ECHA
EUROPEAN CHEMICALS AGENCY

Helsinki, 9 April 2018

Addressee: [redacted]

Decision number: CCH-D-2114394631-45-01/F
Substance name: 3,7-dimethylnona-2,6-dienenitrile
EC number: 263-214-5
CAS number: 61792-11-8
Registration number: [redacted]
Submission number: [redacted]
Submission date: 27/04/2017
Registered tonnage band: 100-1000

DECISION ON A COMPLIANCE CHECK

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

1. **In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.; test method: OECD TG 476 or TG 490) with the registered substance;**

2. **Screening for reproductive/developmental toxicity (Annex VIII, Section 8.7.1.; test method: OECD 421/422) in rats, oral route with the registered substance;**

3. **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;**

4. **Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.; test method: EU B.31./OECD TG 414) in a first species (rat or rabbit), oral route with the registered substance;**

5. **Identification of degradation products (Annex IX, 9.2.3.) using an appropriate test method with the registered substance.**

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 to the REACH Regulation. 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 adequate and reliable documentation.

You have to submit the requested information in an updated registration dossier by ***16 October 2020***. You also have to update the chemical safety report, where relevant. The timeline has been set to allow for sequential testing.

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

Appeal

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

Authorised[1] by Claudio Carlon, Head of Unit, Evaluation E2

[1] As this is an electronic document, it is not physically signed. This communication has been approved according to ECHA's internal decision-approval process.

Appendix 1: Reasons

TOXICOLOGICAL INFORMATION

In accordance with Articles 10(a) and 12(1) of the REACH Regulation, a technical dossier registered at 100 to 1000 tonnes per year must contain, as a minimum, the information specified in Annexes VII to IX to the REACH Regulation. The information to be generated for the dossier must fulfil the criteria in Article 13(4) of the same regulation.

Your registration dossier contains for multiple endpoints adaptation arguments in the form of a grouping and read-across approach under Annex XI, Section 1.5. of the REACH Regulation. ECHA has considered first the scientific and regulatory validity of your read-across approach in general before assessing the individual endpoints (sections 2, 3 and 4).

Grouping of substances and read-across approach

You have sought to adapt information requirements by applying a read-across approach in accordance with Annex XI, Section 1.5, for the endpoints:

- Screening study for reproductive/developmental toxicity (Annex VIII, Section 8.7.1.);
- Sub-chronic toxicity (90-day) study (Annex IX, Section 8.6.2.); and
- Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.).

According to Annex XI, Section 1.5., two conditions shall be necessarily fulfilled. Firstly, there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category. Secondly, it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (read-across approach). ECHA considers that the generation of information by such alternative means should offer equivalence to prescribed tests or test methods.

Based on the above, a read-across hypothesis needs to be provided. This hypothesis establishes why a prediction for a toxicological or ecotoxicological property is reliable and should be based on recognition of the structural similarities and differences between the source and registered substances[2]. This hypothesis explains why the differences in the chemical structures should not influence the toxicological/ ecotoxicological properties or should do so in a regular pattern. The read-across approach must be justified scientifically and documented thoroughly, also taking into account the differences in the chemical structures. There may be several lines of supporting evidence used to justify the read-across hypothesis, with the aim of strengthening the case.

Due to the different nature of each endpoint and consequent difference in scientific considerations (e.g. key parameters, biological targets), a read-across must be specific to the endpoint or property under consideration. Key physicochemical properties may determine the fate of a compound, its partitioning into a specific phase or compartment and largely influence the availability of compounds to organisms, e.g. in bioaccumulation and toxicity tests.
Similarly, biotic and abiotic degradation may alter the fate and bioavailability of compounds as well as be themselves hazardous, bioaccumulative and/or persistent. Thus, physicochemical and degradation properties influence the human health and environmental

[2] Please see for further information ECHA *Guidance on information requirements and chemical safety assessment* (version 1, May 2008), Chapter R.6: QSARs and grouping of chemicals.

properties of a substance and should be considered in read-across assessments. However, the information on physicochemical and degradation properties is only a part of the read-across hypothesis, and it is necessary to provide additional justification which is specific to the endpoint or property under consideration.

The ECHA Read-across assessment framework foresees that there are two options which may form the basis of the read-across hypothesis[3]- (1) (Bio)transformation to common compound(s)- the read-across hypothesis is that different substances give rise to (the same) common compounds to which the organism is exposed and (2) Different compounds have the same type of effect(s)- the read-across hypothesis is that the organism is exposed to different compounds which have similar (eco)toxicological and fate properties as a result of structural similarity (and not as a result of exposure to common compounds).

Finally, Annex XI, Section 1.5. lists several additional requirements, which deal with the quality of the studies which are to be read-across.

You consider to achieve compliance with the REACH information requirements for the registered substance 3,7-dimethylnona-2,6-dienenitrile using data of structurally similar substances citronellyl nitrile (EC No 257-288-8) (hereafter the 'source substance').

You have provided a read-across documentation as a separate attachment in the endpoint summaries.

You use an analogue approach and the read-across hypothesis is based on different compounds which have the same types of effects.

You use the following arguments to support the prediction of properties of the registered substance from data for source substances within the group:

i. The target and source substances have "*similar human health profiles as a result of structural similarity, the same expected mode of action and similar physicochemical properties.*"
    a. According to a profiling carried out using the OECD(Q)SAR toolbox, both the target and source substances "*share structural similarities and also mechanistic actions which are both general and endpoint specific*", hence "*this supports the hypothesis that the target and source substances have similar properties as a result of structural similarity and the same expected mode of action.*" Both substances "*have identical profiling alerts for repeated dose toxicity and developmental and reproductive toxicology*".
    b. You also claim that there are common metabolic pathways: "*the primary route of metabolism for both substances is expected to be initially via epoxidation, epoxide hydration then either aliphatic C-oxidation or O-Glucuronidation*", based on a prediction of TIMES v.2.27.17 (rat in vivo model).

ii. You also provide "*additional supporting data*" from seven "*additional potential source substances*".
    a. The seven source category members "*have >46% structural similarities*" with the target substance. Hence you conclude that "*This high degree of structural similarity increases the confidence...that this category will react in a similar manner in both an in vitro and in vivo test system.*"
    b. Moreover, according to the OECD (Q)SAR toolbox profiling you claim that

[3] Please see ECHA's Read-Across Assessment Framework (https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across).

"*there is no evidence*", that there may be "*concern for repeated dose toxicity or reproductive and/or developmental toxicity based on the available toxicology data*".

As an integral part of this prediction, you propose that the source and registered substance(s) have (i) "*structural similarities*" and (ii) "*common metabolic pathways*" where both substances "*act via the same mode of action*" for the above-mentioned information requirements. ECHA considers that this information is your read-across hypothesis.

ECHA's evaluation and conclusion

(i) "*structural similarities*":

Your proposed adaptation argument is that the similarity in chemical structure and in some of the physico-chemical between the source and registered substance is a sufficient basis for predicting the properties of the registered substance for other endpoints. Structural similarity is a prerequisite for applying the grouping and read-across approach. However similarity in chemical structure and similarity of some of the physico-chemical / toxicological properties does not necessarily lead to predictable or similar human health properties in other endpoints.

ECHA acknowledges that the source substance is structurally similar to the target substance. However, there are also structural differences that you have not accounted for: the target substance has an extra saturated bond to the carbon linked to the nitrile group and on the opposite end to the nitrile group the target substance has a methyl and ethyl group whereas the source substance has two methyl groups. Hence, you fail to present these differences and to explain why these differences in structure allow the possibility to predict similar properties.

This point also applies for the category members used as additional support to the analogue approach. For categories, the read-across hypothesis has to cover the structural variations of the members in order to support a prediction for one member of the category. Furthermore, ECHA notes that one of the category members, 2,6-octadienenitrile,3,7-dimethyl-,(e)- (EC No 226-982-2), is structurally more similar to the target substance than the actual source substance, however you do not provide any explanation why this substance was not used as a source substance in the analogue approach.

Additionally, ECHA notes that the target substance is a multi-constituent (four isomers) however the source substance is a mono-constituent. The additional structures of the target substance and their concentration variations may impact the structural similarity as well as the qualitative and quantitative predictions. Currently, your read-across assessment fails to take such considerations into account.

In the absence of any analysis of the impact of these similarities or dissimilarities on the endpoints concerned, your justification based on structural similarity and similar physico-chemical properties has not established why the prediction is reliable for the human health end-points for which the read across is claimed.

(ii) "*common metabolic pathways*" where both substances "*act via the same mode of action*":

As regards the mechanistic explanations you have provided information on the probable route of metabolism based on prediction using "TIMES v.2.27.17". However you failed to

provide additional supporting information and evidence to substantiate this prediction, such as experimentally-derived information on toxicokinetics (e.g. hydrolysis/metabolism). In the absence of such toxicokinetic information, ECHA considers that the predictions of metabolism are unreliable and do not provide material support for your hypothesis.

As described above, to justify the predictability of the human health properties, you have proposed that there are 'mechanistic actions' similarities, and that this is a basis for predicting the properties of the registered substance. The 'mechanistic action' similarities in Table 1 are listed under the various headings, including "*Estrogen receptor binding*" and "*Dart scheme v. 1.0*". Under the latter you indicate the following: "*not known precedent reproductive and developmental toxic potential*". You also claim that according to the DART scheme "*both substances show no alerts*". However you do not provide an explanation of the methodology for performing the assessment. ECHA is unable to assess this information. Consequently, ECHA considers that this material provides no support for your claim that "*the target and source substances have similar properties as a result of structural similarity and the same expected mode of action*" enable the prediction of relevant human health properties. ECHA considers that there is a failure of adequate and reliable documentation.

With reference to the data matrix provided for the toxicological endpoints ECHA notes that the information does not allow comparison of toxicological profiles of the target and the source substances since with the registered substance there is only one higher tier study available (*in vivo* micronucleus test).

Finally, ECHA has taken into account all of your arguments together. ECHA firstly notes that you have not provided a reasoning as to why these arguments add to one another to provide sufficient basis for read-across. Secondly, the defects of each individual argument indicated above are not mitigated by the other arguments you have provided, and so ECHA considers that the arguments when taken all together do not provide a reliable basis for predicting the properties of the registered substance.

In your comments to the draft decision you indicate your disagreement with ECHA that the target and source substances are dissimilar. You state that "*considering the molecular weight, log_{kow}, consistent profiling for repeated dose toxicity and reproductive endpoints, acute oral and dermal toxicity and negative in vivo micronucleus data the data are consistent across the 2 substances.*" You also claim that "*the 6 constituents of lemonile*" (target substance) "*and the 7 structurally similar source substances share structural similarities and also mechanistic actions which are both general and endpoint specific. This supports the hypothesis that the target (including the 6 constituents) and 7 source substances have similar properties as a result of structural similarity and the same expected mode of action.*"

ECHA notes that for the six constituents of the target substance and the potential seven source substances you have used Cramer and DART (standing for Developmental and Reproductive Toxicity) schemes of the OECD QSAR Toolbox. ECHA notes that the Cramer scheme is just a very preliminary screening tool, and the DART scheme is not exhaustive with respect to potential presence or absence of effects, especially it is not intended to address the repeated dose toxicity (RDT). The lack of alert in the alert-based system is not considered as a lack of activity because the underlying knowledge might have been limited when developing the alert-based system. Furthermore, the "Repeated dose (HESS)" profiler in the Toolbox, which is relevant for the repeated dose toxicity endpoint, indicates that the substance is hepatotoxic. Even if commented, results from HESS profiler are not included in the data matrix for further considerations.

Additionally, ECHA notes that the structural similarity values for selecting the analogues are low in all cases, which resulted in collection of generally not relevant chemicals for structure-activity relationships. Only hydrolysis simulators within the Toolbox were considered, while metabolic simulators within the same software (e.g. "rat S9 liver simulator") were not mentioned. By running those additional metabolic simulators, more metabolites of possible concern were found (triggering structural alerts for DNA binding, carcinogenicity, DART, *in vitro* and *in vivo* mutagenicity), which you failed to discuss.

The (Q)SAR predictions for the different endpoints do not add evidence to your claim because of the following:

i. It is not clear why the NOAEL should be linearly correlated with the log K_{ow} of the compounds;
ii. The experimental endpoints and test materials are not clearly defined; and
iii. The number of analogues is small and not sufficient to derive a robust and statistically significant trends.

Hence the results from the (Q)SARs provided, as part of the additional information provided in your comments, cannot be used instead of the required test as they are not considered as being scientifically valid and adequate for the purpose of risk assessment (Annex XI, Section 1.3.).

Additionally, your proposed adaptation argument is that the toxicological similarity between the source and registered substance in one or multiple endpoints is a sufficient basis for predicting the properties of the registered substance for other (eco)toxicological endpoints. (Note that ECHA does not accept that you have adequately characterised the toxicological properties of these substances, as stated above). However, toxicological similarity in one or multiple endpoints does not necessarily lead to predictable or similar human health properties in other endpoints. You have not established why a prediction for a human health property is reliable. Thus toxicological similarity on certain endpoints is not sufficient to enable the prediction of other human health properties of a substance.

Therefore, ECHA considers that this grouping and read-across approach does not provide a reliable basis whereby the human health effects of the registered substance may be predicted from data for reference substance(s) within the group. Hence, this approach does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. of the REACH Regulation.

ECHA notes that there are specific considerations for the pre-natal developmental toxicity study endpoint which also results in a failure to meet the requirement of Annex XI, Section 1.5. These are set out below in this statement of reasons under the endpoint concerned.

As described above, further elements are needed to establish a reliable prediction for a toxicological or ecotoxicological property, based on recognition of the structural similarities and differences between the source and registered substances. This could be achieved (if it is possible) by a well-founded hypothesis of (bio)transformation to a common compound(s), or that the registered and source substance(s) have the same type of effect(s), together with sufficient supporting information to allow a prediction of human health properties.

1. 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 contains negative results for both these information requirements. Therefore, adequate information *on in vitro* gene mutation in mammalian cells 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 mammalian cells in the dossier that would meet the information requirement of Annex VIII, Section 8.4.3.

You have sought to adapt this information requirement according to Annex VIII, Section 8.4.3., column 2. You provided the following justification for the adaptation "*According to Regulation (EC) No. 1907/2006, Annex VIII, section 8.4.3, an in vitro gene mutation study in mammalian cells is not required if negative results are obtained in Annex VII, Section 8.4.1. and Annex VIII, Section 8.4.2. Since adequate data from reliable in vivo mammalian gene mutation tests are available, testing for this endpoint is not required.*"

However, ECHA notes that your adaptation does not meet the specific rules for adaptation of Annex IX, Section 8.7.2., column 2 because the *in vivo* study available in the dossier is a micronucleus assay and hence the study addresses chromosome aberrations not gene mutations. Consequently, as per Annex VIII, Section 8.4.3., column 2, the *in vitro* gene mutation study needs to be conducted since there is no adequate data from a reliable *in vivo* mammalian gene mutation test available in the technical dossier.

In your comments to the draft decision you indicate that "*given the justified read-across proposed in its registration dossier, the registrant disagrees with the need to conduct the requested study*". However, ECHA notes that for this particular endpoint you have not sought to adapt this information requirement by applying a read-across approach.

Therefore, your adaptation of the information requirement is rejected. Consequently there is an information gap and it is necessary to provide information for this endpoint.

ECHA considers that the *in vitro* mammalian cell gene mutation tests using the *Hprt* and *xprt* genes (OECD TG 476) and the *in vitro* mammalian cell gene mutation tests using the thymidine kinase gene (OECD TG 490) are appropriate to address the standard information requirement of Annex VIII, Section 8.4.3.

In your comments to the draft decision you have also indicated that if you have to perform this study you would choose either OECD TG 476 or OECD TG 490.

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: OECD TG 476 *or* OECD TG 490).

2. Screening for reproductive/developmental toxicity (Annex VIII, Section 8.7.1.)

"Screening for reproductive/developmental toxicity" (test method OECD TG 421 or 422) is a standard information requirement as laid down in Annex VIII, Section 8.7.1. of the REACH Regulation if there is no evidence from available information on structurally related substances, from (Q)SAR estimates or from *in vitro* methods that the substance may be a developmental toxicant. No such evidence is presented in the dossier. Therefore, 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 screening for reproductive/developmental toxicity in the dossier that would meet the information requirement of Annex VIII, Section 8.7.1.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing a study record for a one-generation reproductive toxicity study (OECD TG 415) with the analogue substance(s) citronellyl nitrile (EC no 257-288-8). You also provide the following justification: "*Data is available from a one generation reproductive toxicity study conducted on a structurally similar substance. On the grounds of animal welfare it is therefore considered unnecessary to perform a substance specific screening test on vertebrate animals.*" However, as explained above in Appendix 1 of this decision, under the section of *Grouping of substances and read-across approach*, your adaptation of the information requirement is rejected.

In your comments on the draft decision you have indicated that the "*available experimental data on the 6 source substances... confirm a lack of reproductive toxicity via the oral route.*" However, as already highlighted under the *Grouping and read-across approach* section, your read-across adaptation is rejected. Moreover, ECHA notes that in the technical dossier you have only provided a study record with the analogue substance citronellyl nitrile (EC no 257-288-8), hence the study quality of the OECD TG 421 and 422 studies with the other source substances cannot be assessed. Although the Registrant says that "the data is attached in Appendix I, Tables 5-7", Appendix I, Tables 5-7 contain minimal summary data, and this information is not detailed summary of the objectives, methods, results and conclusions of a full study report providing sufficient information to make an independent assessment of the study minimising the need to consult the full study report. ECHA cannot therefore evaluate the information in these studies.

As additional weight of evidence, in your comments you have also provided two QSAR toolbox reports addressing OECD TG 422_Combined repeated dose toxicity and reproductive/development screening toxicity study for reproductive toxicity endpoint. One QSAR toolbox report simulates read-across and the other QSAR toolbox report documents a QSAR (trend) prediction. Both results are considered not to be acceptable for the purpose of data gap filling. As a general note, reproductive and repeated dose toxicities are considered complex endpoints, which for a number of reasons are not predictable by computational methods, mainly because of exposure duration and due to the internal organism phase, which are critical for the results. There is a failure to provide adequate and reliable documentation, as per Annex XI, 1.3 and 1.5, specifically: The quality of data is unknown because as collected from ECHA Chem, it is not systematically checked (e.g. the indication for reliability is missing in the Toolbox report). Test materials for the source studies are not analysed in the provided report. As your read-across approach and trend analysis are relying on very few chemicals, you should also be able to demonstrate that you have access to the robust study summary of the source studies. Moreover, critical effects are to be known from this test however you failed to provide such information.

Additionally, ECHA notes that the results obtained from the (Q)SARs provided are not scientifically valid because of the following issues:

i. The read-across approach is applied for the OECD TG 422, Combined repeated dose toxicity and reproductive/development screening toxicity study. The selected source

substances are not considered similar to allow prediction of the unknown toxicity. The selected source substances (octene, 1-octene, linalyl acetate, and citral) are not considered similar to the target substance. According to the RAAF, initial similarity needs to be established between the target and the source substances. Ideally, the group of substances should share a common mechanism of action. It is noted that the target is prone to Michael-type addition reactions and indeed is predicted of high toxicity by the Cramer scheme, while the selected source substances are of low toxicity. Additional considerations are needed to assess whether the substance itself or its metabolic products might be toxic. Statistically, it is not appropriate to apply an average operator when the dependent variable varies for more than one log unit. In the proposed approach, refinement is not considered possible because there is only a small number of "analogues". Moreover from the selected source substances there is no analogue with the nitrile group, as the target substance. ECHA notes that the nitrile group is driving the Michael-type reactivity resulting in higher toxicity.

ii. The trend analysis is applied for OECD TG 422, Combined repeated dose toxicity and reproductive/development screening toxicity study. The model is not scientifically valid mainly because of the following issues: unknown type and quality of the endpoint data; missing analysis of the critical effects; the applicability domain is arbitrary to the endpoint of reproductive toxicity; and the analogues have been selected by very low threshold of similarity: the analogues (linalyl acetate, citral, (z)-3,7-dimethylocta-2,6-dienal, and linool) are of various mechanisms of action and are missing the nitrile group. The model is not statistically valid since it is lacking normal distribution of data points, it is a chance correlation between a dot and a cluster, and four points are insufficient for deriving a statistically significant model. The errors of the coefficients are higher than the coefficients themselves which indicates unstable model without sufficient statistical predictivity.

iii. The model is not statistically valid to allow consideration of applicability domain. The large confidence intervals in the toxicity/log K_{ow} plot are the result of poor statistical performance and could not be used as indication of models applicability domain.

iv. The predictions are not adequate for the purpose of data gap filling for risk assessment, PBT assessment, and classification and labelling assessment for all the above-mentioned reasons.

v. The documentation is not sufficient with regards to the dependent variable, its source, type and quality.

As a consequence, the results of the (Q)SARs provided cannot be used instead of testing since the conditions set out in Annex XI, Section 1.3. are not met.

You have argued "For both workers and consumers, no RCRs were above 1. Therefore, safe use was demonstrated..." ECHA notes that RCRs below 1 are not a valid adaptation according to Annex VIII, 8.7.1, column 2 or Annex XI.
As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the information requirement. Consequently there is an information gap and it is necessary to provide information for this endpoint.

According to the test methods OECD TG 421/422, the test is designed for use with rats. On the basis of this default assumption ECHA considers testing should be performed with rats.

ECHA considers that the oral route is the most appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.6.2.3.2. Since the substance to be tested is a liquid, ECHA concludes that testing should be performed by the oral route.

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:
- Reproductive/developmental toxicity screening test (test method: OECD TG 421) *or* Combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (test method: OECD TG 422) in rats by the oral route.

Note for your consideration

For the selection of the appropriate test, please consult ECHA *Guidance on information requirements and chemical safety assessment*, Chapter R.7a, Section R.7.5 and 7.6 (version 6.0, July 2017). You should also carefully consider the order of testing especially the requested screening (OECD TG 421/422) and the developmental toxicity study (OECD TG 414) to ensure unnecessary animal testing is avoided, paying particular attention to ECHA's end point specific guidance document[4].

3. 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.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing a study record for a sub-chronic toxicity (90-day) study (OECD TG 408) with the analogue substance citronellyl nitrile (EC no 257-288-8). However, as explained above in Appendix 1 of this decision, under the section of *Grouping of substances and read-across approach*, your adaptation of the information requirement is rejected.

In your comments to the draft decision you made reference to the NOAEL values obtained for three of the source substances. However, as already highlighted under the *Grouping and read-across approach* section, your read-across adaptation is rejected. Moreover, ECHA notes that you have only provided a study record with the analogue substance, citronellyl nitrile (EC no 257-288-8) in the technical dossier. Hence the quality of the studies with the other source substances cannot be assessed.

You have also provided, as additional weight of evidence and "*to support its lack of repeated dose toxicity concern*", three QSARs: one for OECD TG 407 (28-Day) and two for OECD TG 408 (90-Day) tests. ECHA notes that for the sub-acute and sub-chronic oral toxicity endpoints you have developed your own QSAR models with the help of the OECD QSAR Toolbox.

[4] ECHA Guidance on Information Requirements and Chemical Safety Assessment, Chapter R.7a: Endpoint specific guidance Version 6.0, July 2017, R.7.6.2.3.2, p 486.

Sub-acute oral toxicity:
The analogues have been selected by very low threshold of similarity, as explained above in section 2, Appendix 1 of this decision. ECHA considers that the analogues (octane, 1-octene, alkenes, C8-10, C9 rich, 1-hexene, d-limonene, and 1-ethylpiperidine) are not similar to the substance and are missing the nitrile group. Only one of the analogues has the nitrile group (2-phenylhexanenitrile) which is nevertheless aromatic and triggers alerts for high toxicity.

ECHA notes that the results obtained from the (Q)SAR provided are not valid because of the following issues:

i. The model is not statistically valid since it is lacking normal distribution of data points and while the regression coefficient is nevertheless low, there is no indication for mechanistic similarity.
i. The model is not statistically valid to allow consideration of applicability domain. The large confidence intervals in the toxicity/log K_{ow} plot are result of poor statistical performance and cannot be used as indication for models applicability domain.
ii. The predictions are not adequate for the purpose of data gap filling for risk assessment, PBT assessment, and classification and labelling assessment for all above-mentioned reasons.
iii. Documentation is not sufficient with regards to the dependent variable, its source, type and quality.

As a consequence, the results of the (Q)SARs provided cannot be used instead of testing since the conditions set out in Annex XI, Section 1.3. are not met.

Sub-chronic repeated dose toxicity:

i. ECHA notes that there are two models, with minor differences, for the sub-chronic repeated dose toxicity. ECHA notes that the results obtained from these two models are not valid because of the following issues: The models are not scientifically valid because the endpoint is a mixture of species and protocols, the applicability domain is arbitrary to the endpoint of repeated dose toxicity, and the analogues have been selected by very low threshold of similarity: the analogues (octene, 1-octene, 1-hexene, d-limonene, beta-mycrene) are not similar to the substance and are missing the nitrile group. Acrylonitrile is the only analogue that contains the nitrile group, nevertheless, it triggers alerts for high toxicity.

ii. In the second model 2-methylimidazole was used instead of acrylonitrile. Indeed, these two chemicals are a bit more toxic then the rest of the selection, similarity still being arguable, and they create an artificial correlation (correlation between a dot and a cluster), which does not provide a statistically valid model.

iii. The model is not statistically valid since it is lacking normal distribution of data points: ECHA considers that it is a chance correlation between a dot and a cluster, and that there is no indication for mechanistic similarity (refer to comments about the Cramer and DART schemes under the *Grouping and read-across approach* section above). In addition, both schemes are not relevant for the endpoint of repeated dose toxicity.
iv. The model is not statistically valid to allow consideration of applicability domain. The large confidence intervals in the toxicity/log K_{ow} plot are result of poor statistical performance and could not be used as indication for models applicability domain.
v. The predictions are not adequate for the purpose of data gap filling for risk assessment, PBT assessment, and classification and labelling assessment for all

above-mentioned reasons.

vi. Documentation is not sufficient with regards to the dependent variable, its source, type and quality.

As a consequence, the results of the (Q)SARs provided cannot be used instead of testing since the conditions set out in Annex XI, Section 1.3. are not met.

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

ECHA has evaluated the most appropriate route of administration for the study. Based on the information provided in the technical dossier and/or in the chemical safety report, ECHA considers that the oral route - which is the preferred one as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.5.4.3 - is the most appropriate route of administration. More specifically, the substance is a liquid of very low vapour pressure. Uses with industrial, professional, and consumer spray application are reported in the chemical safety report. However, the reported concentrations are low (<10%). Hence, the test shall be performed by the oral route using the test method EU B.26./OECD TG 408.

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.

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: Repeated dose 90-day oral toxicity study (test method: EU B.26./OECD TG 408) in rats.

4. Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.) in a first species

A "pre-natal developmental toxicity study" (test method EU B.31./OECD TG 414) 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.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing a study record for a one-generation reproductive toxicity study (OECD TG 415) with the analogue substance citronellyl nitrile (EC no 257-288-8). You provided the following justification: "*In a higher tier One-Generation reproduction toxicity study (OECD Guideline 415, Klimisch 1) on the test substance Citronella Nitrile…There was no evidence of adverse treatment related deaths on parental male rats…In addition, acute oral toxicity testing (LD50 = 2000 mg/kg/day), acute dermal toxicity testing (LD50 >5000 mg/kg/day) and the sub-chronic toxicity study (NOAEL 2000 mg/kg/day) on the test substance Hypo-Lem […] confirms that Lemonile will be of very low toxicity. The expected route of exposure to this substance is considered to be dermal rather than oral or inhalation which suggests that systemic exposure via the dermal route would be*

very low."

However, as explained above in Appendix 1 of this decision, under the section of *Grouping of substances and read-across approach*, your adaptation of the information requirement is rejected. Moreover, the pre-natal developmental study cannot be waived on the basis of an existing fertility study with the analogue substance since the one-generation reproductive toxicity study refers to toxicity to reproduction and it does not specifically address the key parameters which are investigated in the developmental toxicity endpoint (for example, examinations of foetuses for skeletal and visceral alterations). Therefore this study fails to meet the requirement of Annex XI, 1.5, that there should be adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3).

Additionally, ECHA notes that you are also explicitly trying to adapt this information requirement according to Annex IX, Section 8.7., column 2, third sub-paragraph. In your adaptation you claim that the acute toxicity studies and 90-day study with the source substances confirm that the registered substance "*will be of very low toxicity*". However, ECHA notes that your adaptation does not meet the specific rules for adaptation of Annex IX, Section 8.7., column 2, third sub-paragraph, because the cumulative conditions of the adaptation requirement, whereby the study does not need to be conducted if *"(i) the substance is of low toxicological activity (no evidence of toxicity seen in any of the tests available), (ii) it can be proven from toxicokinetic data that no systemic absorption occurs via relevant routes of exposure, and (iii) there is no or no significant human exposure*", are not met.

ECHA notes the following observations:

(i) In the technical dossier, there are no 28-day or 90-day repeated dose toxicity studies with the registered substance to determine the absence/presence of toxicity of the registered substance.

(ii) According to the toxicokinetics endpoint there are no specific studies on the absorption, distribution, metabolism and elimination (ADME). In the Endpoint summary for Toxicokinetics, metabolism and distribution, in the 'Additional information section', you state that *"Lemonile is a small organic molecule and the physico-chemical properties suggest it is likely to be absorbed via dermal, inhalation and gastric routes following exposure. Acute and subchronic toxicity data indicate that Lemonile is absorbed following administration by gavage and metabolised by the liver."* Hence, condition (ii) of Annex IX, Section 8.7.2., column 2, third sub-paragraph is not met.

(iii) The registered substance has a significant exposure to both professionals (PROCS 4/8/10/11/13) and consumers (PC 35) and it may be found in sprays and aerosols. Hence condition (iii) of Annex IX, Section 8.7., column 2, third sub-paragraph is not met.

In view of the above observations, two of the three cumulative conditions are not met. Therefore, your adaptation of the information requirement is rejected.

In your comments to the draft decision you indicated that "*given the justified read-across proposed in its registration dossier, the registrant disagrees with the need to conduct the requested study*". However, you also indicate that if you "*would [...] have to perform*" the study, you will conduct "*a Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.) in a first species.*"

As explained above, ECHA has rejected your read-across approach and considers that the pre-natal developmental toxicity study cannot be waived with an existing fertility study,

namely the one-generation reproductive toxicity study performed with the analogue substance, citronellyl nitrile (EC no 257-288-8).

Hence, 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.

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 assumption ECHA considers testing should be performed with rats or rabbits as a first species.

ECHA considers that the oral route is the most appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.6.2.3.2. Since the substance to be tested is a liquid, ECHA concludes that testing should be performed by the oral route.

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 (rat or rabbit) by the oral route.

5. Identification of degradation products (Annex IX, 9.2.3.)

In accordance with Articles 10(a) and 12(1) of the REACH Regulation, a technical dossier registered at 100 to 1000 tonnes per year must contain, as a minimum, the information specified in Annexes VII to IX to the REACH Regulation. The information to be generated for the dossier must fulfil the criteria in Article 13(4) of the same regulation.

The identification of the degradation products is a standard information requirement according to column 1, Section 9.2.3. of Annex IX 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.
The biodegradation section in the technical dossier does not contain any information in relation to the identification of degradation products, nor an adaptation in accordance with column 2 of Annex IX, Sections 9.2 or 9.2.3. or with the general rules of Annex XI for this standard information requirement. "

According to Annex IX, Section 9.2.3., column 2 of the REACH Regulation, identification of degradation products is not needed if the substance is readily biodegradable. ECHA notes that based on the information in the technical dossier, the registered substance is not readily biodegradable in a ready biodegradability test according to OECD 301F.

Furthermore, ECHA notes that you have not provided any justification in your chemical safety assessment (CSA) or in the technical dossier for why there is no need to provide information on the degradation products. ECHA considers that this information is needed in relation to the PBT/vPvB assessment.

In your comments on the draft decision you proposed to address this information requirement by providing further justifications on the likelihood of ultimate biodegradation and low likelihood of degradation products with PBT properties.

ECHA welcomes the inclusion of such justifications in the registration dossier. ECHA agrees that given the results of the extended ready biodegradability studies and the low bioaccumulation potential of the parent substance (log K_{ow} 3.1 - 3.2), together with the consideration in your comments to the draft decision on the potential degradation products there is a low likelihood of bio-accumulative degradation products.

In summary, it is unlikely that the substance or its degradation products would fulfil the criteria set in Annex XIII for PBT/vPvB substances. However, at this point this information is not available in the technical dossier. The information you provided in your comments will evaluated in detail after the deadline to submit the information has passed.

As explained above, the information provided 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 this endpoint.

Regarding appropriate and suitable test method, the methods will have to be substance-specific. When analytically possible, identification, stability, behaviour, molar quantity of metabolites relative to the parent compound should be evaluated. In addition, degradation half-life, log Kow and potential toxicity of the metabolite may be investigated. You will need to provide a scientifically valid justification for the chosen method.

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:

Identification of the degradation products (Annex IX, Section 9.2.3.) by using an appropriate and suitable test method, as explained above in this section.

Note for your consideration

Before providing the above information you are advised to consult the ECHA *Guidance on information requirements and chemical safety assessment* (version 4.0, June 2017), Chapter R.7b., Sections R.7.9.2.3 and R.7.9.4.

Appendix 2: Procedural history

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

The compliance check was initiated on 08 May 2017.

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

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

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

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

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

Appendix 3: Further information, observations and technical guidance

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

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

3. In carrying out the tests required by the present decision, it is important to ensure that the particular sample of substance tested 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. If the registration of the substance covers different grades, the sample used for the new tests must be suitable to assess these.

   Furthermore, there must be adequate information on substance identity for the sample tested and the grades registered to enable the relevance of the tests to be assessed.