EUROPEAN CHEMICALS AGENCY

Helsinki, 23 March 2017

Addressee: [redacted]

Decision number: TPE-D-2114354526-46-01/F
Substance name: Methylene-bis-4,1-(N-phenylene-N'-butylurea)
EC number: 416-600-4
CAS number: 77703-56-1
Registration number: [redacted]
Submission number: [redacted]
Submission date: 09.03.2016
Registered tonnage band: 1000+T

DECISION ON A TESTING PROPOSAL

Based on Article 40 of Regulation (EC) No 1907/2006 (the 'REACH Regulation'), ECHA examined your testing proposal(s) and decided as follows.

Your testing proposal is accepted and you are requested to carry out:

- **Bioaccumulation in aquatic species (Annex IX, Section 9.3.2.; test method: Bioaccumulation in fish: aqueous and dietary exposure, OECD TG 305, aqueous exposure/dietary exposure) using the registered substance.**

You may adapt the testing requested above according to the specific rules outlined in Annexes VI to X and/or according to the general rules contained in Annex XI of the REACH Regulation. In order to ensure compliance with the respective information requirement, any such adaptation will need to have a scientific justification, referring and conforming to the appropriate rules in the respective Annex, and an adequate and reliable documentation.

You are required to submit the requested information in an updated registration dossier by **1 October 2018**. 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. 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, shall 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 Claudio Carlon , Head of Unit, Evaluation E2

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

Appendix 1: Reasons

The decision of ECHA is based on the examination of the testing proposal(s) submitted by you and scientific information submitted by third parties.

- **Bioaccumulation in aquatic species (Annex IX, Section 9.3.2.)**

a) Examination of the testing proposal

Pursuant to Article 40(3)(a) of the REACH Regulation, ECHA may require the Registrant to carry out the proposed test.

"Bioaccumulation in aquatic species, preferably fish" is a standard information requirement as laid down in Annex IX, Section 9.3.2. of the REACH Regulation. Adequate information on this endpoint is not available for the registered substance but needs to be present in the technical dossier to meet the information requirements. Consequently there is an information gap and it is necessary to provide information for this endpoint.

You have submitted a testing proposal for testing the registered substance for a bioaccumulation in aquatic species (Bioaccumulation in Fish: Aqueous and Dietary Exposure, OECD TG 305) with the following justification: "*Based on the estimated log Pow of 5.5 a bioaccumulation potantial of the substance can not be excluded. In conclusion, a bioaccumulation study according to OECD 305 is proposed according to REACH annex IX. In addition, this study study will be used to assess the B/vB criterion in the PBT assessment (please refer to IUCLID section 2.3).*
Test design:
Due to the characteristics of the substance (low water solubility, high log Pow), we propose to conduct a fish dietary feeding study with radioactive labeled test material."

ECHA considers that the proposed study is appropriate to fulfil the information requirement of Annex IX, Section 9.3.2. of the REACH Regulation.

ECHA has considered the available information submitted in the technical dossier and agrees with you that the available information does not meet the conditions set out in Annex XI, Section 1.2. of the REACH Regulation, i.e. the results obtained from the bioaccumulation screening do not allow an assumption/conclusion that the registered substance subject to the present decision has or has not bioaccumulation potential in aquatic species. Hence the available data are not adequate for the purpose of classification and labelling and/or risk assessments.

With regards to the PBT screening assessment, you indicate that the substance is not readily biodegradable (biodegradation of 11% within 28d in carbon dioxide evolution test and <10% within 28d in the CO_2 headspace test. In the PBT assessment you conclude that as the substance is not readily biodegradable, it should be considered to be P. No conclusion on vP can be made based on the available data. You also conclude that based on the toxicological properties the substance does not meet the criteria to be classified as T according to Annex XIII of REACH. ECHA notes that the available biodegradation screening information indicates that the registered substance may have persistent or very persistent (P or vP) properties.

ECHA requested your considerations for alternative methods to fulfil the information requirement for bioaccumulation in aquatic species. ECHA notes that you provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement for which testing is proposed. ECHA has taken these considerations into account.

b) Consideration of the information received during third party consultation

ECHA has received third party information concerning the testing proposal during the third party consultation.

The third party has indicated: *"In the "Considerations of Alternative Methods on Testing Proposals in Your Registration" on the substance methylene-bis-4,1-(N-phenylene-N'-butylurea), Column 2 of REACH Annex IX, section 9.3.2 is cited, stating that a bioaccumulation study does not need to be conducted if "direct and indirect exposure of the aquatic compartment is unlikely". The Registrant argues that "direct or indirect exposure of the aquatic compartment is regarded as low as the substance is used in reactive one or two component adhesives or coatings and is incorporated in a polymer matix (sic), very shorly (sic) after the use as soon as the polymeric matrix is hardened. Even though the bioaccumulation test might not be required due to the arguments based on column 2 of Annex IX given above, the registrant proposes to conduct the study to investigate possible PBT/vBvP properties of the registered substance." In contrast, since the bioaccumulation study can be waived according to Column 2 of REACH Annex IX, section 9.3.2, the bioaccumulation study in an aquatic species would constitute unrequired testing on animals and should not be performed.*

Regarding in vitro methods, the Registrant referred to the Guidance on Information Requirements and Chemical Safety Assessment, Chapter R.7c: Endpoint specific guidance and concluded, citing the Guidance, that "reliable in vitro methods are currently not available for this endpoint". However, an ECHA Partner Expert Group for is currently updating the PBT/vPvB-related Guidance including Chapter R.7c. Most importantly, the part cited by the Registrant, "these methods may become an important part of future test strategies, but their applicability is currently limited due to the lack of standardized protocols and limited validation based on small data sets. Further evaluation work is necessary before they can be recommended for use within an ITS." will be deleted from the guidance and a list of in vitro studies will be included. Possibly, some of the methods to be included in the Guidance would be appropriate to be applied in this case. As animal testing is only applicable as a last resort in the REACH Regulation, applicability of in vitro methods should be checked and, if appropriate, these methods should be applied instead of performing a bioaccumulation study in an aquatic species.

Taken together, we ask ECHA to deny the bioaccumulation testing for methylene-bis-4,1-(N-phenylene-N'-butylurea) because the study is not required according to REACH Annex IX, section 9.3.2 and it is possible that the data can also be produced using alternative methods."

ECHA notes that it is your responsibility as the Registrant to consider and justify in the registration dossier any adaptation of the information requirements in accordance with Annex IX, Section 9.3.2, column 2. This adaptation specifies that in case the substance has

- a low potential for bioaccumulation (for instance a log Kow ≤ 3) and/or a low potential to cross biological membranes, or
- direct and indirect exposure of the aquatic compartment is unlikely,

the bioaccumulation study in aquatic species does not need to be conducted.

However, ECHA notes that the substance has log Kow > 3 that indicates the potential for bioaccumulation. Moreover, the substance has wide dispersive consumer uses and relevant exposure, which have been reported in the Chemical Safety Report (environmental RCR's range from [redacted] in manufacture in aquatic compartments to [redacted] to terrestrial compartment in formulation). Therefore, ECHA does not consider the adaptation of the standard information requirement according to Annex IX, Section 9.3.2, column 2, possible.

Additionally, ECHA notes that there is no evidence in the 3rd party argumentation nor in the dossier about what is meant by 'very shortly' in the claim submitted by the 3rd party: *'incorporated in a polymer matrix, very shortly after the use as soon as the polymeric matrix is hardened'*.

Moreover, you have run QSAR estimation to estimate bioaccumulation potential of the substance (log Kow =5.5) concluding that the calculated BCF is 3428, indicating that the substance is potentially bioaccumulative. The BCF was calculated on an estimated (also calculated) log Kow as a measurement of the Log Kow was technically not feasible. Taken together, you as the registrant came to the conclusion, that a prediction of the bioaccumulation potential based on the calculated BCF is not reliable and an in vivo test as proposed should be conducted.

Lastly, ECHA points out that there are no validated alternative methods available. Therefore, ECHA cannot reject the testing proposal based on 3rd party information.

c) Consideration of your comments

In your comments according to Article 50(1) you requested ECHA "[...] *to change the draft decision for the testing proposal and to include in a first step the proposed in vitro "bi directional permeability assay" using Caco-2 cells. The in vivo test according to OECD 305 should only be done if significant and relevant bioavailability can be proven in the in vitro test. In case no or negligible bioavailability is shown the OECD 305 test should not be conducted and the substance should be classified as not B/vB also result in classification of the substance as not B/vB in course of the PBT assessment*". You further claimed that the requested bioaccumulation test could be waived "*as no exposure to the environment is to be expected*".

ECHA notes that in your chemical safety report (CSR) you have indicated that the registered substance could have wide consumer uses and have reported risk characterisation ratio (RCR) up to [redacted] Thus, ECHA considers that direct or indirect exposure to the environment is likely.

Concerning the testing strategy you have proposed, ECHA notes that according to Annex XI 1.1.2. of the REACH Regulation, data on environmental properties carried out with other tests methods than those referred to in Article 13 (3) can be considered equivalent when the key parameters from the referred test methods are covered and the exposure duration is comparable or longer to the referred test methods. ECHA points out that the proposed in vitro bi-directional permeability assay would provide indication on the potential uptake of the substance, while the requested assay would provide information on bioaccumulation. Bioaccumulation depends not only on the uptake of the substance but also on its transformation and elimination. Furthermore, ECHA notes that in this *in vitro* assay, physiological factors that could facilitate the uptake of the substance in the gut (e.g. mucous and bile salts) are not present. ECHA further notes that the Caco-2 assay is based on a human intestinal cell line, the transferability of this assay to fish remains to be verified, Taxonomic differences in membrane transporters, enzyme complement, and fatty acid composition could influence the absorption process. Finally, ECHA notes that the duration of this assay is very short (2 hours) compared to the OECD 305 TG (uptake phase: 7-14 days and depuration phase: usually 28 days). Thus, the proposed *in vitro* assay would neither fulfil the information requirement of Annex IX, Section 9.3.2. of the REACH Regulation nor constitute a valid adaptation according to column 2 of that Annex or according to Annex XI of the REACH Regulation.

Therefore ECHA considers that a bioaccumulation test is still needed.

ECHA notes that according to ECHA *Guidance on information requirements and chemical safety assessment, Chapter R.7c* (version 2.0, November 2014) bioaccumulation in fish: aqueous and dietary exposure (test method EU C.13. / OECD TG 305) is the preferred test to cover the standard information requirement of Annex IX, Section 9.3.2.

Concerning the test design the OECD 305 guideline states that "*The aqueous exposure test is most appropriately applied to stable organic chemicals with log KOW values between 1.5 and 6.0 but may still be applied to strongly hydrophobic substances (having log KOW > 6.0), if a stable and fully dissolved concentration of the test substance in water can be demonstrated*" and that "*a log KOW above 5 and a water solubility below ~0.01 – 0.1 mg/L mark the range of substances where testing via aqueous exposure may become increasingly difficult.*" Furthermore, ECHA Guidance defines that results obtained from a test with aqueous exposure can be used directly for comparison with the B and vB criteria of Annex XIII of REACH Regulation and can be used for hazard classification and risk assessment. Comparing the results of a dietary study with the REACH Annex XIII B and vB criteria is more complex and has higher uncertainty.

Therefore, the aqueous route of exposure is the preferred route and shall be used whenever technically feasible. As the registered substance has a log Kow of 5.5 and water solubility of 0.05 mg/L, following the receipt of Proposals for Amendment (PfAs) from Member State Competent Authorities (MSCAs) the aqueous exposure route is given as the preferred test design in this decision. The dietary exposure route should only be taken into consideration, if it is demonstrated that maintaining a stable aqueous concentration within the water solubility of the test substance is technically not feasible.

If you decided to conduct the study using the dietary exposure route, you shall provide scientifically valid justification for your decision. You shall also attempt to estimate the corresponding BCF value from the dietary test data by using the approaches given in Annex 8 of the OECD 305 TG. In any case you shall report all data derived from the dietary test as listed in the OECD 305 TG.
d) Outcome

Therefore, pursuant to Article 40(3)(a) of the REACH Regulation, you are requested to carry out the proposed study using the registered substance subject to the present decision:

- Bioaccumulation in fish: aqueous or dietary bioaccumulation fish test (test method: OECD TG 305)

Notes for your consideration

Before conducting testing, you are advised to consult the ECHA Guidance on the information requirements and chemical safety assessment (version 2.0, November 2014), Chapter R.11. PBT/vPvB assessment, in particular to first conclude on whether the registered substance is not persistent (P) and not very persistent (vP) or whether it may fulfil Annex XIII of the REACH Regulation criteria of being P or vP and to consult the PBT assessment for Weight-of-Evidence determination and the integrated testing strategy for bioaccumulation assessment. Also, you need to carefully consider the potential formation of stable degradation products with PBT/vPvB properties.

In addition, you are advised to consult the ECHA Guidance on information the information requirements and chemical safety assessment, Chapters R.4, 5, 6, R.7b and R.7c. If you decide to adapt the testing requested according to the specific rules outlined in Annexes VI to X and/or according to general rules contained in Annex XI of the REACH Regulation, you are referred to the advice provided in practical Guides on "How to use alternatives to animal testing to fulfil your information requirements for REACH registration" and on "How to use and report (Q)SARs".

Due to the low solubility of the substance in water and high octanol-water partition coefficient, you should consult OECD Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures, ENV/JM/MONO (2000)6 and ECHA Guidance, Chapter R7b (table R.7.8-3 summarising aquatic toxicity testing of difficult substances) for choosing the design of the requested test and calculation and expression of the results of the test.

Deadline to submit the requested information in this decision

In the draft decision communicated to you, the time indicated to provide the requested information was 9 months from the date of adoption of the decision. In your comments on the draft decision, you requested an extension of the timeline to 18 to perform the bioaccumulation test requested by ECHA or 21 months if both the in vitro "bi directional permeability assay" using Caco-2 cells you have proposed and the bioaccumulation test requested by ECHA need to be performed. You sought to justify this request by stating that you need extra time for synthesising, purifying and characterising the radiolabelled substance needed in the test. Besides you address that the CROs are actually fully booked, that the test duration is variable due to the unknown duration of the depuration phase and mentioned a possible necessity of performing a pre-test, plus the 1-2 months for updating the CSA accordingly.

ECHA acknowledges that the deadline needs to be adjusted to accommodate the difficulties for testing the substance. In such cases an extra 9 months can be granted. However, ECHA has not accepted your testing strategy for bioaccumulation and therefore will not grant additional time for performing the *in vitro* "bi directional permeability assay" using Caco-2 cells. Therefore, ECHA has set the deadline to 18 months.

Appendix 2: Procedural history

ECHA received your registration containing the testing proposals for examination pursuant to Article 40(1) on 9 March 2016.

ECHA held a third party consultation for the testing proposal(s) from 17 May 2016 until 2 July 2016. ECHA received information from third parties (see Appendix 1).

This decision does not take into account any updates after **28 September 2016**, 30 calendar days after the end of the commenting period.

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 amended the deadline.

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

ECHA received proposals for amendment and modified the draft decision.

ECHA invited you to comment on the proposed amendments.

ECHA referred the draft decision to the Member State Committee.

Your comments on the proposed amendment(s) were taken into account by the Member State Committee.

The Member State Committee reached a unanimous agreement on the draft decision in its MSC-52 written procedure and ECHA took the decision according to Article 51(6) of the REACH Regulation.

Appendix 3: Further information, observations and technical guidance

1. This decision does not imply that the information provided in your registration dossier is in compliance with the REACH requirements. The decision does not prevent ECHA from initiating a compliance check on the registration at a later stage.

2. Failure to comply with the request(s) in this decision, or to fulfil otherwise the information requirement(s) with a valid and documented adaptation, will result in a notification to the Enforcement Authorities of the Member States.

3. In relation to the information required by the present decision, the sample of the substance used for the new test(s) 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 test(s) 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 test(s) must be suitable to assess these grades. Finally there must be adequate information on substance identity for the sample tested and the grade(s) registered to enable the relevance of the test(s) to be assessed.