Helsinki, 14 December 2016

Addressee: [redacted]

Decision number: CCH-D-2114350060-68-01/F
Substance name: TETRAHYDROTHIOPHENE 1,1-DIOXIDE
EC number: 204-783-1
CAS number: 126-33-0
Registration number: [redacted]
Submission number: [redacted]
Submission date: 06.04.2016
Registered tonnage band: 100-1000T

## DECISION ON A COMPLIANCE CHECK

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

1. **In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.; test method: OECD TG 476 or TG 490) with the registered substance;**

2. **Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2.; test method: EU B.26./OECD TG 408) in rats with the registered substance; modified to include urinalysis and a full histopathological examination which is to include immunohistochemical investigation of renal pathology to determine if the pathology is mediated by alpha-2u globulin nephropathy;**

3. **Identification of degradation products (Annex IX, 9.2.3.; test method: Aerobic mineralisation in surface water – simulation biodegradation test, EU C.25./OECD TG 309) with 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 **21 June 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 Leena Ylä-Mononen, Director of Evaluation

[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**

1. **In vitro gene mutation study in mammalian cells (Annex VIII, Section 8.4.3.)**

Pursuant to Articles 10(a) and 12(1) of the REACH Regulation, a technical dossier registered at 100 to 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to IX of the REACH Regulation. The information to be generated for the dossier must fulfil the criteria in Article 13(4) of the same regulation.

An "*in vitro* gene mutation study in mammalian cells" is an information requirement as laid down in Annex VIII, Section 8.4.3. of the REACH Regulation, "if a negative result in Annex VII, Section 8.4.1. and Annex VIII, Section 8.4.2." is obtained.

ECHA notes that the registration dossier contains negative results for the *in vitro* gene mutation study in bacteria and the *in vitro* chromosomal aberration study. Therefore, adequate information *on in vitro* gene mutation in mammalian cells needs to be present in the technical dossier for the registered substance to meet this information requirement.

In the technical dossier you have provided a study record for an "*in vitro mammalian mutation test*" ([redacted] 1983). However, this study does not provide the information required by Annex VIII, Section 8.4.3. because ECHA considers that this study is not appropriate. More specifically, you stated that the study was conducted "*according to a test protocol that is comparable to the appropriate OECD test guideline. It was not compliant with GLP*". The study report executive summary reads: "*Sulfolane was considered by the author of the study report to be mutagenic in both the absence and presence of metabolic activation. The findings are confounded, however, by excessive cytotoxicity at all dose levels. No dose-response was observed in either the toxicity or the increased mutant frequency. When assessed by the criteria in the draft new OECD Test Guideline: "In vitro mammalian cell gene mutation assays using the thymidine kinase gene", the mutant frequency does not exceed the solvent controls by the global evaluation factor (relevant for the plate method) of 90 x10$^{-6}$. Therefore it is considered the test substance is negative for mutagenicity under the conditions of the test*".

ECHA acknowledges that such results would not be considered as positive (i.e. a biologically significant increase in the mutation frequency) according to the current practice. ECHA agrees that no dose response was observed in either cytotoxicity or mutation parameters. The absence of dose-related cytotoxicity is unusual and triggers questions on the appropriateness of this study and on the rationale for setting the highest test concentrations. Moreover, ECHA notes that the level of cytotoxicity reached with the tested substance (i.e. 56 to 75% without metabolic activation and 34 to 68% with metabolic activation) is not in agreement with the previous test guideline (TG) 476 (from 1997), neither with the current TG 490 (from 2015).

Indeed, in both TGs, the highest concentration should aim to achieve between 20 and 10% (but not less than 10%) relative survival, relating to 80 to 90% cytotoxicity. Such low level of survival was not reached for any of the concentrations tested in the study of [redacted] (1983). Hence, ECHA concludes that this study in not appropriate to conclude on the mutagenic property of the registered substance *in vitro* in mammalian cells.

ECHA acknowledges your comment on the draft decision agreeing to conduct the requested *in vitro* gene mutation study in mammalian cells. In addition, you have indicated that you are aware of *in vivo* genotoxicity studies by NTP with negative results but details of the results are not available yet. ECHA would like to note that in order to adapt the requirement for an *in vitro* gene mutation study in mammalian cells with data from an *in vivo* study, the corresponding *in vivo* study would need to address also gene mutations.

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

ECHA considers that the *in vitro* mammalian cell gene mutation tests using the *Hprt* and *xprt* genes (OECD TG 476) and the *in vitro* mammalian cell gene mutation tests using the thymidine kinase gene (OECD TG 490) are appropriate to address the standard information requirement of Annex VIII, Section 8.4.3.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: *In vitro* mammalian cell gene mutation test (test method: OECD TG 476 *or* OECD TG 490).

**2. Sub-chronic toxicity study (90-day), oral route (Annex IX, Section 8.6.2.)**

Pursuant to Articles 10(a) and 12(1) of the REACH Regulation, a technical dossier registered at 100 to 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to IX of the REACH Regulation. The information to be generated for the dossier must fulfil the criteria in Article 13(4) of the same regulation.

A "sub-chronic toxicity study (90 day)" is a standard information requirement as laid down in Annex IX, Section 8.6.2. of the REACH Regulation. Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement.

You have not provided any study record of a sub-chronic toxicity study (90 day) in the dossier that would meet the information requirement of Annex IX, Section 8.6.2.

In the technical dossier you have provided a study record for a "*Twenty-eight day Repeat Dose Oral Toxicity Test of Tetrahydrothiophene-1,1-dioxide in Rats*". However, this study does not provide the information required by Annex IX, Section 8.6.2., because exposure duration is less than 90 days and the number of animals per dose group is significantly lower. Therefore, the sensitivity of a 28-day study is much lower than that of a 90-day study.

You have also provided non-guideline repeated dose toxicity inhalation studies with the exposure duration of 90 to 110 days in rats and guinea pigs. However, these studies do not provide the information required by Annex IX, Section 8.6.2., because these studies are non-guideline, not following good laboratory practice, and relevant parameters were not investigated (e.g. no histopathological examination of the reproductive organs, thymus, pituitary, peripheral nerve, cerebrum, medulla/pons, bone marrow, adrenals, bone marrow, brain (including sections of cerebrum, medulla/pons), parathyroids and peripheral nerve (sciatic or tibial, preferably close to muscle). Furthermore, ECHA notes that the highest dose levels used for these studies were lower than the limit dose levels required by the test guidelines.

ECHA acknowledges your comment on the draft decision. With respect to the "*90-day repeated dose study (Sulfolane toxicity study by oral administration via drinking water pathway to CD rats for 13 weeks)*", it is your responsibility to consider the appropriateness of this study to fulfill the standard information requirements of Annex IX, Section 8.6.2. and to justify the appropriateness accordingly.

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

ECHA has evaluated the most appropriate route of administration for the study. Based on the information provided in the technical dossier and/or in the chemical safety report, ECHA considers that the oral route - which is the preferred one as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 4.1, October 2015) Chapter R.7a, section R.7.5.4.3 - is the most appropriate route of administration. More specifically, even though the information indicates that human exposure to the registered substance by the inhalation route is likely, inhalation studies are available that could provide some (limited) information on the local effects of the substance in the respiratory tract. Hence, the test shall be performed by the preferred oral route using the test method EU B.26./OECD TG 408.

According to the test method EU B.26./OECD TG 408 the rat is the preferred species. ECHA considers this species as being appropriate and testing should be performed with the rat.

In the 28-day repeated dose toxicity study present in your registration dossier, increased number of hyaline droplets and eosinophilic bodies were observed in male rats at doses of 200 and 700 mg/kg bw/d. Increased basophilic staining was also noted in the renal tubules of male rats at doses of 700 mg/kg/day. You assume that those effects appear to be indicative of alpha 2u nephropathy. The fact that these effects were only observed in male rats may indicate that the registered substance may induce alpha-2u-globulin-mediated nephropathy. ECHA accordingly considers that the kidney is a target organ of the registered substance. Since humans do not excrete alpha-2u-globulin and this mode of action is therefeore considered not relevant to humans, the involvement of alpha-2u-globulin in the kidney effects is a key parameter to be assessed in order to establishing the relevance of the kidney effects for hazard and risk assessment.

For these reasons, ECHA considers that urine analysis is required to investigate kidney function (which is optional in paragraphs 3, 30 and 32 of OECD TG 408). Additionally, a full histopathological examination (paragraphs 3, 35 and 36 of OECD TG 408), which is to include immunohistochemical investigation of renal pathology to determine whether the pathology is indeed mediated by alpha-2u globulin.

Therefore, pursuant to Article 41(1) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Repeated dose 90-day oral toxicity study (test method: EU B.26./OECD TG 408) in rats modified to include urinalysis and a full histopathological examination which is to include immunohistochemical investigation of renal pathology to determine if the pathology is mediated by alpha-2u globulin nephropathy.

*Notes for your consideration:*

The "*Report of the Expert Peer Review of Sulfolane Reference Doses for the Alaska Department of Environmental Conservation*" refers to 90-day oral studies (HLS 2001)[2], taking into account the degradation products of the registered substance.

### 3. Identification of degradation products (Annex IX, 9.2.3.)

Pursuant to Articles 10(a) and 12(1) of the REACH Regulation, a technical dossier registered at 100 to 1000 tonnes per year shall contain as a minimum the information specified in Annexes VII to IX of the REACH Regulation. The information to be generated for the dossier must fulfil the criteria in Article 13(4) of the same regulation.

The identification of the degradation products is a standard information requirement according to column 1, Section 9.2.3. of Annex IX of the REACH Regulation. Column 2 of Section 9.2.3. of Annex IX further states that the information does not need to be provided if the substance is readily biodegradable.

ECHA notes that you have sought to adapt this information requirement according to Annex IX, Section 9.2., column 2. You provided the following justification for the adaptation: "*In accordance with column 2 of REACH Annex IX, the simulation testing on ultimate degradation in water and sediment does not need to be conducted as the chemical safety assessment according to Annex I has not indicated a need to investigate further the degradation of the substance.*"

ECHA notes that you have considered the substance to be persistent for the PBT assessment and for the risk assessment (i.e. no biodegradation has been assumed). ECHA further acknowledges that the registered substance per se is not PBT/vPvB because of its low bioaccumulation potential. However, pursuant to Annex XIII of the REACH Regulation "*the identification* [of PBT and vPvB substances] *shall also take account of the PBT/vPvB-properties of relevant constituents of a substance and relevant transformation and/or degradation products*". Your chemical safety assessment does not contain any information on the degradation products and on whether they could be PBT/vPvB or not.

ECHA notes that information on degradation products shall be taken into account for the exposure assessment (Annex I 5.2.4. of the REACH Regulation) and for the hazard assessment (e.g. column 2 of Annex X 9.4 and Annex X 9.5.1 of the REACH Regulation). Finally, information on degradation products is required for the preparation of Section 12 of the safety datasheet (Annex II of the REACH Regulation).

In conclusion, ECHA notes that you have not provided any justification in your chemical safety assessment or in the technical dossier for why there is no need to identify the degradation products. ECHA considers that the information is needed for the PBT/vPvB assessment, for the hazard and exposure assessment and for the compilation of safety data sheets.

As explained above, ECHA considers that you have failed to provide information on the identity of degradation products and to provide a valid adaptation of this standard information requirement. Consequently, there is an information gap and it is necessary to request information for this endpoint.

[2] https://ntp.niehs.nih.gov/ntp/about_ntp/bsc/2011/december/presentations/5_blystone_sulfolane.pdf

Based on the information provided in your registration dossier, ECHA notes that the registered substance is well soluble in water. Aerobic mineralisation in surface water – simulation biodegradation (test method EU C.25. / OECD TG 309) is a validated standard international test laid down in the Test Methods Regulation (EC) No 440/2008 (Sections C.23 and C.24) and therefore meets the requirements of Article 13(3) of the REACH Regulation. This test is appropriate to obtain information on the primary degradation and the formation of major transformation products in water. The analytical methods used will have to be substance-specific. When analytically possible, identification, stability, behaviour, molar quantity of metabolites relative to the parent compound should be evaluated. In addition, degradation half-life, log Kow and potential toxicity of the metabolites may be investigated. As specified in the OECD 309 test guideline, higher concentrations of the test substance (e.g. >100 µg/L) could be used for the identification and quantification of major transformation products to overcome potential analytical limitations.

In your comments to the draft decision, you have proposed to update your dossier to include additional information on degradation data and potential degradation products. You have proposed to apply a weight of evidence approach using available information from the literature but also agreed to conduct additional testing if data from the literature are insufficient to meet the information requirement. You requested ECHA to pause the decision making process until January 2017 so you could have enough time to collect literature data and update your dossier.

ECHA acknowledges that you can provide an adaptation pursuant to Annex XI of the REACH Regulation for identifying degradation products, e.g. if information on the possible degradation pathways for the substance is available in the literature or if it can be predicted. However, ECHA has continued the decision making process considering that the current deadline of 18 months after adoption of the final decision is sufficient to develop your proposed weight of evidence approach and, if needed, to conduct a simulation test.

Therefore, pursuant to Article 41(1)(a) and (3) of the REACH Regulation, you are requested to submit the following information derived with the registered substance subject to the present decision: Identification of the degradation products (Annex IX, Section 9.2.3.) by using the following test method: Aerobic mineralisation in surface water – simulation biodegradation (test method EU C.25. / OECD TG 309).

*Notes for your consideration*

ECHA acknowledges that your chemical safety assessment does not indicate a need to determine an actual value for the degradation rate or for the half-life of the substance, since, as a worst-case assumption, you have considered the registered substance to be persistent. Therefore, the separate issue for simulation testing on ultimate degradation in surface water (Annex IX, Section 9.2.1.2.) that was included in the draft decisions sent for your comments has been removed from this decision. However, as explained above, the requirement to provide information on the degradation products must be fulfilled, which implies conducting a simulation test for this purpose where this information requirement cannot be met by other means. Furthermore, the test does not need to be conducted at a temperature of 12°C as initially indicated since a higher test temperature is acceptable for the purpose of identifying degradation products within the frame provided by the study guideline.

## Appendix 2: Procedural history

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

The compliance check was initiated on 1 June 2016.

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 requests.

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.

You provided comments only on the draft decision. Your comments were not taken into account by the Member State Committee as they were considered to be outside of the scope of Article 51(5).

The Member State Committee reached a unanimous agreement on the draft decision in its MSC-51 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 compliance check decision does not prevent ECHA from initiating further compliance checks on the present registration at a later stage.

2. Failure to comply with the 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 your Member State.

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.