

Helsinki, 3 June 2020

**Addressees**

Registrants of JS\_483-360-5 listed in the last Appendix of this decision

**Date of submission for the jointly submitted dossier subject of a decision**

03/06/2019

**Registered substance subject to this decision, hereafter 'the Substance'**

Substance name: 4-fluoro-1,3-dioxolan-2-one

EC number: 483-360-5

CAS number: 114435-02-8

**Decision number:** [Please refer to the REACH-IT message which delivered this communication (in format TPE-D-XXXXXXXXXX-XX-XX/F)]**DECISION ON A TESTING PROPOSAL**

Based on Article 40 of Regulation (EC) No 1907/2006 (REACH), ECHA requests that you submit the information listed below by the deadline of **8 September 2021**.

**A. Requirements applicable to all the Registrants subject to Annex VII of REACH**

1. In vivo mammalian alkaline comet assay (triggered by Annex VII, Section 8.4., column 2)

**B. Requirements applicable to all the Registrants subject to Annex VIII of REACH**

1. In vivo mammalian alkaline comet assay (Annex VIII, Section 8.4., column 2; test method: OECD TG 489) in rats, oral route, on the following tissues: liver, glandular stomach and duodenum with the Substance

**Conditions to comply with the requests**

Each addressee of this decision is bound by the requests for information corresponding to the REACH Annexes applicable to their own registered tonnage of the Substance at the time of evaluation of the jointly submitted dossier.

To identify your legal obligations, please refer to the following:

- you have to comply with the requirements of Annex VII of REACH, if you have registered a substance at 1-10 tonnes per annum (tpa), or as a transported isolated intermediate in quantity above 1000 tpa;
- you have to comply with the requirements of Annexes VII and VIII of REACH, if you have registered a substance at 10-100 tpa.

Registrants are only required to share the costs of information they are required to submit to fulfil the information requirements for their registration.

The Appendices state the reasons for the requests for information to fulfil the requirements set out in the respective Annexes of REACH.

For certain endpoints, ECHA requests the same study from registrants at different tonnages. In such cases, only the reasoning why the information is required at lower tonnages is provided in the corresponding Appendices. For the highest tonnage, the full reasoning for the request including study design is given. Only one study is to be conducted; the registrants concerned must make every effort to reach an agreement as to who is to carry out the study on behalf of the other registrants under Article 53 of REACH.

The Appendix entitled Observations and technical guidance addresses the generic approach for the selection and reporting of the test material used to perform the required studies and provides generic recommendations and references to ECHA guidance and other reference documents.

You must submit the information requested in this decision by the deadline indicated above in an updated registration dossier and also update the chemical safety report, where relevant, including any changes to classification and labelling, based on the newly generated information.

## **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, has to 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<sup>1</sup> under the authority of Christel Schilliger-Musset, Director of Hazard Assessment

---

<sup>1</sup> 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 A: Reasons for the requirements applicable to all the Registrants subject to Annex VII of REACH**

This decision is based on the examination of the testing proposal you submitted.

**1. In vivo mammalian alkaline comet assay (Annex VII, Section 8.4., column 2)**

Under Annex VII to REACH, further mutagenicity studies must be considered in case of a positive result in an *in vitro* gene mutation study in bacteria.

The ECHA guidance R.7a states that following a positive result in an *in vitro* test, "*adequately conducted somatic cell in vivo testing is required to ascertain if this potential can be expressed in vivo. In cases where it can be sufficiently deduced that a positive in vitro finding is not relevant for in vivo situations (e.g. due to the effect of the test substances on pH or cell viability, in vitro-specific metabolism: see also Section R.7.7.4.1), or where a clear threshold mechanism coming into play only at high concentrations that will not be reached in vivo has been identified (e.g. damage to non-DNA targets at high concentrations), in vivo testing will not be necessary.*"

You have submitted a testing proposal for an *in vivo* mammalian alkaline comet assay.

Your dossier contains a positive result for the *in vitro* gene mutation study in bacteria which raises the concern for gene mutation. The *in vivo* study submitted in your dossier does not address the gene mutation concern.

An appropriate *in vivo* genotoxicity study to follow up the concern on gene mutation is not available for the Substance. You considered it necessary to generate information for this endpoint.

ECHA requested your considerations for alternative methods to fulfil the information requirement for Genetic toxicity *in vivo*. ECHA notes that you provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed. ECHA has taken these considerations into account.

ECHA agrees that an appropriate *in vivo* follow up mutagenicity study is necessary to address the concern identified *in vitro*.

ECHA considers that the proposed test is appropriate to investigate effects on gene mutation *in vivo* as described in the ECHA Guidance<sup>2</sup>.

For the specifications of the study to be performed, see the request B.1.

Under Article 40(3)(a) of REACH, you are requested to carry out the proposed test with the Substance.

---

<sup>2</sup> ECHA Guidance Chapter R.7a, Section R.7.7.6.3

**Appendix B: Reasons for the requirements applicable to all the Registrants subject to Annex VIII of REACH**

This decision is based on the examination of the testing proposal you submitted.

**1. In vivo mammalian alkaline comet assay (Annex VIII, Section 8.4., column 2; test method: OECD TG 489)**

Under Annex VIII to REACH, the performance of an appropriate in vivo somatic cell genotoxicity study must be considered if there is a positive result in any of the in vitro genotoxicity studies in Annex VII or VIII.

The ECHA guidance R.7a states that following a positive result in an *in vitro* test, "*adequately conducted somatic cell in vivo testing is required to ascertain if this potential can be expressed in vivo. In cases where it can be sufficiently deduced that a positive in vitro finding is not relevant for in vivo situations (e.g. due to the effect of the test substances on pH or cell viability, in vitro-specific metabolism: see also Section R.7.7.4.1), or where a clear threshold mechanism coming into play only at high concentrations that will not be reached in vivo has been identified (e.g. damage to non-DNA targets at high concentrations), in vivo testing will not be necessary.*"

You have submitted a testing proposal for an in vivo mammalian alkaline comet assay.

Your dossier contains a positive result for the *in vitro* gene mutation study in bacteria which raises the concern for gene mutation. The *in vivo* study submitted in your dossier does not address the gene mutation concern.

An appropriate *in vivo* genotoxicity study to follow up the concern on gene mutation is not available for the Substance. You considered it necessary to generate information for this endpoint.

ECHA requested your considerations for alternative methods to fulfil the information requirement for Genetic toxicity in vivo. ECHA notes that you provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed. ECHA has taken these considerations into account.

ECHA agrees that an appropriate *in vivo* follow up mutagenicity study is necessary to address the concern identified *in vitro*.

ECHA considers that the proposed test is appropriate to investigate effects on gene mutation *in vivo* as described in the ECHA Guidance<sup>2</sup>.

*Test selection*

According to the ECHA Guidance Chapter R.7a<sup>3</sup>, the transgenic rodent somatic and germ cell gene mutation assays ("TGR assay", OECD TG 488) and the *in vivo* mammalian alkaline comet assay ("comet assay", OECD TG 489) are suitable to follow up a *positive in vitro* result on gene mutation.

---

<sup>3</sup> ECHA Guidance Chapter R.7a, Section R.7.7.6.3

Therefore, the proposed comet assay is a suitable test to follow up the concern on gene mutation for the Substance.

#### *Test design*

You proposed testing in rats by the oral route.

According to the test method OECD TG 489, the test must be performed in rats. Having considered the anticipated routes of human exposure and adequate exposure of the target tissue(s) performance of the test by the oral route is appropriate.

In line with the test method OECD TG 489, the test must be performed by analysing tissues from liver as primary site of xenobiotic metabolism, glandular stomach and duodenum as sites of contact. There are several expected or possible variables between the glandular stomach and the duodenum (different tissue structure and function, different pH conditions, variable physico-chemical properties and fate of the Substance, and probable different local absorption rates of the Substance and its possible breakdown product(s)). In light of these expected or possible variables, it is necessary to analyse both tissues to ensure a sufficient evaluation of the potential for genotoxicity at the site of contact in the gastro-intestinal tract.

You may consider to collect the male gonadal cells collected from the seminiferous tubules (as described by e.g. O'Brien *et al.*<sup>4</sup>) in addition to the other aforementioned tissues, as it would optimise the use of animals. You can prepare the slides for male gonadal cells and store them for up to 2 months, at room temperature, in dry conditions and protected from light. Following the generation and analysis of data on somatic cells, you should consider analysing the slides prepared with gonadal cells. This type of evidence may be relevant for the overall assessment of possible germ cell mutagenicity including classification and labelling according to the CLP Regulation.

Under Article 40(3)(a) of REACH, you are requested to carry out the proposed test with the Substance.

---

<sup>4</sup> O'Brien, J.M., Beal, M.A., Gingerich, J.D., Soper, L., Douglas, G.R., Yauk, C.L., Marchetti, F. (2014) Transgenic Rodent Assay for Quantifying Male Germ Cell Mutant Frequency. *J. Vis. Exp.* (90), e51576, doi:10.3791/51576

### **Appendix C: Procedural history**

ECHA received your registration containing the testing proposals for examination on 10 June 2019.

ECHA held a third party consultation for the testing proposal from 24 July 2019 until 9 September 2019. ECHA did not receive information from third parties.

For the purpose of the decision-making, this decision does not take into account any updates of registration dossiers after the date on which you were notified the draft decision according to Article 50(1) of REACH.

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

ECHA did not receive any comments within the 30-day notification period.

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 D: Observations and technical guidance

1. This testing proposal examination decision does not prevent ECHA from initiating compliance checks at a later stage on the registrations present.
2. Failure to comply with the requests in this decision, or to otherwise fulfil the information requirements with a valid and documented adaptation, will result in a notification to the enforcement authorities of your Member State(s).

3. Test guidelines, GLP requirements and reporting

Under Article 13(3) of REACH, all new data generated as a result of this decision needs to be conducted according to the test methods laid down in a European Commission Regulation or according to international test methods recognised by the Commission or ECHA as being appropriate.

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.

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: 'How to report robust study summaries'<sup>5</sup>.

4. Test material

### *Selection of the test material(s)*

The registrants of the Substance are responsible for agreeing on the composition of the test material to be selected for carrying out the tests required by the present decision. The test material selected must be relevant for all the registrants of the Substance, i.e. it takes into account the variation in compositions reported by all members of the joint submission. The composition of the test material(s) must fall within the boundary composition(s) of the Substance.

While selecting the test material you must take into account the impact of each constituent/impurity is known to have or could have 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.

### *Technical reporting of the test material*

The composition of the selected test material must be reported in the respective endpoint study record, under the Test material section. The composition must include all constituents of the test material and their concentration values. Without such detailed reporting, ECHA may not be able to confirm that the test material is relevant for the Substance and to all the registrants of the Substance.

Technical instructions are available in the manual "How to prepare registration and PPORD dossiers"<sup>6</sup>.

<sup>5</sup> <https://echa.europa.eu/practical-guides>

<sup>6</sup> <https://echa.europa.eu/manuals>

5. List of references of the ECHA Guidance and other guidance/ reference documents<sup>7</sup>

QSARs, read-across and grouping

Guidance on information requirements and chemical safety assessment, Chapter R.6 (version 1.0, May 2008), referred to as ECHA Guidance R.6 in this decision.

ECHA Read-across assessment framework (RAAF, March 2017)<sup>8</sup>

Toxicology

Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

Environmental toxicology and fate

Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7b (version 4.0, June 2017), referred to as ECHA Guidance R.7b in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

PBT assessment

Guidance on information requirements and chemical safety assessment, Chapter R.11 (version 3.0, June 2017), referred to as ECHA Guidance R.11 in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.16 (version 3.0, February 2016), referred to as ECHA Guidance R.16 in this decision.

OECD Guidance documents

Guidance Document on aqueous-phase aquatic toxicity testing of difficult test chemicals – No 23, referred to as OECD GD 23.

Guidance Document on Standardised Test Guidelines for Evaluating Chemicals for Endocrine Disruption – No 150, referred to as OECD GD 150.

---

<sup>7</sup> <https://echa.europa.eu/guidance-documents/guidance-on-information-requirements-and-chemical-safety-assessment>

<sup>8</sup> <https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across>



**Appendix E: List of the registrants to which the decision is addressed and the corresponding information requirements applicable to them**

<b>Registrant Name</b>	<b>Registration number</b>	<b>(Highest) Data requirements to be fulfilled</b>
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]

Note: where applicable, the name of a third party representative (TPR) may be displayed in the list of recipients whereas the decision is sent to the actual registrant.