EUROPEAN CHEMICALS AGENCY

Decision number: TPE-D-0000003264-79-04/F Helsinki, 16 October 2013

DECISION ON A TESTING PROPOSAL SET OUT IN A REGISTRATION PURSUANT TO ARTICLE 40(3) OF REGULATION (EC) NO 1907/2006

For isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol, CAS No 25265-77-4 (EC No 246-771-9), registration number: [redacted]

Addressee: [redacted]

The European Chemicals Agency (ECHA) has taken the following decision in accordance with the procedure set out in Articles 50 and 51 of Regulation (EC) No 1907/2006 concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH Regulation).

I. Procedure

Pursuant to Article 40(1) of the REACH Regulation, ECHA has examined the following testing proposal submitted as part of the registration dossier in accordance with Articles 10(a)(ix) and 12 (1)(e) thereof for isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol, CAS No 25265-77-4 (EC No 246-771-9), by [redacted] (Registrant):

- Developmental toxicity / teratogenicity study (OECD 414) for the related substance 2,2,4-trimethyl-1,3-pentanediol diisobutyrate [redacted]

This decision is based on the registration dossier as submitted with submission number [redacted], for the tonnage band of 1000 tonnes or more per year. This decision does not take into account any updates after 18 January 2013 the date upon which ECHA notified its draft decision to the Competent Authorities of the Member States pursuant to Article 51(1) of the REACH Regulation.

This decision does not imply that the information provided by the Registrant in his registration dossier is in compliance with the REACH requirements. The decision does not prevent ECHA to initiate a compliance check on the present dossier at a later stage.

On 27 April 2010, pursuant to Article 40(1) of the REACH Regulation, ECHA initiated the examination of the testing proposal set out by the Registrant in the registration dossier for the substance mentioned above.

ECHA held a third party consultation for the testing proposal from 16 June 2011 until 1 August 2011. ECHA did receive information from third parties (see section III below).

On 12 June 2012 ECHA sent the draft decision to the Registrant and invited him to provide comments within 30 days of the receipt of the draft decision. That draft decision was based on the registration dossier as submitted with submission number [redacted]

On 7 July 2012 ECHA received comments from the Registrant. On 25 July 2012 the Registrant updated his registration dossier (submission number [redacted]).

ECHA considered the Registrant's comments and dossier update received.
On the basis of the comments and dossier update, Section II was amended. The Statement

of Reasons (Section III) was changed accordingly.

On 18 January 2013 ECHA notified the Competent Authorities of the Member States of its draft decision and invited them pursuant to Article 51(1) of the REACH Regulation to submit proposals to amend the draft decision within 30 days of the receipt of the notification.

Subsequently, one Competent Authority of a Member State submitted a proposal for amendment to the draft decision.

On 21 February 2013 ECHA notified the Registrant of the proposal for amendment to the draft decision and invited him pursuant to Article 51(5) of the REACH Regulation to provide comments on the proposal for amendment within 30 days of the receipt of the notification.

ECHA reviewed the proposal for amendment received and decided not to amend the draft decision.

On 4 March 2013 ECHA referred the draft decision to the Member State Committee.

On 13 March 2013 the Registrant provided comments on the proposed amendment. The Member State Committee took the comments of the Registrant into account.

A unanimous agreement of the Member State Committee on the draft decision was reached on 8 April 2013 in a written procedure launched on 27 March 2013. ECHA took the decision pursuant to Article 51(6) of the REACH Regulation.

II. Testing required

The Registrant shall carry out the following proposed test pursuant to Article 40(3)(c) of the REACH Regulation using the indicated test method and the registered substance subject to the present decision:

- Pre-natal developmental toxicity study in rats or rabbits, oral route (Annex IX, 8.7.2; test method: EU B.31/OECD 414)

while the originally proposed test for a Developmental toxicity / teratogenicity study /OECD 414 proposed to be carried out using the analogue substance 2,2,4-trimethyl-1,3-pentanediol diisobutyrate [redacted] is rejected pursuant to Article 40(3)(d) of the REACH Regulation.

Pursuant to Articles 40(4) and 22 of the REACH Regulation, the Registrant shall submit to ECHA by **16 June 2015** an update of the registration dossier containing the information required by this decision.

Data from a second pre-natal developmental toxicity study on another species is a standard information requirement according to Annex X, 8.7.2 of the REACH Regulation. The Registrant should firstly take into account the outcome of the pre-natal developmental toxicity on a first species and all other relevant available data to determine if the conditions are met for adaptations according to Annex X, 8.7 column 2, or according to Annex XI. If the Registrant considers that testing is necessary to fulfil this information requirement, he should include in the update of his dossier a testing proposal for a pre-natal developmental toxicity study on a second species.

At any time, the Registrant shall take into account that there may be an obligation to make every effort to agree on sharing of information and costs with other Registrants.

III. Statement of reasons

The decision of ECHA is based on the examination of the testing proposal submitted by the Registrant for the registered substance proposed to be performed with the analogue substance 2,2,4-trimethyl-1,3-pentanediol diisobutyrate [redacted], on the submitted read-across justification and scientific information submitted by third parties.

Pre-natal developmental toxicity study

a) Read-across approach

In relation to the testing proposal subject to the present decision, the Registrant has proposed to use a read-across approach in accordance with Annex XI, 1.5, and to perform the test on another substance than the registered substance. ECHA has considered first the scientific validity of the proposed read-across before assessing the testing proposed.

In the section 1.2 "Composition of the substance, Analog chemical", page 3 of the CSR, the Registrant gives the following justification for the read-across:
"TXIB™ is the isobutyric ester of [redacted]™ (isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol). Therefore its molecular structure and properties are closely related to isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol. The only structural difference is that TXIB™ is the diester of 2,2,4-trimethyl-1,3-pentanediol, whereas isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol is the monoester. Both isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol and TXIB™ have been shown to hydrolyze to 2,2,4-trimethyl-1,3-pentanediol [redacted]. The former was demonstrated by an *in vitro* hydrolysis study and the latter by an *in vivo* metabolic profile. These studies are discussed in Sect. 5.1.3 of this CSR."

However, ECHA notices that in the section 5.1.3 "Summary and discussion of toxicokinetics" page 16 of the CSR, the Registrant provides information which shows that both [redacted] and TXIB are not fully metabolized to [redacted] Specifically, the Registrant states that "*In vitro*, isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol [[redacted]] undergoes partial hydrolysis in whole rat or human blood to produce the expected diol, [redacted]." Furthermore, the Registrant provides that "Approximately 35.5% of the starting material did not undergo hydrolysis in either rat or human blood under conditions of this study" and in the IUCLID dossier section 7.1.1. Basic toxicokinetics, the study of Boatman 1989 concludes that "The results of these *in vitro* studies suggest that the 1-substituted and 3-substituted isomers of [redacted] may differ in their metabolic uptake, distribution, metabolism, and elimination." Also, with regard to the *in vivo* study, the Registrants shows in the section 5.1.3."Summary and discussion of toxicokinetics" page 16 of the CSR that "Disposal of TXIB™ appears to involve some non-absorption and partial hydrolysis in the gut based on the occurrence of both TXIB-3-[14] C and a mono-isobutyrate ester of [redacted] in the feces".

On the basis of the information provided by the Registrant, showing that [redacted] and also TXIB are not fully metabolized to [redacted], ECHA concludes that it cannot be excluded that these parent chemicals could be responsible for the observed systemic toxicity as well. The two studies cited above failed to demonstrate a complete metabolic convergence of the target and source chemicals and the fact that the systemic toxicity is only due to the effect of the common [redacted] metabolite. Since un-reacted [redacted] and TXIB could be responsible for the observed systemic toxicity, different substance specific effects cannot be excluded.

Finally, the *in vitro* study with [redacted] gives half-life of the 1-isomer in minutes (19.6 in rat and 17.3 minutes in human blood), while the description of the *in vivo* study with TXIB does not indicate kinetic parameters. [redacted], administered separately is recovered in the urine 88% (and 2% in the feces) within 48 hours of the dosing. Thus, the hydrolysis rates of the

esters cannot be compared.

ECHA concluded that based on the data submitted in the dossier [redacted] (the subject of the draft decision sent to Registrant on 12 June 2012), the requirements of the REACH Regulation for the acceptance of read-across as they are listed in Annex XI section 1.5, were not met. Due to the absence of a scientifically convincing justification of the read-across hypothesis the requirement for adequate and reliable documentation of the applied method were not fulfilled.

Consequently, as the proposed read-across approach to 2,2,4-trimethyl-1,3-pentanediol diisobutyrate (TXIB) had failed to satisfy the requirements of Annex XI section 1.5. of the REACH Regulation, the testing proposal on the read-across substance is rejected.

In his official comments on the draft decision, to further support the proposed read-across strategy, the Registrant explained that the read-across hypothesis from the related diisobutyrate (TXIB) was based upon the Registrant's assertion that following oral administration, TXIB will hydrolyze to produce the component monoisobutyrate esters of [redacted]. The newly proposed hypothesis based on TXIB hydrolysis to Texanol differs from the hypothesis inferred from the previous submission which was based on sharing the common metabolite [redacted]. However, the available in vivo study with TXIB does not indicate any kinetic parameters. The Registrant acknowledged in his comments *"that further in vivo data are needed to better understand the metabolism and toxicokinetics of TXIB, compared to [redacted] and to provide adequate documentation and conclusive evidence that TXIB is a suitable precursor for [redacted] in animal studies."*

Therefore, to address the robustness of the read-across hypothesis and to obtain sufficient evidence that 2,2,4 -trimethyl-1,3 -pentanediol diisobutyrate (TXIB) undergoes extensive ester hydrolysis *in vivo* to produce the corresponding 1-substituted and 3-substituted monoisobutyrates, the Registrant intends to conduct a study in *"four separate groups of rats (n=10/group) that are given a single dose of either TX [redacted] or TXIB by oral gavage or by intravenous injection. Gavage studies will demonstrate bioavailability and potential GI metabolism, while IV studies will be used to evaluate the absolute metabolism of TXIB to TX. Goals of this experiment are to a) measure and document the rate of hydrolysis of TXIB to the corresponding monoesters, b) quantify the 1- and 3-substituted monoesters produced, and c) determine if additional metabolites of TXIB are produced, that are not associated with either the 1- or 3-substituted monoester. Blood samples will be collected at 10-minute intervals(5 rats sampled per timepoint) for one hour post-dosing, and plasma will be isolated via centrifugation. Concentrations of TXIB, TX (1- and 3-substituted), [redacted], and Isobutyric acid will be quantitated in plasma samples using a GC/FID or GC/MS method, as appropriate, by comparison to standard reference materials. The study will be conducted in compliance with Good Laboratory Practice (GLP) guidelines. Beyond issues related to ester hydrolysis, an additional consideration that will be addressed in a metabolism and toxicokinetic study is the net concentrations of TX and Isobutyric acid that will be delivered following administration of TXIB. Since TXIB is composed of approximately [redacted] TX plus [redacted] Isobutyric acid (IBA), the "effective equivalent dose of TX" will need to be calculated by reducing the administered dose of TXIB by approximately [redacted]. In addition, the concentration of IBA will need to be measured following TXIB administration, to determine whether the additional IBA that is liberated via hydrolysis of TXIB will affect the toxicokinetics and/or toxicodynamics of TX."*

ECHA notes that in order for ECHA to assess the read-across it has to be demonstrated that the hydrolysis of TXIB to [redacted] is rapid enough to prevent significant systemic exposure to TXIB and that hydrolysis to [redacted] is the only metabolic pathway, as well as lack of influence of IBA on toxicokinetics. Such demonstration should include information on the extent and the fast rate of hydrolysis of TXIB to the corresponding monoesters, on the

presence/absence of other metabolites than the [redacted] components, on any influence of IBA on toxicokinetics and IBA possible effects on the toxicodynamics of [redacted]. It is at the Registrant's discretion to initiate any such investigations to acquire sufficient data to substantiate his read-across hypothesis.

However, as the Registrant's assumptions are not confirmed by scientific data, the dossier does not contain at the moment sufficient information to demonstrate that properties of the registered substance may be predicted from the properties of the proposed read-across substance. As adequate and reliable documentation of the proposed read-across strategy as required by Annex XI, 1.5 is not yet provided by the Registrant, the testing proposal for conducting test on a read-across substance cannot be approved by ECHA as an appropriate mean to fulfil the information requirements of the substance subject to the present decision.

Pursuant to Article 40(3)(d) of the REACH Regulation, ECHA rejects the proposal to carry out the test on a substance other than the registered substance. Nevertheless, it is necessary to consider whether the test proposed shall be performed in order to meet the information requirements.

b) Information requirement

Pursuant to Article 40(3)(c) of the REACH Regulation, ECHA may require the Registrant to carry out one or more additional tests in case of non-compliance of the testing proposal with Annexes IX, X or XI of the REACH Regulation.

A pre-natal developmental toxicity study for a first species is a standard information requirement as laid down in Annex IX, section 8.7.2 of the REACH Regulation. The 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.

The Registrant did not specify the species and route to be used for testing. According to the test method EU B.31/OECD 414, the rat is the preferred rodent species, the rabbit the preferred non-rodent species and the test substance is usually administered orally. ECHA considers these default parameters appropriate and testing should be performed by the oral route with the rat or the rabbit as a first species to be used.

c) Consideration of the information received during third party consultation

ECHA received third party information concerning the testing proposal during the third party consultation. For the reasons explained further below the information provided by third parties is not sufficient to fulfil this information requirement.

The third party has proposed a nonlinear classification ANN QSAR Model for prenatal developmental toxicity. The third party has indicated that their information is confidential and this information is not provided to the registrant.

The result from the QSAR classification model (i.e. "toxic" or "non-toxic") is not suitable for the purposes of classification and labelling and/or risk assessment for the endpoint for which testing has been proposed to meet the information requirement (Annexes IX or X, 8.7).

In addition, the submitted information does not exclude the possibility that the registered substance might be outside the applicability domain of the model. The (Q)SAR Model Reporting Format (QMRF) does not provide sufficient information to deduce whether the training set was constructed from studies that cover the information requirements of the OECD 414 guideline, or important study aspects, such as the species, dose selection and

number of animals used. Finally, the submitted QPRF does not contain any indication on the adequacy in relation to a defined regulatory purpose of the Testing Proposal.

Therefore, ECHA concludes that on this occasion, the information submitted does not meet the conditions for the adaptation on the basis of QSAR models set out in Annex XI, Section 1.3. Therefore, it cannot constitute an acceptable adaptation to standard information requirements.

d) Outcome

Therefore, pursuant to Article 40(3)(c) of the REACH Regulation, the Registrant is required to carry out the following study: Pre-natal developmental toxicity study in rats or rabbits, oral route (test method: EU B.31/OECD 414), using the registered substance isobutyric acid, monoester with 2,2,4-trimethylpentane-1,3-diol, CAS No 25265-77-4 (EC No 246-771-9).

ECHA also notes that if further investigations of the registrant, conducted on his own discretion, provide sufficient data to substantiate read-across strategy, an adaptation of the standard information requirement to be validly applied has to be done in accordance with the rules of Annex XI, 1.5 and adequate and reliable documentation has to be provided.

When considering the need for a testing proposal for a prenatal developmental toxicity study in a second species, the Registrant should take into account the outcome of the pre-natal developmental toxicity study on the first species and all available data to determine if the conditions are met for adaptations according to Annex X, 8.7. column 2, or according to Annex XI; for example if the substance meets the criteria for classification as toxic for reproduction Category 1B: May damage the unborn child (H360D), and the available data are adequate to support a robust risk assessment, or alternatively, if Weight of Evidence assessment of all relevant available data provides scientific justification that the study in a second species is not needed.

At any time, the Registrant shall take into account that there may be an obligation to make every effort to agree on sharing of information and costs with other Registrants.

e) Deadline to provide the requested information

On basis of the Registrant's comments an additional 8 months to the initial deadline of 12 months have been granted. Therefore, the deadline was extended from 12 to 20 months.

IV. Adequate identification of the composition of the tested material

The process of evaluation of testing proposals set out in Article 40 of the REACH Regulation aims at ensuring that the new study meets real information needs. Within this context, the Registrant's dossier was sufficient to confirm the identity of the substance to the extent necessary for evaluation of the testing proposal. The Registrant must note, however, that this information, or the information submitted by other registrants of the same substance, has not been checked for compliance with the substance identity requirements set out in Section 2 of Annex VI of the REACH Regulation.

In relation to the proposed test, the sample of substance used for the new study must be suitable for use by all the joint registrants. Hence, the sample should have a composition that is within the specifications of the substance composition that are given by the joint registrants. It is the responsibility of all joint registrants of the same substance to agree to the test proposed (as applicable to their tonnage level) and to document the necessary information on their substance composition.

In addition, it is important to ensure that the particular sample of substance tested in the new study 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 by each registrant. If the registration of the substance by any registrant covers different grades, the sample used for the new study must be suitable to assess these grades.

Finally there must be adequate information on substance identity for the sample tested and the grade registered to enable the relevance of the study to be assessed.

V. General requirements for the generation of information and Good Laboratory Practice

ECHA reminds registrants of the requirements of Article 13(4) of the REACH Regulation that ecotoxicological and toxicological tests and analyses shall be carried out in compliance with the principles of good laboratory practice (GLP).

According to Article 13(3) of the REACH Regulation, tests that are required to generate information on intrinsic properties of substances shall be conducted in accordance with the test methods laid down in a Commission Regulation or in accordance with other international test methods recognised by the Commission or the European Chemicals Agency as being appropriate. Thus, the Registrant shall refer to Commission Regulation (EC) No 440/2008 laying down test methods pursuant to Regulation (EC) No 1907/2006 as adapted to technical progress or to other international test methods recognised as being appropriate and use the applicable test methods to generate the information on the endpoints indicated above.

VI. Information on right to appeal

An appeal may be brought against this decision to the Board of Appeal of ECHA under Article 51(8) of the REACH Regulation. Such appeal shall be lodged within three months of receiving notification of this decision. Further information on the appeal procedure can be found on the ECHA's internet page at **http://echa.europa.eu/appeals/app_procedure_en.asp.** The notice of appeal will be deemed to be filed only when the appeal fee has been paid.

[redacted]
Jukka Malm
Director of Regulatory Affairs