

Groundwater metabolites – guidance on efficacy aspects

In February 2003 an EC guidance document on the assessment of the relevance of metabolites in groundwater of substances regulated under Directive 91/414 was published. A copy of this guidance document is available from the CRD website:

http://www.pesticides.gov.uk/psd_pdfs/registration_guides/data_reqs_handbook/Supporting/RelevantMetabolites.pdf

This guideline provides further advice on the efficacy aspects of determining relevant groundwater metabolites, and should be read alongside the EC document.

What is a relevant metabolite?

A metabolite is 'relevant' if there is reason to assume that it has comparable intrinsic properties to the active substance in terms of its biological activity (defined as 50% or more of the activity of the active substance against the target organism) or if it has certain toxicological properties that are considered severe and unacceptable. If a metabolite has **any** of these characteristics it is treated like the parent active substance. Where such a metabolite exceeds the maximum permissible concentration (0.1 µg/l) for groundwater, a non-inclusion decision would be triggered at Community level for the active substance or a non-authorisation decision would be triggered at national level for specific uses of products containing that substance.

Efficacy input

During the evaluation of an active substance, the environmental fate assessment may identify representative scenarios that predict contamination of groundwater by the active substance or individual metabolite(s) at > 0.1 µg/l. Any metabolite, which might be expected to be at >0.1 µg/l in groundwater, is further assessed. This is essentially a 3-stage assessment involving (i) biological activity screening, (ii) genotoxicity hazard screening, and (iii) toxicity hazard screening. Any metabolite that does not pass all three stages is 'relevant' and thus unacceptable at groundwater contamination levels > 0.1 µg/l.

Passing the three stages does not imply non-relevance – it simply means that further assessment is required. Please note that there may be substances which may give reason for ecotoxicological concern at levels even below the default limit value provided for drinking water. This aspect is considered in the context of the ecotoxicological assessment of the active substance, which is outside the scope of this document.

Efficacy input will vary depending on whether the evaluation is part of a review of an existing active substance or is an application for a new active substance.

New Active Substances

The CRD efficacy evaluation will include a new section (under 'Undesirable side-effects') entitled 'Effect of metabolites in groundwater'. This section will include consideration of both the biological activity of any metabolites and the potential risk to crops. Applicants may also choose to include a similar section in their Biological Assessment Dossier (which ever format is chosen), particularly where their case is based on data previously summarised in the BAD (e.g. under preliminary data).

Determination of Biological Activity

The purpose of the first stage of the assessment process is to identify metabolites which have a comparable target activity to the parent active ingredient and to deal with cases where the parent molecule is a precursor of the active substance. For activity to be 'comparable' the metabolite must have 50% or more of the activity of the active substance.

Metabolites highlighted by the CRD evaluation (or applicants) in groundwater at levels > 0.1 µg/l. will be assessed by efficacy specialists to determine whether they have comparable biological activity to the parent molecule.

An assessment of biological activity may be addressed by the applicant in a number of ways.

- Structure-activity relationships. These may be used for extrapolating between the parent molecule (i.e. usually the active substance) and its metabolites. For example if the parent molecule has a particular chemical grouping that confers pesticidal activity and this is absent from the metabolite then a case may be appropriate to demonstrate that the metabolite is not pesticidally active.
- Preliminary screening data with the parent substance and its metabolites. Screens may have been undertaken for a number of reasons including the evaluation of pesticidal activity, crop safety or any potential risks to following crops.
 - For fungicides and insecticides the biological activities of the parent and its metabolites may be addressed in screening experiments against a range of organisms close to the target, including the target organism.
 - For herbicides generally pre- and post-emergence selectivity across a range of representative crop species is ascertained from glasshouse or semi-field testing, with NOEL or ED₁₀ values provided for a range of species. The biological activities of any metabolites may be assessed similarly.
- In addition metabolite screening data may be provided under the ecotoxicological sections. For example the parent molecule and its metabolites may be tested against a range of non-target organisms e.g. plants or arthropods.

All of these sources of information may be used for addressing the biological activity of metabolites. 'Relevant' metabolites are those with a comparable or higher biological activity than the parent. The case or data submitted must demonstrate that the biological activity of a metabolite is clearly less than 50% of the activity of the parent molecule otherwise its biological activity is considered to be 'comparable'. **It is essential that data on both the biological activity of the parent and its metabolites are provided to enable this comparison.**

Risk to crops

As part of the efficacy risk assessment the **risk to crops** from any metabolites should be assessed. This should be based on whether the metabolites have any significant biological activity at the environmental concentrations predicted. Field trials designed to assess the phytotoxicity of the active substance may provide useful supporting evidence on whether the degradation products have any phytotoxic effect on the target crop. However, the highest risk of damage to crops from **metabolites in groundwater** is likely to be when metabolites with herbicidal activity are present in groundwater and are applied as irrigation water to crops more sensitive than the target crop.

Reviews of Existing Active Substances

Under the review of existing active substances there is no specific consideration of efficacy until the re-registration process. During the review prior to Annex I listing, efficacy specialists may still be contacted to assess the biological activity of any metabolites appear in groundwater at > 0.1 ug/l. In these cases the risk to crops is not generally considered.

Determination of Biological Activity

Although efficacy trials may not be available for the review of an active substance much of the information cited in the section on New Active Substances may be relevant. As examples the following may provide useful information on whether metabolites are 'comparable' in activity to the parent substance;

- Structure-activity relationships may be described within the sections on the toxicity of the active substance and its metabolites. This can provide an insight into the likelihood of pesticidal activity.
- Biological screening data may be submitted by the applicant to address the activity of the metabolites against target (and related) species.
- Ecotoxicology data may be submitted to address the effect of the metabolites on non-target species.

All of these sources of information can be used by the Efficacy evaluator to assess the activity of the metabolites. However, as for new active substances the issue is whether the biological activity of a metabolite is clearly less than 50% of the activity of the parent molecule. **It is essential to be able to make this comparison that data on both the biological activity of the parent and its metabolites are provided.**

6. Examples

Example 1: New Herbicide

The product in this example is a pre and post emergence herbicide. The Environmental Fate evaluation has highlighted one metabolite as likely to appear in groundwater at > 0.1 µg/l.

Data provided on succeeding crops has indicated that the crops most sensitive to the parent molecule pre-emergence are sugar beet and oilseed rape with a NOEL of 0.35 g as/ha for both crops. The NOEL for the metabolite is 15.5 and 6.75 g as/ha for sugar beet and oilseed rape respectively.

The most sensitive crop post-emergence was lettuce with a glasshouse NOEL of 0.006 g as/ha (parent), beans and oilseed rape produced damage from doses of 0.2 g as/ha (parent). There was no specific crop data for the metabolite but ecotoxicological studies showed that the metabolite is at least 128 times less active against *Lemna* when used post-emergence.

In terms of the biological activity of the metabolite the *Lemna* data provided would be sufficient to demonstrate that it has less than 50% of the activity of the parent and thus its activity is not 'comparable'.

In this case the assessment has also considered the direct risk to crops from the metabolite. In some cases the risk to crops from a metabolite in irrigation water will need to be considered:

An example of how to approach this follows:

1. Calculate the possible rate of application

An average irrigation event is around 25 mm. This is equivalent to 250,000 l water/ha. Say for example the metabolite referred to previously appears in groundwater at 0.15 µg/l. This would mean that 0.038 g/ha of the metabolite would be applied per irrigation event.

2. Determine whether this rate is likely to cause crop damage.

In this example the ecotoxicology data have indicated that lettuce is the most sensitive crop post-emergence with a NOEL of 0.006 g as/ha for the parent. Ecotoxicology data have also determined that the metabolite is 128 less active than the parent. This would mean an approximate sensitivity of 0.768 g as/ha for the metabolite on lettuce. To achieve this dose would require 20.2 irrigation events.

So although the metabolite in this example is herbicidally active, it is less active than the parent, by over 10 times pre-emergence and 100 times post emergence. If the metabolite accumulated at the predicted levels irrigation would need to be applied to a single crop around 20 times, or more in quick succession, for any affect to be seen and this volume is most unlikely to be applied.

So in this example the risk to crops from irrigation water containing the metabolite would be considered to be very low and no further data would be required.

Example 2: Review of an existing insecticide

The Environmental Fate assessment of this application has identified that five metabolites of insecticide A (IV, IX, XIII, XV and XIX) may leach to groundwater to concentrations above 0.1 µg /l. Efficacy have been asked to consider the biological activity of these metabolites.

Insecticide A belongs to a well defined group of active substances. The notifier has examined structure activity relationships and has made the case that none of the metabolites, except metabolite II, have the chemical grouping that infers activity, and thus do not have the potential for biological activity. The notifier has referenced toxicology and ecotoxicology studies to demonstrate that many of the metabolites have biological activity significantly less than Insecticide A itself. These aspects will have been considered elsewhere in the evaluation and may have demonstrated lack of activity sufficiently.

A screening study has also been submitted to assess the insecticidal activity of some of the Insecticide A metabolites. This comprised insecticide screening data where two types of screen were conducted, primary screens (single treatment) or super screens (multiple doses). The test substances were metabolites II, III, IV, VIII, IX, X, and XI. This study did not include metabolites XIII, XV and XIX from the list highlighted by F&B.

Test species were the Southern armyworm (*Spodoptera eridania*), Southern corn rootworm (*Diabrotica undecimpunctata howardi*) and in some cases (metabolites III, IV, X and XI) the cotton aphid (*Aphis gossypii*). Aphids would be expected to be the most appropriate test species for screening trials as these are the primary target pest species. With this the case the value of the data on Southern armyworm and Southern corn rootworm is of limited value. The notifier has made the case that only the active substance and its primary metabolite (II) have any biological activity due to their similar chemical structure. In the screening study, however, metabolite II had no activity on either the Southern armyworm or the Southern corn rootworm, and its activity on cotton aphid was not determined. The parent molecule was not included at all as a test substance.

The screening data provided did not include the parent molecule as a standard reference against which to compare the activity of the metabolites tested. In addition the metabolite most likely to have biological activity i.e. metabolite II had no activity on the two test species selected. Thus it is impossible to compare the activity of metabolites IV and IX with either the parent or indeed metabolite II. Metabolites XIII, XV and XIX were not included as test substances at all. So this study is of limited value in determining whether any of the metabolites highlighted are relevant or not. In a case such as this CRD may have to contact the applicant for further details.

In terms of herbicidal activity there is no evidence, from ecotoxicology studies, of the parent molecule having any effect on plant species. It is reasonable to assume that this will also be true for the metabolites although no specific study was submitted.

The examples presented here hopefully provide some information on how to address both the biological activity and the risk to crops from metabolites in groundwater. For review substances in particular the assessment will need to be made on **all** available information and the extent of this will vary.

Summary

The assessment of relevant groundwater metabolites is not part of the 91/414 Annex III Efficacy data requirements. However, efficacy specialists do get involved in assessing the biological activity of metabolites, and hence identifying those that are considered relevant and may require further assessment. Various methods can be used by the applicant, including reasoned cases based on structure/activity relationships and use of screening data. The latter includes the standard pre- and post-emergence screening tests, and data generated during the initial development in identifying candidate actives and determining their biological activity. The applicant may choose to include such information under a separate heading in the Biological Assessment Dossier, or in relevant parts of the Environmental Fate submission.