~~Confidential~~

Helsinki, 24 January 2024

Addressees
Registrants of JS_28198-05-2_[redacted] as listed in Appendix 3 of this decision

Date of submission of the dossier subject to this decision
08 November 2017

Registered substance subject to this decision ("the Substance")
Substance name: 1,4-bis[(4-butylphenyl)amino]-5,8-dihydroxyanthraquinone
EC/List number: 248-895-9

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

DECISION ON A COMPLIANCE CHECK

Under Article 41 of Regulation (EC) No 1907/2006 (REACH), you must submit the information listed below by ***4 May 2026***.

Requested information must be generated using the Substance unless otherwise specified.

Information required from all the Registrants subject to Annex VII of REACH

1. Skin sensitisation (Annex VII, Section 8.3.)
   a) *in vitro/in chemico* skin sensitisation information on molecular interactions with skin proteins (OECD TG 442C), inflammatory response in keratinocytes (OECD TG 442D) and activation of dendritic cells (OECD TG 442E) (Annex VII, Section 8.3.1.); and
   b) only if the *in vitro/in chemico* test methods specified under point a) above are not applicable for the Substance or the results obtained are not adequate for classification and risk assessment, *in vivo* skin sensitisation (Annex VII, Section 8.3.2.; test method: EU B.42./OECD TG 429).

2. *In vitro* gene mutation study in bacteria (Annex VII, Section 8.4.1.; test method: Bacterial reverse mutation test, OECD TG 471).

3. Long-term toxicity testing on aquatic invertebrates (triggered by Annex VII, Section 9.1.1., Column 2; test method: EU C.20./OECD TG 211)

4. Growth inhibition study aquatic plants (Annex VII, Section 9.1.2.; test method: EU C.3./OECD TG 201)

The reasons for the requests are explained in Appendix 1.

Information required depends on your tonnage band

You must provide the information listed above for all REACH Annexes applicable to you in accordance with Articles 10(a) and 12(1) of REACH. The addressee of the decision and its corresponding information requirements based on registered tonnage band are listed in Appendix 3.

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 requirements for testing and reporting new tests under REACH, see Appendix 4.

Appeal

This decision, when adopted under Article 51 of REACH, may be appealed to the Board of Appeal of ECHA within three months of its notification to you. Please refer to http://echa.europa.eu/regulations/appeals for further information.

Failure to comply

If you do not comply with the information required by this decision by the deadline indicated above, ECHA will notify the enforcement authorities of your Member State.

Authorised[1] under the authority of Mike Rasenberg, Director of Hazard Assessment

Appendix 1: Reasons for the requests
Appendix 2: Procedure
Appendix 3: Addressees of the decision and their individual information requirements
Appendix 4: Conducting and reporting new tests under REACH

[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 for the requests

Reasons common to several requests

0.1. Weight of evidence adaptation rejected

1 You have adapted the following standard information requirements by using weight of evidence in accordance with Annex XI, Section 1.2.:

- Skin sensitisation (Annex VII, Section 8.3.)
- *In vitro* gene mutation study in bacteria (Annex VII, Section 8.4.1.)

2 Annex XI, Section 1.2. states that there may be sufficient weight of evidence from several independent sources of information enabling, through a reasoned justification, a conclusion on the information requirement, while the information from each single source alone is insufficient to fulfil the information requirement.

3 The justification must have regard to the information that would otherwise be obtained from the study that must normally be performed for this information requirement.

4 According to ECHA Guidance R.4, a weight of evidence adaptation involves an assessment of the relative values/weights of the different sources of information submitted. The weight given is based on the reliability of the data, consistency of results/data, nature and severity of effects, and relevance and coverage of the information for the given regulatory information requirement. Subsequently, relevance, reliability, coverage, consistency and results of these sources of information must be balanced in order to decide whether they together provide sufficient weight to conclude on the corresponding information requirement.

0.1.1. Lack of documentation justifying the weight of evidence adaptation

5 Annex XI, Section 1.2. requires that adequate and reliable documentation is provided to describe a weight of evidence approach.

6 You have not included a justification for your weight of evidence adaptation, which would include an adequate and reliable (concise) documentation as to why the sources of information provide sufficient weight to conclude on the information requirements under consideration.

7 Beside this critical deficiency common to all information requirements under consideration, your weight of evidence approach has additional deficiencies.

8 Additional deficiencies justifying the rejection that are common to all information requirements under consideration are addressed under section 0.1.2.2. below. Deficiencies that are specific for each of the information requirements are addressed under requests 1 and 2.

0.1.2. Reliability assessment of the read-across adaptations

9 For the information requirements listed above you have provided information on structurally similar substances as part of an analogue read-across approach.

10 In addition, you have provided information derived from experimental data from a group of substances using the OECD QSAR Toolbox and flagged the information as QSAR for the information requirements under consideration. As the group of substances are used as source substances to predict the respective toxicological properties of the Substance, we understand that you have also relied on a category read across approach.

11 Therefore, ECHA understands that you intend to predict the toxicological properties of the Substance of the information requirements under consideration, by using both analogue

and category read-across approach under Annex XI, Section 1.5. as part of your weight of evidence adaptation.

12 ECHA has considered the scientific and regulatory validity of your read-across approaches in general before assessing the specific standard information requirements in the following sections.

13 Annex XI, Section 1.5. specifies two conditions which must be fulfilled whenever a read-across approach is used. 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.

14 Additional information on what is necessary when justifying a read-across approach can be found in the Guidance on IRs and CSA, Chapter R.6. and related documents (RAAF, 2017; RAAF UVCB, 2017).

15 A common deficiency between your analogue and category read-across approach is identified under section 0.1.2.1. below. The deficiencies that are specific to the analogue and category read across approach respectively are identified further below under sections 0.1.2.2 and 0.1.2.3.

0.1.2.1. Absence of read-across documentation

16 Annex XI, Section 1.5. requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must include an explanation why the properties of the Substance may be predicted from information on the source substances.

17 You have not provided documentation to explain why information submitted on the source substances is relevant for the Substance and why the properties of the Substance may be predicted from information on those source substances.

18 In the absence of such documentation, the properties of the Substance cannot be reliably predicted from the data on the source substances.

0.1.2.2. Reliability assessment of your analogue read-across approach

0.1.2.2.1. Identification of source substances

19 You predict the relevant toxicological properties of the Substance from information obtained from the following source substances:

20 For skin sensitisation (Annex VII, Section 8.3.):

- 2,2'',6,6''-Tetra-tert-butyl-4,4''-methylenediphenol, EC 204-279-1 (source substance 1);
- Bis(4-octylphenyl)amine, EC 202-965-5 (source substance 2).

21 For in vitro gene mutation study in bacteria (Annex VII, Section 8.4.1.):

- 1-(2-methyl-4-(2-methylphenylazo)phenylazo)-2-naphthol, EC 201-635-8 (source substance 3);
- 1-(2,4-dimethylphenylazo)-2-naphthol, EC 221-490-4 (source substance 4).

22 You provide no reasoning for the prediction of toxicological properties.

23 ECHA understands that your read-across hypothesis assumes that different compounds have the same type of effects. You predict the properties of your Substance to be quantitatively equal to those of the source substances.

0.1.2.2.2. Prediction for toxicological properties

0.1.2.2.2.1. Missing supporting information to compare properties of the substances

24 Annex XI, Section 1.5. requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must provide supporting information to scientifically justify the read-across explanation for prediction of properties. The set of supporting information should strengthen the rationale for the read-across in allowing to verify the crucial aspects of the read-across hypothesis and establishing that the properties of the Substance can be predicted from the data on the source substance(s) (Guidance on IRs and CSA R.6., Section R.6.2.2.1.f.).

25 As indicated above, your read-across hypothesis is based on the assumption that the structurally similar source substance(s) cause the same type of effect(s). In this context, relevant, reliable and adequate information allowing to compare the properties of the source substances is necessary to confirm that the substances cause the same type of effects. Such information can be obtained, for example, from bridging studies of comparable design and duration for the Substance and of the source substances.

26 For the selected analogue substances, you provide the studies used in the prediction in the registration dossier. Apart from those studies, you do not provide any read-across justification explaining why the properties for the source substances can be used to predict the properties of the Substance. In addition, you do not include any information on the Substance that would confirm that the source substances and the Substance cause the same type of effects.

27 In the absence of such information, you have not established that the Substance and the source substances are likely to have similar properties. Therefore you have not provided sufficient supporting information to scientifically justify the read-across.

0.1.2.2.2.2. Inadequate or unreliable source studies

28 According to Annex XI, Section 1.5., if the grouping concept is applied then in all cases the results to be read across must have adequate and reliable coverage of the key parameters addressed in the corresponding study that shall normally be performed for a particular information requirement.

29 Specific reasons why the studies on the source substances do not meet these criteria are explained under requests 1 and 2. Therefore, no reliable predictions can be made for these information requirements.

0.1.2.2.3. Conclusion

30 For the reasons above, you have not established that relevant properties of the Substance can be predicted from data on the source substances. Therefore, the information from your analogue read across approach submitted under your weight of evidence adaptation is not considered reliable.

0.1.2.3. Reliability assessment of your category read-across approach

0.1.2.3.1. Scope of the grouping of substances

31 In this decision, the following abbreviations are used for the category members:

32 For skin sensitisation:

- Cat. member No. 1: 4,4',4''-(1-methylpropanyl-3-ylidene)tris[6-tert-butyl-m-cresol] (EC no. 217-420-7, CAS RN 1843-03-4);
- Cat. member No. 2: 1,4-bis(mesitylamino)anthraquinone (EC no. 204-155-7, CAS RN 116-75-6);
- Cat. member No. 3: Bis(4-(1,1,3,3-tetramethylbutyl)phenyl)amine (EC no. 239-816-9, CAS RN 15721-78-5);
- Cat. member No. 4: Phenol, dodecyl-, sulfurized, calcium salts)EC no. 272-486-4, CAS RN 68855-45-8);
- Cat. member No. 5: Benzene, 1,4-bis(1-methylethyl)-, homopolymer (9CI) (EC no. 687-440-2, CAS RN 25822-43-9);
- Cat. member No. 6: 6,6'-di-tert-butyl-4,4'-butylidenedi-m-cresol (EC no. 201-618-5, CAS RN 85-60-9).

33 For in vitro gene mutation in bacteria:

- Cat. member No. 7: 1,3,4,6,8,13-hexahydroxy-10,11-dimethylphenanthro[1,10,9,8-opqra]perylene-7,14-dione (EC no. 208-941-0, CAS RN 548-04-9);
- Cat. member No. 8: 2,2',4,4',5,5'-hexahydroxy-7,7'-dimethyl-[1S,1'-bianthracene]-9,9',10,10'-tetrone (CAS RN 602-06-2);
- Cat. member No. 9: 4-[[2,3,4,4,6-pentakis(4-hydroxyphenoxy)-1,3,5-triaza-2,4lambda5,6-triphosphacyclohex-4-en-1-yl]oxy]phenol (EC no. , CAS RN 23788-22-9);
- Cat. member No. 10: 2,2',6,6'-tetrabromo-4,4'-isopropylidenediphenol (EC no. 201-236-9, CAS RN 79-94-7);
- Cat. member No. 11: Psi,psi-carotene(EC no. 207-949-1, CAS RN 502-65-8).

34 You justify the grouping of the substances by the following statement: "[the selected substances are the] *nearest neighbours compared by prediction descriptors*."

35 You have provided no definition of the applicability domain of your category.

0.1.2.3.1.1. Incomplete description of the applicability domain of the category

36 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" (Guidance on IRs and CSA, Section R.6.2.4.1.). 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" (Guidance on IRs and CSA, Section R.6.2.1.2.). Therefore, to reliably predict properties within a category the applicability domain should be described. Such description must cover the borders of the category, define unambiguous inclusion- and exclusion criteria, and include a justification for these.

37 You describe the applicability domain of the substances covered by the grouping as: "*Category members are single chemicals or mixtures and are selected based on the profile of the target chemical. Only chemicals having experimental data are listed in the category*." You have not provided a description of the structural requirements and ranges of physico-chemical, environmental fate, toxicological or ecotoxicological properties for the category.

38 This applicability domain does not introduce unambiguous inclusion/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.

0.1.2.3.2. Predictions for toxicological properties

39 You predict the properties of the Substance from information obtained from several source substances. The list of source substances corresponding to the prediction under the respective standard information requirement under consideration is provided under section 0.1.2.3.1.

40 You provide no specific reasoning as to why the selected category members provide relevant information for the prediction of the respective toxicological properties.

41 ECHA understands that your read-across hypothesis assumes that different compounds have the same type of effects. You predict the properties of your Substance to be quantitatively equal to those of the source substances.

0.1.2.3.2.1. Missing supporting information to compare properties of the substances

42 Annex XI, Section 1.5. requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must provide supporting information to scientifically justify the read-across explanation for prediction of properties. The set of supporting information should strengthen the rationale for the read-across in allowing to verify the crucial aspects of the read-across hypothesis and establishing that the properties of the Substance can be predicted from the data on the source substances (Guidance on IRs and CSA R.6., Section R.6.2.2.1.f.).

43 As indicated above, your read-across hypothesis is based on the assumption that the structurally similar source substances cause the same type of effect(s). In this context, relevant, reliable and adequate information allowing to compare the properties of the source substances is necessary to confirm that the substances cause the same type of effects. Such information can be obtained, for example, from bridging studies of comparable design and duration for the Substance and of the source substances.

44 For the selected category members, you provide reference to the data that is used in the prediction in the registration dossier. Apart from the reference to those data, you do not provide any read-across justification explaining why the properties for the source substances can be used to predict the properties of the Substance. In addition, you do not include any information on the Substance that would confirm that both the Substance and the source substances cause the same type of effects.

45 In the absence of such information, you have not established that the Substance and the source substances are likely to have similar properties. Therefore you have not provided sufficient supporting information to scientifically justify the read-across.

0.1.2.3.2.2. Missing robust study summaries

46 Annex XI, Section 1.5. requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must include robust study summary for each source study used in the adaptation.

47 Robust study summaries must provide a detailed summary of the objectives, methods, results, and conclusions of a full study report providing sufficient information to make an independent assessment of the study (Article 3(28)).

48 ECHA understands that your category read-across approach relies on experimental data. You have not provided robust study summaries of the tests conducted with the category members, whose results are the basis for your prediction.

49 You have not provided detailed information on the methods, results and conclusions, allowing for an independent assessment of the source studies. Therefore, you have failed to provide a robust study summary for each source study used in your category read across approach as required by Annex XI, Section 1.5.

0.1.2.3.3. Conclusion

50 For the reasons above, you have not established that relevant properties of the Substance can be predicted from data on the source substances. Therefore, the information from the category read across approach submitted under your weight of evidence adaptation is not considered reliable.

51 As indicated further above under section 0.1.1., additional issues of your weight of evidence adaptation, including those related to the analogue and category read across approaches that have been submitted thereunder, are addressed under requests 1 and 2.

Reasons related to the information under Annex VII of REACH

1. Skin sensitisation

52 Skin sensitisation is an information requirement under Annex VII, Section 8.3. Under Section 8.3., Column 1, the registrants must submit information allowing (1) a conclusion whether the substance is a skin sensitiser and (2) whether it can be presumed to have the potential to produce significant sensitisation in humans (Cat. 1A).

1.1. Information provided

53 You have adapted this information requirement by using Annex XI, Section 1.2. (weight of evidence) based on the following:

(i) A prediction from OECD QSAR toolbox v3.3, Prediction Report (2017);

(ii) A maximization test of Magnusson and Kligman with the source substance 1, EC 204-279-1;

(iii) A modified Landsteiner test with the source substance 2, EC 202-965-5.

1.2. Assessment of the information provided

1.2.1. Assessment whether the Substance causes skin sensitisation

54 In addition to the deficiencies identified in Section 0.1., ECHA identified endpoint specific issues addressed below.

55 Information that can be used to support a weight of evidence adaptation for the information requirements of Annex VII, Section 8.3. includes similar information to that investigated by the internationally recognised in vitro, in chemico and/or in vivo test methods on skin sensitisation. The key parameters of such test methods address each of the 3 key events of skin sensitisation, either individually or in an integrated approach as follows:

(1) investigation of cell proliferation in the draining lymph nodes (local lymph node assay), or

(2) investigation of local responses in animals or humans (guinea pig assays or human studies), or

(3) investigation of molecular interaction with proteins, inflammatory response in keratinocytes and activation of dendritic cells (in vitro and in chemico assays).

56 The source of information (i) may provide information on the key parameters (1) and (2).

57 The sources of information (ii) and (iii) provide information on key parameter (2).

58 You have not provided any information on key parameter 3.

59 However, the reliability of these sources of information is affected by the following deficiencies:

1.2.1.1. Reliability issues of the analogue read-across approach

60 In addition to the deficiencies identified in section 0.1.2.2, ECHA identified endpoint specific issues addressed below.

1.2.1.1.1. Reliability issues of the provided studies (ii) and (iii)

61 EU Method B.6/OECD TG 406 sets out the following specifications:

b) a dose level selection rationale is provided;

In studies (ii) and (iii), no dose level selection rationale was provided. Therefore, it is not possible to understand how these dose levels were selected.

c) the induction concentration is the highest causing mild-to-moderate irritation to the skin;

In studies (ii) and (iii), no information was provided whether the concentration used for induction (0.5% in study (ii), none reported in study (iii)) caused mild-to-moderate irritation.

d) the challenge dose is the highest non-irritation concentration;

In studies (ii) and (iii), no information was provided whether the challenge concentration was the highest non-irritating concentration. Therefore, it is not clear that the animals could not have been exposed to a higher concentration.

e) the appropriate number of animals is included in the study: minimum 10 in test group and 5 in control, if negative results 20 in test group and 10 in control group highly recommended;

In study (iii), no information was provided on the number of animals used. Therefore, it is not possible to evaluate the statistical power of the study.

f) positive and negative controls are included to establish the sensitivity and reliability of the experimental technique;

In studies (ii) and (iii), no information was provided on positive and negative control groups. Therefore, it is not possible to evaluate the acceptability and performance of the tests.

62 Based on the above, the studies (ii) and (iii) submitted in your analogue read across approach, as currently reported in your dossier, cannot be considered reliable sources of information that could contribute to the conclusion on local responses in animals or humans (key parameter 2) investigated by the required study.

1.2.1.2. Reliability issues of the category read-across approach

63 In addition to the deficiencies identified in section 0.1.2.3, ECHA identified endpoint specific issues addressed below.

1.2.1.2.1. Read-across hypothesis contradicted by existing data (source i)

64 Annex XI, Section 1.5. requires that whenever read-across is used adequate and reliable documentation of the applied method must be provided. Such documentation must provide supporting information to scientifically justify the read-across explanation for prediction of properties. The set of supporting information must strengthen the rationale for the read-across in allowing to verify the crucial aspects of the read-across hypothesis and establishing that the properties of the Substance can be predicted from the data on the source substance(s) (Guidance on IRs and CSA R.6, Section R.6.2.2.1.f.).

65 The observation of differences in the toxicological properties between the source substances and the Substance would contradict the hypothesis that the properties of the Substance can be predicted from the data on the source substances. An explanation why such differences do not affect the read-across hypothesis must be provided and supported by scientific evidence.

66 As indicated above, your read-across hypothesis is based on the assumption that the structurally similar Substance and source substances cause the same type of effect(s).

67 However, the results of the information on skin sensitisation obtained with the source substances in the category vary. Specifically, you indicate that 1 category member out of 6 is considered to be a skin sensitiser (Cat. member No. 1).

68 The available set of data on the Substance and on the source substances indicates differences in the relevant toxicological property of the substances. This contradicts your read-across hypothesis whereby the Substance and source substances cause the same type of effect(s). However, you have not supported and scientifically justified why such differences in the relevant toxicological property do not affect your read-across hypothesis.

1.2.1.3. Conclusion

69 While you may have provided information on key parameters 1 and 2, the sources of information (i), (ii) and (iii) have deficiencies affecting their reliability thereby preventing drawing the conclusion on these key parameters.

70 Therefore, it is not possible to conclude, based on any source of information alone or considered together, whether the Substance causes skin sensitisation.

71 Based on the above, your weight of evidence adaptation under Annex XI, Section 1.2. is rejected.

1.2.2. No assessment of potency

72 To be considered compliant and enable a conclusion in cases where the substance is considered to cause skin sensitisation, the information provided must also allow a conclusion whether it can be presumed to have the potential to produce significant sensitisation in humans (Cat. 1A).

73 As the currently available data does not allow to conclude whether the Substance causes skin sensitisation (see section 1.2.1. above), this condition cannot be assessed.

74 Therefore, the information requirement is not fulfilled.

75 In your comment to the draft decision, you agree to perform the requested study.

1.3. Study design

76 To fulfil the information requirement for the Substance, information on molecular interaction with skin proteins and inflammatory response in keratinocytes and activation of dendritic cells (OECD TG 442C and OECD TG 442D and OECD TG 442E) must be provided. Furthermore an appropriate risk assessment is required if a classification of the Substance as a skin sensitiser (Cat 1A or 1B) is warranted.

77 In case no conclusion on the skin sensitisation potency can be made for the Substance based on the existing data or newly generated data, in vivo skin sensitisation study must be performed and the murine local lymph node assay (EU Method B.42/OECD TG 429) is considered as the appropriate study for the potency estimation.

2. *In vitro* gene mutation study in bacteria

78 An in vitro gene mutation study in bacteria is an information requirement under Annex VII, Section 8.4.1.

2.1. Information provided

79 You have adapted this information requirement by using Annex XI, Section 1.2. (weight of evidence) based on the following:

(i) A prediction from OECD QSAR Toolbox version 3.3, Prediction Report (2017);

(ii) An *in vitro* gene mutation in bacteria(1978) with the source substance 3, EC 201-635-8;

(iii) An in vitro gene mutation study in mammalian cells (2006) with the source substance 4, EC 221-490-4.

2.2. Assessment of the information provided

80 In addition to the deficiencies identified in section 0.1., ECHA identified endpoint specific issue(s) addressed below.

81 Information that can be used to support weight of evidence adaptation for the information requirement of Annex VII, Section 8.4.1. includes similar information that is produced by the OECD TG 471. OECD TG 471 requires the study to investigate the following key parameter:

(1) Detection and quantification of gene mutations (base pairs, substitution or frame shift) in cultured bacteria including data on the number of revertant colonies

82 The source of information (iii) does not provide information on detection and quantification of gene mutations in cultured bacteria.

83 The sources of information (i) and (ii) may provide relevant information on the detection and quantification of gene mutations in cultured bacteria. However, the reliability of these sources of information is affected by the following deficiency:

2.2.1. Reliability issues of the analogue read-across approach

84 In addition to the deficiencies identified in section 0.1.2.2, ECHA identified endpoint specific issues addressed below.

2.2.1.1. Reliability issues of the provided study (ii)

85 OECD TG 471 sets out the following specifications:

a) the test is performed with 5 strains: four strains of *S. typhimurium* (TA98; TA100; TA1535; TA1537 or TA97a or TA97) and one strain which is either *S. typhimurium* TA102 or *E. coli* WP2 uvrA or *E. coli* WP2 uvrA (pKM101);
b) the maximum dose tested induces a reduction in the number of revertant colonies per plate compared to the negative control, or the precipitation of the tested substance. If no precipitate or limiting cytotoxicity is observed, the highest test dose corresponds to 5 mg/plate or 5 µl/plate;
c) at least 5 doses are evaluated, in each test condition;
d) triplicate plating is used at each dose level;
e) a concurrent negative control is included in each assay and the number of revertant colonies per plate for the concurrent negative control is inside the historical control range of the laboratory;
f) the mean number of revertant colonies per plate is reported for the treated doses and the controls;
g) negative results are confirmed in a repeat experiment with modification of study parameters to extend the range of conditions assessed, or a justification why confirmation of negative results is not considered necessary is provided.

86 In study (ii):

a) the test was performed with the strains TA100, TA98, TA1535, TA1537 and TA1538 (i.e., the strain S. typhimurium TA102 or E. coli WP2 uvrA or E. coli WP2 uvrA (pKM101) is missing);
b) the maximum dose tested did not induce a reduction in the number of revertant colonies per plate compared to the negative control, or the precipitation of the tested substance and it was less than 5 mg/plate or 5 µl/plate;
c) 1 dose was evaluated in absence and in presence of metabolic activation (i.e., less than 5 doses);
d) triplicate plating was not used at each dose level;
e) a concurrent negative control was not included in the study;
f) the mean number of revertant colonies per plate for the treated doses and the controls was not reported;
g) no repeat experiment was performed to confirm the negative results and no justification was provided.

87 Based on the above, the study (ii) submitted in your analogue read across approach, as currently reported in your dossier, cannot be considered a reliable source of information that could contribute to the conclusion on detection and quantification of gene mutations in cultured bacteria investigated by the required study.

2.2.2. Conclusion

88 While you have provided information on detection and quantification of gene mutations in cultured bacteria, the sources of information (i) and (ii) have deficiencies affecting their reliability thereby preventing drawing the conclusion on this key parameter.

89 Therefore, it is not possible to conclude, based on any source of information alone or considered together, on the information requirement for in vitro gene mutation study in bacteria.

90 On this basis, your weight of evidence adaptation under Annex XI, Section 1.2. is rejected.

91 Therefore, the information requirement is not fulfilled.

92 In your comment to the draft decision, you agree to perform the requested study.

3. Long-term toxicity testing on aquatic invertebrates

93 Short-term toxicity testing on aquatic invertebrates is an information requirement under Annex VII, Column 1, Section 9.1.1. However, under Column 2, long-term toxicity testing on aquatic invertebrates may be required by the Agency if the substance is poorly water soluble, i.e. solubility below 1 mg/L.

3.1. Triggering of the information requirement

94 You have provided information which indicates that the Substance is poorly water soluble (predicted water solubility of 0.00000000458 mg/L at 25° C using WSKOW v1.42 solubility).

95 Therefore, the Substance is poorly water soluble and information on long-term toxicity on aquatic invertebrates must be provided.

3.2. Information requirement not fulfilled

96 You have provided a waiver for the short-term toxicity study on aquatic invertebrates based on "*substance insolubility*" but no information on long-term toxicity on aquatic invertebrates for the Substance.

97 Therefore, the information requirement is not fulfilled.

98 In your comments to the draft decision, you propose to reassess the solubility of the Substance in water by performing a water solubility study as per OECD TG 105. You explain that you will conduct long-term toxicity testing on aquatic invertebrates only if the results of the new water solubility study confirm that the solubility of the Substance is below 1 mg/L.

99 As this strategy relies on data which is yet to be generated for the substance, no conclusion on the compliance of the proposed approach can be made. You remain responsible for complying with this decision by the set deadline.

3.3. Study design

100 The Substance is difficult to test due to the low water solubility (0.00000000458 mg/L) and adsorptive properties (estimated Log K_{ow} 12.23). OECD TG 211 specifies that, for difficult to test substances, you must consider the approach described in OECD GD 23 or other approaches, if more appropriate for your substance. In all cases, the approach selected must be justified and documented. Due to the properties of Substance, it may be difficult to achieve and maintain the desired exposure concentrations. Therefore, you must monitor the test concentration(s) of the Substance throughout the exposure duration and report the results. If it is not possible to demonstrate the stability of exposure concentrations (i.e. measured concentration(s) not within 80-120% of the nominal concentration(s)), you must express the effect concentration based on measured values as described in OECD TG 211. In case a dose-response relationship cannot be established (no observed effects), you must demonstrate that the approach used to prepare test solutions was adequate to maximise the concentration of the Substance in the test solution.

4. Growth inhibition study aquatic plants

101 Growth inhibition study on aquatic plants is an information requirement under Annex VII to REACH (Section 9.1.2.).

4.1. Information provided

102 You have adapted this information requirement by using Column 2 of Annex VII, Section 9.1.2. To support the adaptation, you have provided the following justification: "*aquatic toxicity is unlikely to occur as the substance is highly insoluble (solubility: 0.000000004581mg/L at 25° C) in water*".

4.2. Assessment of the information provided

4.2.1. The provided adaptation does not meet the criteria of Annex VII, Section 9.1.2., Column 2

103 Under Annex VII, Section 9.1.2., Column 2, the study may be omitted if aquatic toxicity is unlikely, for instance if the Substance is highly insoluble in water. Guidance on IRs and CSA, Section R.7.8.5 explains that there is no scientific basis to define a cut off limit for solubility below which toxicity is unlikely. Therefore, the justification must demonstrate very low water solubility and low likelihood to cross biological membranes. For the latter, the indicators used for low likelihood of a high bioaccumulation potential (Guidance on IRs and CSA, Figure R.11-4) must be considered, including:

- physico-chemical indicators of hindered uptake due to large molecular size (*e.g.* D_{max} > 17.4 Å and MW > 1100 or MML > 4.3 nm) or high octanol-water partition

coefficient (log K_{ow} > 10) or low potential for mass storage (octanol solubility (mg/L) < 0.002 x MW), and
- supporting experimental evidence of hindered uptake (no chronic toxicity for mammals and birds, no chronic ecotoxicity, no uptake in mammalian toxicokinetic studies, very low uptake after chronic exposure).

104 Unless it can reliably be demonstrated that aquatic toxicity is unlikely to occur, the Substance must be considered as poorly water soluble.

105 Your registration dossier provides:

- information on the solubility of the Substance in water (0.00000000458 mg/L based on QSAR);

- Even though the water solubility of the Substance is low, you have provided no information such as toxicokinetic or experimental proof of absence (or very low) uptake in relevant toxicological or ecotoxicological studies to show hindrance of uptake.

106 As an outcome, you have not demonstrated that toxicity is unlikely to occur, and your adaptation is rejected and the Substance must be considered as poorly water soluble.

107 Therefore, the information requirement is not fulfilled.

108 In your comment to the draft decision, you agree to perform the requested study.

4.3. Study design

109 OECD TG 201 specifies that, for difficult to test substances, OECD GD 23 must be followed. As already explained above, the Substance is difficult to test. Therefore, you must fulfil the requirements described in "Study design" under request 3.3.

References

The following documents may have been cited in the decision.

Guidance on information requirements and chemical safety assessment (Guidance on IRs & CSA)

Chapter R.4 Evaluation of available information; ECHA (2011).
Chapter R.6 QSARs, read-across and grouping; ECHA (2008).
Appendix to Chapter R.6 for nanoforms; ECHA (2019).
Chapter R.7a Endpoint specific guidance, Sections R.7.1 – R.7.7; ECHA (2017).
Appendix to Chapter R.7a for nanomaterials; ECHA (2017).
Chapter R.7b Endpoint specific guidance, Sections R.7.8 – R.7.9; ECHA (2017).
Appendix to Chapter R.7b for nanomaterials; ECHA (2017).
Chapter R.7c Endpoint specific guidance, Sections R.7.10 – R.7.13; ECHA (2017).
Appendix to Chapter R.7a for nanomaterials; ECHA (2017).
Appendix R.7.13-2 Environmental risk assessment for metals and metal compounds; ECHA (2008).
Chapter R.11 PBT/vPvB assessment; ECHA (2017).
Chapter R.16 Environmental exposure assessment; ECHA (2016).

Guidance on data-sharing; ECHA (2017).
Guidance for monomers and polymers; ECHA (2023).
***Guidance on intermediates*;** ECHA (2010).
All guidance documents are available online: https://echa.europa.eu/guidance-documents/guidance-on-reach

Read-across assessment framework (RAAF)

RAAF, 2017 Read-across assessment framework (RAAF); ECHA (2017).
RAAF UVCB, 2017 Read-across assessment framework (RAAF) – considerations on multi- constituent substances and UVCBs; ECHA (2017).

The RAAF and related documents are available online:
https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across

***OECD Guidance documents* (OECD GDs)**

OECD GD 23 Guidance document on aquatic toxicity testing of difficult substances and mixtures; No. 23 in the OECD series on testing and assessment, OECD (2019).
OECD GD 29 Guidance document on transformation/dissolution of metals and metal compounds in aqueous media; No. 29 in the OECD series on testing and assessment, OECD (2002).
OECD GD 150 Revised guidance document 150 on standardised test guidelines for evaluating chemicals for endocrine disruption; No. 150 in the OECD series on testing and assessment, OECD (2018).
OECD GD 151 Guidance document supporting OECD test guideline 443 on the extended one-generation reproductive toxicity test; No. 151 in the OECD series on testing and assessment, OECD (2013).

Appendix 2: Procedure

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

ECHA followed the procedure detailed in Articles 50 and 51 of REACH.

The compliance check was initiated on 31 October 2022.

The deadline of the decision is set based on standard practice for carrying out OECD TG tests. It has been exceptionally extended by 12 months from the standard deadline granted by ECHA to take into account currently longer lead times in contract research organisations.

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 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 REACH.

Appendix 3: Addressee(s) of this decision and their corresponding information requirements

In accordance with Articles 10(a) and 12(1) of REACH, the information requirements for individual registrations are defined as follows:

- the information specified in Annex VII to REACH, for registration at 1-10 tonnes per year (tpa), or as a transported isolated intermediate in quantity above 1000 tpa;
- the information specified in Annexes VII and VIII to REACH, for registration at 10-100 tpa;
- the information specified in Annexes VII, VIII and IX to REACH, for registration at 100-1000 tpa;
- the information specified in Annexes VII to X to REACH, for registration at more than 1000 tpa.

Registrant Name	Registration number	Highest REACH Annex applicable to you
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]

Where applicable, the name of a third-party representative (TPR) may be displayed in the list of recipients whereas ECHA will send the decision to the actual registrant.

Appendix 4: Conducting and reporting new tests for REACH purposes

1. Requirements when conducting and reporting new tests for REACH purposes

1.1 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 (https://echa.europa.eu/practical-guides).

(4) Under the introductory part of Annexes VII/VIII/IX/X to REACH, where a test method offers flexibility in the study design, for example in relation to the choice of dose levels or concentrations, the chosen study design must ensure that the data generated are adequate for hazard identification and risk assessment.

1.2 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 all constituents of each Test Material and their concentration values and other parameters relevant for the property to be tested, in this case purity and particle size distribution.

With that detailed information, ECHA can confirm 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 (https://echa.europa.eu/manuals).