~~Confidential~~

Helsinki, 22 July 2021

Addressees
Registrants of 265-182-8/64742-79-6 listed in the last Appendix of this decision

Date of submission for the jointly submitted dossier subject of a decision
04/04/2019

Registered substance subject to this decision, hereafter 'the Substance'
Substance name: Gas oils (petroleum), hydrodesulfurized
EC number: 265-182-8
CAS number: 64742-79-6

Decision number: Please refer to the REACH-IT message which delivered this communication (in format TPE-D-XXXXXXXXXX-XX-XX/F)

DECISION ON A TESTING PROPOSAL

Under Article 40 of Regulation (EC) No 1907/2006 (REACH), you must submit the requested information listed in A.1. (B.1.), B.2., B.3. below by **02 May 2023** *from the date of the decision*; and all other requested information listed below by **29 April 2025** *from the date of the decision*.

The requested information must be generated using the Substance unless otherwise specified.

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

1. Same In vivo mammalian alkaline comet assay combined with in vivo mammalian erythrocyte micronucleus test (triggered by Annex I, Section 0.5 in conjunction with Annex VIII, Section 8.4., column 2) , as requested below in B.1.;

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

1. In vivo mammalian alkaline comet assay test method: OECD TG 489) combined with in vivo mammalian erythrocyte micronucleus test (test method: OECD TG 474) in rats, oral route (triggered by Annex I, Section 0.5 in conjunction with Annex IX, Section 8.4., column 2). For the comet assay the following tissues shall be analysed: liver, glandular stomach and duodenum.
2. Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2.; test method OECD TG 408) in rats;
3. Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.; test method OECD TG 414) in a first species (rat or rabbit), oral route;

C. Information required from the Registrants subject to Annex X of REACH

1. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.; test method OECD TG 414) in a second species (rat or rabbit), oral route;
2. Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.; test method: OECD TG 443) in rats, oral route specified as follows:
   - Ten weeks premating exposure duration for the parental (P0) generation;
   - Dose level setting shall aim to induce systemic toxicity at the highest dose

level;

- Cohort 1A (Reproductive toxicity);
- Cohort 1B (Reproductive toxicity) with extension to mate the Cohort 1B animals to produce the F2 generation which must be followed to weaning;
- Cohorts 2A and 2B (Developmental neurotoxicity); and
- Cohort 3 (Developmental immunotoxicity).

You must report the study performed according to the above specifications. Any expansions of the study design must be scientifically justified.

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

- Appendix entitled "Reasons common to several requests";
- Appendices entitled "Reasons to request information required under Annexes VIII to X of REACH", respectively.

Information required depends on your tonnage band

You must provide the information listed above for all REACH Annexes applicable to you, and in accordance with Articles 10(a) and 12(1) of REACH:

- the information specified in Annexes VII and VIII to REACH, for registration at 10-100 tpa;
- the information specified in Annexes VII to X to REACH, for registration at more than 1000 tpa.

You are only required to share the costs of information that you must submit to fulfil your information requirements.

For certain endpoints, ECHA requests the same study from registrants at different tonnages. In such cases, only the reasoning why the information is required at lower tonnages is provided in the corresponding Appendices. For the tonnage where the study is a standard information requirement, the full reasoning for the request including study design is given. Only one study is to be conducted; the registrants concerned must make every effort to reach an agreement as to who is to carry out the study on behalf of the other registrants under Article 53 of REACH.

How to comply with your information requirements

To comply with your information requirements you must submit the information requested by this decision in an updated registration dossier by the deadline indicated above. You must also **update the chemical safety report**, where relevant, including any changes to classification and labelling, based on the newly generated information.

You must follow the general testing and reporting requirements provided under the Appendix entitled "Requirements to fulfil when conducting and reporting new tests for REACH purposes". In addition, you should follow the general recommendations provided under the Appendix entitled "General recommendations when conducting and reporting new tests for REACH purposes". For references used in this decision, please consult the Appendix entitled "List of references".

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.

Approved[1] under the authority of Christel Schilliger-Musset, Director of Hazard Assessment

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

Appendix on reasons common to several requests

Assessment of the Grouping of substances and read-across approach, in light of the requirements of Annex XI, Section 1.5.

Despite the proposals for testing you submitted, you have submitted information on analogue substances for the following standard information requirements seeking to apply a read-across approach in accordance with Annex XI, Section 1.5:

- Sub-chronic toxicity study (90-day), (Annex IX, Section 8.6.2.)
- Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.)
- Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.).

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

Annex XI, Section 1.5. specifies two conditions which must be fulfilled whenever a read-across approach is used:
(i)there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category (addressed under 'Scope of the grouping');
(ii)it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (addressed under 'Assessment of prediction(s)').

1. Scope of the grouping

a. Description of the grouping

In your registration dossier you have provided a read-across justification document in IUCLID Section 13 (named "[redacted])").

More specifically, in Section 3 ("[redacted]") of that category justification document, you provide the following reasoning for the grouping the substances: "*The complex and variable composition of OtherGO substances implies that it is not possible to define precisely their physical-chemical, toxicological and environmental properties, but they will fall into a range defined by the properties and the concentrations of the individual hydrocarbon constituents. […]Where limited or no data exist for OtherGO substances, read-across is conducted from similar substances in the Vacuum Gas Oils, Hydrocracked Gas Oils & Distillate Fuels (VHGO) category.*"

You define the structural basis for the grouping as relying on the fact that "*Figures 10 and 11 respectively show the average values for the carbon number and the hydrocarbon class profiles, […] for samples of the source VHGO category, together with the equivalent data already described for the target OtherGO category. These data show that the carbon number and the hydrocarbon class profiles are similar for the source and target categories and support the use of read-across where applied.*" ECHA understands that this is the applicability domain of the grouping and will assess your predictions on this basis.

b. Assessment of the grouping

ECHA has identified the following issues:

i. Group members are not characterised

Annex XI, Section 1.5 of REACH provides that "*substances whose physicochemical, toxicological and ecotoxicological properties are likely to be similar or follow a regular pattern as a result of structural similarity may be considered as a group.*" According to the ECHA Guidance, "*in identifying a category, it is important that all potential category members are described as comprehensively as possible*", because the purity profile and composition can influence the overall toxicity/properties of the potential category members[2]. Therefore, qualitative and quantitative information on the compositions of the category members must be provided to confirm the category membership and, for UVCBs, qualitative compositional information of the individual constituents of the category members; as well as quantitative characterisation in the form of information on the concentration of the individual constituents of these substances; to the extent that this is measurable.[3]

You propose to read across from similar substances in the "[redacted]" (hereafter referred to as "VHGO category"), which contains UVCB substances and you provide average carbon number profiles and various hydrocarbon class profiles for the VHGO category.

You did not provide the substance identity information, purity profile and composition for any of the similar substances belonging to the VHGO category. Therefore no qualitative or quantitative comparative assessment of the compositions of the different category members can be completed. Therefore, both the membership of the category and whether the analogue substances and the Substance are of similar composition cannot be confirmed. Consequently it is not possible to establish if there is structural similarity, nor to establish the basis for prediction of properties based on the similarities and differences in chemical compositions.

ii. Applicability domain of the categories is not defined

According to the ECHA Guidance, a category (grouping) hypothesis should address "*the set of inclusion and/or exclusion rules that identify the ranges of values within which reliable estimations can be made for category members for the given endpoint".*[4] Particularly, "*the applicability domain of a (sub)category would identify the structural requirements and ranges of physico-chemical, environmental fate, toxicological or ecotoxicological properties within which reliable estimations can be made for the (sub)category members*".[5] Therefore, to reliably predict properties within a category, the applicability domain must be described including the borders of the category, for which chemicals the category does not hold and a justification for the inclusion and/or exclusion rules.

You describe the applicability domain of the substances covered by the grouping as "*the carbon number and the hydrocarbon class profiles are similar for the source and target categories*". *Where limited or no data exist for OtherGO substances, read-across is conducted from similar substances in the Vacuum Gas Oils, Hydrocracked Gas Oils & Distillate Fuels (VHGO) category*." You do not provide a listing of the substances belonging to the VHGO category. Instead you provide a document titled "*PAH hypothesis for toxicity testing of petroleum UVCB substances and read across assessments*", which does not provide a basis for the definition of the VHGO category beyond restricting it to the continuum of petroleum substances.

[2] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.4.1
[3] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.5.5
[4] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.4.1
[5] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.1.2

You have not defined the applicability domain with unambiguous inclusion or exclusion criteria which would identify the structural requirements and ranges of physico-chemical, environmental fate, toxicological or ecotoxicological properties within which reliable estimations can be made for the (sub-)category members. You specified the identity of the Substance. It is not possible to identify the limits to the range of substances belonging to the VHGO category within which you propose that read-across is possible.

Because you have not defined which substances belong to the VHGO category, it is not possible to:
(1) establish which of the analogue substances used in your dossier are members of the VHGO category, and
(2) if they are not members of the aforementioned category, it is not possible to establish on what basis the structural similarity and prediction of properties are made for the Substance.
Accordingly, it is not possible to establish that there is structural similarity between the Substance and substances belonging to the VHGO category and to establish a basis for prediction of the properties of the Substance based on similarities and dissimilarities in composition of the analogue substances.

c. Assessment of the prediction

In the "[redacted]" you state that *"Toxicological data do not exist for all OtherGO substances.[…] In some cases, the OtherGO category value has been assigned using read-across from substances in the Vacuum Gas Oil, Hydrocracked Gas Oil & Distillate Fuels (VHGO) category, which are chemically similar to OtherGO substances (see Section 3)."*

ECHA understands that you

a. predict the properties of the Substance using a read-across hypothesis which assumes that different compounds have the same type of effects. The properties of your Substance are predicted to be quantitatively equal to those of the source substance because of "*similar physical/ chemical properties and chemical composition*"; and
b. as described in your "*PAH hypothesis for toxicity testing of petroleum UVCB substances and read across assessments*" document, you intend to predict the properties for the Substance from information obtained from the source substances, indicated for each endpoint, belonging to the VHGO category, because they in turn belong to the continuum of petroleum substances. You have provided the following reasoning for the prediction of toxicological properties in the "*PAH hypothesis for toxicity testing of petroleum UVCB substances and read across assessments*" document. "*Based on the existing data across the continuum of petroleum substances (PS), Concawe hypothesises that higher tier toxicological effects such as genotoxicity, repeated dose systemic toxicity, reprotoxicity (developmental and fertility) and carcinogenicity are associated with the level and types of polycyclic aromatic hydrocarbons (PAHs).*"
In addition you provide evidence (a) that specific PAHs are highly toxic, (b) that the toxicity of various hydrocarbon streams is correlated with the contents of specific classes of PAHs, and (c) that particular non-PAH hydrocarbon streams are not toxic.

Furthermore you state in your dossier that "*Compositional and physico-chemical data show that VGOs/HGOs/Distillate Fuels are very similar to Other Gas Oils. It is considered appropriate, therefore, to read across from the VGOs/HGOs/Distillate Fuels data to Other Gas Oils.*"

Similarly as above, ECHA has identified the following issues:

i. Absence of a well-founded hypothesis to establish a reliable prediction

To fulfil the two conditions set out in Annex XI, Section 1.5., a read-across hypothesis must be provided, establishing why a prediction for a toxicological property is reliable. This hypothesis must be based on recognition of the structural similarities and differences between the substances[6]. It should explain why the differences in the chemical structures should not influence the toxicological properties or should do so in a regular pattern.

Your read-across hypothesis relies on the structural and physico-chemical similarity between the Substance and substances belonging to the VHGO category.

- Need of structural similarity

ECHA considers that you have no detailed justification for the above mentioned substances, because:
While structural similarity is a prerequisite for applying the grouping and read-across approach, it does not necessarily lead to predictable or similar human health properties. Likewise, physico-chemical similarity does not provide a reliable basis for predicting the human health properties of a substance. You have not provided a well-founded hypothesis to establish a reliable prediction for the toxicological properties, based on recognition of the structural similarities and differences between the members of the different categories (*"VGOs/HGOs/Distillate Fuels")*.
For none of the endpoints, nor individual study, have you justified on the structural similarity/ properties with the Substance, beyond the Section 3 of your "[redacted]" document, as addressed above.

- Need of prediction

For none of the endpoints, nor individual study, have you established in a well-documented justification how the results from the studies you submitted can allow prediction of the intrinsic properties of the Substance.

Thus, you have not provided a well-founded hypothesis to establish a reliable prediction for the toxicological properties, based on recognition of the structural similarities and differences between the substances belonging to the VHGO category and the Substance.

On the contrary, ECHA understands that you do not rely on these supporting studies to predict the relevant properties of the Substance from data of other substances since you have submitted several proposals for testing, subject of the current decision (see in the next appendices).

ii. Absence of supporting information to compare properties of source and target substances

To demonstrate that the structurally similar members of the several categories across the continuum of petroleum substances (PS) cause the same type of effect(s) namely that "*higher tier toxicological effects such as genotoxicity, repeated dose systemic toxicity, reprotoxicity (developmental and fertility) and carcinogenicity are associated with the level and types of polycyclic aromatic hydrocarbons (PAHs)*", you must provide relevant, reliable and adequate supporting information[7]; in this case, information allowing to compare the properties of the substances belonging to the VHGO category with those of the Substance and to confirm that those substances cause the same type of effects.

[6] Guidance on information requirements and chemical safety assessment, Chapter R.6: QSARs and grouping of chemicals.
[7] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.2.1.f

More specifically, in a document entitled "[redacted]", you have proposed that reproductive toxicity is primarily associated with the presence of condensed PAH of three or more rings, and that the substance with the highest content of PAH of three or more rings will be the most reprotoxic.

Your hypothesis is based on the analysis of:

a. *in vivo* data, *inter alia*, from [redacted], 1994, [redacted] 2012, [redacted], 2013 and [redacted], 2013;
b. analysis of *in vitro* data (assays in mouse embryonic stem cell test (EST), CALUX Reporter Gene Assays and zebrafish embryotoxicity test (ZET) *inter alia,* ([redacted], 2017 and 2018) aiming at comparing potencies, defining the role of endocrine activities and whether ZET can allow prediction of PNDT, respectively;
c. the characterisation of several >3-ring PAHs as being highly toxic; and
d. information on some substances with low content of >3-ring PAHs which are of low toxicity.

ECHA has assessed the various elements of your hypothesis:

a. The *in vivo* analyses appear to be based upon principally dermal studies. ECHA considers that these dermal studies do not provide adequate information about the hazardous properties of the substance, since the oral route of exposure is the most appropriate route (see points B.2. and B.3., and C.1. below): repeated dose and reproductive toxicity studies must be performed by the "most appropriate route of administration, having regard to the likely route of human exposure." Since the systemic availability of chemical components of a substance after repeated oral and dermal administration is expected to be significantly different, the toxicological properties of a substance could be significantly different when comparing between repeated oral and dermal administration.
   Therefore ECHA considers that your hypothesis, as based principally on dermal studies, is not a reliable basis for predicting the properties of the Substance after oral administration.
b. You have provided information from *in vitro* studies which use DMSO extracts of the substances (rather than the substances themselves). Testing of DMSO extracts in *in vitro* studies does not provide a reliable basis for predicting the properties of the Substance.
   The EST method does not provide a reliable basis to support read-across because:
   (i) the consequence of testing DMSO extracts, as opposed to the substances themselves, on the outcome of *in vivo* and *in vitro* testing is not explained. Thus testing DMSO extracts does not provide a basis for reliably predicting the properties of the Substance;
   (ii) in line with the tests of Annex XI, Section 1.4, ECHA agrees with your statement that the *in vitro* tests '*cannot and should not be interpreted as screening assays for toxicity*' and considers that the individual *in vitro* results have no clear relation to *in vivo* toxicity assays or endpoints. Consequently, any relationship between substances in *in vitro* test results has an unclear relationship to any *in vivo* toxicity assays on the test substances. There is thus no adequate basis for using *in vitro* data to predict that the results of *in vivo* tests can be read across from one substance to another.

c. ECHA notes that some >3-ring PAHs are highly toxic.

d. You have provided information on C9-C12 (predominantly) aliphatics (CAS RN 64742-81-0), C9-C16 (predominantly) aliphatics (CAS RN 8008-20-6 MIL-T-83133A), C20-C30 Highly refined base oils (CAS RN 8042-47-5 and 8012-95-1), C8-C26 GTL (gas-to-liquid) gas oil (CAS RN 848301-67-7) and C18-C50 GTL base oil (CAS RN 848301-69-9). ECHA understands that your hypothesis is that the >3-ring PAHs are responsible for toxicity, and the information from these substances is aimed at showing that substances without >3-ring PAHs are of low (and known) toxicity. Merely providing information is not a

demonstration of why the structural variation in and the abundance of the constituents of the above substances provides a basis for predicting the properties of the Substance, when related to the structures of the constituents (and their variation in abundance) for the Substance. In particular, the characterisation of the constituents of these substances is crude, and does not allow resolution of individual constituents.

Summarising, ECHA considers that the PAH hypothesis does not provide a basis for predicting the "*human health hazard endpoints*" of the Substance. In conclusion you have not established that the substances belonging to the VHGO category are likely to have similar properties. Therefore, you have not provided sufficient supporting information to strengthen the rationale for the read-across.

2. Conclusions on the grouping of substances and read-across approach

As explained above, you have not established that relevant properties of the Substance can be predicted from data on the analogue substances.

Therefore, your adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. and your grouping and read-across approach between the Substance and substances belonging to the VHGO category is rejected.

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

This decision is based on the examination of the testing proposals you submitted.

1. In vivo mammalian alkaline comet assay combined with in vivo mammalian erythrocyte micronucleus test

Under Annex VIII, Section 8.4, column 2 of REACH, appropriate *in vivo* mutagenicity studies shall be considered in case of a positive result in any of the genotoxicity studies in Annex VII or VIII.

You have provided *in vitro* studies on analogue substances and on the Substance with negative results, except two with an ambiguous result.

However, you conclude that *in vitro* studies are not appropriate to address the mutagenicity properties of the Substance due to the difficulties with its solubility, and that *in vivo* studies are necessary to confirm the intrinsic properties of the Substance.

For that same reason, ECHA agrees that further information is necessary for producing the chemical safety report and that this information can only be obtained by performing an *in vivo* mutagenicity study in accordance with Annex IX to REACH (Section 0.5, Annex I to REACH), to address gene mutation and chromosomal aberration.

For the specifications of the study to be performed, see the request B.1.

Appendix B: Reasons for the requirements applicable to all the Registrants subject to Annex IX of REACH

This decision is based on the examination of the testing proposals you submitted.

1. In vivo mammalian alkaline comet assay combined with in vivo mammalian erythrocyte micronucleus test

Under Annex IX, Section 8.4, column 2 of REACH, the information requirement for an appropriate *in vivo* somatic cell genotoxicity study is triggered if 1) there is a positive result in any of the *in vitro* genotoxicity studies in Annex VII or VIII and 2) there are no appropriate results already available from an *in vivo* somatic cell genotoxicity study.

You have provided (1) *in vitro* studies on analogue substances and the Substance with negative results, except two with an ambiguous result, and (2) one *in vivo* study on an analogue substance belonging to the OtherGO category (see below).

Your dossier contains key and supporting studies:

i. One key study performed according to the Ames test (OECD TG 471) with the analogue substance Distillates (petroleum), hydrotreated middle (EC number 265-148-2, CAS RN 64742-46-7) and that show a negative result; several other (7) supporting *in vitro* studies performed according to (a modified procedure of) the Ames test (OECD TG 471) with either the Substance, or the analogue substance Distillates (petroleum), hydrotreated middle (EC number 265-148-2, CAS RN 64742-46-7) belonging to the OtherGO category, or substances belonging to other categories;
ii. One key *in vitro* sister chromatid Exchange (SCE) test (OECD TG 479) with an analogue substance (Distillates (petroleum), hydrodesulfurized middle (EC number 265-183-3, CAS RN 64742-80-9) belonging to the OtherGO category and which shows an ambiguous result;
iii. One key *in Vitro* Mammalian Cell Gene Mutation Test (OECD TG 476) with the analogue substance (Distillates (petroleum), hydrodesulfurized middle (EC number 265-183-3, CAS RN 64742-80-9) and which shows a negative result; one supporting *in Vitro* Mammalian Cell Gene Mutation Test (OECD TG 476) with the analogue substance (Distillates (petroleum), hydrodesulfurized middle (EC number 265-183-3, CAS RN 64742-80-9) and which shows an ambiguous result;
iv. One key *in vivo* cytogenicity/ bone marrow chromosome aberration (OECD TG 475) with an analogue substance (Distillates (petroleum), hydrodesulfurized middle (EC number 265-183-3, CAS RN 64742-80-9) and which shows a negative result;
v. A testing proposal for a Combined OECD TG 422/OECD TG 474 study.

However, you conclude that *in vitro* studies are not appropriate to address the mutagenicity properties of the Substance due to the difficulties with its solubility, and that *in vivo* studies are necessary to confirm the intrinsic properties of the Substance.

The *in vivo* study (see iv. above), provided on an analogue substance, does not provide compliant information on cytogenicity: there is, e.g. no information on (a) the mitotic index determined in at least 1000 cells/ all treated animal (including positive controls), untreated or vehicle/solvent negative control animal; on (b) at least 200 metaphases analysed for each animal for structural chromosomal aberrations including and excluding gaps.

For these reasons, ECHA considers that you have adequately demonstrated the need to perform further testing to better address the lack of data for the endpoint of mutagenicity *in vitro/ in vivo* and agrees that further information is necessary for producing the chemical safety report and that this information can only be obtained by performing an *in vivo* mutagenicity study in

accordance with Annex IX to REACH (Section 0.5, Annex I to REACH), to address gene mutation and chromosomal aberration.

Standard and modified Ames tests

You have submitted a sequential testing programme proposing to conduct new standard and modified Ames tests with the Substance. While ECHA acknowledges your intentions, it notes that a test covering an endpoint of Annex VII as you proposed does not fall within the scope of the examination of a testing proposal and that it is at your discretion to conduct such tests.

Combined OECD TG 422/OECD TG 474 study

As part of your "[redacted]" (November 2018) attached in the IUCLID dossier, section 7.5.3, you have submitted a proposal to test the Substance in a combined OECD TG 422/OECD TG 474 (*in vivo* micronucleus) study, oral route "*if any positive* in vitro *results*". You explained that *"due to the difficulties with solubility, it was considered that this endpoint [*in vitro *genotoxicity] was best addressed using an* in vivo *test. Further this would be incorporated into the planned OECD TG 422 test so as to minimise the use of animals. If the OECD TG 474 does not meet the full criteria for acceptance when conducted as part of other studies […], then a stand-alone study will be required."*

Your proposal for such a combined test cannot be accepted because testing proposals can be only made for the provision of the information specified in Annexes IX and X to REACH. A test covering an endpoint of Annex VIII (combined repeated dose toxicity study with reproductive toxicity screening test (OECD TG 422)), as you proposed, does not fall within the scope of the examination of a testing proposal under Articles 40 and 10(a)(ix) of REACH.

Therefore, this part of the proposal is out of scope of the evaluation of your testing proposal. It is at your discretion to conduct such combined tests without compromising the validity of the test requested. In this regard, you must pay attention to the dosing schemes of the different studies, which may jeopardise the regulatory validity of each separate information requirement.

Test selection
According to ECHA Guidance Chapter R.7a, section R.7.7.6.3, information on the capability to induce gene mutations, structural chromosome aberrations (clastogenicity) and numerical chromosome aberrations (aneugenicity) is required to be able to evaluate the mutagenic potential of a substance.

The proposed *in vivo* micronucleus test (according to TG OECD 474) while it investigates *in vivo* chromosomal mutagenicity (as the study detects both structural and numerical chromosomal aberrations) is not suitable to address gene mutation.

According to the ECHA Guidance, the *in vivo* mammalian alkaline comet assay ("comet assay", OECD TG 489) is a genotoxicity indicator test that is suitable to address both gene mutation and chromosomal aberration, also for substances of low systemic bioavailability. However, the comet assay is not appropriate for detecting aneugens (numerical chromosome aberrations).

As indicated above, the *in vivo* mammalian erythrocyte micronucleus test ("MN" test, OECD TG 474) is a mutagenicity test that provides evidence on *in vivo* chromosomal mutagenicity, as the study detects both structural and numerical chromosomal aberrations.

Consequently, as also indicated in the ECHA Guidance, it is possible to combine the comet assay and the MN test into a single study. The combined study can help reduce the number of

tests performed and the number of animals used while addressing both structural and numerical chromosomal aberrations and gene mutations.

Therefore, the comet assay combined with the MN test is an appropriate study for the Substance.

Test design

According to the test method OECD TG 489, the test must be performed in rats. Therefore, the combined test (OECD TG 489 and OECD TG 474) must be performed in rats. Having considered the anticipated routes of human exposure and adequate exposure of the target tissue(s), performance of the test by the oral route is appropriate.

In line with the test method OECD TG 489, the test must be performed by analysing tissues from liver as primary site of xenobiotic metabolism, glandular stomach and duodenum as sites of contact. There are several expected or possible variables between the glandular stomach and the duodenum (different tissue structure and function, different pH conditions, variable physico-chemical properties and fate of the Substance, and probable different local absorption rates of the Substance and its possible breakdown product(s)). Because of these expected or possible variables, you must analyse both tissues to ensure a sufficient evaluation of the potential for genotoxicity at the site of contact in the gastro-intestinal tract.

The combination of OECD TGs 489 and 474 should not impair the validity of and the results from each individual study. Careful consideration should be given to the dosing, and tissue sampling for comet analysis alongside the requirements of tissue sampling for the mammalian erythrocyte micronucleus test(see OECD TG 489, e.g. Bowen *et al*. 2011[8]).

Germ cells

A subsequent germ cell genotoxicity study (TGR/OECD TG 488, or chromosome aberrations on spermatogonia/OECD TG 483) may still be required under Annex IX of REACH, in case 1) an *in vivo* genotoxicity test on somatic cell is positive, and 2) no clear conclusion can be made on germ cell mutagenicity.

Therefore, you may consider to collect the male gonadal cells from the seminiferous tubules in addition to the other aforementioned tissues in the comet assay, as it would optimise the use of animals. You can prepare the slides for male gonadal cells and store them for up to 2 months, at room temperature, in dry conditions and protected from light. Following the generation and analysis of data on somatic cells in the comet assay, under Annex IX, Section 8.4., column 2 you should consider analysing the slides prepared with gonadal cells.

This type of evidence may be relevant for the overall assessment of possible germ cell mutagenicity including classification and labelling according to the CLP Regulation.

Outcome

According to Article 40(3)(b) of the REACH Regulation, you are requested to carry out the proposed test under modified conditions, as explained above, with the Substance.

2. Sub-chronic toxicity study (90-days)

A sub-chronic toxicity study (90 day) is a standard information requirement in Annex IX, Section 8.6.2. to REACH.

[8] Bowen D.E. *et al.* (2011). Evaluation of a multi-endpoint assay in rats, combining the bone-marrow micronucleus test, the comet assay and the flow-cytometric peripheral blood micronucleus test. *Mutation Research*, 722 7–19

You have submitted a testing proposal for a sub-chronic toxicity study (90 day) in rats by the dermal route according to OECD TG 411 with the Substance.

ECHA notes that you provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed. ECHA has taken these considerations into account.

You stated that "*this test is proposed to be conducted on one member of the OtherGO category, with the results then read-across to other category members. Brief justification is given below, with additional support for the category given in the category justification document (attached to the category object and to Section 13 of the dossier) and in the document "OtherGO strategy Nov 2018" attached below.*"

Your technical dossier does not contain information on a repeated dose toxicity (90-day) study (Annex IX, Section 8.6.2.) with the Substance, but instead you submitted several studies with analogue substances. As explained above in the Appendix on reasons common to several requests, your adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. and your grouping and read-across approach is rejected.

Consequently it is necessary to provide information for this endpoint, and the proposed study fulfils the information requirement of Annex IX, Section 8.6.2. of REACH.

Route of administration proposed

According to Annex IX, Section 8.6.2., column 2, testing by the dermal route is appropriate if:
(1) skin contact in production and/or use is likely; AND
(2) the physicochemical properties suggest a significant rate of absorption through the skin; AND
(3) one of the following conditions is met:

- toxicity is observed in the acute dermal toxicity test at lower doses than in the oral toxicity test; or
- systemic effects or other evidence of absorption is observed in skin and/or eye irritation studies; or
- *in vitro* tests indicate significant dermal absorption; or
- significant dermal toxicity or dermal penetration is recognised for structurally-related substances.

You proposed testing by the dermal route, and argued in the "[redacted].*pdf*" document, section 7.5.3 of the IUCLID dossier, that: "*that oral exposure is not a relevant route to inform the risk management measures" and that "dermal application is the preferred route of exposure […] where technically possible.*"

In this document, you referred to the criteria listed in Annex IX, Section 8.6.2., column 2 considering that they are met:

(1) "*as demonstrated by the use descriptors and exposure scenarios, dermal (And occasionally inhalation) is a predominant route of exposure (i.e. more than just 'likely')*". This applies to workplace (professional use as fuels and lubricants) and to a lesser extent to consumer (formulated lubricants);

(2) *"The UVCB nature of PS makes some of these parameters difficult to determine (e.g. Log Pow); […] although a PS in its entirety is unlikely to cross the dermal barrier, the physical-chemical characteristics of UVCB does not prevent the penetration of smaller molecules.*

Replicating the dermal route of administration ensures that those molecules systemically available to the rat are those that have the potential to cross the human dermal barrier." Finally you further referred to the possible dermal penetration of polycyclic aromatic hydrocarbons (PAH), components of the Substance, considered to be responsible for the toxicity;

(3)

- regarding the 1st indent, you indicated "*PS are generally of low/no acute toxicity, […] not all in-vitro dermal absorption tests are directly appropriate because of solubility issues with some test systems. However, there are some studies, […] which assessed the penetration of the test materials using radiolabelled marker molecules, which provide further prove for the dermal penetration of the PS tested.*"
- regarding the 4th indent, you referred to "*a robust historical data base across the spectrum of PS that has allowed successful risk management for many years, in which the majority of studies use the dermal or inhalation route. […] gas oils […] can generally be characterized by a series of systemic effects […]: increased liver weight, decreased thymus weight, and reductions in certain haematological parameters […] and that irritating effects can be controlled […]. When OtherGO are not derived from carcinogenic feed stocks, these systemic target organ effects are not observed, as indicated by the note described in Concawe's Classification and Labelling report (*[redacted]*). On the basis of these dermal and inhalation studies, certain categories of PS (such as the gas oils and some of the unrefined heavier oil products) carry GHS classifications for target organ toxicity (STOT RE 2 and STOT RE 1) for bone marrow, spleen and thymus.*"

You concluded that "*the existing data base clearly demonstrates that systemic toxicity occurs after dermal exposure with some PS, particularly those containing poly aromatic hydrocarbons. The physical-chemical properties of PS cannot be based on a single measurement, as complex UVBCs they contain long hydrocarbon chains with low potential to penetrate the dermis but also small molecules (like PAHs) which do penetrate the dermis*;" "*Dermal is the most relevant exposure route, and is sufficiently robust, to identify any potential hazards from repeated exposures to petroleum products to be able to adequately manage the potentially associated risks. […] The strategy document can be found in Annex 13.*"

In the other document available in section 7.5.3 of the IUCLID dossier "[redacted]" (November 2018), you stated that "*the dermal route is the most appropriate having regard to likely route of human exposure.*"

ECHA has identified the following issue with the route of administration proposed:
ECHA agrees that condition (1) is met, whereas the condition (2) is not met, since you reported in your justification for the "[redacted]" document (November 2017), that "*no measured data are available on the dermal absorption of gas oils*" and that "*systemic tissue changes in repeated dose toxicity studies […] indicates that some absorption across the skin is possible",* to then conclude: "*systemic exposure is limited".* You did not provide any measured data on water solubility and partition coefficient claiming that the "*standard tests for this endpoint are intended for single substances and are not appropriate for this complex substance*." Without providing any measured data, your claim that the physicochemical properties suggest a significant rate of absorption through the skin cannot be justified.

Therefore the dermal route is not appropriate without further assessing whether one of the conditions under (3) is met, for which in any case:

- 1st indent: you did not provide any studies conducted with the Substance which would demonstrate that toxicity is observed in the acute dermal toxicity test at lower doses

than in the oral toxicity test; On an analogue substance, part of the OtherGO category (Gas oils (petroleum), hydrodesulfurized middle, EC number 265-183-3, CAS RN 64742-89-0), acute dermal LD50 is > 2000 mg/kg bw, while moderate-severe skin irritation was recorded following application of the neat substance (Gas oils (petroleum), hydro Distillates (petroleum), hydrotreated middle, EC number 265-148-2, CAS RN 64742-46-7);

- 2nd indent: no systemic effects or other evidence of absorption are reported in skin and/or eye irritation studies performed with an analogue substance of the OtherGO category (Gas oils (petroleum), hydrodesulfurized middle, EC number 265-183-3, CAS RN 64742-89-0);
- 3rd indent: no *in vitro* test submitted on the Substance;
- 4th indent: you did not provide any robust study summaries to the studies performed with the analogue substances in the registration dossier. Without assessing the analogue approach, in the absence of robust study summaries, reporting on exact dosages and composition of test substances, the relevance of data cannot be assessed and thus cannot be used for supporting a proposed dermal route.

ECHA concludes that the proposed dermal route is not an appropriate route of administration for testing and to fulfil the information requirement.

Route of administration required

ECHA has evaluated the choice of most appropriate route:

- The oral route is the default one because it is assumed to maximise systemic availability (internal dose) of most substances, and you have not demonstrated a greater absorption by the dermal route. By contrast, the physicochemical characteristics of numerous components of the Substance indicate that there will be very limited dermal absorption.
- Further, you argued that *"a number of the PS are associated with dermal irritation",* and this also argues against the dermal route, since dermal irritation will tend to limit the applied dose and systemic availability of the substance.

In your comments on the draft decision you maintained your position and argued that the dermal route is *"the most appropriate in relation to the human exposure to petroleum substances"* and "to represent the hazard to humans". You requested further clarification *"on the rationale justifying the oral route […] is the likely route of human exposure".* However, you did not provide any scientific evidence substantiating this statement or any further scientific arguments supporting your claim regarding the dermal route of administration. Finally, the OECD TG 408 explicitly recommends the oral route of administration when no other argument supports another route. Therefore, based on the information currently available, the study is requested via the oral route of administration.

ECHA therefore concludes that the oral route is the most appropriate route of administration.

Species

According to OECD TG 408, the rat is the preferred species.

Selection of the substance tested

In your comments, you argued that you now consider that the Substance is no longer the most representative substance for the OtherGO category (worst-case based on the content of PAHs with 3-7 aromatic rings). However, you have not substantiated your comments with any detailed analytical information supporting your statement. Your choice of substance to test was based on the considerations you set out in your read-across justification and ECHA considered

this reasoning plausible in the context of the read-across. Therefore, based on the information currently available, the study is requested on the Substance.

Outcome

According to Article 40(3)(b) of the REACH Regulation, you are requested to carry out the proposed test under modified conditions, as explained above, with the Substance.

3. Pre-natal developmental toxicity study

A pre-natal developmental toxicity (PNDT) study (OECD TG 414) in one species is a standard information requirement under Annex IX, Section 8.7.2. to REACH.

You have submitted a testing proposal for a PNDT study according to OECD TG 414 with the Substance in the rabbit, by the dermal route.

You provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed. ECHA has taken these considerations into account.

Furthermore, you stated "*It is requested that this OECD PNDT is conducted in the rabbit. As the Concawe testing strategy progresses […], there will be a wealth of data covering most of the aspects of rodent pre and post-natal development and a PNDT study in the rabbit, Concawe request the opportunity to develop a weight of evidence and read-across argument that will negate the need for conducting two PNDT studies (covering two species). In order to do this then, the first PNDT will need to be conducted in the rabbit.*" *"This single testing proposal should not be viewed in isolation, it should be considered as part of Concawe's overall testing proposal, following the overall Concawe testing strategy for petroleum substances and the OtherGO category […] and for more detailed information on the proposed testing for this category is referred to the document "*[redacted]*" attached."*

Your technical dossier does not contain information on a pre-natal developmental toxicity study (Annex IX, Section 8.7.2.) with the Substance, but instead you submitted several studies with analogue substances. As explained above (in the Appendix on reasons common to several requests), your adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. and your grouping and read-across approach is rejected.

Consequently it is necessary to provide information for this endpoint, and the proposed study fulfils the information requirement of Annex IX, Section 8.7.2. of REACH.

Regarding your comment about "*opportunity to develop a weight of evidence and read-across argument*", please refer to point C.1. below.

Route of administration proposed

You proposed testing by the dermal route, referring to the criteria listed in the Annex IX, Section 8.6.2, column 2 of REACH and justified the selection of the dermal route of administration because (i) dermal and inhalation routes are the predominant routes of exposure for workers; and (ii) systemic effects have been observed following repeated dermal administration to rats of similar petroleum substances and thus confirming dermal penetration. Relying on several sub-chronic, pre-natal developmental and chronic toxicity studies performed on rats via dermal exposure with other petroleum substances, you concluded that investigation of the potential for systemic toxicity for this type of substances can be achieved using the dermal route of application.

ECHA disagrees for the following reasons:

- your considerations based on the criteria listed in the Annex IX, Section 8.6.2, column 2 address route of administration for a sub-chronic toxicity study and are not applicable for a pre-natal developmental toxicity study, which is addressed under Annexes IX and X, Section 8.7.2.;
- As explained under point B.2. above, you have not provided any evidence that there is particularly high dermal penetration with the Substance, that the dermal route produces a higher relevant internal dose, nor have you argued that dermal application leads to a specific toxicity, or leads to more potent toxicity as compared with oral application.

Route of administration required

The most appropriate route for reproductive toxicity studies is via oral administration, as the default route recommended in the test method guideline (OECD TG 414). In addition, the OECD GD 43 outlines that "*the dermal route of exposure is not recommended for reproductive toxicity studies. The technical difficulties associated with reproductive toxicity testing by administration by the dermal route outweigh the advantages of mirroring the human exposure. Other studies, such as ADME studies should be undertaken to facilitate extrapolation from the oral to dermal route, if this is required*". Based on the considerations above, the test proposed must be conducted by oral route, since it is the most appropriate route of administration to investigate reproductive toxicity[9].

In your comments on the draft decision, you maintained that the dermal route is the most appropriate to represent the hazard to humans. However, you did not provide any scientific evidence substantiating this statement or any further scientific arguments supporting your claim regarding the dermal route of administration. Therefore, based on the information currently available, the study is requested via the oral route of administration.

Species

You may select between the rat or the rabbit because both are preferred species under the OECD TG 414.

Selection of the substance tested

In your comments, you argued that you now consider that the Substance is no longer the most representative substance for the OtherGO category (worst-case based on the content of PAHs with 3-7 aromatic rings). However, you have not substantiated your comments with any detailed analytical information supporting your statement. Your choice of substance to test was based on the considerations you set out in your read-across justification and ECHA considered this reasoning plausible in the context of the read-across. Therefore, based on the information currently available, the study is requested on the Substance.

Outcome

Under Article 40(3)(b) of the REACH Regulation, you are requested to carry out the proposed test under modified conditions, as explained above, with the Substance.

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

Appendix C: Reasons for the requirement applicable to all the Registrants subject to Annex X of REACH

This decision is based on the examination of the testing proposals you submitted.

1. Pre-natal developmental toxicity study

Pre-natal developmental toxicity (PNDT) studies (OECD TG 414) on two species is a standard information requirement under Annex X to REACH.

As outlined under point B.3., ECHA has approved your testing proposal for a pre-natal developmental toxicity study in a first species according to OECD TG 414.

However your technical dossier does not contain information on a pre-natal developmental toxicity study in a second species (Annex X, Section 8.7.2.).

Consequently there is an information gap and it is necessary to provide information for this endpoint, despite your statement *"As the Concawe testing strategy progresses […], there will be a wealth of data covering most of the aspects of rodent pre and post-natal development and a PNDT study in the rabbit, Concawe request the opportunity to develop a weight of evidence and read-across argument that will negate the need for conducting two PNDT studies (covering two species)."*

Because no information is currently available in your dossier, ECHA cannot assess whether your proposed future read across and grouping and/or weight of evidence arguments will allow you to predict the above information requirement. Therefore, ECHA cannot conclude on the statement and on whether these approaches are reliable and allow that you fulfil the information requirement.

A PNDT study according to the OECD TG 414 study should be performed in rabbit or rat as the preferred second species, depending on the species tested in the first PNDT study (required under point B.3.), and the oral route is the most appropriate route of administration to investigate reproductive toxicity. For the selection of the appropriate species you are advised to consult ECHA Guidance R.7a.

In your comments on the draft decision, you argued that the dermal route is the most appropriate to represent the hazard to humans. However, you did not provide any scientific evidence substantiating this statement or any further scientific arguments supporting your claim regarding the dermal route of administration. Therefore, based on the information currently available, the study is requested via the oral route of administration.

In your comments, you argued that you now consider that the Substance is no longer the most representative substance for the OtherGO category (worst-case based on the content of PAHs with 3-7 aromatic rings). However, you have not substantiated your comments with any detailed analytical information supporting your statement. Your choice of substance to test was based on the considerations you set out in your read-across justification and ECHA considered this reasoning plausible in the context of the read-across. Therefore, based on the information currently available, the study is requested on the Substance.

Outcome

According to Article 40(3)(c) of the REACH Regulation, you are requested to carry out the additional test, as indicated above, with the Substance.

2. Extended one-generation reproductive toxicity study

The basic test design of an extended one-generation reproductive toxicity study (EOGRTS) is a standard information requirement under Annex X to the REACH Regulation. Furthermore, column 2 of Section 8.7.3. defines when the study design needs to be expanded.

You have submitted a testing proposal for an EOGRTS according to OECD TG 443 by the oral route in rats with 10-week premating exposure duration.
You have provided the following justification and specification of the study design according to the criteria described in Column 2 of Section 8.7.3, Annex X: "*Concawe intend to take a holistic approach […] and this involves a program to support grouping and read across with a targeted testing strategy. The test proposed for this CAS number is part of that strategy. This CAS number (64742-79-6) has been selected from the OtherGO group because it represents the highest percentage of poly-aromatic hydrocarbons (PAH) with more than 3 cyclic rings. This test is proposed to be conducted on one member of the OtherGO category, with the results then read-across to other category members. Brief justification is given below, with additional support for the category given in the category justification document (attached to the category object and to Section 13 of the dossier) and in the documents "*[redacted]*" and "*[redacted]*" attached."Extended One Generation Reproductive Toxicity Study (OECD TG 443) with the registered substance; Gas oils (petroleum), hydrodesulfurized (CAS RN 64742-79-6, EC No 265-182-8). In addition, it is anticipated that these results will allow read-across to the to the following CAS numbers: Distillates (petroleum), hydrotreated middle (CAS RN 64742-46-7, EC No 265-148-2) and Distillates (petroleum), hydrodesulfurized middle (CAS RN 64742-80-9, EC No 265-183-3)."*

You provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed. ECHA has taken these considerations into account.

Your technical dossier does not contain information on an extended one-generation reproductive toxicity study (Annex IX, Section 8.7.3.) with the Substance, but instead you submitted several studies with analogue substances. As explained above (in the Appendix on reasons common to several requests), your adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. and your grouping and read-across approach is rejected.

Consequently it is necessary to provide information for this endpoint, and the proposed study fulfils the information requirement of Annex IX, Section 8.7.3. of REACH.

The following refers to the specifications of this required study.

Premating exposure duration and dose-level setting

You proposed "*Premating exposure duration for parental (P0) animals - 10 weeks*". ECHA agrees with your proposal. In this specific case a ten-week exposure duration is supported by the lipophilicity of the Substance to ensure that the steady state in parental animals has been reached before mating, as advised in the ECHA Guidance R.7a.

In order to be compliant and not to be rejected due to too low dose levels, the highest dose level must aim to induce systemic toxicity, but not death or severe suffering of the animals, to allow comparison of reproductive toxicity and systemic toxicity. The dose level selection should be based upon the fertility effects with the other cohorts being tested at the same dose levels. A descending sequence of dose levels should be selected in order to demonstrate any dose-related effect and to establish NOAELs.

If there is no existing relevant data to be used for dose level setting, it is recommended that results from a range-finding study (or range finding studies) are reported with the main study. Also you must provide a justification with your study report that demonstrate that the dose level selection meets the conditions described above.

Cohorts 1A and 1B

Cohorts 1A and 1B belong to the basic study design and shall be included.

Extension of Cohort 1B

If the Column 2 conditions of Section 8.7.3., Annex X are met, Cohort 1B must be extended by mating the Cohort 1B animals to produce the F2 generation.

You proposed to include an extension of Cohort 1B and provided a justification following Column 2 criteria: "*some of the 4 –7 ring aromatic substances present in the OGO may lead to interactions with the estrogen receptor and may be associated with foetal death and resorption. There are no indications of developmental effects in a similar group (SRGO) nor robust evidence of endocrine disruptor activity. However, it is recognized that the database is limited and the potential role of PACs is not fully understood and hence it is requested that the study includes a cohort to fully evaluate the effects on the developing offspring.*"

ECHA agrees that the criteria to extend the Cohort 1B are met, because:

The use of the Substance reported in the joint submission leads to significant exposure of consumers because the Substance is used by consumers as lubricants.

- Substances in the OtherGO category are UVCBs. There is no *in vivo* toxicokinetic data of OtherGO substances. However, there is evidence to suggest that PAH, small molecules and short chain aliphatic molecules penetrate the dermis (to a limited extent). There is no data determining steady-state, as you stated "*it is difficult to apply standard methodology for assessing absorption, distribution, and metabolism*".

- In addition, there are indications for endocrine-disrupting modes of action because information from existing data have shown that "*in vitro EST work found a correlation between 3–7 ring PAH content and a reduction in embryo differentiation. The work has suggested that it is the 4–7 PAH's that are mostly, if not solely, responsible for embryotoxicity and Ah-receptor binding. The mode of action for PAH embryotoxicity is not fully understood and additional work is ongoing to fully understand the role of individual PAHs or specific types of PAHs. However, for the purposes of hazard identification, based on the available data, the following statements can be made about the reproductive toxicity potential of aromatic constituents: (i) <3-ring polycyclic aromatics exert no effect on reproductive organs and no selective developmental effects; while (ii) ≥3-ring polycyclic aromatics (mainly 4-7 ring PAHs) with specific structures (not necessarily present in gas oils) are associated with systemic toxicity (effects on liver, thymus and blood forming organs but not reproductive organs) and are potentially mutagenic and dermal carcinogens. In developmental studies they produce foetal death and resorption. Recent* in vitro *work suggests that ≥3 ring polycyclic aromatics can alter embryo development.*"

Therefore, Cohort 1B must be extended. The F2 generation must be followed to weaning, allowing assessment of nursing and lactation of the F1 parents and postnatal development of F2 offspring. Investigations for F2 pups must be similar to those requested for F1 pups in OECD

TG 443 and described in OECD GD 151. It is recommended to aim to 20 litter per dose group in order to have similar statistical power for investigations than in P0 generation.

Cohorts 2A and 2B

The developmental neurotoxicity Cohorts 2A and 2B need to be conducted in case of a particular concern on (developmental) neurotoxicity.

You proposed to include Cohorts 2A and 2B and provided justification following the Column 2 criteria: "*Neurotoxicity endpoints are not fully addressed in the existing database, a slight delay in startle response was observed in one read-across study which may be indicative of a neurotoxic effect*"

ECHA agrees that the criteria to include Cohorts 2A and 2B are met, because existing information on analogue substances (substances from the OtherGO category, or other categories) derived from an *in vivo* study using diesel fuel ([redacted], 1984), show that "*there was a statistically significant increase in auditory startle of 1 – 2 mSecs. The significance of this change is difficult to interpret as other studies yielded differences of 17 – 20 mSec. This was the only parameter examined.*"

ECHA concludes that the developmental neurotoxicity cohorts 2A and 2B need to be conducted because there is a particular concern on (developmental) neurotoxicity.

Cohort 3

The developmental immunotoxicity Cohort 3 needs to be conducted in case of a particular concern on (developmental) immunotoxicity.

You proposed to include Cohort 3 and provided justification following Column 2 criteria: "*OtherGO are classified as 'May cause damage to blood, thymus, and liver through prolonged or repeated exposure' and these organs are associated with the immune system.*"

ECHA agrees that the criteria to include Cohort 3 are met, because you reported that "*in two dermal studies conducted with cracked gas oils there were effects on haematology, thymus, spleen, lymph nodes, and bone marrow. It should be noted that groups affected also had severe skin reaction so were subject to notable toxicity and stress. However, it cannot be definitively stated that the observations were secondary to this. This group carries the classification 'H373: May cause damage to blood, thymus, and liver through prolonged or repeated exposure' and there is a concern that the effects on the developing foetus and neonate have not been fully assessed. Blood, thymus and liver are target organs of OtherGO toxicity and hence immunotoxicity investigations are required.*"

ECHA concludes that the developmental immunotoxicity Cohort 3 needs to be conducted because there is a particular concern on (developmental) immunotoxicity.

Species and route selection

You proposed testing by oral route in rats. ECHA agrees with your proposal.

You should consider performing a dose-range finding study to identify the best administration, and decide between dietary ("*chosen in consultation with contract research organisation experts*") and gavage. You considered that "*dietary administration will lead to a continued steady exposure and the peak-and-trough toxicokinetics often observed with oral gavage dosing should be avoided. In addition, the procedure of gavage administration may result in mild stress, which may influence some of the sensitive outcomes in this type of study*."

ECHA agreed with the test design you proposed in your testing proposal (i.e. the addition of extra cohorts). In your comments on the draft decision, you requested the possibility to adapt the proposed test design based on *"additional data generated as part of the testing programme"*. However, no additional information is submitted with your comments. Therefore, based on the information currently available, the study is requested with the design described above.

Selection of the substance tested

In your comments, you argued that you now consider that the Substance is no longer the most representative substance for the OtherGO category (worst-case based on the content of PAHs with 3-7 aromatic rings). However, you have not substantiated your comments with any detailed analytical information supporting your statement. Your choice of substance to test was based on the considerations you set out in your read-across justification and ECHA considered this reasoning plausible in the context of the read-across. Therefore, based on the information currently available, the study is requested on the Substance.

Outcome

Under Article 40(3)(a) of the REACH Regulation, you are requested to carry out the proposed test with the Substance.

Appendix D: Requirements to fulfil when conducting and reporting new tests for REACH purposes

A. Test methods, GLP requirements and reporting

1. Under Article 13(3) of REACH, all new data generated as a result of this decision must be conducted according to the test methods laid down in a European Commission Regulation or to international test methods recognised by the Commission or ECHA as being appropriate.
2. Under Article 13(4) of REACH, ecotoxicological and toxicological tests and analyses must be carried out according to the GLP principles (Directive 2004/10/EC) or other international standards recognised by the Commission or ECHA.
3. Under Article 10(a)(vi) and (vii) of REACH, all new data generated as a result of this decision must be reported as study summaries, or as robust study summaries, if required under Annex I of REACH. See ECHA Practical Guide on How to report robust study summaries[10].

B. Test material

Before generating new data, you must agree within the joint submission on the chemical composition of the material to be tested (Test material) which must be relevant for all the registrants of the Substance.

1. Selection of the Test material(s)
   The Test material used to generate the new data must be selected taking into account the following:
   - the variation in compositions reported by all members of the joint submission,
   - the boundary composition(s) of the Substance,
   - the impact of each constituent/ impurity on the test results for the endpoint to be assessed. For example, if a constituent/ impurity of the Substance is known to have an impact on (eco)toxicity, the selected Test material must contain that constituent/ impurity.
2. Information on the Test material needed in the updated dossier
   - You must report the composition of the Test material selected for each study, under the "Test material information" section, for each respective endpoint study record in IUCLID.
   - The reported composition must include the careful identification and description of the characteristics of the Tests Materials in accordance with OECD GLP (ENV/MC/CHEM(98)16) and EU Test Methods Regulation (EU) 440/2008 (Note, Annex), namely all the constituents must be identified as far as possible as well as their concentration. Also any constituents that have harmonised classification and labelling according to the CLP Regulation must be identified and quantified using the appropriate analytical methods,
   - Considering the toxicity of PAHs, you must report the total percentage and, as far as technically feasible, the individual percentages and identification of each PAH in the test material to assess whether it is representative of the Substance.

This information is needed to assess whether the Test material is relevant for the Substance and whether it is suitable for use by all members of the joint submission. Technical instructions on how to report the above is available in the manual on How to prepare registration and PPORD dossiers.[11]

[10] https://echa.europa.eu/practical-guides
[11] https://echa.europa.eu/manuals

Appendix E: Procedure

ECHA started the testing proposal evaluation in accordance with Article 40(1) on 8 February 2019, following the necessary clarification of the identity of your substance.

ECHA held a third party consultation for some of the testing proposals from 28 February 2019 until 15 April 2019. ECHA did not receive information from third parties.

ECHA held a second third party consultation for the testing proposal from 14 May 2020 until 29 June 2020. ECHA did not receive information from third parties.

This testing proposal examination decision does not prevent ECHA from initiating compliance checks at a later stage on the registrations present.

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.

In your comments on the draft decision, you made several remarks not relating to the Substance and the studies requested in the present decision, but concerning the OtherGO category:

- you argued that since the Substance is part of the OtherGO category, there is a *"need to fully clarify the OtherGO category prior to [the] request further […] testing."*
- you made further comments regarding the strategy you wish to follow for the "*complicated UVCB nature of petroleum substances*", without providing additional information or data to support your arguments, such as *"the need to further clarify the Other GO category prior to request [further test]", "pending endorsement of the proposed category, any request for higher tier testing on any of the OtherGO substances […] may prove unnecessary"*.
- you indicated your understanding "*that no testing will be requested on the remaining OtherGO substances prior to the full clarification of the category*".
- you highlighted your difficulty of "*applying ECHA's RAAF… for UVCB substances*" and you request ECHA to provide some methodology to compensate the lack of ECHA Guidance, or to "*approve the testing strategy proposed by the registrants"*.
- You provided statements related to the UVCB status and collaboration with ECHA linked to clarifying the identification (methods) of the OtherGO substances.

However, these remarks are not relevant for the present decision. The present decision addresses exclusively the testing proposals you submitted specifically on the Substance. The purpose of the present decision is therefore not to assess your grouping strategy or real information needs for other substances of the OtherGO category. This will be assessed after the submission of the information requested in this decision and in decisions on other substances of the category. This assessment will also take account of other data that you may generate in order to support lower tier information on all the substances of the category.

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

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

ECHA invited you to comment on the proposed amendments and referred the modified draft decision to the Member State Committee.

You did not provide comments on the proposed amendment(s) within the deadline set.

The Member State Committee reached a unanimous agreement on the draft decision in its MSC-74 written procedure and ECHA took the decision according to Article 51(6) of the REACH Regulation.

Appendix F: List of references - ECHA Guidance[12] and other supporting documents

Evaluation of available information
Guidance on information requirements and chemical safety assessment, Chapter R.4 (version 1.1., December 2011), referred to as ECHA Guidance R.4 where relevant.

QSARs, read-across and grouping
Guidance on information requirements and chemical safety assessment, Chapter R.6 (version 1.0, May 2008), referred to as ECHA Guidance R.6 where relevant.

Read-across assessment framework (RAAF, March 2017)[13]

RAAF - considerations on multi-constituent substances and UVCBs (RAAF UVCB, March 2017)[13]

Physical-chemical properties
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Toxicology
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

Data sharing
Guidance on data-sharing (version 3.1, January 2017), referred to as ECHA Guidance on data sharing in this decision.

OECD Guidance documents[14]
Guidance Document on aqueous–phase aquatic toxicity testing of difficult test chemicals – No 23, referred to as OECD GD 23.

Guidance document on transformation/dissolution of metals and metal compounds in aqueous media – No 29, referred to as OECD GD 29.

Guidance Document on Standardised Test Guidelines for Evaluating Chemicals for Endocrine Disruption – No 150, referred to as OECD GD 150.

Guidance Document supporting OECD test guideline 443 on the extended one-generation reproductive toxicity test – No 151, referred to as OECD GD 151.

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

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

You must provide the information requested in this decision for all REACH Annexes applicable to you.

Registrant Name	Registration number	(Highest) Data requirements to be fulfilled
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]

Note: where applicable, the name of a third party representative (TPR) may be displayed in the list of recipients whereas the decision is sent to the actual registrant.