment and modified the draft decision.

ECHA invited you to comment on the proposed amendment(s).
ECHA referred the draft decision to the Member State Committee.

Your comments on the proposed amendment(s) were taken into account by the Member State Committee.

The Member State Committee reached a unanimous agreement on the draft decision during its MSC-50 meeting and ECHA took the decision according to Article 51(6) of the REACH Regulation.

Deadline to submit the requested information in this decision

In the draft decision communicated to you, the time indicated to provide the requested information was 42 months from the date of adoption of the decision. In your comments on the draft decision, you requested an extension of the timeline to 48 months. You sought to justify this request by submitting additional evidence of scheduling provided by your selected Contract Research Organisation for the conduction of the studies and the time needed to select the representative sample. ECHA acknowledges your arguments and therefore, has granted the request and set the deadline to 48 months.

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

3. In relation to the information required by the present decision, the sample of substance used for the new test(s) must be suitable for use by all the joint registrants. Hence, the sample should have a composition that is suitable to fulfil the information requirements 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 substance tested in the new test(s) is appropriate to assess the properties of the registered substance, taking into account any variation in the composition of the technical grade of the substance as actually manufactured or imported by each registrant. If the registration of the substance by any registrant covers different grades, the sample used for the new test(s) must be suitable to assess these grades. Finally there must be adequate information on substance identity for the sample tested and the grade(s) registered to enable the relevance of the test(s) to be assessed.