EUROPEAN CHEMICALS AGENCY

Decision number: CCH-D-2114355524-49-01/F Helsinki, 7 April 2017

**DECISION ON A COMPLIANCE CHECK OF A REGISTRATION PURSUANT TO ARTICLE 41(3) OF REGULATION (EC) NO 1907/2006**

**For 3-(4-tert-butylphenyl)propionaldehyde, EC No 242-016-2 (CAS No 18127-01-0), registration number:** [redacted]

**Addressee:** [redacted]

The European Chemicals Agency (ECHA) has taken the following decision in accordance with the procedure set out in Articles 50 and 51 of Regulation (EC) No 1907/2006 concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH Regulation).

I. Procedure

Pursuant to Article 41(1) of the REACH Regulation ECHA has performed a compliance check of the registration for 3-(4-tert-butylphenyl)propionaldehyde, EC No 242-016-2 (CAS No 18127-01-0), submitted by [redacted] (Registrant).

This decision is based on the registration as submitted with submission number [redacted], for the tonnage band of 10-100 tonnes per year.

This decision does not take into account any updates after 19 January 2016.

The substance subject to the present decision is provisionally listed in the Community rolling action plan (CoRAP) for start of substance evaluation in 2017.

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

The compliance check was initiated on 1 September 2015.

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

ECHA took into account your comments submitted thereupon, and these are reflected in the Section III, statement of reasons.

You were notified that the draft decision does not take into account any updates after 02 November 2015. You updated your registration with submission number [redacted] on 19 January 2016. In your update the tonnage band was changed from 100-1000 tonnes per year to 10-100 tonnes per year. At this tonnage level the originally requested 90 day sub-chronic toxicity study is no longer required and has been removed from this draft decision. Given the exceptional circumstances of the change in tonnage band (no more need for subchronic toxicity test), ECHA has taken into account the update of 19 January 2016 when processing this decision.

On 19 January 2017 ECHA notified the Competent Authorities of the Member States of its draft decision and invited them pursuant to Article 51(1) of the REACH Regulation to submit proposals for amendment of the draft decision within 30 days of the receipt of the notification.

As no proposal for amendment was submitted, ECHA took the decision pursuant to Article 51(3) of the REACH Regulation.

II. Information required

**A. Information in the technical dossier derived from the application of Annexes VII to XI**

Pursuant to Articles 41(1), 41(3), 10(a)(vi) and/or (vii), 12(1)(d), 13 and Annexes VII, VIII, IX of the REACH Regulation the Registrant shall submit the following information using the indicated test methods and the registered substance subject to the present decision:

1. 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) using one of the following strains: E. coli WP2 uvrA, or E. coli WP2 uvrA (pKM101), or S. typhimurium TA102, as specified in section III.A.3 below;
2. In vitro cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2., test method: OECD TG 473) or in vitro micronucleus study (Annex VIII, Section 8.4.2., test method: OECD TG 487);
3. Screening study for reproductive/developmental toxicity (Annex VIII, Sections 8.6.1 and 8.7.1.; test method: OECD TG 422) in rats, oral route; and
4. Provided that both studies requested under 1. and 2. have negative results; In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.; test method: OECD TG 476 or OECD TG 490),

Note for consideration by you:

*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 to and conforming with the appropriate rules in the respective Annex, and an adequate and reliable documentation.*

*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 the Member States.*

**B. Deadline for submitting the required information**

Pursuant to Articles 41(4) and 22(2) of the REACH Regulation you shall submit to ECHA by **16 April 2018** an update of the registration dossier containing the information required by this decision, including, where relevant, an update of the Chemical Safety Report. The timeline has been set to allow for sequential testing as appropriate.

III. Statement of reasons

Pursuant to Article 41(3) of the REACH Regulation, ECHA may require the Registrant to submit any information needed to bring the registration into compliance with the relevant information requirements.

EUROPEAN CHEMICALS AGENCY

## A. Information in the technical dossier derived from the application of Annexes VII to XI

Pursuant to Articles 10(a)(vi) and/or (vii), 12(1)(d) of the REACH Regulation, a technical dossier for a substance manufactured or imported by the Registrant in quantities of 10 to 100 tonnes per year shall contain as a minimum the information specified in Annexes VII to VIII of the REACH Regulation.

### 1. 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.

According to Article 13(3) of the REACH Regulation, tests required to generate information on intrinsic properties of substances shall be conducted in accordance with the test methods recognised by the Commission or ECHA.

Section 1.1.2 of Annex XI provides that existing data on human health properties from experiments not carried out according to GLP or the test methods referred to in Article 13(3) may be used if the following conditions are met:

1. Adequacy for the purpose of classification and labelling and/or risk assessment;
2. Adequate and reliable coverage of the key parameters foreseen to be investigated in the corresponding test methods referred to in Article 13(3);
3. Exposure duration comparable to or longer than the corresponding test methods referred to in Article 13(3) if exposure duration is a relevant parameter; and
4. Adequate and reliable documentation of the study is provided.

According to paragraph 13 of the current OECD 471 test guideline (updated 1997) at least five strains of bacteria should be used. These should include four strains of S. typhimurium (TA1535; TA1537 or TA97a or TA97; TA98; and TA100) that have been shown to be reliable and reproducibly responsive between laboratories. These four S. typhimurium strains have GC base pairs at the primary reversion site and it is known that they may not detect certain oxidising mutagens, cross-linking agents and hydrazines. Such substances may be detected by E.coli WP2 strains or S. typhimurium TA102 which have an AT base pair at the primary reversion site.

You have provided an Ames study with an assigned reliability score of 1 on the registered substance and an additional Ames test reported with a reliability 4 conducted with Florhydral. The tests used five different strains of S. typhimurium TA 1535, TA 1537, TA 1538, TA 98 and TA 100. However, since the test was conducted, significant changes have been made to OECD guideline 471, including new strains to allow for the coverage of new key parameters, the detection of certain oxidising mutagens, cross-linking agents and hydrazines. This means that the study does not meet the current guidelines, nor can it be considered as providing adequate coverage of the key parameters foreseen to be investigated according to the criteria in Annex XI, 1.1.2. of the REACH Regulation.

ECHA concludes that a test using E. coli WP2 uvrA, or E. coli WP2 uvrA (pKM101), or S. typhimurium TA102 has not been submitted by you and that the test using one of these is required to conclude on in vitro gene mutation in bacteria.

As explained above, 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.

In the comments to the draft decision you have submitted a read-across approach which purports to justify the read-across from Lilial (CAS 80-54-6) to the registered substance. This is addressed in section III.B.3, and fails to meet the requirements of Annex XI, 1.5. However, you have provided Ames test data on a different read-across source substance, florhydral (CAS 125109-85-5), and your justification in the comments to the draft decision does not mention florhydral. Therefore, the comments to the draft decision contain no explanation why this read-across from florhydral is possible.

In the endpoint study record in the registration dossier, you argue that read-across to florhydral is possible because "The Target Substance and Source Substance have been characterised in using the categories and databases present in the OECD QSAR toolbox . From the profiling in this table , it can be seen that the two substances share structural similarities and also 'mechanistic action' similarities which are both general and endpoint specific. Therefore read across is justified." However, you have not provided any profiling or a table. The read-across to florhydral fails to meet the requirements of Annex XI, 1.5 for the reasons as set out in Section III.B.2 below.

Finally, you consider the study to be of not assignable reliability, and for this reason also ECHA considers it cannot be reliable information. Based on all these reasons, the read-across to florhydral is rejected.

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* with one of the following strains: E. coli WP2 uvrA, or E. coli WP2 uvrA (pKM101), or S. typhimurium TA102 following OECD test guideline 471.

**2. 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 sought to adapt this information requirement in accordance with Annex XI, Section 1.5 of the REACH Regulation by providing an In Vitro Chromosome Aberration Test in Chinese Hamster V79 Cells using Florhydral (β-methyl-3-(1-methylethyl)benzenepropanal (EC: 412-050-4, CAS: 125109-85-5)) as the test substance, and an in vivo bone marrow chromosome aberration test using Florhydral (β-methyl-3-(1-methylethyl)benzenepropanal (EC: 412-050-4, CAS: 125109-85-5)) as the test substance.

However, ECHA notes that this adaptation does not meet the general rules for adaptation of Annex XI, 1.5.

The registered substance (target) is a cinnamyl substituted aldehydes with a para-substituted aryl alkyl chains whereas the source substance is ortho substituted aryl aldehyde. The source substance used was nearly 99% pure and no impurities were reported whereas the registered substance contains 4.3% 3-(3-tert-butylphenyl)propanal as an impurity. The study followed OECD 473 and was conducted under GLP, which is appropriate to fulfil the information requirement for chromosomal aberration in vitro.

You have provided QSAR Toolbox empirical and mechanistic chemical profiles of the source and target substances to support the read-across approach. In the documentation, you have set out three arguments in support of his read across approach: (1) the source and the target are structurally similar, (2) they have comparable physico-chemical and key toxicological properties and (3) they have mechanistic similarities based on a number of predictions or alerts. ECHA understands that together, this constitutes the hypothesis that underlies the prediction of the properties of the registered substance, and that therefore the available toxicological data from the source substance can allegedly be used to predict the in vitro chromosome aberration property of the registered substance.

However, first, the application of the group concept requires inter alia that human health effects can be predicted from the data of the reference substance. ECHA agrees that both the source and the target substances are structurally similar (aryl and alkyl substituted aromatic aldehydes). Structural similarity is a prerequisite for applying the grouping and read-across approach, but structural similarity per se does not always lead to predictable or similar human health properties. Hence, further elements are needed such as a well-founded hypothesis of (bio)transformation to a common compound(s), or that different compounds have the same type of effect(s), to allow a prediction of human health properties that does not underestimate risks.

Second, likewise similarities in physico-chemical and human health properties are a prerequisite for establishing the grouping and read-across approach. However, very few health properties are compared: acute toxicity (oral and dermal) and genetic toxicity in bacteria. ECHA does not accept that the similar properties provide a basis to predict the human health properties of the substance, particularly in view of this paucity of comparable information on human health properties. ECHA notes that there are many substances with similar acute toxicity, but have markedly different properties with regard to genetic toxicity.

Third, ECHA notes you have provided a list of mechanistic predictions or alerts. ECHA considers that it is unclear how these predictions/ alerts were generated and what value these predictions/ alerts have in terms of predicting the human health properties of the registered substance. For these reasons ECHA cannot place any reliance on these predictions.

Having regard to all the reasons above, and considering also the cumulative weight of all the arguments together, ECHA considers that the requirement of Annex XI, 1.5, that human health effects may be predicted from data for reference substance(s) within the group by interpolation to other substances in the group (read-across approach), has not been met. Therefore, the adaptation of the information requirement suggested by you cannot be accepted.

As explained above, 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.

In the comments to the draft decision you have submitted a read-across approach for the read-across source substance Lilial. This is addressed in section III.B.3, and fails to meet the requirements of Annex XI, 1.5. You have not provided any studies using this source substance, and so this read-across justification does not apply for this endpoint.

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: OECD TG 473) or *in vitro* mammalian cell micronucleus study (test method: OECD TG 487).

### 3. 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. Therefore, adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement provided that both studies requested under III.B.1. and 2. have negative results.

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.

The technical dossier does not contain an adaptation in accordance with column 2 of Annex VIII, Section 8.4.3. or with the general rules of Annex XI for this standard information requirement.

As explained above, 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.

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.

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) provided that both studies requested under III B. 1. and 2. have negative results.

*Further observations on the read-across proposed in the comments to the draft decision*

You have provided a document ( [redacted] ) as part of the comments to the draft decision, and this contains an annex I which has a read-across justification from source substance Lilial to the registered substance. In this document the you indicate that you will read-across *in vitro* gene mutation test in bacteria, *in vitro* cytogenicity study in mammalian cells and *in vitro* gene mutation study in mammalian cells from the source substance "Lilial". ECHA notes that the reliability of the studies conducted with Lilial cannot be evaluated since the (robust) study summaries are not included in either the dossier or your comments to the draft decision.

The observations presented below are only preliminary and are based on the information provided by your comments.

From your comments ECHA understands that the hypothesis for using "Lilial" as a source substance to "Bourgeonal" (the registered substance) is based on (bio) transformation to common compound, i.e. formation of a known metabolite, 4-tert-butylbenzoic acid (TBBA), which is a common metabolite for both Bourgeonal and Lilial (parent substances).

In figures 1 and 2 of the read-across justification document, have presented the proposed metabolic routes of the source and the target substances in rat hepatocytes. In table 1 of the comments, you have given the concentrations of each detected metabolite following 4 hour incubation in rat hepatocytes to 100 uM Bourgeonal or Lilial.

ECHA observes that after four hours incubation

- the concentration of TBBA is 2 – 4 % of the original dose (100 uM) of the parent substances (i.e. 2.2 uM for Lilial and 4.2 uM for Bourgeonal),
- the concentration of Bourgeonal is 0.5 uM whereas Lilial was not detected,
- the concentration of O-glucuronides of alcohols is 4-7 fold higher for Lilial compared to Bourgeonal, and an O-glucuronide of hydroxylated alcohols can be found for Bourgeonal whereas it is not present for Lilial, and
- the concentrations of other metabolites including hippuric acid and various glucuronide conjugates were comparable between Bourgeonal and Lilial.

ECHA notes that based on the study results approximately 70 % (Bourgeonal) and 85 % (Lilial) of the original doses of parent substances were detected as metabolites after 4 hours incubation. Only 4 % and 2 % TBBA, the common metabolite, was formed from the parent substances, Bourgeonal and Lilial, respectively. Thus the majority of the metabolites formed were structurally different metabolites.

ECHA considers that although TBBA has been shown to be reproductive toxicant causing testicular toxicity, you have not provided any endpoint-specific justification to demonstrate that TBBA is also responsible for the genotoxicity of the substances and that the different metabolites will not impact the (geno)toxicity. ECHA therefore considers that there is not a sufficient basis to predict the genotoxicity of the registered substance from the data of the source substance.

ECHA further notes that the metabolism profiles of Bourgeonal and Lilial are different as Bourgeonal was detected after 4 hours incubation (although at very small concentration) whereas Lilial was not detected, the formation of the claimed main metabolite TBBA is 2 fold higher in case of Bourgeonal, and the concentration of O-glucuronides of alcohols is higher for Lilial compared to Bourgeonal. ECHA considers that these differences in metabolism may impact the toxicological profiles of the substances.

To ECHA's understanding the metabolites were measured after 4 hours incubation. No detailed data on the rate of metabolism has been provided, and therefore the rate of disappearance of the parent substances is not known. In addition, no data on the rate of formation of the metabolites has been provided. ECHA considers that based on the data provided the rate of the metabolism cannot be verified, and thus the impact of the parent substances on toxicity cannot be ruled out.

ECHA additionally concludes that due to shortcomings described above, i.e. lack of endpoint-specific read-across justification, different metabolic profiles of the substances and lack of data on the rate of metabolism, there is not an adequate basis for predicting the properties of the registered substance from the data of the source substance.

As supportive evidence the you refer to: *"data generated in a GLP urine bioanalysis study confirmed that the predicted metabolite 4-tert-butylbenzoic acid (TBBA) was found in the urine of male rats, and that "the concentrations of TBBA found in the urine are proportional to the dose of each of the parent compounds further supporting the hypothesis of similar metabolism for each substance".*

ECHA observes that based on this study, similar levels of TBBA are formed between Bourgeonal and Lilial in proportion to the initial doses. However, no detailed data on the methods and results, such as the rate of disappearance of the parent substances and formation of the metabolites and their concentrations, have been provided. ECHA considers that based on this data, the rate and completeness of the metabolism cannot be verified.

Comparison of physico-chemical and toxicological effects across other endpoints

You additionally refer to similar results in toxicity testing for the source and the target substances. According to the comments "*The available Genetoxicity data for the in vitro Ames assay for both frame shift and base-pair substitution are all negative and in agreement for the "Target" and "Source" substances".* You also state that the acute toxicity profiles are similar for Bourgeonal and Lilial. You have also listed the physico-chemical properties of the two substances.

ECHA considers that based on the Ames test results only no conclusion on the potential of the substances to cause chromosomal aberrations can be drawn and thus this data is insufficient to conclude on the similarities of genotoxic properties. Further, similar acute toxicity and physico-chemical properties alone are not sufficient to conclude on the similar genotoxicity profiles of the substances.

Characterisation of "Target" and "Source" substances (GC-MS)

According to the comments "*The available analytical data including purity information for both Bourgeonal and Lilial are given in Table 2. The data indicates that under the REACH guidance both Bourgeonal and Lilial are mono-constituent substances (>80% purity) with both substances being >95% pure".* From table 2 Bourgeonal has 4.3% of 3-(3-tert-butylphenyl) propanal (CAS 1023288-21-2) whereas Lilial has 0.9% of 3-(3-tert-butylphenyl)-2-methylpropanal (CAS 62518-65-4) as impurity. According to the you "*the impurities will have minimal influence on the individual toxicity profile of the parents or on the comparative toxicology between the parent substances".*

ECHA considers that there is insufficient information to conclude that, e.g., the impurity found in Bourgeonal is of "*minimal influence*" for the genotoxic profile of the substance.

Link of structural similarity and differences with the proposed prediction

You have provided in the comments an output from the OECD [Q]SAR Toolbox in which the structural and mechanistic similarities between Bourgeonal and Lilial are listed.

ECHA agrees that both the source and the target substances present very similar properties based on the OECD QSAR Toolbox predictions. However, that alone is not sufficient to conclude that they have similar genotoxic properties. ECHA notes that you have not addressed the impact of the structural differences (methyl group in Lilial) on toxicokinetic and toxicological properties of the substances. Therefore the structural similarity is not an adequate basis for why the source substance may be used to predict the properties of the registered substance, as required by Annex XI, 1.5.

Reliability and adequacy of source studies

In the read-across justification document you conclude that "*all four studies*", i.e. the genotoxicity studies and the *in vivo* urine bioanalysis study "*are considered to be suitable for read-across to Bourgeonal*".

The dossier does not contain (robust) study summaries of the studies and nor are these described in the comment. Therefore adaptation according to Annex XI, 1.5 fails because ECHA is unable to evaluate their reliability and adequacy.

Bias that influences the prediction

Based on your analysis "*The choice of analogue was based on structural similarity from the OECD [Q]SAR Toolbox when criteria for structural similarity to Bourgeonal by Dice atom centred fragments was set at > 70%. Under these criteria the structural similarity of Lilial to Bourgeonal was 75.9%".*

You further justify that "*the identical main metabolites (carboxylic acid and benzoic acid derivative "TBBA") formed in the urine after 5-Day dosing and the similar physico-chemical data, Lilial is considered to be acceptable as the read-across substance of choice".*

ECHA agrees that the proposed source substance can be considered similar regarding the structure. However, structural similarity alone is insufficient to conclude on the acceptability of the read across. Moreover, the metabolism of the substances is different as described above.

Based on the above considerations, i.e. lack of endpoint-specific read-across justification, different metabolic profiles of the substances, lack of data on the rate of disappearance of the parent substances and formation of the metabolites, insufficient data on impurities and structural differences and lack of robust study summaries, ECHA concludes that you have not provided adequate and reliable information to demonstrate that the proposed read-across approach provides a basis for predicting the properties of the registered substance from date for the source substance for the endpoints in consideration.

ECHA therefore concludes that the criteria of Annex XI, Section 1.5., are not met, and consequently information provided using the read-across source substance (Lilial) is not appropriate to fulfil the information requirement of the substance subject to the present decision.

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

The requested study according to OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction/Developmental Toxicity Screening Test) fulfils both standard information requirements Annex VIII, 8.6.1. (short-term repeated dose toxicity study (28 days) as well as Annex VIII, 8.7.1. (screening for reproductive/ developmental toxicity). Both standard information requirements are addressed here below: See section 4.1. for reproductive toxicity screening and section 4.2. for 28-day repeated dose toxicity.

***4.1 Screening study for reproductive/developmental toxicity (Annex VIII, Section 8.7.1)***

"Screening for reproductive/developmental toxicity" 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 by submitting a "Toxicity Study by Oral Gavage Administration to Sexually Mature Male CD Rats for 5 Days".

Although this study provides information on gonadal toxicity in male rats it is insufficient to fulfil the information requirement of a screening study according to OECD 421/422, because the study is of too short duration, only six male rats/ dose were used and the study does not provide or provides only limited information on reproductive toxicity other than gonadal toxicity in male rat and thus does not allow a reliable assumption/decision on reproductive toxicity. Therefore, it does not provide adequate and reliable coverage of the key parameters foreseen to be investigated in a screening study for reproductive/developmental toxicity (Annex XI, 1.1.2) and this study does not meet the information requirements for the screening study for reproductive/developmental toxicity.

As supporting information, you have also sought to adapt this information requirement in accordance with Annex XI, Section 1.5 of the REACH Regulation with an OECD Guideline 415 (One-Generation Reproduction Toxicity Study) performed on the read-across source substance 3-p-cumenyl-2-methylpropionaldehyde (CAS: 103-95-7, also called cyclamen aldehyde).

You have provided QSAR Toolbox empirical and mechanistic chemical profiles of the source and target substances to support the read-across approach. In the documentation, you have set out three arguments in support of his read across approach: (1) the source and the target are structurally similar, (2) they have comparable physico-chemical and key toxicological properties and (3) they have mechanistic similarities based on a number of predictions or alerts. ECHA understands that together, this constitutes the hypothesis that underlies the prediction of the properties of the registered substance, and that therefore the available toxicological data from the source substance can be used to predict the human health properties of the registered substance.

First, ECHA agrees that both the source and the target substances are structurally similar (aryl and alkyl substituted aromatic aldehydes) with some similarities in the physico-chemical and toxicological properties. Structural similarity is a prerequisite for applying the grouping and read-across approach, but ECHA does not accept in general or this specific case that structural similarity per se is sufficient to enable the prediction of human health properties of a substance, since structural similarity does not always lead to predictable or similar human health properties. Hence, further elements are needed such as a well-founded hypothesis of (bio)transformation to a common compound(s), or that different compounds have the same type of effect(s), to allow a prediction of human health properties that does not underestimate risks.

Second, ECHA likewise agrees that similarities in physico-chemical and human health properties may be a prerequisite for establishing the grouping and read-across approach ("Substances whose physicochemical, toxicological and ecotoxicological properties are likely to be similar or follow a regular pattern..."). However, very few health properties are compared: acute toxicity (oral and dermal), genetic toxicity in bacteria. On this basis, ECHA does not accept in general or in this specific case that the similar properties provide a basis to predict the human health properties of the substance, particularly in view of this paucity of comparable information on human health properties. ECHA notes that there are many substances with similar acute toxicity, but markedly different reproductive toxicity.

Third, ECHA notes that you have provided a list of mechanistic predictions or alerts. ECHA considers that it is unclear how these predictions/ alerts were generated and what value these predictions/ alerts have in terms of predicting the human health properties of the registered substance. For these reasons ECHA cannot place any reliance on these predictions. Having regard to all the reasons above, and considering also the cumulative weight of all the arguments together, ECHA considers that the requirement of Annex XI, 1.5, that human health effects may be predicted from data for reference substance(s) within the group by interpolation to other substances in the group (read-across approach), has not been met. Consequently, for this supporting study, the adaptation cannot be accepted.

Further, looking at the differences in toxicities of florhydral after 28 days and the registered substance after 5 days, it is obvious that the toxicity profiles of these two substances are markedly different. Already at a dose of 100 mg/kg bw, the registered substance produced only after five days marked toxicity in the liver, the kidneys and severe toxicity in the testicles. In comparison, Florhydral, used as the test article in the subacute study, "was generally well tolerated in rats and did not affect health to an important degree at dose levels up to 300 mg/kg/day under the prevailing study conditions. Moreover,no treatment-related macroscopic and microscopic findings could be detected." Also, no effects were reported for testes. ECHA concludes that you have not addressed the obvious differences in toxicity between the source substance and the registered substance and did not explain why those differences would not be significant. ECHA considers that these differences in toxicity contradict your hypothesis of similar behaviour, and for this reason also, ECHA considers that the requirement of Annex XI, 1.5, that human health effects may be predicted from data for reference substance(s) within the group by interpolation to other substances in the group (read-across approach), has not been met.

As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the standard information requirements of Annex VIII, Sections 8.7.1. Consequently there is an information gap and it is necessary to provide information for this endpoint.
In the comments to the draft decision, you propose to perform the study by dermal route. You base the request on 1) dermal exposure of the registered substance and 2) *in vivo* skin absorption data of Lilial (source substance) and similar physico-chemical properties of the registered substance and Lilial.

1) ECHA observes that the registered substance is indeed used as a fragrant in cosmetic products. However, according to the CSR, the substance is also used in products such as insecticides, repellents, and air freshener aerosols and thus inhalation exposure is likely. In addition, inhalation exposure occurs e.g. during manufacture of the substance, professional use, and formulation of fragranced end-products.

2) ECHA observes that based on the read-across justification document, the water solubility and partition coefficient values are in a range that favour dermal absorption, and in section 5.1.3. of the CSR, you have estimated that both the oral and dermal route absorption occur at a rate of 50% (value used for the CSA).

ECHA notes the skin absorption study conducted with Lilial has not been included in the comments nor in the technical dossier, and therefore ECHA is not able to make any conclusions regarding actual human dermal absorption of Lilial. ECHA considers that the estimate for dermal route absorption is not based on measurements of dermal absorption, and therefore the estimate has high uncertainty.

ECHA considers that dermal absorption occurs with the registered substance, and the dermal route is a likely route of exposure. However, ECHA notes that the substance is classified as Skin Irrit.2 and Skin Sens. 1B, that these toxicological properties would tend to minimise the applied dose and cause suffering in experimental animals, and therefore the dermal route is not considered the most appropriate route of exposure. ECHA considers that oral exposure is the most appropriate route because it will maximise the systemic exposure. Therefore, the test should be conducted via oral route.

There is also a data gap for a short-term repeated dose toxicity study (Annex VIII, Section 8.6.1), as shown in Section III.B.4.2 below. The standard information requirements of Annex VIII, 8.6.1 and 8.7.1, can both be fulfilled by providing an OECD 422 study, and less animals are required for the conduct of an OECD 422 study, as compared to separately conducting a 28-day study (OECD 407) and an OECD 421 or 422 study. For reasons of animal use, and taking due account of the potential complications in using this study, ECHA therefore requires that this information requirement be met by an OECD 422 study. Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit information derived with the registered substance subject to the present decision: Combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (test method: OECD TG 422) in rats by the oral route.

***4.2 Short-term repeated dose toxicity study (28 days) (Annex VIII, Section 8.6.1)***

Under REACH Annex VIII 8.6.1. a short-term repeated dose toxicity study (28 days), one species, male and female, is a standard information requirement. Currently the dossier does not contain such a study conducted with the registered substance.

You have sought to adapt this information requirement in three different ways.

The justification of the first adaptation given by you is that the study is not scientifically necessary using the following arguments: "*The physicochemical characteristics of Bourgeonal (log Pow 3.2) and the molecular mass are in a range suggestive of absorption from the gastro-intestinal tract subsequent to oral ingestion. This assumption of oral absorption is confirmed by the data from the existing oral studies on bourgeonal and specific structural analogues were associated with marked systemic toxicity and testicular/epididymal toxicity, and the urine contained TBBA, a known metabolite biomarker of testicular toxicity in rats.*
*The screening study also highlighted specific target organ responses which have led to classification as a repeat dose specific organ toxicity classification of STOT RE 2 based with regard to 3.9.2 of CLP (EC 1907/2006) the observed effects can be considered using guidance values (Table 3.9.3) and a classification can be suggested STOT RE 2.*
*Furthermore, given the results of the existing read across one generation reproduction toxicity study which suggests an NOAEL of 25 mg/kg bw/d, the repeat dose effects of the substance should be considered covered by the reproduction toxicity data.*
*As an objective of Regulation EC No. 1907/2006 is to reduce, replace or refine animal testing, based on the above information and information in this dossier it is not warranted to perform further studies in animals."*

ECHA notes that this adaptation does not meet the specific rules for adaptation of Annex VIII, Section 8.6.1., column 2 because there is no column 2 adaptation in the event of severe toxicity or self-classification of the substance with STOT RE 2(R48).

Second, you have also provided a key study for repeated dose toxicity, a "*Toxicity Study by Oral Gavage Administration to Sexually Mature Male CD Rats for 5 Days*". Although this study provides information about toxicity in male rats it is insufficient to fulfil the information requirement of a repeated dose toxicity, because the study is of too short duration, only six male rats/ dose were used. Therefore, it does not provide adequate and reliable coverage of the key parameters foreseen to be investigated in a 28-day study (Annex XI, 1.1.2) and this study does not meet the information requirements for a 28-day study.

Third, as supportive information, you have sought to adapt this information requirement in accordance with Annex XI, Section 1.5 of the REACH Regulation by providing a 28 days study summary conducted with a read across substance (OECD Guideline 407 (Repeated Dose 28-Day Oral Toxicity in Rodents) using Florhydral (β-methyl-3-(1-methylethyl)benzenepropanal (EC: 412-050-4, CAS: 125109-85-5)) as the test substance, but with a non-assignable reliability (4).

ECHA considers that this is essentially the same read-across approach as was used for the Screening study for reproductive/developmental toxicity (Annex VIII, Section 8.7.1) in Section III.B.4.1. ECHA considers that the adaptation fails to meet the requirements of Annex XI, 1.5, for the same reasons as set out in Section III.B.4.1 above. Therefore the information requirement for a 28-day study on the registered substance has not been met.

As explained above, the information provided on this endpoint for the registered substance in the technical dossier does not meet the standard information requirements of Annex VIII, Sections 8.6.1. Consequently there is an information gap and it is necessary to provide information for this endpoint.

There is also a data gap for Annex VIII, 8.7.1 (see Section III.B.4.1 above). The standard information requirements of Annex VIII, 8.6.1 and 8.7.1, can both be fulfilled by providing an OECD 422 study, and less animals are required for the conduct of an OECD 422 study, as compared to separately conducting a 28-day study (OECD 407) and an OECD 421 or 422 study. For reasons of animal use, and taking due account of the potential complications in using this study, ECHA therefore requires that this information requirement be met by an OECD 422 study.

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

From the comments, ECHA acknowledges your willingness to perform the reproduction/developmental toxicity screening test (OECD 422). You propose to perform the study by dermal route. In the summary section of the toxicokinetics endpoint you have estimated that both the oral and dermal route absorption occur at a rate of 50%. You base the estimation on the octanol/water partition coefficient (log Kow of 3.2) and the fact that the molecular weight (190 Da) of Bourgeonal are in ranges which favour oral and dermal absorption.

ECHA considers that this information alone is not sufficient to conclude that the dermal absorption will indeed be so high. ECHA notes there may be dermal absorption occurring with the registered substance, and it is a likely route of exposure. However, ECHA notes that the substance is classified as Skin Irrit.2 and Skin Sens. 1B, that these toxicological properties would tend to minimise the applied dose and cause suffering in experimental animals, and therefore the dermal route is not considered the most appropriate route of exposure. ECHA considers oral exposure is the most appropriate route because it will maximise the systemic exposure.

Based on the information provided in the technical dossier and the chemical safety report the conditions for testing by the dermal route are not met. The properties of the registered substance and its uses indicate that human exposure by the inhalation route is possible as the uses include spraying and charging/discharging the substance from one vessel to another. However, the substance is a liquid of low vapour pressure and exposure to vapours is expected to be low. Moreover, according to ECHA Guidance R.7.a, chapter R.7.5.4.3, the oral route is the default route of exposure concerning repeated dose toxicity testing. Hence, ECHA considers that the oral route is the most appropriate route of administration.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit information derived with the registered substance subject to the present decision: 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 consideration:*

You can use the results of the Combined Repeated Dose Toxicity Study with the Reproduction/Developmental Toxicity Screening Test (test method: OECD 422) to adapt the information if the results of that study show severe toxicity effects according to the criteria for classifying the substance as STOT RE 1 or 2.

In carrying out the studies 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. If the registration of the substance covers different grades, the sample used for the new studies must be suitable to assess these.

Furthermore, there must be adequate information on substance identity for the sample tested and the grade(s) registered to enable the relevance of the studies to be assessed.

## IV. Information on right to appeal

An appeal may be brought against this decision to the Board of Appeal of ECHA under Article 51(8) of the REACH Regulation. Such an appeal shall be lodged within three months of receiving notification of this decision. Further information on the appeal procedure can be found on ECHA's internet page at http://www.echa.europa.eu/regulations/appeals. The notice of appeal will be deemed to be filed only when the appeal fee has been paid.

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.