Meeting the requirements for toxicological information in applications for authorisation of Plant Protection Products under Regulations (EC) No 1107/2009 and (EC) No 284/2013: a guide for applicants

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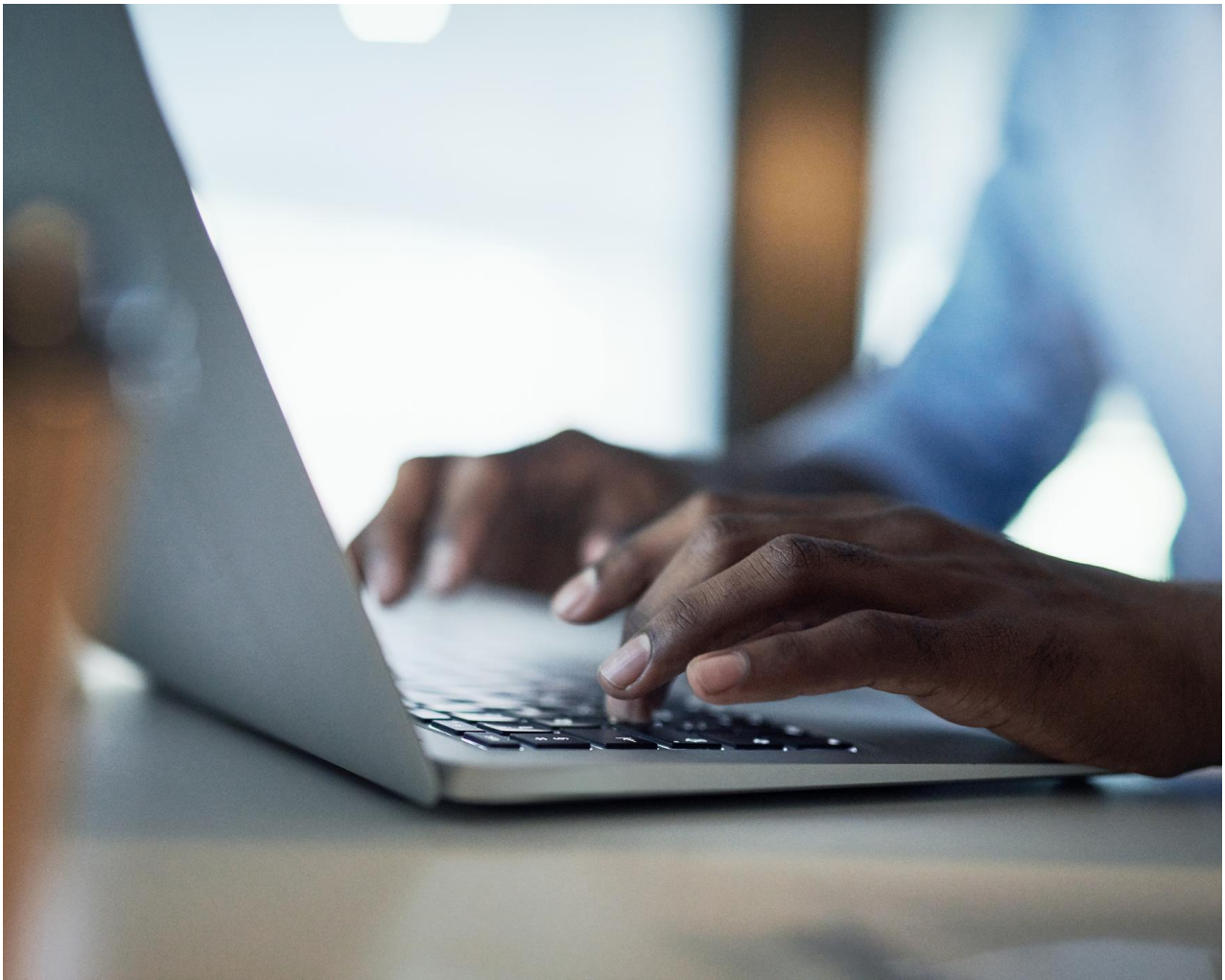

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Introduction

An applicant for authorisation of a Plant Protection Product (PPP) under Regulation (EC) No 1107/2009 is required to submit to the relevant regulatory authority (HSE in the case of GB) information on some aspects of the toxicity of the product: acute oral, dermal and inhalation toxicity, skin and eye irritation potential and skin sensitisation potential. Information on the dermal absorption of the active substance in the PPP is also required.

1. Regulation (EC) No 284/2013 section 7 sets out the toxicological information that applicants must submit to support their applications for authorisation. The primary purpose of this information is to establish the toxicity of the PPP after single exposures so that appropriate measures can be implemented to protect against any potential consequences arising from these forms of toxicity in those who might encounter the product: distributors, users, bystanders, residents, consumers or workers. In considering authorisation, other known information on the toxicology of the active substance(s) and co-formulants shall also be taken into account (for example, repeated-dose toxicity, carcinogenicity, reproductive toxicity; see paragraph 5 below in relation to information on co-formulants), but specific analyses of the PPP as a whole for these potential effects are not required.

2. The information provided should also permit the applicant to propose a classification of the PPP in accordance with Regulation (EC) No 1272/2008 (CLP), where appropriate (Para 1.10 of Reg (EC) No 284/2013). The label that ensues from the classification informs the PPP's users of its hazards and of appropriate risk management measures.

3. Regulation (EC) No 284/2013 offers options, with associated criteria, for applicants to take in meeting the information requirements. Of particular importance is that, before conducting a study on vertebrate animals, applicants must consider if other methods (for example, *in vitro* tests, computational (*in silico*) predictions) are available for use as acceptable substitutes for animal testing. For each test conducted on vertebrate animals, an explanation must be provided of why it was necessary and why alternative approaches were not appropriate, in accordance with Article 33 (3)(c) of Regulation (EC) No 1107/2009. Article 62 (1) of this Regulation also stipulates that testing on vertebrate animals for the purposes of this Regulation shall be undertaken only where no other methods are available.

4. Available toxicological data on co-formulants should be submitted and assessed by the applicant (section 7.4. of Regulation (EC) No 284/2013). This information shall include, but is not limited to: where applicable, registration numbers issued under Regulation (EC) No 1907/2006 (REACH); the study summaries included in the REACH technical

(registration) dossier; an SDS that complies with Article 31 of REACH. A REACH-compliant SDS should also be submitted for the PPP.

5. The information requirement for each aspect of toxicity listed in paragraph 1 above and the options for meeting the requirement are outlined below. These are presented in a tiered sequence to be consistent with the provisions of Article 62 (1) of Regulation (EC) No 1107/2009, i.e., with the requirement to minimise unnecessary animal testing.

Acute oral toxicity

6. Sufficiently reliable information to establish the acute oral toxicity of the PPP and enable the applicant to propose a classification, if appropriate, is required. There are three ways in which this requirement might be satisfied, with important conditions applying to each approach:

    a. by “bridging” – prediction of the acute oral toxicity of the PPP by reading across from such data available on another product of similar composition;

    b. by the “calculation method” – prediction of the acute oral toxicity of the PPP from the acute oral toxicity data of its individual components;

    c. via the results of an acute oral toxicity study on the PPP in experimental animals. The study might be a pre-existing one (perhaps originally conducted for a different purpose), or one undertaken specifically for the PPP authorisation application.

7. Further details on the appropriate conditions for each approach are given below. [Note that at present there are no internationally-recognised *in vitro* tests for assessing acute oral toxicity.]

8. In relation to the requirement for information on acute oral toxicity, there are options for the applicant to use an alternative approach to acute oral toxicity assessment that is recognised under the CLP Regulation (1272/2008); i.e., without undertaking an acute oral toxicity test on the PPP for which authorisation is being sought. Applicants should first consider if one of these alternative approaches is appropriate.

9. An alternative approach under Annex 1 to the CLP Regulation is bridging, in which an applicant will rely upon acute oral toxicity data generated in an existing study on vertebrate animals conducted with another closely-related formulation.

10. As outlined below (paragraphs 13 to 15) for an acute oral toxicity study on the specific PPP for which authorisation is sought, the same principle applies to using “bridging” data from a test on a different substance – if the test that generated the data to be used for bridging was commissioned after 14 June 2011, and not previously evaluated under Regulation (EC) No 1107/2009, the applicant would be required to explain why it was necessary to rely on the results of an *in vivo* test for the purposes of an authorisation application. On receipt of an application in which the bridging approach is used, HSE will give consideration to the legitimacy of the bridging argument, based on the chemical composition of the PPP for which authorisation is sought and the material on which acute toxicity data has been generated. Where the test that generated the data to be used for bridging has previously been evaluated under Regulation (EC) No 1107/2009, no such justification is required to re-use the test data.

11. Another alternative approach is the calculation method (additivity). The calculation method involves a prediction of the toxicity of a whole mixture from a consideration of the toxicity of its individual components. If this approach is used, under paragraph 7.1.1 of the Annex to Regulation (EC) No 284/2013, the applicant must provide the calculations. Given the duty under section 62 of Regulation (EC) No 1107/2009 to test on animals only where no other methods are available, the use of an alternative approach is encouraged by HSE. Acute oral toxicity of all components of the PPP must be provided or reliably predicted. Under Annex 1 to Regulation (EC) No 1272/2008 "all components" means the relevant ingredients of a mixture, i.e. those that are present in concentrations of 1% or greater, unless there is a reason to suspect that an ingredient present at a concentration of less than 1% is still relevant for classifying the mixture for acute toxicity. Ingredients that are presumed not to be acutely toxic (for example, water, sugar or common food ingredients, such as xanthan gum) would not be considered relevant for classifying the mixture for acute toxicity[1]. Information on the acute oral toxicity of a component should be obtained from its safety data sheet (SDS). Its harmonised classification should also be checked, as, if available and if more severe, it has precedence over the SDS self-classification. The most recent version of the SDS should be used, which should be compliant with the SDS stipulations of REACH; Article 31 (9) of REACH specifies under what circumstances suppliers must update their SDS. HSE interpret the lack of acute oral toxicity classification (in section 2 of the SDS or in the harmonised entry) as lack of hazard and not as an unknown (data gap). With classification information (or lack thereof) of all relevant components, a calculation of the predicted acute oral toxicity can be made. As lack of classification is interpreted as lack of hazard, the additivity formula with no unknowns described in paragraph 3.1.3.6.1 of Annex I of the CLP Regulation should be used. Note that if all components have an LD50 > 2000 mg/kg bw or are not classified, the calculation method will not apply, but it can be presumed that the PPP will also have an LD50 > 2000 mg/kg bw.

12. If neither of the above approaches is feasible for the PPP concerned (e.g. no suitable bridging or no SDSs provided for some co-formulants), another approach is to use the results of an acute oral toxicity study on the PPP in experimental animals. However, if such an approach is considered, it is crucial to keep in mind the stipulations of Articles 33 and 62 of Regulation (EC) No 1107/2009. The use of this approach should be only in situations where it is judged that no other methods are available by which to adequately assess the acute oral toxicity of the PPP; and an explanation of why it was not possible to avoid animal testing should be provided.

[1] Please refer to section 3 of Annex I to the CLP Regulation (1272/2008) and the associated *Guidance on the Application of the CLP Criteria* for information on the levels of components that should be taken into account in the consideration of mixture toxicity; i.e., generic concentration limits, specific concentration limits, cut-off levels for inclusion of a component in calculations.

13. As the implementation date for Regulation (EC) No 1107/2009 was 14 June 2011, these provisions apply to the use of data from a test on vertebrate animals that were commissioned after that date – regardless of the purpose for which the study was originally undertaken.

14. If the approach taken is to use the results of an acute oral toxicity study on the PPP in experimental animals and a justification for this approach has not been provided, or is considered by HSE to be insufficient, the application will be rejected at the sift stage. HSE will also consider investigating whether there has been a contravention of Article 62(1) of Regulation (EC) No 1107/2009 and therefore whether an offence has been committed under regulation 23 of the Plant Protection Products Regulations 2011.

15. So, although acute oral toxicity information is required relating to a PPP for which authorisation is sought, this should not be generated by experimental animal testing where an alternative approach can be justified. There are criteria to be satisfied for each of the possible approaches. A particular issue is the desirability of minimising the use of experimental animals in satisfying this information requirement and the need to justify its use.

16. Note that if an experimental animal study has already been done and the result indicates a degree of toxicity (acute oral in this case) substantially greater than anticipated, then such data (meeting the definition of “adverse data” under Article 56 of Regulation (EC) No 1107/2009) will be used in HSE’s evaluation.

Acute dermal toxicity

17. Sufficiently reliable information to establish the acute dermal toxicity of the PPP and enable the applicant to propose a classification, if appropriate, is required on a case-by-case basis (for example, it might be possible to justify non-submission of information if there is evidence that none of the components of the PPP is likely to be absorbed through the skin to any significant extent). One consideration in deciding how to meet this information requirement is that, in general, the acute dermal toxicity of an individual chemical or a product is unlikely to be higher than its acute oral toxicity, because the speed and extent of absorption into the systemic circulation is usually greater in the latter case.

18. Therefore, there are several ways in which this requirement might be satisfied, with important conditions applying to each approach:

    a. by “bridging” – prediction of the acute dermal toxicity of the PPP by reading across from such data (dermal or oral) available on another product of similar composition;

    b. by the “calculation method” – prediction of the acute dermal toxicity of the PPP from the acute dermal toxicity data of its individual components;

    c. by extrapolation from the results of an acute *oral* toxicity study on the PPP in experimental animals. The study might be a pre-existing one, or one undertaken specifically for the PPP authorisation application;

    d. via the results of an acute dermal toxicity study on the PPP in experimental animals. The study might be a pre-existing one, or one undertaken specifically for the PPP authorisation application.

19. Further details on the appropriate conditions for each approach are given below. [Note that at present there are no internationally-recognised *in vitro* tests for assessing acute dermal toxicity.]

20. In relation to the requirement for information on acute dermal toxicity, there are options for the applicant to use an alternative approach to acute dermal toxicity assessment that is recognised under the CLP Regulation (1272/2008); i.e., without undertaking an acute dermal toxicity test on the PPP for which authorisation is being sought. Applicants should first consider if one of these alternative approaches is appropriate.

21. One such alternative is the bridging approach, in which an applicant will rely upon acute dermal (or oral) toxicity data generated in an existing study on vertebrate animals conducted with another closely-related formulation. The conditions for the use of this approach are outlined in paragraph 10.

22. Another such approach is the calculation method (additivity). If this approach is used, section 7.1 of Regulation (EC) No 284/2013 states that the applicant shall justify that it is appropriate and shall provide information on the acute dermal (or oral) toxicity of **all relevant** components of the PPP. See paragraph 11 for other conditions that apply to the use of this approach.

23. Another approach is to use the results of an acute *oral* toxicity study on the PPP to predict its dermal toxicity (route-to-route extrapolation). The conditions outlined in paragraphs 13 – 15 would also apply to the use of the results from an acute oral toxicity study on experimental animals in this scenario.

24. Only if none of the above approaches can be used and it has been determined that information is needed, the final approach is to use the results of an acute dermal toxicity study on the PPP in experimental animals. See paragraphs 13 – 15 for the conditions that apply to the use of results from tests on experimental animals.

25. So, although acute dermal toxicity information might be required relating to a PPP for which authorisation is sought, this need not be generated by experimental animal testing. There are criteria to be satisfied for each of the possible approaches. A particular issue is the desirability of minimising the use of experimental animals in satisfying this information requirement and the need to justify its use.

Acute inhalation toxicity

26. Sufficiently reliable information to establish the acute inhalation toxicity of the PPP and enable the applicant to propose a classification, if appropriate, is required. Section 7.1.3. of Regulation (EC) No 284/2013 lists the criteria that determine if an acute inhalation toxicity test on the formulation should be provided – a test is **only** required if the PPP meets one of these exposure-based criteria:

    a. it's a gas or liquefied gas;

    b. it's a smoke-generating PPP or fumigant;

    c. it's used with fogging/misting equipment;

    d. it's a vapour-releasing PPP;

    e. it's supplied in an aerosol dispenser;

    f. it's in the form of a powder or granules containing a significant proportion of particles of diameter < 50 µm (> 1% on a weight basis);

    g. it's to be applied from aircraft in cases where inhalation exposure is relevant;

    h. it contains an active substance with a vapour pressure > 1×10^{-2} Pa and is to be used in enclosed spaces;

    i. it's to be applied by spraying.

27. An applicant could therefore present a waiver for the test. If the test can be waived, hazard classification for this endpoint should still be addressed by applying the calculation method of the CLP Regulation (see paragraph 35 below). It is noted that where the calculations lead to classification of the product despite minimal/negligible inhalation exposure, the applicant should consider whether it is appropriate to apply the predicted classification in line with the provisions of Art 9(5) of the CLP Regulation (i.e. the form or physical state in which the product is placed on the market and can be reasonably expected to be used may be irrelevant to the acute inhalation toxicity hazard).

28. If the test cannot be waived, then there are three ways in which the information requirements might be satisfied, with important conditions applying to each approach:

    a. by "bridging" – prediction of the acute inhalation toxicity of the PPP by reading across from such data available on another product of similar composition;

b. by the “calculation method” – prediction of the acute inhalation toxicity of the PPP from the acute inhalation toxicity data of its individual components;

c. via the results of an acute inhalation toxicity study on the PPP in experimental animals. The study might be a pre-existing one, or one undertaken specifically for the PPP authorisation application.

29. Further details on the appropriate conditions for each approach are given below. [Note that at present there are no internationally-recognised *in vitro* tests for assessing acute inhalation toxicity.]

30. As noted above, applicants should first decide if information on acute inhalation toxicity is needed for the PPP to be considered for authorisation. One of the criteria that require such information to be provided is that the PPP *is to be applied by spraying*. This would seem to encompass most PPPs. However, in deciding if this criterion applies, applicants could consider the following questions:

    a. will the supplied product (for which authorisation is sought) be sprayed, or will it be diluted before spraying? If the latter, then it is not the PPP (for which authorisation is sought) that “is to be applied by spraying” and therefore the criterion is <u>not</u> met;

    b. as a particular case of (a), is the product extensively diluted (≥ 1 in 100) with water before spraying? Such dilutions, where water forms by far the major component, are likely to have very low acute inhalation toxicity;

    c. if it is the PPP that is to be applied by spraying, then what droplet size is produced by the sprayer and how does this relate to the size of particles that are respirable (can reach the deep lung)? The droplet size specified in OECD test guidelines 403 and 436 for acute inhalation toxicity is approximately 3 µm; it is possible that in spray application the droplet size might be appreciably greater – and therefore an experimental test result would be misleading.

31. The OECD has published a document that explains some of the considerations to be taken into account in deciding upon the usefulness of an acute inhalation toxicity test[2].

32. Taking into account considerations such as those above and in the OECD publication, HSE anticipates that, in many cases, applicants will be able to provide a justification for not submitting a study on acute inhalation toxicity.

[2] OECD (2016). Guidance document on considerations for waiving or bridging of mammalian acute toxicity tests. Series on Testing and Assessment No. 237, ENV/JM/MONO(2016)32.

33. If applicants determine that the test cannot be waived, there are options for the applicant to take, using an alternative approach to acute inhalation toxicity assessment that is recognised under the CLP Regulation (1272/2008); i.e., without undertaking an acute inhalation test on the PPP for which authorisation is being sought. Applicants should first consider if one of these alternative approaches is appropriate.

34. One such approach is the bridging approach, in which an applicant will rely upon acute inhalation toxicity data generated in an existing study on vertebrate animals conducted with another closely-related formulation. The conditions for the use of this approach are outlined in paragraph 10.

35. Another alternative approach is the calculation method (additivity). If this approach is used, section 7.1 of Regulation (EC) No 284/2013 states that the applicant shall justify that it is appropriate and shall provide information on the acute inhalation toxicity of **all relevant** components of the PPP. See paragraph 11 for other conditions that apply to the use of this approach.

36. If neither of the above approaches is feasible for the PPP to be authorised, another approach is to use the results of an acute inhalation toxicity study on the PPP in experimental animals (normally by head/nose-only exposure). See paragraphs 13 – 15 for the conditions that apply to the use of results from tests on experimental animals.

37. So, although acute inhalation toxicity information might be required relating to a PPP for which authorisation is sought, this need not be generated by experimental animal testing. There are criteria to be satisfied for each of the possible approaches. A particular issue is the desirability of minimising the use of experimental animals in satisfying this information requirement and the need to justify its use.

Skin irritation

38. Sufficiently reliable information is required to establish the skin irritation potential (including reversibility) of the PPP and enable the applicant to propose a classification, if appropriate.

39. The OECD has published an 'IATA' (integrated approach to testing and assessment) for skin corrosion and irritation[3]. This describes the use of existing test data, physico-chemical properties and non-test methods (such as bridging and additivity) in a weight-of-evidence approach to decide upon the predicted effects on human skin and the need for new tests.

40. The EU pesticides regulatory regime recognises this OECD IATA and, therefore, there are multiple ways in which this requirement for information on skin irritation potential might be satisfied:

    a. by a consideration of the pH of the PPP;

    b. via findings of severe skin irritation or corrosion in an acute dermal study on the PPP, if this study has been done and its conduct was justified;

    c. by "bridging" – prediction of the skin irritation potential of the PPP by reading across from such data available on another product of similar composition;

    d. by the "calculation method" – prediction of the skin irritation potential of the PPP from the skin irritation data of its individual components;

    e. via the results of validated *in vitro* skin irritation / corrosion tests with the PPP;

    f. via the results of an existing skin irritation study on the PPP that uses experimental animals.

41. Further details on the appropriate conditions for each approach (a – f) are given below. [Note that there are now internationally-recognised *in vitro* test methods for assessing skin irritation that can provide a full replacement for *in vivo* tests.]

42. If the pH of the PPP to be authorised is ≤ 2 or ≥ 11.5, it can be presumed that it will be corrosive to skin – (a). No further tests are needed and the appropriate classification should be applied. In some cases, if it can be shown that the acid/alkaline reserve in combination with other sources of information (e.g. in vitro tests) justifies a lower

[3] OECD (2014). New guidance document on an integrated approach on testing and assessment (IATA) for skin corrosion and irritation. Series on testing and assessment No. 2003, ENV/JM/MONO(2014)19.

classification (even where the formulation has a pH in the extreme ranges described above), this will be taken into account.

43. If an acute dermal toxicity study on the PPP resulted in severe skin irritation or corrosion, further information to assess skin irritation potential is not needed – (b). See paragraphs 13 – 15 for the conditions that apply to the use of results from tests on experimental animals. [Note that this could apply also in bridging from local skin effects seen in an acute dermal study on a similar entity.]

44. Beyond (a) and (b), there are options for the applicant to use an alternative approach to skin irritation assessment that is recognised under the CLP Regulation (1272/2008); i.e., without undertaking an *in vivo* skin irritation test on the PPP for which authorisation is being sought.

45. One such alternative is the bridging approach, in which an applicant will rely upon skin irritation data generated in an existing study on vertebrate animals or with *in vitro* test systems conducted with another closely-related formulation – (c). The conditions for the use of this approach are outlined in paragraph 10.

46. Another such approach is the calculation method – (d). If this approach is used, section 7.1 of Regulation (EC) No 284/2013 states that the applicant shall justify that it is appropriate and shall provide information on the skin irritation potential of **all relevant** components of the PPP. See paragraph 11 for other conditions that apply to the use of this approach.

47. Another approach is to undertake new tests on the PPP concerned. At the time Regulation (EC) No 284/2013 was written, outlining the data requirements, the EU Test Method Regulation (EC No 440/2008) described a sequential test strategy for the assessment of acute dermal irritation and corrosion; this strategy recommended that *in vitro* tests be conducted before any tests were undertaken on live animals but did not recognise negative results from the non-animal tests as being conclusive – in other words, tests in animals were required to confirm a negative result. The same strategy was adopted in the data requirements for PPP (Regulation (EC) No 284/2013).

48. In 2014, the OECD published an IATA for skin corrosion and irritation (guidance document 203). The IATA can be applied to both individual substances and mixtures. If a weight-of-evidence analysis of existing test data, physico-chemical properties and non-test methods (such as bridging and additivity) is inconclusive with regards to skin irritation potential, the IATA describes the test methods that can be used to generate more data.

49. The recommended test method(s) to generate more data depend in part upon the skin irritation classification categories and sub-categories of the United Nations Globally Harmonised System (GHS) that have been adopted into regional implementing

legislation. In the EU's implementing legislation, the CLP Regulation (1272/2008), the categories used for skin corrosion are category 1 (sub-categories 1A, 1B, 1C) or non-corrosive; and for skin irritation, category 2 or not classified. By following the tiered strategy in the IATA, *in vitro* test methods, either singly or in combination, can be used to predict the skin irritation potential of substances and mixtures and determine an appropriate classification under CLP. All these methods have OECD test guidelines and regulatory acceptance.

50. Therefore, in the EU these *in vitro* tests can now be used to fully replace tests on experimental animals, provided the test material is within the scope and applicability domain of the tests used and there are no chemical-specific limitations to those tests (for example, the tests do not allow the testing of gases and aerosols) – (e).

51. Another approach to meeting the information requirement is to use the results of an existing skin irritation study on the PPP in experimental animals – (f). See paragraphs 13 – 15 for the conditions that apply to the use of results from tests on experimental animals. Of particular importance in justifying the use of results from a study on animals is the availability of *in vitro* alternatives at the time the test was conducted, and the application of a tiered assessment strategy, taking into account, for example, findings in acute dermal toxicity studies and physico-chemical properties.

52. So, although information on skin irritation potential is required relating to the concerned PPP, this should not be generated by experimental animal testing on PPPs for which the available alternatives are appropriate. Whilst the data requirement in Regulation (EU) 284/2013 mentions the use of information from skin irritation studies on experimental animals, the science has progressed and there are now OECD test guidelines for *in vitro* tests that can fully replace *in vivo* tests for regulatory purposes. Therefore, we would now expect there to be few, if any, instances when new *in vivo* skin irritation tests on PPPs are necessary or justifiable.

Eye irritation

53. Sufficiently reliable information is required to establish the eye irritation potential (including reversibility) of the PPP and enable the applicant to propose a classification, if appropriate.

54. Regulation (EC) No 284/2013 requires that a weight-of-evidence approach be applied to assess eye damage potential before tests on animals are undertaken with the PPP to be authorised.

55. There are multiple ways in which this requirement for information on the potential of the PPP to be able to produce serious eye damage and irritation might be satisfied:

    a. by a consideration of the pH of the PPP;

    b. by a consideration of the skin corrosion potential (predicted or observed) of the PPP;

    c. by “bridging” – prediction of the eye irritation potential of the PPP by reading across from such data available on another product of similar composition;

    d. by the “calculation method” – prediction of the eye irritation potential of the PPP from data on the eye irritation potential of its individual components;

    e. via the results of validated *in vitro* and *ex vivo* eye irritation / corrosion tests with the PPP, either alone or in combination;

    f. via the results of an eye irritation study on the PPP that uses experimental animals.

56. Further details on the appropriate conditions for each approach are given below (a – f). [Note that there are now internationally-recognised *in vitro* and *ex vivo* test methods for assessing eye damage that can provide a partial replacement for *in vivo* tests.]

57. If the pH of the PPP to be authorised is ≤ 2 or ≥ 11.5, it can be presumed that it will cause serious eye damage. No further tests are needed and the appropriate classification should be applied.

58. If the available information indicates that the PPP is corrosive to skin, it can be presumed that the PPP will cause serious eye damage/irritation; further information to assess eye damage/irritation potential is then not needed. See paragraphs 13 – 15 for the conditions that apply to the use of results from tests on experimental animals.

59. Beyond (a) and (b), there are options for the applicant to use an alternative approach to eye damage/irritation assessment that is recognised under the CLP Regulation

(1272/2008); i.e., without undertaking an *in vivo* eye irritation test on the PPP for which authorisation is being sought.

60. One such alternative is the bridging approach, in which an applicant will rely upon eye damage/irritation data generated in an existing study on vertebrate animals or with *in vitro* test systems conducted with another closely-related formulation – (c). The conditions for the use of this approach are outlined in paragraph 10.

61. Another such approach is the calculation method, as applicable – (d). If this approach is used, section 7.1 of Regulation (EC) No 284/2013 states that the applicant shall justify that it is appropriate and shall provide information on the eye damage/irritation potential of all components of the PPP. See paragraph 11 for other conditions that apply to the use of this approach.

62. Another approach is to undertake new tests on the PPP to be authorised. Regulation (EC) No 284/2013 describes a sequential test strategy for the assessment of eye damage, in which *in vitro* / *ex vivo* tests are conducted before any tests are undertaken on live animals.

63. In 2017, the OECD published an 'IATA' for serious eye damage and irritation[4]. The EU (and GB) pesticides regulatory regime recognises this OECD IATA and it can be applied to both individual substances and mixtures. If a weight-of-evidence analysis of existing test data, physico-chemical properties and non-test methods (such as bridging and additivity) is inconclusive with regards to eye damage/irritation potential, the IATA describes the test methods that can be used to generate more data.

64. The *in vitro* test method(s) with OECD test guidelines that are included in the IATA to generate more data are able, when used singly or in combination, to differentiate between chemicals that cause serious eye damage (CLP Category 1) and those that do not cause eye irritation (not classified under CLP) – (e).

65. Therefore, for chemicals that either cause serious eye damage or do not cause eye irritation, these *in vitro* tests can now be used to fully replace tests on experimental animals. The test material should be within the scope and applicability domain of the tests used. *In vitro* / *ex vivo* tests cannot, currently, identify eye irritants that would be classified in CLP category 2.

66. Another approach to meeting the information requirement is to use the results of an existing or new eye irritation study on the PPP in experimental animals – (f). New eye irritation studies on animals should be conducted **only as a last resort**, when it is not possible to reach a conclusion on the PPP's eye irritation potential through the use of

[4] OECD (2017). Guidance document 263 on an integrated approach on testing and assessment (IATA) for serious eye damage and eye irritation.

existing data and *in vitro* / *ex vivo* tests. See paragraphs 13 – 15 for the conditions that apply to the use of results from tests on experimental animals. Of particular importance in justifying the use of results from a study on animals is the availability of *in vitro* alternatives at the time the test was conducted, and the application of a tiered test strategy. Applicants should **not** proceed directly to a test in animals without having first undertaken a weight-of-evidence assessment and appropriate *in vitro* tests; the results of *in vivo* eye damage tests conducted from 2015 onwards and in isolation, without reference to a tiered, weight-of-evidence approach, will not be considered by HSE as fulfilling the information provision requirement for eye irritation in PPP authorisation.

67. So, although information on eye damage/irritation potential is required relating to a PPP for which authorisation is sought, this should not be generated exclusively by experimental animal testing and frequently it might well be appropriate to not conduct an *in vivo* test. A particular issue is the desirability of minimising the use of experimental animals in satisfying this information requirement and the need to justify its use. In all cases where test data are generated, the test strategy must follow a tiered approach.

Skin sensitisation

68. Sufficiently reliable information is required to establish the skin sensitisation potential of the PPP and enable the applicant to propose a classification, if appropriate. There are multiple ways in which this requirement might be satisfied, with important conditions applying to each approach:

    a. if the active substance(s) or any co-formulant is known to have sensitising properties and is present at a level ≥ the relevant concentration limit, no further information is needed;

    If no component triggers classification of the product, then the following approaches should be considered in a sequential manner:

    b. prediction of the skin sensitisation potential of the PPP from the skin sensitisation test data of its individual components;

    c. use of a pre-existing (commissioned before December 2021 – 6-month implementation period after the date of publication of OECD Guideline 497) *in vivo* skin sensitisation study on the PPP of interest, if available;

    d. “bridging” – prediction of the skin sensitisation potential of the PPP by reading across from a pre-existing (commissioned before December 2021) *in vivo* study available on another product of similar composition;

    e. application to the PPP of one of the defined approaches within the recently adopted and published (June 2021) OECD DASS (Defined Approaches for Skin Sensitisation) 497 guideline[5];

    f. via the results of a new skin sensitisation test on the PPP in experimental animals.

69. Further details on the appropriate conditions for each approach are given below.

70. If any component of the PPP to be authorised, whether it be an active substance or a co-formulant, is known to have sensitising properties and is present at a level ≥ the relevant concentration threshold, no further information is needed – (a). For components that meet the criteria of Category 1 / 1B of the CLP Regulation, the relevant generic concentration threshold is 1 %; for Category 1A, it is 0.1 %. Some chemicals have a specific concentration limit (SCL), which takes precedence over the generic limits. If any known sensitising component is present at a level ≥ the relevant

[5] Guideline No. 497: Guideline on Defined Approaches for Skin Sensitisation (oecd-ilibrary.org)

concentration threshold, it is presumed that the PPP will be a skin sensitiser, and no further information needs to be provided.

71. If no component triggers classification of the product, the next option is to consider the skin sensitisation potential of **all relevant** components of the PPP – (b). Co-formulants that are presumed not to be skin sensitisers (for example some polymers or chemicals with high Log Pow unlikely to cross the skin) would be considered not relevant. Specifically in relation to polymers, if an applicant can show that a specific component meets the REACH definition of a polymer and can provide a declaration that none of its monomers, reactants and starting materials are known skin sensitisers, then such a component could be dismissed as not relevant. For skin sensitisation HSE requires actual experimental or human experience data (as described in section 11 of the SDS or the associated REACH registration dossier where available or publicly-available information) on all **relevant** co-formulants present ≥ 1 % for products for which a negative classification has been proposed by the applicant when applying the calculation method (no data/ unknown is **not** acceptable). The reasons for a different approach for this endpoint compared to the other acute endpoints are multiple: skin sensitisation is a serious, insidious and delayed effect; it has a very low GCL (generic concentration limit for mixtures) of 1 % under the CLP Regulation; it is not an additive endpoint; and it has a significant impact on the risk assessment/management of the product. If no component is a known skin sensitiser, or if known skin sensitisers are present at a level lower than the relevant concentration limits, it can be presumed that the PPP will not be a skin sensitiser.

72. If skin sensitisation information is not available on all relevant components, another option is to use a pre-existing (commissioned before December 2021) *in vivo* skin sensitisation study generated on the PPP of interest– (c).

73. If such pre-existing i*n vivo* skin sensitisation study on the PPP is not available, another option is the bridging approach, in which an applicant will rely upon skin sensitisation data generated in a pre-existing (commissioned before December 2021) study on vertebrate animals conducted with another closely-related formulation – (d). To be considered acceptable by HSE, the *in vivo* study on a related formulation needs to have been commissioned before at latest 6 months after the publication of the OECD DASS Guideline 497 in June 2021 (see paragraph 74 below). For *in vivo* studies commissioned after December 2021, HSE will consider only <u>previously accepted</u> studies submitted for other applications.

74. If bridging is not possible, the *hazard identification module* (“2 out of 3”) or one of the *hazard potency modules* (ITSv1 and ITSv2) within the recently adopted and published OECD DASS 497 guideline should be applied to the PPP – (e). In June 2021, the OECD has published guideline 497 for Defined Approaches to Skin Sensitisation (DASS). The DASS include three approaches: a hazard identification module based on

validated *in vitro/in chemico* tests (OECD TG 442C, 442D, 442E) and two hazard characterisation/potency modules (ITSv1 and ITSv2) based on a combination of *in vitro/in chemico* tests and QSAR predictions. Although QSAR models cannot be applied to mixtures (i.e. mixtures are out of domain), in some instances, conclusive potency scores can still be obtained from the two *in vitro/in chemico* tests of ITSv1 and ITSv2 through a data interpretation procedure specified in the guideline. It is noted that a fully quantitative skin sensitisation risk assessment could not be performed even when applying the potency modules of the DASS, as the potency scores obtained are arbitrary numbers of no biological value.

75. If the DASS cannot be applied to the PPP to be authorised (e.g. PPP is out of domain of the *in vitro/in chemico* tests), produce inconclusive results or a fully quantitative risk assessment is required to refine the evaluation, another approach is to use the results of a “new” (post-December 2021) skin sensitisation study on the PPP in experimental animals - (f). Overall, a “new” *in vivo* skin sensitisation study (preferably a Local Lymph Node Assay -LLNA) commissioned at the latest 6 months after the publication of the OECD DASS 497 guideline in June 2021 will only be accepted by HSE when 1) no component triggers classification of the product; 2) skin sensitisation information is not available on all co-formulants; 3) a pre-existing (pre-December 2021) *in vivo* skin sensitisation study is not available; 4) bridging is not possible; 5) the DASS is either not feasible or produces inconclusive results; or 6) or a fully quantitative risk assessment is required to refine the evaluation.

76. New or pre-existing *in vivo* skin sensitisation studies on PPPs providing positive results and leading to classification of the product (especially where other approaches appear to indicate no skin sensitisation potential) will also be accepted by HSE as they constitute “adverse” data under Art 56 of Regulation (EC) No 1107/2009.

77. A combination of different alternative approaches (as described at step (b), (c) and (d)) will also be accepted by HSE using a Weight-of-Evidence analysis.

78. As with any guidance, scientifically justified exceptions to the sequence of approaches described above will be carefully and thoroughly considered by HSE, on a case-by-case basis.

Dermal absorption

79. A study on dermal absorption of the PPP to be authorised shall be conducted when dermal exposure is a significant exposure route, and the use of default absorption values does not provide an estimate that risk is acceptable.

80. If a test is required, data from dermal absorption studies, preferably on human skin in an *in vitro* test system, shall be reported. Studies should be performed on representative PPPs at both in-use dilutions (if applicable) and concentrates.

81. Therefore, a tiered approach should be taken to meet the data requirement for dermal absorption. The following strategy, in order of refinement, is recommended in EFSA's Guidance on Dermal Absorption (2017)[6]:

    a. an initial exposure assessment can be undertaken with default values or data on closely-related products;

    b. *in vitro* studies on human skin;

    c. data on rats (or other experimental animals);

    d. 'triple pack' approach: *in vivo* data in animals (usually rats) are corrected for the ratio of absorption between rats and humans *in vitro*.

82. The default values to be applied to different formulation categories in the absence of experimental data are given in section 6.1 of the EFSA 2017 guidance.

83. When the applicant for authorisation of a PPP wishes to rely on data from a closely-related formulation, the conditions outlined in section 6.2 of the EFSA 2017 guidance should be met.

84. In the context of 'information on experimental animals', HSE will accept such in vivo dermal absorption studies only for applications submitted before 25 August 2018 (implementation date of the EFSA 2017 guidance) which have been performed in line with the EFSA 2012 guidance[7]. This is because the older EFSA dermal absorption guidance permits the conduct of in vivo studies, using a tiered approach. However, for applications submitted after 25 August 2018, HSE will accept only *existing* (i.e. performed before August 2018) in vivo studies, but not *new* animal assays. If the use of the in vitro dermal absorption study leads to unacceptable risks and no authorisation, then, on a case-by-case basis, HSE will explore with the applicant other possible

[6] Buist *et al*. (2017). Guidance on dermal absorption. EFSA Journal 15(6): 4873.
[7] EFSA (2012). Microsoft Word - Guidance on Dermal Absorption status 23 4 2012 _2_.doc (europa.eu)

refinement options, including the conduct of an in vivo study if necessary and justified. It is anticipated that this would be a rare occurrence.

85. Taking together the information requirements of Regulation (EC) No 284/2013 and the EFSA guidance on dermal absorption (2017), a new study on experimental animals to investigate dermal absorption should not normally be conducted. In the exceptional circumstance that a new animal study is considered to be necessary because an *in vitro* assay is not technically feasible, the stipulations of Articles 33 and 62 of Regulation (EC) No 1107/2009 must be borne in mind. A tiered approach, as described in points (a) to (d) above, should always be considered first.

APPENDIX

Examples of acceptable and unacceptable justifications for the submission or non-submission of data, including tests on vertebrate animals

Please note that all justifications will be assessed case-by-case.

An example of a justification for the submission of an acute oral toxicity test on vertebrate animals that will not be accepted by HSE is:

> ***"The test was available, having been done to meet the requirements of another regulatory regime."***

An example of an acceptable justification for the use of data on the acute *oral* toxicity of a PPP to meet the information requirement for acute dermal toxicity might be:

> ***"An acute oral toxicity study with the PPP is available, with an LD50 > 2000 mg/kg bw. On this basis, and considering the components of the PPP, we predict that the acute dermal toxicity of the PPP would not be greater than its acute oral toxicity and therefore would also be > 2000 mg/kg bw. Therefore, we propose that the conduct of an acute dermal toxicity study is not justified on animal welfare or scientific grounds."***

An acceptable justification for doing an acute dermal toxicity study on the full PPP could be:

> ***"The active substance is known to have significant acute dermal toxicity and the PPP contains a significant percentage of a co-formulant that is known to facilitate dermal absorption; hence it was considered judicious to examine if the formulation produced an exacerbation of the acute dermal toxicity of the active substance."***

An example of an unacceptable justification for doing an acute dermal toxicity study could be:

> ***"The test was done to examine the reliability of the prediction arising from the calculation method."***

An example of an acceptable justification for the non-submission of data on the acute inhalation toxicity of a PPP might be:

> ***"The product for which authorisation is being sought will not be sprayed as it is supplied. Before being sprayed, the product is intended to be diluted 1 in 200 with water. Therefore, an acute inhalation test on the product as supplied would not be representative of the product as used, and so we consider that the criterion 'to be applied by spraying' in the data requirements is not met by the concentrated product. Water is the major component of the dilution (99.5 %); consequently, it can be predicted that the acute inhalation toxicity of the diluted product would be very low. On this basis, we conclude that an acute inhalation toxicity test on either the product as supplied (for which we are seeking authorisation) or the in-use spray dilution is not justified on either scientific or animal-welfare grounds. The product does not meet any of the other criteria listed in section 7.1.3 of Regulation (EC) 284/2013.***

An example of an acceptable justification for the non-submission of test data on skin sensitisation might be:

> ***"The active substance in the PPP to be authorised has a harmonised classification for skin sensitisation Category 1. The active substance is present in the PPP at a concentration of 20%. Therefore, since a known sensitiser is present in the PPP at a concentration that is greater than the generic concentration limit of 1% for Category 1 sensitisers, we propose that test data on the PPP to be authorised does not need to be provided."***

Another example of an acceptable justification for the non-submission of test data on skin sensitisation might be:

> ***"We have not conducted a skin sensitisation test with the PPP to be authorised. However, reliable data on the skin sensitisation potential of all the components of the PPP was available from tests that were compliant with the relevant OECD test guidelines (LLNA or guinea-pig tests). All the components were negative when tested at high-enough concentrations, as defined in the test guidelines, to give meaningful negative results. On this basis, we conclude that the PPP is unlikely to be a skin sensitiser, and we propose that it is not necessary to test the PPP itself."***

Version history

Date	*Changes*
Sept 2021	Version 1 – first time document has been published.
March 2022	Version 2 – minor changes to skin sensitisation chapter

Further information

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