~~Confidential~~

Helsinki, 02 November 2023

Addressees
Registrant(s) of 113_JS_CHDG as listed in Appendix 3 of this decision

Date of submission of the dossier subject to this decision
04/11/2022

Registered substance subject to this decision ("the Substance")
Substance name: D-gluconic acid, compound with N,N''-bis(4-chlorophenyl)-3,12-diimino-2,4,11,13-tetraazatetradecanediamidine (2:1)
EC/List number: 242-354-0

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 the deadline of ***12 May 2025***.

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

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

1. Bioaccumulation in aquatic species (triggered by Annex VIII, Section 9.3., Column 2.; test method: EU C.13/OECD TG 305), aqueous or dietary exposure.

The reasons for the decision(s) 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 addressees of the decision and their corresponding information requirements based on registered tonnage band are listed in Appendix 3.

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

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. In addition, the studies relating to biodegradation and bioaccumulation are necessary for the PBT assessment. However, to determine the testing needed to reach the conclusion on the persistency and bioaccumulation of the Substance you should consider the sequence in which these tests are performed and other conditions

described in this Appendix.

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 decision
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 decision

Contents

0. Reasons common to several requests

0.1. Triggers for further testing to clarify PBT properties of the Substance

1 The information requirements described in this section apply to the following endpoint(s): Section 1. Bioaccumulation in aquatic species.

2 Further testing to clarify degradation and bioaccumulation properties is triggered by the chemical safety assessment (CSA) if the substance is a potential PBT/vPvB substance (Annex VIII, Sections 9.2. and 9.3, Column 2; Annex XIII, Section 2.1). This is the case if the Substance itself or any of its constituent or impurity present in concentration ≥ 0.1% (w/w) or relevant transformation/degradation product meets the following criteria:

- it is potentially persistent or very persistent (P/vP) as:
  - it is not readily biodegradable (*i.e.* < 60 degradation in an OECD 301B study), and
- it is potentially bioaccumulative or very bioaccumulative (B/vB) as:
  - for some groups of substances (e.g. organometals, ionisable substances, surfactants) other partitioning mechanisms may drive bioaccumulation (e.g. binding to protein/cell membranes) and high potential for bioaccumulation cannot be excluded solely based on its potential to partition to lipid;

3 Your registration dossier provides the following:

- the Substance fulfils the 'very persistent' criterion (vP) of Annex XIII, Section 1.2.1, point (b), as its degradation half-life in fresh water sediment is higher than 180 days. The degradation half-lifes measured in OECD 308 study provided in your dossier were: DT50 >1000 days and DT50 > 277 days for Calwich Abbey and Swiss Lake systems, respectively);
- You conclude that the Substance will dissociate completely in aqueous solution at relevant pH values forming double protonated chlorhexidine and single deprotonated gluconic acid. On that basis the Substance is an ionisable substance and therefore high potential for bioaccumulation cannot be excluded based on available information;

4 The information above indicates that the Substance is a potential PBT/vPvB substance.

0.1.1. B/vB assessment

5 You further conclude under section 2.3 of your IUCLID dossier that your Substance is not B/vB with the following justification: *"(…) Based on radioactivity measurements, the bioaccumulation factor was determined to be 40 - 42 L/kg. Uptake and depuration rates were not determined. For a log Pow of 1.58, (partition coefficient of Chlorhexidine base CAS 55-56-1) the BCF(earthworm) is calculated to be 9.5 L/kg. Due to this result, Chlorhexidine digluconate is not expected to accumulate in terrestrial biota."* In support of your conclusion you provide the following additional information:

i. assessment of the Substance potential to cross biological membranes;

ii. the bioaccumulation studies on the Substance:
   1) key study: Freitag D. et al, (1982)
   2) key study: Freitag D. et al, (1985)
   3) Freitag D. et al, (1985)

4) Geyer et all (1981);
5) QSAR calculation of $BCF_{(earthworm)}$;

iii. QSAR prediction of BCF for a read-across substance: chlorhexidine (EC 200-238-7, CAS 55-56-1);

6 We have assessed this information and identified the following issues:

0.1.1.1. Low potential to cross biological membranes

7 Guidance on IRs and CSA, Section R.7.8.5. explains that there is no scientific basis to define molecular characteristics that would render a substance unlikely to cross biological membranes. In this context, the indicators used for low likelihood of a high bioaccumulation potential (Guidance on IRs and CSA, Section R.11, 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).

8 Your registration dossier provides:

- physico-chemical indicators which you consider supportive of hindered uptake (cross-sectional diameters of chlorhexidine base: $D_{effective}$ and $D_{maximum}$ of 1.1 nm and 2.1 nm respectively),
- a conclusion of low likelihood to cross biological membranes based on hindered uptake of the Substance (substantiated with the above physico-chemical indicators and the molecular weight of chlorhexidine (505.5 g/mol),
- a repeated-dose toxicity: chronic oral study in beagle dogs, TG452, (1978-1982) that support systemic exposure.

9 Available information on the Substance do not support that the Substance is unlikely to cross biological membranes because effects supporting systemic exposure were observed e.g. in dogs: NO(A)EL = 0.5 mg chlorhexidine base/kg bw/d (equivalent to 0.89 chlorhexidine digluconate/kg bw/d).

0.1.1.2. The provided studies and $BCF_{(earthworms)}$ QSAR do not meet the information requirement

First issue

10 As explained in the Guidance on IRs and CSA, section R.11.4.1.2.11., if there is a reliable aqueous bioaccumulation study available, such as an aqueous exposure OECD TG 305 study, the result from this test can be directly related to the criteria for B and vB.

11 For the study performed according to the OECD TG 305 the following results must be provided:

- the uptake rate constant (k_1) and loss rate constants including the depuration rate constant (k_2) and/or
- the steady-state bioconcentration factor (BCF_{SS}), and/or
- the kinetic bioconcentration factor (BCF_K), and/or
- the biomagnification factor (BMF).

12 You provided two key bioaccumulation studies ii.1) and ii.2) providing the BCF values of 40 and 42 L/kg and two supporting studies ii.3) and ii.4) with Chlorella fusca showing the following:

13 Neither uptake nor depuration rate constants were determined. Furthermore, there is no available information that would prove that steady-state was reached during the three days of exposure. Therefore, the estimated BCF cannot be compared neither to BCF_{SS} or BCF_k and therefore, cannot be directly to compared with B and vB criteria from Annex XIII of REACH. No justification was provided in this respect why this would not be the case. In addition, none of the supporting studies 3) and 4) provide any information on the above key parameters.

Second issue

14 Guidance on IRs and CSA, Sections R.11.4.1.2.2 and R.11.4.1.2.5 explain that in normal cases where experimental information on bioaccumulation is needed, a bioaccumulation test with fish (OECD 305) is preferred due to the better possibilities of comparing the results from such test with the B/vB criteria. However, only in specific cases, where fish bioaccumulation test is not expected to reflect sufficiently the bioaccumulation potential, testing of bioaccumulation potential in soil or sediment might then provide the necessary information for deriving conclusions on the B/vB-assessment.

15 No alternative using algae is provided as being appropriate.

16 You provided a QSAR calculation of $BCF_{earthworm}$ = 9.5 L/kg [study ii.5)]. Based on this result you concluded that the Substance is not expected to accumulate in terrestrial biota. However, you have not justified how the predicted bioaccumulation in earthworms is expected to reflect sufficiently the aquatic bioaccumulation potential in fish and why it is relevant for deriving conclusion on B/vB criteria. There is no indication that this would be the case here.

17 You also provided two supporting studies ii.3) and ii.4) using algae, *Chlorella fusca* without explanation why this would be appropriate for bioaccumulation.

18 Based on the above, the available studies do not provide information which could be compared to B and vB criteria and do not allow to conclude if the substance meets or not these criteria.

0.1.1.3. Assessment of QSAR prediction

19 Under Appendix C of the OECD Guidance document on the validation of (Q)SAR models (ENV/JM/MONO(2007)2) and ECHA Guidance R.6.1.6.3., adequate and reliable documentation must include a (Q)SAR Model Reporting Format document (QMRF). In addition, the information specified in or equivalent to the (Q)SAR Prediction Reporting Format document (QPRF) must be provided to have adequate and reliable documentation of the applied method.

20 You have provided the study iii. QSAR prediction of BCF for chlorhexidine (EC 200-238-7, CAS 55-56-1) – an estimation by ACD/LogD Suite Program, Version 8, estimating a BCF = 1.77.

21 You have not provided any above information about the model. In addition, the supporting study ii.5) QSAR calculation of $BCF_{(earthworm)}$ does not contain any information about the model either.

22 In absence of such information, ECHA cannot establish that the model and predictions are adequate for the purpose of classification and labelling and/or risk assessment.

0.1.1.4. Conclusion

23 The currently available information from your PBT assessment is not adequate to conclude that the Substance is not a potential PBT/vPvB substance and the chemical safety assessment (CSA) indicates the need for further investigation of the PBT/vPvB properties.

24 The selection of the requested test(s) and the test design are addressed in Appendix 1, section 1.

Reasons related to the information under Annex VIII of REACH

1. Bioaccumulation in aquatic species

25 Under Annex VIII, Section 9.3., Column 2, further information on bioaccumulation or further testing as described in Annex IX must be generated if the chemical safety assessment (CSA) in accordance with Annex I indicates the need to investigate further the bioaccumulation properties of the substance.

1.1. Triggering of the information requirement

a) Therefore, this information requirement is triggered in case if for example additional information on bioaccumulation as set out in Annex XIII, point 3.2.2, is required to assess PBT or vPvB properties of the substance in accordance with subsection 2.1 of that Annex.

26 As already explained in Appendix 1. on Reasons common to several requests, section 0.1, the Substance is a potential PBT/vPvB substance.

1.2. Information provided

27 The studies provided in your dossier (the assessment of the Substance potential to cross biological membranes, the bioaccumulation studies and QSAR predictions of bioaccumulation potential) are rejected for the reasons explained in section 0.1.1 above (Reasons common to several requests).

28 Therefore, the information requirement is not fulfilled.

29 Therefore, the chemical safety assessment (CSA) indicates the need for further investigation on bioaccumulation in aquatic species.

30 In your comments to the initial draft decision you agree with the request.

1.3. Study design and test specification

31 Bioaccumulation in fish: aqueous and dietary exposure (Method EU C.13 / OECD TG 305) is the preferred test to investigate bioaccumulation (Guidance on IRs and CSA, Section R.7.10.3.1.). Exposure via the aqueous route (OECD TG 305-I) must be conducted unless it can be demonstrated that:

- a stable and fully dissolved concentration of the test material in water cannot be maintained within ± 20% of the mean measured value, and/or
- the highest achievable concentration is less than an order of magnitude above the limit of quantification (LoQ) of a sensitive analytical method.

32 This test set-up is preferred as it allows for a direct comparison with the B and vB criteria of Annex XIII of REACH.

33 You may only conduct the study using the dietary exposure route (OECD 305-III) if you justify and document that testing through aquatic exposure is not technically possible as indicated above. You must then estimate the corresponding BCF value from the dietary test data according to Annex 8 of the OECD 305 TG and OECD Guidance Document on Aspects of OECD TG 305 on Fish Bioaccumulation (ENV/JM/MONO(2017)16).

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).

All Guidance on REACH is 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 16 November 2021.

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

You have provided comments during the decision-making phase which were found to address the information required in the draft decision. The information provided with your comments is also included in your updated registration dossier. Therefore the following original requests were removed from this decision:

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

2. Short-term toxicity testing on fish (Annex VIII, Section 9.1.3.; test method: EU C.1./OECD TG 203)

3. Simulation testing on ultimate degradation in surface water (triggered by Annex VIII, Section 9.2.; test method: EU C.25./OECD TG 309) at a temperature of 12°C. Non-extractable residues (NER) must be quantified and a scientific justification of the selected extraction procedures and solvents must be provided.

4. Soil simulation testing (triggered by Annex VIII, Section 9.2.; test method: EU C.23./OECD TG 307) at a temperature of 12°C. Non-extractable residues (NER) must be quantified and a scientific justification of the selected extraction procedures and solvents must be provided.

5. Sediment simulation testing (triggered by Annex VIII, Section 9.2.; test method: EU C.24./OECD TG 308) at a temperature of 12°C. Non-extractable residues (NER) must be quantified and a scientific justification of the selected extraction procedures and solvents must be provided.

6. Identification of degradation products (triggered by Annex VIII, Section 9.2; test method: using an appropriate test method.

The deadline in the initial draft decision took into account the above requests. As these requests have been removed from the decision, ECHA amended the deadline to 15 months. This deadline is set based on standard practice for carrying out OECD TG tests. It has been exceptionally extended by 6 months from the standard deadline granted by ECHA to take into account currently longer lead times in contract research organisations

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: Addressees 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]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[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[2].

(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.

[2] https://echa.europa.eu/practical-guides

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[3].

2. General recommendations for conducting and reporting new tests

2.1. Strategy for the PBT/vPvB assessment

Under Annex XIII, the information must be based on data obtained under conditions relevant for the PBT/vPvB assessment. You must assess the PBT properties of each relevant constituent of the Substance present in concentrations at or above 0.1% (w/w) and of all relevant transformation/degradation products. Alternatively, you would have to justify why you consider these not relevant for the PBT/vPvB assessment.

You are advised to consult Guidance on IRs & CSA, Sections R.7.9, R.7.10 and R.11 on PBT assessment to determine the sequence of the tests needed to reach the conclusion on PBT/vPvB. The guidance provides advice on 1) integrated testing strategies (ITS) for the P, B and T assessments and 2) the interpretation of results in concluding whether the Substance fulfils the PBT/vPvB criteria of Annex XIII.

In particular, you are advised to first conclude whether the Substance fulfils the Annex XIII criteria for P and vP, and then continue with the assessment for bioaccumulation. When determining the sequence of simulation degradation testing you are advised to consider the intrinsic properties of the Substance, its identified uses and release patterns as these could significantly influence the environmental fate of the Substance. You must revise your PBT assessment when the new information is available.

[3] https://echa.europa.eu/manuals