EUROPEAN CHEMICALS AGENCY

Helsinki, 19 August 2019

Addressee: [redacted]

Decision number: CCH-D-2114479848-24-01/F
Substance name: N-(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)-2-[(4-nitrophenyl)azo]-3-oxobutyramide
EC number: 258-221-5
CAS number: 52846-56-7
Registration number: [redacted]
Submission number: [redacted]
Submission date: 29/05/2015
Registered tonnage band: 10-100

DECISION ON A COMPLIANCE CHECK

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

1. **In vitro 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) with the registered substance;**

2. **In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.; test method: OECD TG 476 or TG 490) with the registered substance, provided that the study requested under 1. has a negative result;**

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

You have to submit the requested information in an updated registration dossier by ***28 February 2022***. You shall also update the chemical safety report, where relevant.

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

Appeal

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

Authorised[1] by Wim De Coen, Head of Unit, Hazard Assessment.

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

Appendix 1: Reasons

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

Grouping of substances and read-across approach

Your registration dossier contains for multiple endpoints adaptation arguments in the form of a grouping and read-across approach under Annex XI, Section 1.5. of the REACH Regulation.

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

- *in vitro* cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2.);
- *in vitro* gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.);
- sub-acute toxicity study (28 days; Annex VIII, Section 8.6.1.); and
- screening for reproductive/developmental toxicity (Annex VIII, Section 8.7.1.);

ECHA has considered first the scientific and regulatory validity of your read-across approach in general before assessing the information request for the individual endpoints.

According to Annex XI, Section 1.5., two conditions shall be necessarily fulfilled. Firstly, there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category.

Secondly, it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (read-across approach). ECHA considers that the generation of information by such alternative means should offer equivalence to prescribed tests or test methods.

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

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

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

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

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

A. Scope of the category

You have provided a category documentation in the CSR (sections 1.1 Category definition and its members, 1.2 Category justification, and 1.3 Data matrix).

You have defined the structural basis for the category/grouping, including its boundaries, as follows: "*The pigments grouped in this category are structurally similar and contain a substituted phenyl moiety, an azo moiety, an acetoacetyl moiety and a benzimidazolone moiety, which is connected to the central part of the molecule via an amide bound [...]. Differences between the various Acetolone Pigments are due to the different identity of the substituents of the phenyl ring.*" There are 4 substitution sites (R^1, R^2, R^3 and R^4) at the phenyl ring, and the allowed substituents "*may vary between* [redacted] Category member substance substituents at these sites are: [redacted]

You have identified the following substances as 'Acetolone pigments' category members:

[1] pigment yellow (PY) 120: dimethyl 5-[[1-[[(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)amino]carbonyl]-2-oxopropyl]azoterephthalate (EC No 249-955-7, CAS No 29920-31-8);

[2] PY 194: N-(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)-2-[(2-methoxyphenyl)azo]-3-oxobutyramide (EC No 279-914-9, CAS No 82199-12-0);

[3] PY 181: N-[4-(aminocarbonyl)phenyl]-4-[[1-[[(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)amino]carbonyl]-2-oxopropyl]azo]benzamide (EC No 277-873-1, CAS No 74441-05-7);

[2] Read-Across Assessment Framework (RAAF). 2017 (March) ECHA, Helsinki. 60 pp. Available online: Read-Across Assessment Framework (https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across)
[3] Read-across assessment framework (RAAF) - considerations on multi-constituent substances and UVCBs. 2017 (March) ECHA, Helsinki. 40 pp. Available online: https://echa.europa.eu/publications/technical-scientific-reports

[4] PY 180: 2,2'-[Ethylenebis(oxyphenyl-2,1-eneazo)]bis[N-(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)-3-oxobutyramide (EC No 278-770-4, CAS No 77804-81-0);

[5] PY 154: N-(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)-3-oxo-2-[[2-(trifluoromethyl)phenyl]azo]butyramide (EC No 268-734-6, CAS No 68134-22-5);

[6] PY 151: 2-[[1-[[(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)amino]carbonyl]-2-oxopropyl]azo]benzoic acid (EC No 250-830-4, CAS No 31837-42-0);

[7] PY 175: dimethyl 2-[[1-[[(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)amino]carbonyl]-2-oxopropyl]azo]terephthalate (EC 252-650-1, CAS No 35636-63-6);

[8] pigment orange (PO) 36: 2-[(4-chloro-2-nitrophenyl)azo]-N-(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)-3-oxobutyramide (EC No 235-462-4, CAS No 12236-62-3);

[9] PO 62: N-(2,3-dihydro-2-oxo-1H-benzimidazol-5-yl)-2-[(4-nitrophenyl)azo]-3-oxobutyramide (EC 258-221-5, CAS No 52846-56-7);

ECHA notes the following shortcomings with regards to your category definition.

B. Prediction of toxicological properties

You have provided the following hypothesis for the prediction of toxicological properties:
"The category hypothesis is that all members share a very similar chemical structure [...], which is the basis for similar physical-chemical properties. These properties
- very low solubility in water but also in organic solvents and lipophilic matrices
- non-degradability
lead to inert behaviour and negligible bioavailability [...]."

In addition, you have provided the following justification: *"Additionally, experimental data and physical-chemical properties further indicate that the bioavailability of the Acetolone Pigments after oral, dermal or inhalative exposure is negligible. Lacking bioavailability is probably the reason for the absence of any relevant mammalian toxicity. None of the category members showed a toxic effect after single oral or inhalational exposure, no skin or eye irritation, no skin sensitizing effect, no toxicity after repeated oral exposure and no mutagenic properties."*

ECHA understands from this hypothesis that you base your predictions on the assumption that different compounds have similar toxicological properties as a result of structural similarity and similar physio-chemical properties. As an integral part of this prediction, you assume absence of toxicity due to the fact that the category members have negligible bioavailablility.

ECHA notes the following shortcomings with regards to prediction of toxicological properties.

Structural dissimilarities

Structural similarity is a prerequisite for applying the grouping and read-across approach according to REACH Annex XI, Section 1.5. As outlined in Read-Across Assessment Framework (RAAF) 2017 (March), section 3.2, in order to meet the provisions in Annex XI, Section 1.5. to predict human health effects from data for a reference substance within the group by interpolation to other substances in the group, ECHA considers that structural similarity alone

is not sufficient. It has to be justified why such prediction is possible in view of the identified structural differences and the provided evidence has to support such explanation. In particular, the structural similarities must be linked to a scientific explanation of how and why a prediction is possible.

In the applicability domain section of your category documentation you identified elements of structural similarity among the category members as well as structural differences, namely the allowed different phenyl ring substituents. You have not, however, provided any considerations on these structural differences and in particular on the potential impact of these structural differences on toxicological properties.

Thereby, ECHA concludes that you have not addressed the obvious structural differences between the source substances and the target substance and did not explain why those differences would not lead to differences in the toxicity profile of target and source.

Consistency between the available data and the read-across hypothesis

According to the ECHA Guidance on information requirements and chemical safety assessment Chapter R.6.2, Section R.6.2.2.2, (version 1.0, May 2008) *"a demonstration of consistent trends in the behaviour of a group of chemicals is one of the desirable attributes of a chemical category and one of the indicators that a common mechanism for all chemicals is involved"*.

In your read-across hypothesis, you assume that due to structural similarity among the category members, they have low solubility and are not prone to transformation (i.e. non-degradability), which in turn leads to inert behaviour and negligible bioavailability. ECHA considers that your claim of negligible bioavailabilityis not demonstrated. Studies showing no toxicity up to the limit dose cannot be considered as a proof of low bioavailability. In your comments to the draft decision you accept that a proof of the hypothesis is not yet available. More specifically, you propose *"extensive investigations in chemico, in vitro and, finally, in vivo to determine the bioavailability of a number of representative pigments [...] including the acetolone pigment."*

Furthermore, your claim of negligible bioavailability is not supported by the available data because one provided reproductive/developmental toxicity screening test (according to OECD TG 422) showed effects. More specifically, a statistically significant reduction in sperm motility has been reported with the category member PO 36. In your comments to the draft decision you indicated that this *"is an isolated finding of reduced sperm motility in one single study which, however, was considered as non-adverse by the authors of the study."* Furthermore, you comment that since sperm motility is not a standard parameter in a screening study according to OECD TG 422, and a number of factors influence sperm motility and its measurement, interpretation of the results should be very careful. You conclude the following: *"consideration of all available information led the authors of the dossier to the conclusion, that the observation is most likely not due to an effect of the pigment but a unique, spurious result of unknown origin, which does not devalue the plausible hypothesis of "no bioavailability"."* However, you also state that, *"though it cannot be explicitly excluded to be an effect of the registered substance, it can also not be excluded, that it is a completely incidental finding."* ECHA notes that *"effects on sperm motility which might be test item-related were noted in all dose groups"* by the author of the study, and that no other studies with seminological investigations have been provided for any other category member substances. Comparison of the category members with regard to this particular toxicological property is therefore not possible. ECHA considers these statistically significant sperm motility findings toxicologically relevant, irrespective if the authors considered them as non-adverse,

and as evidence that the tested substance is bioavailable, and is not inert. The finding is in contradiction with your claim that "*lacking bioavailability is probably the reason for the absence of any relevant mammalian toxicity*."

Regarding the further intended supporting bioavailability investigations indicated in your comments to the draft decision, ECHA notes that it is in your discretion to generate and provide any data to support of your category hypothesis.

ECHA therefore concludes that, as the observation on sperm motility cannot be excluded as a test item-related effect based on the information currently available to ECHA, your read-across hypothesis is not supported by sufficient information and even contradicted. Concequently, this hypothesis can not be verified nor accepted as basis of any reliable predictions.

Data density

A number of factors contributes to the robustness of a category. According to the ECHA Guidance on information requirements and chemical safety assessment Chapter R.6.2, Section R.6.2.1.5., (version 1.0, May 2008), one of these factors is the density and distribution of the available data across the category. In order to derive reliable prediction of the properties of the members of the category, adequate and reliable information covering the range of structural variations identified among the category members needs to be available.

You claim that there is "*no toxicity after repeated oral exposure and no mutagenic properties*" for the category member substances. ECHA has the following observations:

Low data density

1. As regards genotoxicity, 5 of the 9 included category members (PY 120, PY 181, PY 180, PY 151 and PO 62) were tested only in a bacterial reverse mutation assay. In addition, 3 out of 9 included category members (PY 194, PY 154 and PO 36) were tested in *in vitro* test for mammalian chromosomal aberrations or *in vivo* in a micronucleus study, and 2 out of 9 included category members (PY 175 and PO 36) were tested in an *in vitro* test for mammalian gene mutation.

   In your comments to the draft decision, you state that "*none of the available results of 14 in vitro mutagenicity studies indicates any effect of the nine different test materials in these systems. This is in line with the basic hypothesis for Acetolone-category formation of "no bioavailability"*." You therefore consider that, "*as all of the mutagenicity results indicate an identical behavior of all substances (i.e. no effects) any sub-set can be considered representative*."

2. As regards repeated dose toxicity, 7 out of 9 included category members (PY 120, PY 194, PY 181, PY 180, PY 151, PY 175 and PO 62) do not have reliable data provided on oral toxicity. One of the included category members (PO 36) have been tested in a combined repeated dose toxicity study with the reproductive/ developmental toxicity screening test (OECD 422), and another included category member (PY 154) in a sub-acute toxicity study (28-day) (OECD 407) and in a reproductive/developmental toxicity screening test (OECD 421).

   In your comments to the draft decision you state that, in absence of any adverse

effects, the available provided screening studies with PY 154 and PO 36 "*confirm each other and support the hypothesis of very limited bioavailability of pigments as do many other studies on pigments.*" Furthermore, you refer to number of repeated dose toxicity studies and reproductive toxicity studies available in the ECHA database on structurally variable types of pigments indicating no adverse effects.

3. As regards reproductive toxicity, a reproductive/developmental toxicity screening test (OECD 421) with included category member PY 154, and a combined repeated dose toxicity study with the reproductive/developmental toxicity screening test (OECD 422) with included category member PO 36 have been provided. ECHA notes that statistically significant reduction in sperm motility have been reported in the screening study with PO 36 (see 'Consistency between the available data and the read-across hypothesis') while no other studies with seminological investigations have been reported for any other category member substances.

   Therefore, ECHA notes that comparison of the category members with regard to this particular toxicological property is not possible. Furthermore, you did not consider the observed effect in selection of the No-Observed-Adverse-Effect-Level (NOAEL) nor addressed it other way in interpretation of the results.

ECHA notes the justifications you provided in the comments to the draft decision why you consider the tested substances to be representative of the other category members with regard to genetic toxicity properties and repeated dose toxicity.

ECHA notes, however, that i) as discussed above, further data for confirming your category hypothesis is required, ii) the data available in your dossier does not allow comparison of individual toxicological endpoints across the category member substances, and iii) you have not provided the details of the referred additional studies in your documentation and explained the relevance of such analogue substance data to your category structures. ECHA stresses that any information supporting your category hypothesis should be included and discussed in your documentation in order for ECHA to take it into account in evaluation of your case.

Hence, considering the applicability domain of the category and the distinct structural differences between the members of the category, ECHA still considers that there are too few data points (i.e. low data density) in the current data matrix for demonstrating absence of effects due to no bioavailability and making the suggested predictions for the listed toxicological endpoints.

Missing data

ECHA notes that bioavailability with regards to human health endpoints has not been investigated in any of the provided studies.

In your comments to the draft decision you acknowledge that no data from specific studies on pre-natal developmental toxicity have been provided. You also note that 28 reproductive toxicity studies (according to OECD TG 421/422 or 414) performed for different pigment classes did not show any adverse effect. You consider that absence of adverse effects in these studies, in combination with the assumed little or no bioavailability, are sufficient reasons to waive the developmental *in vivo* studies. You consider conducting further studies as "*scientifically not justified as unexpected adverse effects can be excluded with high probability.*" Furthermore, you argue that "*performance of animal studies for purely formal reasons without the expectations of added scientific knowledge is considered unethical and a*

breach of Art. 25 of the REACH Regulation." However, you have not provided the details of the referred studies in your documentation and explained the relevance of such analogue substance data to your category structures. ECHA stresses that comments and references provided are of general nature, and any specific information supporting your category hypothesis should be included and discussed in your documentation in order for ECHA to take it into account in evaluation of your case.

To adapt a standard information requirement, the REACH Regulation provides either specific adapation possibilities for repeated dose toxicity (Annex VIII, Section 8.6.1., column 2) and reproductive toxicity (Annex VIII, Section 8.7.1., column 2) or general adaptation possibilities of Annex XI, Section 1, when testing does not appear scientifically necessary. However, the information currently provided is not sufficient to fulfll any of those adaptation possibilities. Hence, the requests cannot be considered as unethical and breach of Art. 25 of the REACH Regulation.

Conclusion

Overall, ECHA considers that the currently provided supporting data do not establish a scientifically credible link between structural similarity and the predicted toxicological endpoints, and is not sufficient to predict human health properties of the registered substance.

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

In your comments to the draft decision, you propose to postpone the definitive decision and the current requests until plan to generate proposed supporting information to your category hypothesis has been discussed and agreed with ECHA, including the review of the first results of any such proposed investigations. ECHA notes that no testing plan has been agreed and future studies with uncertain results cannot justify delaying the decision-making procedure.

1. 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 according to Annex XI, Section 1.5. of the REACH Regulation by providing

- Read-across key study: "*in vitro* mammalian chromosome aberration test" (OECD TG 473) with the analogue substance PY 154 (EC no 268-734-6), [redacted] 2012 (study report), rel. 1.
- Read-across key study: "*in vitro* mammalian chromosome aberration test" (OECD TG 473) with the analogue substance PY 194 (EC no 279-914-9), [redacted] 2011 (study report), rel. 1
- Read-across key study: "mammalian erythrocyte micronucleus test" (according to

OECD 474) with the analogue substance PO 36 (EC no 235-462-4), [redacted]1981 (study report), rel. 2.

However, as explained above in Appendix 1, section "Grouping of substance and read-across approach" of this decision, your adaptation of the information requirement is rejected.

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

ECHA considers that the *in vitro* mammalian chromosome aberration test (test method OECD TG 473) and the *in vitro* mammalian cell micronucleus test (OECD TG 487) are appropriate to address the standard information requirement of Annex VIII, Section 8.4.2. of the REACH Regulation.

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

2. 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.

Adequate information *on in vitro* gene mutation in mammalian cells will however need to be present in the technical dossier for the registered substance to meet this information requirement provided that the study requested under 1 has a negative result. ECHA set the deadline for provision of the information to allow for sequential testing.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing

- Read-across key study: "*in vitro* gene mutation study in mammalian cells" (OECD TG 476) with the analogue substance PY 175 (EC no 252-650-1), [redacted]2005 (study report), rel. 1
- Read-across key study: "*in vitro* gene mutation study in mammalian cells" (OECD TG 476) with the analogue substance PO 36 (EC no 235-462-4), [redacted]2012 (study report), rel. 1

However, as explained above in Appendix 1, section "Grouping of substance and read-across approach" of this decision, your adaptation of the information requirement is rejected.

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

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 the study requested under 1. has a negative result.

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

A "short-term repeated dose toxicity study (28 days)" is a standard information requirement as laid down in Annex VIII, Section 8.6.1. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing the following sources of information:

- Read-across key study: "screening for reproductive /developmental toxicity" in rat, oral route (OECD TG 422; GLP) with the analogue substance PO 36 (EC no 35-462-4), [redacted]2012 (study report), rel. 1.
- Read-across key study: "sub-acute toxicity study (28 days)" in rat, oral route (OECD 407; GLP) with the analogue substance PY 154 (EC No 268-734-6), [redacted]2012 (study report), rel. 1
- Read-across supporting study: "sub-acute toxicity study (30 days)" in rat, oral route, (no guideline; non-GLP) with the analogue substance PY 151 (EC No 250-830-4), [redacted]1976 (study report), rel. 3.
- Read-across supporting study: "sub-acute toxicity study (30 days)" in rat, oral route (no guideline; non-GLP), with the analogue substance PY 120 (EC No 249-955-7), [redacted] 1971 (study report), rel. 3.
- Read-across supporting study: "sub-acute toxicity study (19 days)" in rat, oral route, (no guideline; non-GLP) with the analogue substance PO 36 (EC no 35-462-4), [redacted]1964 (study report/publication), rel. 3.

ECHA has first evaluated your adaptation according to Annex XI, Section 1.5. of the REACH Regulation. However, as explained above in Appendix 1, section 'Grouping of substances and read-across approach' of this decision, your adaptation of the information requirement according to Annex XI, Section 1.5. is rejected.

Second, ECHA notes that you considered the three provided short-term repeated dose toxicity studies with analogue substances not reliable (non-GLP, reliability 3) as they relevantly deviate from the current test guidelines. ECHA has therefore not considered these studies.

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

When there is no information available neither for the 28-day repeated dose toxicity endpoint (EU B.7, OECD TG 407), nor for the screening study for reproductive/developmental toxicity

(OECD TG 421 or TG 422), the conduct of a combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (OECD TG 422) is preferred to ensure that unnecessary animal testing is avoided. Such an approach offers the possibility to avoid carrying out a 28-day study according to OECD TG 407, because the OECD TG 422 can at the same time fulfil the information requirement of REACH Annex VIII, 8.6.1 and that of REACH Annex VIII, 8.7.1.[4]

ECHA has evaluated the most appropriate route of administration for the study. Based on the information provided in the technical dossier and/or in the chemical safety report, ECHA considers that the oral route - which is the preferred one as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.5.4.3 - is the most appropriate route of administration. More specifically, you have adapted repeated dose toxicity studies by the inhalation route with the following justification *"exposure of humans via inhalation is considered unlikely taking into account the vapour pressure of the substance and/or the possibility of exposure to aerosols, particles or droplets of an inhalable size."* ECHA notes that the registered substance contains particles of inhalable size (D50 1.6 µm). However, no specific information on uses indicating inhalation exposure has been provided. Hence, the test shall be performed by the oral route using the test method OECD TG 422. Furthermore, because an effect on reproductive toxicity was observed with the structurally related substance PO 36 using oral gavage administration, testing should be performed using oral gavage administration.

According to the test method 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.

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

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

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

You have not provided any study record of a screening for reproductive/developmental toxicity in the dossier that would meet the information requirement of Annex VIII, Section 8.7.1.

You have sought to adapt this information requirement according to Annex XI, Section 1.5. of the REACH Regulation by providing study records of two "screening for reproductive/developmental toxicity" studies with analogous substances:

[4] ECHA Guidance, Section R.7.6.2.3.2., pages 484 to 485 of version 6.0 – July 2017. (https://echa.europa.eu/documents/10162/13632/information_requirements_r7a_en.pdf)

- Read-across key study: "screening for reproductive / developmental toxicity" in rat, oral route (OECD TG 421; GLP) with the analogue substance PY 154 (EC no 268-734-6), [redacted]2012 (study report), rel. 1.

- Read-across key study: "screening for reproductive / developmental toxicity" in rat, oral route (OECD TG 422; GLP) with the analogue substance PO 36 (EC no 235-462-4), [redacted]2012 (study report), rel. 1.

However, as explained above in Appendix 1, section "Grouping of substances and read-across approach" of this decision, your adaptation of the information requirement according to Annex XI, Section 1.5. is rejected.

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

When there is no information available neither for the 28-day repeated dose toxicity endpoint (EU B.7, OECD TG 407) (as explained above under section 3.), nor for the screening study for reproductive/developmental toxicity (OECD TG 421 or TG 422), the conduct of a combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (OECD TG 422) is preferred to ensure that unnecessary animal testing is avoided. Such an approach offers the possibility to avoid carrying out a 28-day study according to OECD TG 407, because the OECD TG 422 can at the same time fulfil the information requirement of REACH Annex VIII, 8.6.1 and that of REACH Annex VIII, 8.7.1.[5]

ECHA considers that the oral route is the most appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 6.0, July 2017) Chapter R.7a, Section R.7.6.2.3.2. Since the substance to be tested is a solid, ECHA concludes that testing should be performed by the oral route. Furthermore, because an effect on reproductive toxicity (as indicated below under "Notes for your consideration") was observed using oral gavage administration, testing should be performed using oral gavage administration.

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.

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 (gavage).

Notes for your consideration

Due to the observed effect on reproductive toxicity (sperm motility) in the screening study provided for the structurally related substance PO 36, investigations for testicular effects should be included in the screening study performed with the registered substance for addressing this effect.

[5] ECHA Guidance, Section R.7.6.2.3.2., pages 484 to 485 of version 6.0 – July 2017. (https://echa.europa.eu/documents/10162/13632/information_requirements_r7a_en.pdf)

ECHA considers the assessment of sperm parameters in accordance with paragraph 39 of the OECD TG 408 (including sperm motility) would ensure the optimal use of animal testing in your case, and can be included in a screening study without compromising the validity of the test as a whole.

Appendix 2: Procedural history

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

The compliance check was initiated on 29 June 2018.

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

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

ECHA took into account your comments and did not amend the request(s) and/or the deadline.

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

As no amendments were proposed, ECHA adopted the decision under Article 51(3) of the REACH Regulation.

Appendix 3: Further information, observations and technical guidance

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

2. Failure to comply with the requests in this decision will result in a notification to the enforcement authorities of your Member State.

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

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

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