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MANUAL FOR SUMMARISING AND EVALUATING THE ENVIRONMENTAL ASPECTS OF PESTICIDES

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This research was carried out on behalf and for the account of the Directorate General for Environmental Protection, Directorate of Chemicals, Safety and Radiation Protection and Directorate for Drinking Water, Water and Agriculture, in the framework of project 679101 "Quality objectives and risks".

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PREFACE AND ACKNOWLEDGEMENTS

This Manual gives guidance in evaluating the environmental aspects of pesticides. It does not give guidance in assessing the risks of a pesticide for the environment. This will be dealt with in an update of this Manual. The Manual is primarily meant for reviewers of the Department of Evaluating Environmental Aspects of Pesticides (BBM) of the Toxicology Advisory Centre (ACT).

This first Manual does not cover all relevant tests. For example, instructions on analysis in drinking water, and on field studies are not available yet. The current discussion on how to evaluate potential side-effects of new pesticides, prior to their introduction on the market, is dynamic on both the national and the international level. It is the intention of the authors to update this Manual once a year. Therefore critical readers are kindly requested to send their remarks to the authors.[1]

The report also reflects some of the discussions within ACT on the use of quality indicators. Especially with respect to this, the report presents a state of the art, and therefore the authors hope that it will contribute to further discussions.

Several people have contributed to this report. The members of the Advisory Toxicology Group II have reviewed the whole report, and their specialisms in particular:

Dr. R.A. Baumann	(LOC/RIVM, organic chemistry)
Ir. W.W.M. Brouwer	(Plant Protection Service, agricultural aspects and behaviour in soil)
Drs. H. Canton	(ECO/RIVM, aquatic ecotoxicology)
Dr. M. van den Hoop	(LAC/RIVM, inorganic chemistry)
Ir. A.M.A. v.d. Linden	(LBG/RIVM, behaviour in soil)
Drs. R. Luttik	(ACT/RIVM, avian ecotoxicology)
Dr. Ir. J. Notenboom	(ECO/RIVM, ecotoxicology and behaviour in water)
Dr. M. Schrap	(RIZA, aquatic ecotoxicology).

In addition the following specialists have been consulted:

Dr. P. van Beelen	(ECO/RIVM, terrestrial ecotoxicology)
Ing. R. Faasen	(RIZA, aquatic ecotoxicology and behaviour in water)
Dr. P. Oomen	(Plant Protection Service, beneficial insects and mites)
Dr. Ir. W. Peynenburg	(ECO/RIVM, behaviour in water)
Dr. J. Struijs	(ECO/RIVM, behaviour in water)
Dr. J.W. Tas	(ACT/RIVM, bioconcentration in waterorganisms)
Ir. M. Vaal	(ECO/RIVM, aquatic ecotoxicology)

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AUTHORS AND RECORD HISTORY

Drs. H. Tibosch and L. Wijkhuizen-Maślankiewicz started the writing of this Manual in the autumn of 1993. Their work was taken over by drs. H. Mensink and drs. M. Montforts. Ir. J. Linders has been supervising as the assessment expert of ACT.

The Manual has been adopted by the Toxicology Advisory Group II at the meetings of January 10; January 17; March 27; and April 3, 1995.

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SUMMARY

This Manual gives guidance in summarising and evaluating the environmental aspects of agricultural and non-agricultural pesticides. Actually this is the part of the pesticide registration process in the Netherlands preceding the risk assessment. The requirements of the Dutch government for admission of pesticides to the market are included in the Manual. As they are not reported exhaustively, it is advised to consult the CTB Application Form, if felt necessary.

This Manual focusses primarily on instructions with respect to studies on behaviour in water and soil, ecotoxicity for aquatic and terrestrial organisms, and bioconcentration for aquatic organisms. The instructions cover both the intrinsic reliability of test reports provided by the industry, and the summarised description of these reports in an Advisory Report of RIVM. The latter is prepared by the Department of Evaluating Environmental Aspects of Pesticides on behalf of the Dutch Board for the Authorisation of Pesticides (CTB). This Board has the responsibility for admitting or refusing pesticides on the Dutch market in view of the Pesticides Act.

A distinction has been made between the reliability and usefulness of tests. The first refer to intrinsic aspects of a test —methodology, description—, the latter to the use of such a test for a particular purpose —e.g. risk assessment. In this Manual the emphasis is on the reliability, and it offers a check-list per environmental item to discriminate between reliable, less reliable and not reliable tests. These check-lists are presented as so-called summary tables, and they are the core of the Manual.

This Manual does not claim to be complete. Many environmental issues are currently in discussion both nationally and internationally. Therefore this Manual represents the state of art. It will be updated regularly to cope with these national and international developments. The risk assessment is also not included and will be dealt with in a follow-up.

SAMENVATTING

Deze handleiding geeft richtlijnen voor het samenvatten en beoordelen van diverse milieu-aspecten van landbouwkundige en niet-landbouwkundige bestrijdingsmiddelen. Het betreft dat deel van de toelatingsprocedure in Nederland dat de risico-schatting voorafgaat. De eisen van de Nederlandse overheid voor de toelating van bestrijdingsmiddelen zijn opgenomen in deze handleiding. Omdat ze niet integraal zijn opgenomen wordt het aangeraden het CTB Aanvraagformulier te raadplegen, indien gewenst.

De handleiding richt zich vooral op richtlijnen wat betreft het gedrag in water en bodem, de ecotoxiciteit voor aquatische en terrestrische organismen, en de bioconcentratie in aquatische organismen. De richtlijnen hebben betrekking op zowel de intrinsieke betrouwbaarheid van de door de fabrikant aangeleverde testrapporten als op de beknopte beschrijving van deze rapporten in een zogeheten RIVM Adviesrapport. Dit laatste wordt opgesteld door de afdeling Beoordeling Bestrijdingsmiddelen Milieu (BBM) ten behoeve van het College voor de Toelating van Bestrijdingsmiddelen (CTB). Dit College heeft de verantwoordelijkheid voor het al dan niet toelaten van bestrijdingsmiddelen op de Nederlandse markt in het kader van de Bestrijdingsmiddelenwet.

Er is onderscheid gemaakt tussen de betrouwbaarheid en de bruikbaarheid van testen. Het eerste verwijst naar de intrinsieke aspecten van een test —methodologie, rapportage—, het laatste verwijst naar het nut van een dergelijke test voor een specifiek doel —bijvoorbeeld risico-schatting. De nadruk ligt in deze handleiding op de betrouwbaarheid, en het geeft een check-list per milieu-aspect om op grond hiervan onderscheid te kunnen maken tussen betrouwbare, minder betrouwbare en onbetrouwbare testen. Deze check-listen worden weergegeven als zogenoemde resumé-tabellen; zij vormen de kern van de handleiding.

De handleiding is niet compleet. Talrijke milieu-aspecten zijn momenteel zowel nationaal als internationaal in discussie. Daarom is de handleiding een reflectie van de huidige stand van zaken. Ze zal regelmatig worden bijgesteld om de nationale en internationale ontwikkelingen bij te kunnen houden. De risico-schatting is eveneens niet opgenomen. Dit zal gebeuren in een volgende versie.

1. INTRODUCTION

Status of the Manual

This Manual gives guidance in summarising and evaluating environmental aspects of pesticides. In course of time, several criteria have been developed about what to summarise and how. No integrated report on this information has been available until now. This Manual tries to meet this demand. It comprises both agricultural and non-agricultural pesticides. In this Manual it is reflected that the expertise on summarising and evaluating agricultural pesticides is larger than on non-agricultural pesticides (disinfectants, industrial biocides, preservatives, household products, wood preservatives and protectors, antifoulings).

The Manual is one of the quality system documents of ACT/RIVM (ACT/H/007). The Manual claims to function as a tool with respect to:

- reproducibility of the evaluation process;
- traceability of the evaluation process;
- proper documentation.

The Manual has no legality with respect to the admission of pesticides in The Netherlands. No rights can be founded on the contents of the Manual.

Advisory Report

An Advisory Report (AR) is a document that is prepared by the Toxicology Advisory Centre, on behalf of the Board for the Authorisation of Pesticides (CTB). All the information about a pesticide that has been supplied to the CTB (i.e. a dossier), is summarised in the AR, as to make an evaluation for the requested use of the pesticide. When several companies notify the same active ingredient, all dossiers will be summarised and evaluated in one AR. ARs are prepared per active ingredient and they consist of Summaries[2] of the test reports, and an RIVM Conclusion. Only the Summaries has been dealt with in this Manual.

The department BBM —of Evaluating Environmental Aspects of Pesticides— evaluates the environmental aspects. The department BAV —of Evaluating Public Health Aspects of Pesticides— evaluates the public health aspects. In this Manual, when referring to the AR, only the environmental part is considered.

The summarising and evaluating is carried out with the assumption that the substances are used in accordance with the labels (usage instructions).

Summary

Various test reports have to be submitted by the manufacturer in order to get access to the market. A Summary includes in a concise way the most relevant information of a particular test.

The information in the test reports is recorded in a database system. Currently a relational database-system, called TOXIS, is used. In TOXIS there are Fixed Fields and Free Text Fields. The first contain the relevant test conditions and main results.

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[2] The structure and contents of an AR are denoted with specific terms. Specific terms, e.g. Header and Free Text, are written with a capital. All terms with a capital are included in the glossary (Appendix 9).

These items form the Header of the summarised test (see Figure 1.1). The latter —Free Text Field— contains the Description of the test. The Description itself contains successively:

1. the Methodology (as far as not reported in the Header);
2. the Results (as presented by the author);
3. the Remarks (critical comments on the test, made by the reviewer).

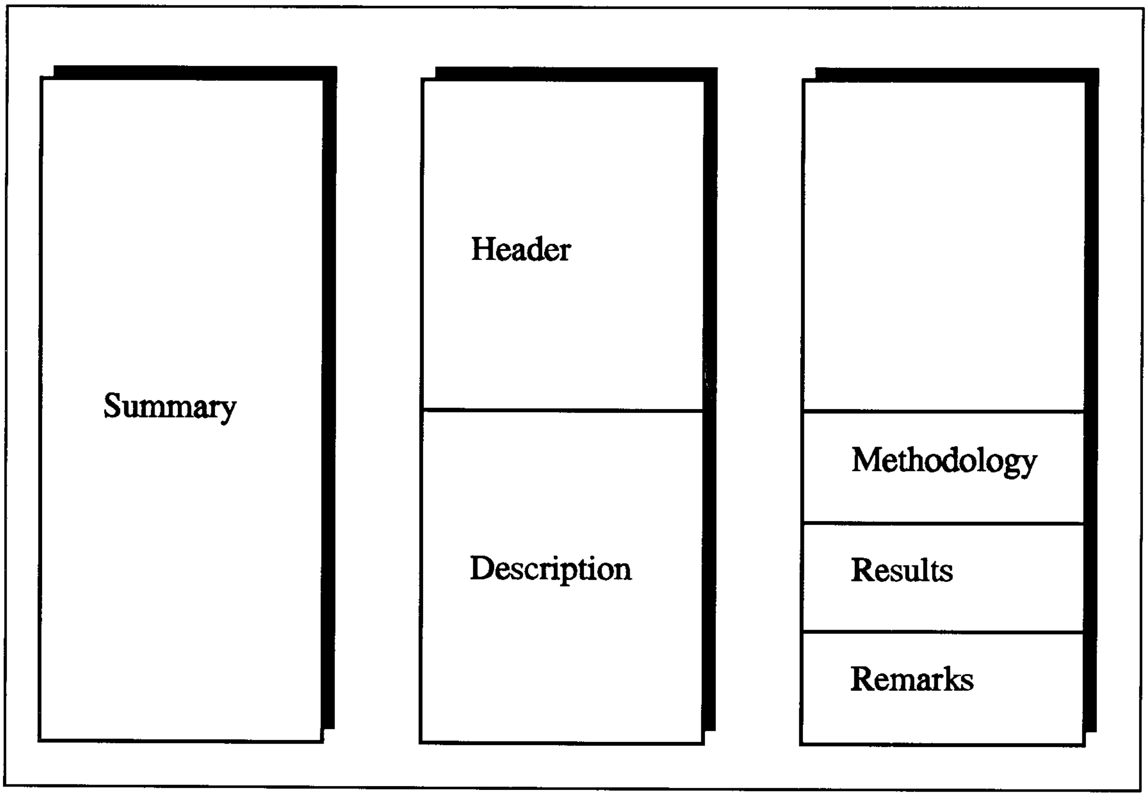

Figure 1.1. The structure of a Summary.

RIVM Conclusion

An RIVM Conclusion contains the major steps of the risk assessment: effect assessment (incl. dose-response assessment), exposure assessment, and risk assessment. The RIVM Conclusions are written conform the Final Environmental File (FEF) as presented in Appendix 8. The RIVM Conclusions are written in English and in Dutch.

Since March 1994, the risk assessment of pesticides is carried out with USES 1.0 (Uniform System for the Evaluation of Substances), a decision-support system of governmental ministries and institutes (RIVM, DGM, & WVC[3], 1994). USES 1.0 includes the risk assessment of both agricultural and non-agricultural pesticides. In ARs finished before March 1994, only two models have been used for the risk assessment, one for calculating the expected leaching and accumulation in the soil,

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[3] In August 1994 the name of the Ministry of Welzijn, Volksgezondheid en Cultuur (WVC; Welfare, Public Health and Culture) has been changed into Volksgezondheid, Welzijn en Sport (Health, Welfare and Sports).

and the other for calculating the hazard for aquatic organisms. These models are PESTLA 1.1 (Boesten & Van der Linden, 1991) and SLOOT.BOX (Linders et al., 1990), respectively. PESTLA 1.1 and SLOOT.BOX are now integrated in USES 1.0.

The hazard quotients calculated by USES 1.0, are classified in accordance with the 'CTB decision trees' (Van Vliet, 1992; CTB, 1993; Luttik et al., 1993).

Additional questions

The RIVM Conclusion is concluded with Additional Questions (AQs). AQs are posed when the delivered information is considered insufficient. For AQs see Appendix 6.

Final Environmental File

The Dutch RIVM Conclusions (in TOXIS) are edited to give the Final Environmental File (FEF; Dutch: Milieufiche) (free text, e.g. in WordPerfect or Word). See Appendix 8 for the stencil of the English FEF.

Instructions

The instructions comprise both official Guidelines and the BBM directives on summarising and evaluating test reports. The CTB application form and the OECD Guidelines are the starting points for the instructions. According to the CTB application form the desired tests may be performed in accordance with various (inter)national Guidelines like those of the OECD. The BBM directives contain decisions on items the official Guidelines do not handle, directives on (re)calculating test results, and directives on the way of reporting various information.

The instructions are of a technical nature: it is stated which information has to be dealt with, and in which way, and how to apply this information in the models and decision schemes without an extensive explanation of all the rationales. The latter can be found in CTB (1993); Brouwer et al. (1993); Canton et al. (1991); Linders et al. (1994); Luttik et al. (1993); RIVM, VROM & WVC (1994); and the various (inter)national Guidelines.

In this way the instructions function as a checklist for preparing the Summaries and the RIVM Conclusion. It should also be noted that this Manual is not a cookery-book (which would result in uniform ARs): expert judgement remains *crucially* important in the process of evaluating the environmental aspects of pesticides.

Updates

The Manual represents the state of the art. The contents will be updated because of changes in scientific viewpoints, and (inter)national developments on the requirements for the registration of pesticides. These updates will be adopted by the Toxicology Advisory Group (TAG, i.e. a panel of specialists). Once a year these updates are integrated into this Manual and the whole document is revised.

Structure of the Manual

The Manual is built up as follows: Chapter 2 gives the procedural context of an AR; Chapter 3 discusses the Reliability Indicators and sources of information; Chapter 4 gives instructions for summarising and evaluating tests. The appendices contain additional information and a glossary.

In Chapter 4 the sections that deal with a specific environmental compartment or with a specific group of organisms are introduced with a grey box. This box contains the requirements of the CTB with respect to this compartment or group of organisms, as formulated in the application form (Anonymous, 1995).

2. PROCEDURES

The procedural context of the summarising and evaluating is explained in this chapter. Applications concerning the production and sales of pesticides are registered by the CTB, which is the institute in charge of the admission procedure. These applications have to fulfil certain requirements: besides general information about e.g. the use of the pesticide, a complete set of information (a dossier) concerning physico-chemical properties, behaviour in soil and water, and toxicological properties has to be submitted. Not until these requirements are fulfilled, the CTB sends the dossier to ACT/BBM.

The task of ACT/BBM then is to summarise the dossier and to make an evaluation with respect to the environmental aspects. An overview of the procedure is presented in Figure 2.1.

First, the application is registered in the ACT database system TOXIS (see also Chapter 3): the documentalist does the basic registration after which the reviewer registers the data for research and the planning for the Toxicology Advisory Group (TAG, i.e. a panel of specialists). Then the information is summarised and evaluated, and a draft AR is sent to the specialists of several RIVM laboratories:

1. Laboratory of Organic Analytical Chemistry (LOC) or Laboratory of Inorganic Analytical Chemistry (LAC) for reviewing the physico-chemical data and the methods of analysis;
2. Laboratory for Soil and Groundwater Research (LBG) for reviewing the data on behaviour in soil;
3. Laboratory of Ecotoxicology (ECO) for reviewing the data on behaviour in water, and on ecotoxicology.
4. Toxicology Advisory Centre (ACT) for reviewing the data on avian toxicity.

The relevant test reports are also sent to the specialists. After the first draft has been reviewed by the specialists, a second draft is sent to the assessment expert (Dutch: beoordelingsdeskundige) of ACT who has the responsibility for the AR. The final draft then, is sent to the members of the TAG, at least two weeks before the TAG meeting. The final draft, including a proposal for the Additional Questions, will be reviewed in this meeting. Then the AR is finalised. The Dutch RIVM Conclusion is also printed in the form of a Final Environmental File (FEF, Dutch: Milieufiche) for the CTB.

The time for drafting an AR is limited since 01-01-94. Within four months after receiving the dossier for registration from the CTB, the AR should be completed and handed over.

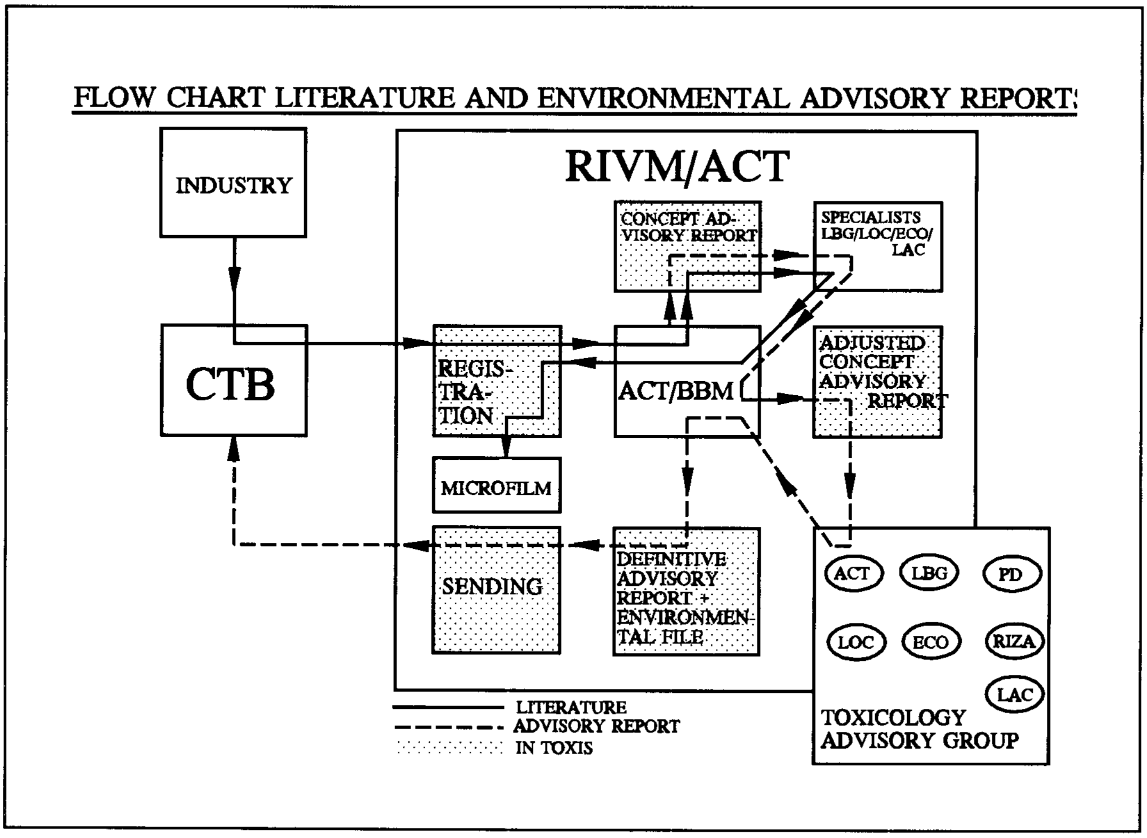

Figure 2.1 Flow chart of literature and Advisory Reports

Details on procedures, responsibilities, and Quality Assurance can be found in the ACT Standard Operating Procedures (SOPs) for the TAG (Dutch: adviesgroep), the reviewer (Dutch: behandelaar), the assessment expert (Dutch: beoordelingsdeskundige), the use of databases, the TOXIS administration, and the registration and archiving of dossiers.

3. INFORMATION AND RELIABILITY

3.1 Information sources

Industry

Industrial information is defined as that information that is supplied by industry and that is announced in an accompanying letter. ACT receives this information from the CTB. This information is checked, summarised and evaluated. Information sent in late is only included if the final draft of an AR has not been finished yet.

Dutch, English, German, and French studies have to be summarised; studies in other languages do not have to be summarised.

Previous (RIVM) evaluations

Previous evaluations (prepared by e.g. RIVM, DGM, or LNV) can be found in the CTB suspension files in case an AR has not been stored in the TOXBANK database (the predecessor of TOXIS). Only the data that fill up gaps in the industrial information, or contradict this information, have to be taken from these old evaluations (before 1984). In the Summary of each study a remark has to be made that the data are derived from a previous evaluation; if these data are used in the RIVM Conclusion this qualification about the source of information is not repeated.

If stored in TOXBANK, the existing AR is always the basis for further summarising and evaluating (in this way it concerns an update). Check with the assessment expert if the update will be recorded in TOXBANK, and if the TOXBANK summaries have to be converted (manually) to TOXIS.

An incomplete description of a test in the existing AR or old evaluation, only has to be completed if the study is considered useful, and if the original source is available.

Other literature

The WHO/IPCS, OESO, RIVM, and other evaluating bodies have made several public evaluations. This information can be included in an AR in case the information provided by the industry is considered as insufficient, i.e. when there is no reliable information available, or when the new information gives cause for a different point of view on the substance.

Information supplied by members of the Toxicology Advisory Group (TAG) is also summarised and evaluated.

Additional information

The following additional information is used for preparing an AR:

- data sheets on functions and uses, supplied by the CTB;
- monitoring data, if directly available;
- documents on the risk assessment procedure ('Handboek Normen en Criteria' (CTB, 1993); ESPE-2 (Luttik et al., 1993); USES 1.0 (RIVM, VROM & WVC, 1994)).

3.2 Reliability of information in TOXIS

All the studies that are summarised and evaluated in an AR, are given a Reliability Index (RI) as a measure for the *reliability*. Indicators is used as the plural form of Index (another option could have been indices), because it is more in accordance with international discussions on the quality of sources (Vermeire, 1992).

Indicators are currently only available with respect to the *reliability* of a test, whereas indicators with respect to *usefulness* and *quality* (for definitions, see below) have to be developed in the near future. The definitions are:

Reliability:	the intrinsic reliability of a test with respect to the methodology and the description. Synonym: *betrouwbaarheid (Dutch).*
Usefulness:	the extent to which a test is appropriate for a particular purpose (e.g. standard setting procedures, risk assessment). Synonyms: relevance, *bruikbaarheid (Dutch).*
Quality:	the degree of excellence of a test as determined by both its reliability and its usefulness. Synonym: *kwaliteit (Dutch).*

From these definitions it follows that reliability + usefulness = quality. There are four Reliability Indicators:

RELIABILITY INDEX (RI)	DEFINITION	DESCRIPTION
1	reliable	the methodology and the description are in accordance with the instructions in this Manual
2	less reliable	the methodology and/or the description are less in accordance with the instructions in this Manual
3	not reliable	the methodology and/or the description are not in accordance with the instructions in this Manual
4	no original data	the RI has not been determined as the original data are not available[4]

The Reliability Index (RI) is found in the Header of every summarised test in an AR. It is an obligatory record for the reviewer. The RI has consequences for the use of the information in the AR:

1. Reliable data are included in the RIVM Conclusion of the AR.
2. Less reliable data may be included in the RIVM Conclusion, if reliable data are lacking.
3. Unreliable data may be included in the RIVM Conclusion when reliable or less

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[4] this can refer to e.g. reviews and old (RIVM) evaluations. If RIVM/ACT has made agreements with other laboratories or institutes with respect to evaluation criteria, evaluations of the latter may be assigned an RI of 1,2, or 3, even in spite of the absence of original data.

reliable data are lacking.

4. This information may be summarised and included in the RIVM Conclusion if other data are lacking.

Although usefulness indicators are not yet developed, there are already instructions on the usefulness of data (see Chapter 4.2). Data considered less useful are in general not used for RIVM Conclusions.

3.3 Reliability of information in TOXBANK and previous (RIVM) evaluations

In the TOXBANK database one will find Indicators at the end of the Description of the Summary, written as (cat. X), with X = 1, 2, or 3. However, the Indicators used here refer to a mixture of reliability and usefulness, so they are Quality Indicators rather than Reliability Indicators. There are three TOXBANK Indicators:

INDEX	RELIABILITY & USEFULNESS
1	reliable, and useful
2	less reliable, and useful
3	not reliable, and useful or not useful

This categorisation has the following consequences for the use of the information in the (TOXBANK) AR:

1. reference is summarised and evaluated, and included in the RIVM Conclusion.
2. reference is summarised and evaluated, but not included in the RIVM Conclusion. Only when the concerning studies of category 1 are lacking, this study is also included in the RIVM Conclusion, with a remark on the value of the delivered data.
3. reference is not included in the AR except when no data are available.

Currently these Indicators are converted to the RIs in TOXIS. For reasons of convenience the TOXBANK Indicators 1 and 2 become Reliability Indicators 1 and 2, whereas TOXBANK Index 3 is converted to the temporary Reliability Index 0. Later on, it will be updated into Reliability Index 3 or 4.

The information in the previous RIVM evaluations —not in TOXBANK— is not categorised with indicators. If necessary, the reviewer should check the original source.

4. INSTRUCTIONS ON SUMMARISING AND EVALUATING THE TEST REPORTS

4.1 General information

Quality instructions

The reviewer should aim at proper documentation and at a consistent way of reporting, which increases the reproducibility of an evaluation. It is also important that it is made clear how a report was summarised, and what (crucial) decisions were made in the evaluation. Conclusions should be traceable. For example, when it is concluded that the calculated concentration in shallow groundwater amounts up to 5 mg/litre, it should be clear on which figures from which tests this value has been based. It should also be clear why comparable figures from other tests could not be used.

All comments of the reviewer can be written in the Remarks section of the Description. It is strongly advised to keep a personal journal to note solutions for new problems, or solutions that deviate from the Manual's instructions. These can be discussed in a TAG meeting (and might be adopted as updated instructions).

Registration of references in TOXIS

All information used is registered in the reference system of TOXIS. The references have to be registered before Summaries are recorded in TOXIS. The reference menu in TOXIS has no Fixed Fields. Therefore a standard sentence is used for the registration of the provided reports:

- (Title). Unpublished report of (name testing company) (date), (Report no.) , Provided by (name applying company), (date of receival).

Application forms are registered as:

- Application form Folidol E-605 stuif, No. 3568-2 TT, May 18, 1979.

Data sheets on functions and uses are registered as:

- Data sheet on functions and uses of ..., Plant Protection Service, Wageningen, (date)

Letters or previous evaluations with relevant information are included in the reference list.

Order of Summaries in an Advisory Report

Preferably studies with active ingredient (in order of purity) first, then with formulations, and then with metabolites. Preferably studies with higher reliability first. The TOXIS database does not provide the functionality of changing the order of tests in the output generated for the reviewer. It is therefore wise to pay attention to the order of studies when recording information in TOXIS.

Structure of a Summary

The main information is recorded in a Header, and the details are placed in a Description (see Figure 1.1). This Description also contains the Results and Remarks. Comments made by the authors of the test report are placed in the Results. The Remarks section is used by the reviewer to report critical comments. In the Remarks section of the Description all reasons for lowering the Reliability Index should be mentioned. It should always be elucidated whether a result is used for an RIVM Conclusion or not, and if the latter is the case, why a result is not used. In the Instructions below a standard sentence is given. Any result that is used for an RIVM Conclusion is quoted in the Remarks:

- .... is used for RIVM Conclusions (in case of lower Reliability Index, add: because better data are not available).

GLP

Test reports should have a GLP certificate. If this GLP statement is lacking, check with the assessment expert.

Rounding off

Results have to be rounded off correctly; only after the last calculation the result is rounded off. Numbers of ≤9999 are written as two-digit figures (= 1 - 9), and numbers >10000 as three-digit figures. Examples: 0.0347 becomes 0.035; 1.645E-8 becomes 1.6E-8; 288 becomes 290; 11253 becomes 11300.

Units

All quantities should be expressed in units of the S.I. system (Système International d'Unités). For recalculating other commonly used units, first consult Appendix 1. Some exceptions are the dosage in [kg/ha] instead of [g/m^2], and the matric suction (or moisture tension) of the soil (pF) in [log(cm)] instead of [Pa].

Statistics

Correct statistical procedures need to be followed. The methods that are suitable for evaluating the test results are dealt with in the coming (sub)sections.

The standard deviation of an arithmetic mean must be based on the standard deviation of the sample (σ_{n-1}). Lotus123 calculates a standard deviation of the population (σ_n). To recalculate σ_n to σ_{n-1} use:

$$\sigma_{n-1} = \sigma_n \times \frac{\sqrt{n}}{\sqrt{n-1}}$$

Linear regression analysis needs to be performed with at least five data points. The next requirement for validity is: $r^2 \geq 0.7$. When $r^2 < 0.7$, the result is not valid.

4.2 Summary tables

The summary tables are the core of the instructions. The summary tables structure the abundance of information and help assigning a Reliability Index (RI; see subsection 3.2) to the tests.

Table 1 is an example. It starts with the 'description' including the relevant test conditions, followed by the 'results' with the relevant test results and it ends by 'pay attention' including those items that should be checked, but need not necessarily be included in the Summary.

In the summary tables you find the requirements which have to be met for a study; the items refer to the *reliability* of a test. Items that refer to the *usefulness* rather than to the reliability are given in the footnotes of the table. It is, however, felt by the authors that one may dispute whether certain test items fall within reliability or usefulness. One may e.g. argue whether the item on the λ of the light source in a photolysis test in water (see Table 8), implies that a test with λ <290 nm is less reliable or that such a test is less useful as the λ does not reflect natural conditions. In this way the summary tables keep on fostering discussions. The tables should therefore not be seen as too compelling.

If items reported are less or not in accordance with the summary tables, the reliability of a study is expected to decrease. In the column with the heading 'Reliability lower?' (see Table 1) this is indicated by a Y(es) or a E(xpert judgement):

Y. Y(es) indicates that solely based on not fulfilling this requirement for this item, the reliability of the entire study is expected to decrease. This can be reflected in assigning an RI of 2 to a test, or even assigning an RI of 3. It is up to expert judgement in the latter case, to decide how many "Y"-items are required for assigning an RI of 3 to a particular test.

E. E(xpert judgement), indicates that no clear guidance can be given. The reviewer can consult a specialist.

It should always be stated clearly in a Summary under Remarks why a certain RI has been assigned, so that this can be verified.

Some standard sentences have been constructed to increase the conformity. These are listed in the texts that accompany the summary tables. They are provided with the symbol ♦ .

Table 1: Example of a summary table

	Items	**Notes**	**Reliability lower?**
Description	**These items should always be included in the test description in a Summary.**	These notes explain the requirements which have to be met for a reliable test (i.e. with an adequate methodology and description). If items in a study deviate from these requirements, check in the next column ("reliability lower?") whether the reliability with respect to that particular item may decrease. ☆ Y(es) ➪ ➪ ➪ ➪ ➪ ➪ ➪ ➪ ➪ ➪ This note indicates that the reliability can be considered to decrease.	Y
	These items should only be included, if a test is not performed according to a Guideline.	☆ E(xpert judgement) ➪ ➪ ➪ ➪ ➪ This note indicates that no clear guidance can be given here. The assignment of an RI is up to the reviewer.	E
Results	**These results should always be included, under Results.**	X in this column indicates a cut-off value. When there is no concensus, expert judgement should be used to establish one. Cut-off values that are used in the Dutch effect and risk assessment are given here between square brackets (e.g. the application rate should not exceed X times the recommended rate [X=2]).	
Pay atten.	The items here should not necessarily be included into a Summary, but should be checked. These items —if deviating from the requirements— can be included under Remarks.		

4.3 General information about the compound and metabolites

The general information about the compound comprises information on identity, names, detection, analysis, use, behaviour, and occurrence of chemicals, substances, and formulations. The "Handleiding TOXIS" (Aalbers et al., 1994) provides instructions on how to record this information in TOXIS.

Names, substances and formulations

All trade names (T), chemical names (C) (standard: IUPAC), and other names (O) of the active ingredient(s) (a.i.) are recorded under Chemical Identity. Industrial codes may be used instead of the complete chemical names if that is the only information the industry has provided. The use of trivial names is encouraged. Names of formulations are *not* recorded under Chemical Identity

The various substances are recorded under Substances. Preferably the following order is used:
1. analytically pure;
2. technical product;
3. purity unknown.

Radioactive substances should be recorded as mentioned above. The position of the label should be described in the Description.

All formulations that are used in tests have to be recorded under Formulations.

Physico-Chemical properties

The next physico-chemical properties are at least required for the risk assessment with USES 1.0, and for the interpretation of test results: log K_{ow}, solubility in water, pK_a, and vapour pressure.

The dimensionless Henry's Law Constant H (see the formula below) has to be used for the calculation of the accumulation in the furrow and leaching to the shallow groundwater in the RIVM Conclusion.

$$H = \frac{P \times M}{S \times R \times T}$$

H = Henry's Law Constant [-]
P = vapour pressure [Pa]
M = molar weight [$g.mol^{-1}$]
R = 8.3144 [$J.mol^{-1}.K^{-1}$]
S = water solubility [$g.m^{-3}$]
T = temperature in Kelvin [K]

USES 1.0 calculates a Henry's Law Constant H' in [$Pa.m^3.mol^{-1}$], and an air-water partition coefficient, which is the Henry's Law Constant H.

Analysis and detection

Reports about the most relevant analytical methods are summarised in this section. A requirement of the CTB for the applying firm is the description of a method for the analysis of residues in surface water (see the application form section G.2.5 (Anonymous, 1995).

Analytical methods, referred to in test reports, are briefly summarised in the Description, and it is not necessary to repeat them under Detection.

If only a reference for the description of a method is given by the author, while this description is important for the Summary, this reference may be checked (even though the industry did not send it).

Additional information about analytical methods can be found in the archive of LOC/RIVM.

Function and uses

The following information should be given in this record:

- formulations, for which the request for registration is made;
- crop;
- mode of application;
- time of application;
- application rate, frequency of applications and interval period;
- mode of action (e.g. herbicide).

These data are provided by the CTB. Use the trivial names of substances and formulations in this record.

Metabolites and metabolisation scheme

Structural formulas should be depictured uniformly. If only a code and a structural formula of a metabolite are given by a firm, the chemical name has to be recorded only, if it is formed in an amount $\geq 5\%$ in a certain medium (e.g water, soil). The chemical name is given by the expert from LOC/RIVM or LAC/RIVM.

The use of trivial names is encouraged.

The metabolisation schemes in water and soil are recorded in TOXIS in the record Chemobiokinetics. Use for the active ingredient the primary name, and for the metabolites the codes (mA, mB, etc.).

Metabolism in plants

Test reports about the metabolism in plants are not summarised, except plant-soil studies, in which the uptake from the soil, or the degradation on the surface of the plant (e.g. leaf), is described.

4.4 Behaviour in soil

4.4.1 General information

The study of the behaviour of a pesticide in soil is important for those pesticides that are applied directly on or in soil, or those that may reach the soil after application. The information that is important in this context, concerns the transformation, leaching and accumulation behaviour. Two parameters are required, namely the time in which 50% of the pesticide has transformed in soil, and the degree of sorption of the pesticide in soil. The transformation rate and the sorption should be determined in at least three soil types; the transformation route should be determined in at least one soil type; at least one column leaching study with aged residue should be carried out. Sorption constants are required for at least three different soil types relevant for The Netherlands. If a compound is charged or dissociated under naturally occurring conditions (pH range 4-9), the sorption constants are required for the situation in which the sorption is expected to be relatively low (generally pH range 7-8).

Sorption and transformation constants are required for parent compounds and all major (i.e. formed in quantities of 10% —molar fraction— or more) transformation products.

Photochemical transformation on the leaf or on the soil surface can play a role of importance, depending on the nature of the compound and the way of application. For example, when a compound is not ploughed into the soil, it is important to know the photostability of the compound. In some cases, data about the photolysis in water are sufficient (Anonymous, 1995).

The instructions in Tables 2, 3, 4, 5, and 6 are based on the Appendix G.1. of the CTB Application Form (Anonymous, 1995), and on OECD and BBA Guidelines.

For an introduction in the use of soil science for the evaluation of the behaviour of substances in the soil, the reviewer is referred to Van Gestel (1986).

Classification of the soil type

The classification of the soil type, given by the authors, should be checked with the American Soil Classification System (see Appendix 2) (USDA, 1951), and this US-classification is reported in the Header. If verification is not possible, the classification given by the authors is used (if necessary a literal translation from Dutch, French or German). It should clearly be stated in the Description, which classification was used.

If it concerns soil types not representative for the Dutch situation, like paddy soil and volcanic soil, this should be clearly stated.

The sizes of soil particles are fastened down in the different classification systems. The sizes are however not identical in the different systems. Pay attention to this when classifying a soil according to the American Soil Classification system.

pH measurement

The pH of the soil can be measured in the water phase (pH-H_2O) of the soil, or after a solution of KCl or $CaCl_2$ was added (pH-KCl and pH-$CaCl_2$, respectively). pH-KCl and pH-$CaCl_2$ are always lower than pH-H_2O as more protons in the soil solution can be measured.

It is assumed that the pH-$CaCl_2$ (0.01 N) gives the best estimate for the soil solution, and is therefore the most convenient value with respect to bioavailability for plants. It is assumed that the pH-KCl (1.0 N) gives the best estimate for the sorption of pesticides to soil particles.

Record in the TOXIS Header the pH-KCl, if available. If not, record the pH-$CaCl_2$ or the pH-H_2O (in this order of preference). It should always be indicated in the Description, which pH is used.

Use of substance codes and formulation codes in soil studies

In the Header always the substance code is given, even if it was applied as a formulation. In the Description this should then clearly be stated: e.g. Sub. 1 was applied as formulation X (Y % a.i.).

Useful formulas

1) % organic matter (o.m.) = 1.7 × % organic carbon (o.c.)
2) 1 kg a.i./ha = 1.4 mg a.i./kg dw soil (assuming the compound homogeneously mixed over a soil depth of 5 cm, and a dry bulk density of 1.4 kg/dm^3)

Residues after multiple applications

If an a.i. is applied more than once during a season, the concentration that is reached in the soil after the last application (according to the label) is used for the evaluation of the studies on the behaviour in soil, and for the risk evaluation for the leaching to the shallow groundwater. To calculate the concentration of the a.i. in the soil after multiple applications the reader is referred to Appendix 2.

4.4.2 Transformation in the top soil (see Table 2)

DT_{50} values should be based on transformation. Transformation means the compound is converted to smaller or larger molecules by biological, microbiological, and/or chemical action. Degradation means the compound is converted to smaller molecules by biological, microbiological, and/or chemical action. Dissipation means that the compound "disappears": this can be by transformation, volatilisation, leaching, plant uptake, or run-off. Mineralisation means the compound degrades to inorganic compounds (e.g. H_2O, CO_2).

If raw data are available, the DT_{50} values always have to be recalculated. The calculation of a reliable DT_{50} value has to meet the following conditions.

1. At least five time points including the value on $t = 0$ (to enable adequate regression analysis) within the first 100 days of the study (because after 100 days the biological activity of the soil may have declined substantially (Anderson, 1987)). Within this period only data showing mole fractions of $\geq 10\%$ are taken into account; at lower fractions e.g. diffusion may influence the transformation rate.

2. At least three (additional to point 1, see above) time points are needed to ensure that there is a lag phase. The lag phase is not included in the calculation of the DT_{50}.

3. To check whether the curve shows first-order kinetics, or consists of a successive serie of two first-order processes, the 'hockey stick' model is used (see Appendix 3). All available time points are used for this check, including those outside the first 100 days (showing molar fractions $\geq 10\%$). In the case of a 'hockey stick' curve, a period with a higher transformation rate is followed by a period with a lower transformation rate, resulting in a hinge point in the transformation curve. With the 'hockey-stick' model a calculation is performed to estimate whether this model gives a better fit compared to the single log-linear model. In case of a significant hinge point ($p \leq 0.05$) within 50 - 100 days after application of the substance only the time points up to the hinge point will be used for the calculation of the DT_{50}. In case of a significant hinge point before 50 days, or in case the hinge point is after 100 days and the % residues are high (molar fraction >50%) after 100 days, both periods should be mentioned (hinge point and slopes), and expert judgement is required to establish the DT_{50}.

4. The DT_{50} value is calculated with a log-linear regression model using all selected time points, provided that first-order kinetics appear to be valid. If not, the DT_{50} is determined graphically (Linders et al., 1994). If $r^2 < 0.7$, the regression is not valid.

The recalculated DT_{50} value is recorded in the Header. The DT_{50} value calculated by the authors is mentioned in the Results. In the Remarks is stated

- DT_{50} in Header derived from data given by the author.

In the Remarks should always be stated, if the transformation followed first-order kinetics or not, and if the DT_{50} was calculated or determined graphically. In case DT_{50} values are based on CO_2-production or dissipation, this is stated in the Header (Header field 'remarks') and in the Remarks. In case the DT_{50} was extrapolated, this should also be mentioned in the Remarks. The DT_{90} value can be used to check whether the transformation followed first-order kinetics (as a rule of thumb: DT_{90} is c. three times the DT_{50}).

DT_{50} values are converted to 20 °C (automatically by TOXIS) using the equation:

$$DT_{50}\ (20^\circ C) = DT50\ (T) \times e^{(0.08\times(T-20))}$$

T = temperature (°C) at which the study was conducted

In the Remarks all deviations from the instructions (in this section and in Table 2) are stated. DT_{50} values determined in soils with %o.m. >15 are preferably not used for RIVM Conclusions because these soils are not considered useful for Dutch risk assessment. If, however, the results from these soils are within the range of (or near) results from other soils with %o.m. <15, they are selected for the RIVM Conclusions. In this way the requirement that reliable results of three soil types should be available can be met in a satisfying way without taking the trouble of additional testing.

If DT_{50} values are used in the RIVM Conclusion, in the Remarks is stated:
- DT_{50} value(s) used for the RIVM Conclusion is (are).. .
- Converted DT_{50} value(s) (20 °C) is (are) .. .

Storage conditions of sampled soil, that is not immediately used for transformation studies, should be as follows: in the laboratory at 4 °C for at most three months (to avoid anaerobic conditions); in the open or in a glasshouse under well-drained conditions (to avoid desiccation).

The pF of the soil can be represented as a function of the soil water content (see Appendix 2). The soil water content can be expressed as volume water per volume soil (v/v) or as weight water per weight soil (w/w). The moisture content in the pF-graph in Appendix 2 is v/v. This should be multiplied with the the bulk density (default 1400 kg/m^3 soil) to obtain the moisture content w/w.

If a firm wants to have a purified mixture of isomers registered (e.g. a mixture of only trans-isomers), the transformation rates of mixtures of other isomers (e.g. cis and trans-isomers) can be used. It should be investigated if the transformation rates of the isomers might differ substantially from each other. The expert of the LOC should be consulted with respect to the potential behaviour of the isomers. To know whether the optic centre is the point of impact in the molecule can be of help. If the transformation of a mixture does not show first-order kinetics, this might be an indication that the different isomers have different transformation rates. If there are

Table 2: Soil aerobic transformation studies (top soil)

	Items	Notes	Reliability lower ?
Description	**1. test type**		
	1.1 aerobic	1.1 aerobic, non-sterile conditions are required	1.1 Y
	1.2 sterile	1.2 method of sterilisation should be given	1.2 E
	2. test substance and position of label		
	3. vehicle		
	4. soil	4. top soil should be used; no enrichment with e.g. alfalfa	4. Y
	4.1 soil type	4.1 US-class. and other relevant data (paddy, etc.)	
	4.2 pH		
	4.3 CEC		
	4.4 % o.m.		
	4.5 storage conditions	4.5 if there is no immediate use, storage in the lab or in the open should be appropriate (see text)	
	5. weight of soil sample	5. weight soil sample should be ≥X g. [X= 25]	5. E
	6. temperature		
	7. application	7. report way of application	
	7.1 rate	7.1 within X times [X= 2] the recommended application rate higher or lower [5]	7.1 Y
	8. moisture content	8. in an aerobic study: pF 2 - 3; soils with pF ≥4.2 get RI of 3[6]	8. Y
	9. light condition	9. incubation in the dark[7]	9. Y
	10. test system	10. should be closed with volatile traps	10. E
	11. incubation time	11. preferred until 90% transformation or up to 100 days	11. E
	12. sampling frequency	12. ≥5 time points are needed for adequate regression analysis	12. Y
	13. extraction/analysis method	13. This should be appropriate for the substance and the metabolites, and the recovery of the substance should be >X% [X= 70] and <Y% [Y= 110]	13. E
Results	**1. DT_{50}[8]; %a.i. at the end of incubation**		
	2. total recovery	2. the recovery at every time point should be >X% [X= 80] (recovery of radiolabel or the sum of compounds)	2. E
	3. kinetic order	3. check 1st order kinetics with Hockey-stick model	
	4. bound residue	4. maximum and time, amount after 100 days, and amount at end	
	5. produced CO_2	5. maximum and time, amount after 100 days and amount at end	
	6. metabolites:	6. identified and quantified separately	6. Y
	6.1 ≥ 5%:	6.1 chemical name, maximum and time, amount after 100 days, and amount at end	
	6.2 < 5%: number	6.2 number of metabolites <5%	
	6.3 ≥10%: DT_{50}	6.3 if no reliable DT_{50} is calculated, transformation studies are required (AQs).	
Pay att.	1. the dissipation type	1. this should be transformation	1. E
	2. the agricultural history soil	2. no prior use of compounds that may have lead to adapted microorganisms in the previous five years	2. E
	3. storage	3. BBA/Speyer soils before 1982 were probably stored too dry	3.2 Y

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[5] If the application rate is more than two times too high: inhibition (or stimulation) of the soil organisms is possible, leading to a higher (lower) DT_{50}; if the application rate is more than two times too low: inhibition of the soil organisms might occur at the normal application rate. It is also possible that adaptation mechanisms come into play, leading to inaccurate results.

[6] In Appendix 3 the required moisture content per soil type is given. Soils should not become too wet or too dry (pF should be in the range 2 - 3), also during pretreatment.

[7] Unless it has been shown that soil phototransformation is of no importance.

[8] Only the following DT_{50} values are selected for risk assessment in the RIVM Conclusion: performed in a laboratory (not in the field); aerobic; in the upper soil at pF 2 - 3; in a soil with a %o.m. <15; in a soil relevant for the Dutch situation (e.g. loamy sand or clay, not paddy or volcanic soil); at a dosage not higher and not lower than two times the recommended dosage; at a temperature within the range 15 - 25°C. DT_{50} values determined in soils with %o.m. >15 are selected, if these DT_{50} values are within the range found in soils with a %o.m. <15.

any indications that the purified mixture might show a behaviour different from the other mixtures, transformation studies with the separate isomers are required.

4.4.3 Sorption and mobility in unsaturated soil (see Tables 3, 4, and 5)

Sorption may be studied through experiments with:
- soil-water suspensions (also called adsorption, batch or slurry experiments);
- soil columns (also called leaching experiments);
- thin or thick layer chromatography (TLC) experiments.

In the respective sections below further instructions are given.

If a pesticide has one or more acid or basic groups, the soil pH influences its mobility. As in Dutch soils the negative charges of soil particles prevail, it is important to verify whether a pesticide is dissociated (i.e. negatively charged, due to the loss of H^+), protonated (i.e. positively charged, due to the take up of H^+), or neutral, given a specific soil pH. The soil pH determines the extent of dissociation or protonation of such pesticides, and therefore their mobility. The potency of a pesticide to protonate or dissociate is expressed by its pK_a. Generally, negatively charged pesticides in a rather basic soil are assumed to be more mobile than neutral or protonated pesticides.

Rf, $K_{s/l}$ or K_{om} values have to be recalculated from the raw data, if available. The value calculated by the author is mentioned in Results. In the Remarks is stated

♦ Results in Header derived from data given by the author.

TOXIS calculates the $K_{s/l}$ and K_{om} automatically, using the leaching data in the Header.

Soil, sorption and mobility, column leaching studies (see Table 3)

In soil column leaching experiments the penetration depth of the substance is generally determined. This depth is estimated as the depth of the bottom of the layer above which half of the substance recovered (in soil column plus leachate) is present. With the penetration depth the sorption coefficient can be calculated. The penetration depth (Xp) is established as follows:

a) The substance is applied on top of the soil column without homogeneous mixing:
- If 50% or more of the applied compound is found in the column Xp is established as the distance from the top of the column to the bottom of the layer in which the 50% is reached.
- If more than 50% is in the leachate and the leachate is not analysed in fractions, Xp is defined as $\geq$ column length; consequently the K_{om} cannot be calculated accurately and a $\leq$ sign is added (and the reliability is lowered).

Table 3: Soil, sorption and mobility, column leaching studies

	Items	Notes	Reliability lower?
Description	**1. test substance + position of label**		
	2. vehicle		
	3. soil[9]		
	3.1 soil type	3.1 US-class. and other relevant data (paddy etc.)	
	3.2 pH		
	3.3 CEC and %clay		
	3.4 % o.m.		
	4. aged or unaged treated soil	4. aged soil: amount of a.i. and metabolites present after ageing should be determined	4. Y
	5. ageing time	5. give duration, in general equal to the DT_{50} of the most relevant compound	5. E
	6. application rate		
	7. way of application:		
	7.1 unaged: e.g. spraying		
	7.2 aged: volume/weight of soil sample	7.2 it is important that the applied amount of aged soil is homogeneously mixed	7.2 E
	8. soil column		
	8.1 length	8.1 length min. X cm [X= 20]	8.1 E
	8.2 diameter	8.2 ϕ min. X mm [X= 25]	8.2 E
	9. water saturation prior to leaching	9. column should be at field capacity before the test substance is applied	9. E
	10. waterflux	10. maximum X [X=10] cm/day; see CTB Application form for details	10. Y
	11. leaching time		
	12. samples:[10]		
	12.1 column slices	12.1 column should be sliced: min. X layers [X= 5], with max. thickness of X cm [X= 5 cm]	12.1 Y
	12.2 leachate	12.2 should be analysed in at least X fractions [X= 5]	12.2 Y
	13. extraction/analysis method	13. recovery should be >X% [X= 70]	13. E
Results	**1. penetration depth**[11]		
	2. total recovery (maximum, and time)	2. recovery of the activity should be >X% [X= 80]	2. E
	3. percentage a.i. in leachate		
	4. distribution of a.i. over the column/leachate		
	5. metabolites	5. number and percentages	
Pay attent.	1. pK_a of the substance	1. if $2<pK_a<6$: sorption should be determined at pH-range 7-8 (3 soils)	
	2. handling of soil	2. no manipulation with exception of sieving (2 mm) is allowed. According to the BBA IV 4-2 Guidelines the sieve diameter is 1 mm	2. E
	3. volatilisation	3. there should be no major loss due to volatilisation	3. E

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[9] For some pesticides (e.g. those containing a phosphate moiety) the amounts of sesqui-oxides/hydroxides in soil might explain the amounts sorbed. For some pesticides (e.g. those with a positive charge) the amounts of clay might explain the amounts sorbed.

[10] In general both column and leachate should be analysed in layers or fractions, but:
- if <25% in leachate: the column should be sliced, but the leachate need not be analysed in fractions;
- if >75% in leachate: the column need not be sliced, but the leachate should be analysed in fractions;
- in all other cases: both column and leachate should be analysed in fractions.

[11] Only the following Xp values are selected for the calculation of $K_{s/l}$ and the hazard assessment in the RIVM Conclusion: performed in a laboratory; in a soil relevant for the Dutch situation (e.g. no paddy or volcanic soil); in a soil with a % o.m. 0.5 - 15.

- If more than 50% is found in the leachate and the leachate is analysed in fractions, Xp equals the column length; the thickness of the water layer (D) used in the formula below is adapted. In this case we do not need the thickness of the totally applied water layer, but only the thickness of the water layer that leached 50% of the substance. The thickness of this water layer is calculated from the amount of leachate that contains the first 50% of the substance. The volume of this leachate is divided by the column area to give the thickness of the water layer.

b) The substance is homogeneously mixed with soil and applied in a soil layer on top of the soil column (this applies also to soil with aged residue):

- If 50% or more is found in the applied soil layer, the Xp is established as half the height of the applied soil layer, resulting in a $K_{s/l}$ value with a "≥" sign.
- If 50% is found in a layer below the applied layer, the penetration depth is established as the distance to the bottom of the segment in which the 50% limit of the penetrated substance is reached minus half the thickness of the applied soil layer.
- If more than 50% is found in the leachate and the leachate is analysed in fractions, Xp equals the column length minus half the thickness of the applied soil layer; the thickness of the water layer (D) used in the formula below is adapted in the same way is described under a).

The penetration depth and the $K_{s/l}$ are related as follows:

$$Xp = \frac{D}{\rho \times K_{s/l} + \theta}$$

Xp = *the penetration depth of the substance* [*cm*]
D = *the thickness of the applied water layer* [*cm*]
ρ = *the dry soil bulk density* (*default* 1.4 kg/dm^3)
$K_{s/l}$ = *the solid-liquid partition coefficient* [dm^3/kg]
θ = *the moisture volume fraction* (*default* 0.4)

If the water movement is followed using a tracer:

$$X_p = \frac{\theta \times X_{p,\ tracer}}{\rho \times K_{s/l} + \theta}$$

X_p = *the penetration depth of the substance* [*cm*]
$X_{p,\ tracer}$ = *the penetration depth of the tracer* [*cm*]
θ = *the moisture volume fraction* (*default* 0.4) [-]
ρ = *the dry soil bulk density* (*default* 1.4) [kg/dm^3]
$K_{s/l}$ = *the soil-liquid partition coefficient* [dm^3/kg]

In the TOXIS Header the percentage of the *substance* (not the r.a.) that is found in the leachate is recorded, as wel as Xp. If no substance is found in the leachate, the percentage leaching in the Header is '<X' (X is the detection limit). If the results of a study are based on r.a. and it is not sure that it concerns only the substance, the results are not recorded in the Header.

The K_{om} value is derived from the $K_{s/l}$ value with:

$$K_{om} = \frac{100 \times K_{s/l}}{\%\ o.m.}$$

K_{om} = *the organic matter partition coefficient* $[dm^3/kg]$
$K_{s/l}$ = *the solid-liquid partition coefficient* $[dm^3/kg]$
$\%\ o.m.$ = *the percentage organic matter of the soil*

When results are used for the RIVM Conclusion, in the Remarks is stated:

- The next calculated $K_{s/l}$ values are used for RIVM conclusions:..
- K_{om}-value(s) used for RIVM Conclusion is (are) ..

In BBA-37 studies, the leaching time is 2 days. In case in BBA-37 studies the percentages organic matter (o.m.) are not reported, the following values are used for the soils 2.1 (sand), 2.2 (loamy sand), 2.3 (sandy loam): 1%, 4%, and 2%, respectively. In general BBA-37 studies get the Reliability Index 2, because the columns are not sliced. The results are only used for the RIVM Conclusion if better data are not available.

In general, studies with freshly applied residues are used. In some cases studies with aged residue may be used to obtain a K_{om}, if the penetration depth of the substance can be determined in a reliable way. After ageing, the soil must be analysed for individual substances. Instructions for determining $K_{s/l}$ values from aged-residue studies (for a.i. and metabolites) are under development. For summarising aged-residue tests follow Table 3. Aged-residue tests —to obtain qualitative information on the behaviour of metabolites— are included in Table 3.

Soil, sorption and mobility, TLC studies (see Table 4)

TLC stands for both soil *thin* layer chromatography and soil *thick* layer chromatography. Soil thin layer: <0.5 - 1 mm; soil thick layer: 5 - 10 mm. In soil thin/thick layer studies the retardation factor Rf is determined. The Rf is the ratio between the elution distance of the substance and the elution distance of the developing phase. The Rf is used to derive the solid-liquid partition coefficient for the substance: $K_{s/l}$.

$K_{s/l}$ values determined in TLC experiments are considered less reliable because of difficulties in the exact determination of the relative rates of movement. The sorption may be underestimated, due to handling of the soil, possible influence of the support material, and a probable non-equilibrium situation. Therefore, the highest Reliability Index these studies can get is 2.

The elution distance (peak) of the substance is found in the centre of the spot on the TLC plate. In case both central and frontal Rf values are given, the central value is included in the Header; the frontal value is only included in the Results if it is at least a factor two higher than the central value.

The retardation factor (Rf) is calculated using the following equation:

$$Rf = \frac{\text{distance of substance and site of application}}{\text{distance of waterfront and site of application}}$$

The $K_{s/l}$ value is derived from the Rf using the following equation:

$$K_{s/l} = \frac{\theta\ (1 - Rf)}{\rho \times Rf}$$

$K_{s/l}$ = *the solid-liquid partition coefficient* [dm^3/kg]
θ = *the moisture volume fraction* (*default* = 0.4)
ρ = *the dry soil bulk density* (*default* = 1.4 kg/dm^3)
Rf = *the retardation factor*

How to calculate the K_{om} from the $K_{s/l}$ is described in the previous section on column leaching experiments.

Table 4: Soil, sorption and mobility, TLC studies

	Items	Notes	Reliability lower?
Description	**1. test substance + position of label**		
	2. vehicle		
	3. soil[12]		
	3.1 soil type (US-class.)		
	3.2 pH		
	3.3 CEC and %clay		
	3.4 % o.m.		
	4. temperature		
	5. application rate		
	6. thickness/surface layer		
	7. developing phase (pH etc.)		
	8. extraction/analysis method	8. recovery should be >X% [X= 70]	8. E
Results	**1. Rf**[13]	1.	
	1.1 a.i.	1.1 Rf should be based on a.i. (so not on total of a.i and metabolites)	1.1 Y
	1.2 minimal Rf	1.2 if Rf <X [X= 0.3], $K_{s/l}$ is based on Rf <X	1.2 Y
	2. Rf of reference compound		
Pay attention	1. adsorption to glass or support materials		
	2. soil handling	2. no manipulation, with exception of sieving (0.5 mm for the 0.5 mm layer or 2 mm for a thick layer) allowed	2. E
	3. shape of the spot	3. tails may indicate overloading	3. E
	4. analysis	4. Rf should not be determined via a bioassay	4. Y

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[12] For some pesticides (e.g. those containing a phosphate moiety) the amounts of sesqui-oxides/hydroxides in soil might explain the amounts sorbed. For some pesticides (e.g. those with a positive charge) the amounts of clay might explain the amounts sorbed.

[13] Only the following Rf values are selected for hazard assessment in the RIVM Conclusion: reliable data from column leaching or adsorption studies are lacking; in a soil relevant for Dutch situation (e.g. no paddy or volcanic soil); in a soil with % o.m. 0.5 - 15.

Soil, sorption and mobility, adsorption studies (see Table 5)

In soil adsorption studies in general a $K_{s/l}$ is derived by measuring the amount of r.a. that is present in the liquid phase.

In soil column leaching experiments the distribution constant (in general indicated as K_d) is indicated as $K_{s/l}$, because it concerns the distribution between the solid phase and the liquid phase. In the adsorption/desorption studies the distribution constant is indicated as K_F as they are derived from the Freundlich equation:

$$S = K_F C^{1/n}$$

$1/n$ = *Freundlich exponent* [-]
S = *amount sorbed* [*mg/kg*]
C = *equilibrium concentration in water* [*mg/l*]
K_F = *Freundlich constant* [$dm^{3/n}/kg$]

The $K_{s/l}$ is derived from the partitioning between the concentrations in the solid and liquid phase. Because the adsorption is generally not irrespective of the amount of substance present, a correction factor is introduced: the Freundlich exponent 1/n. The correction results in a 'different' $K_{s/l}$: the K_F ($K_{s/l} = K_F^{1/n}$).

For matters of convenience, we accept the K_F as a $K_{s/l}$ for further calculations, provided that the Freundlich exponent is within the range 0.7 - 1.1. K_F values with 1/n outside the range 0.7 - 1.1 are not used for K_{om} calculations and get the Reliability Index 2 (Boesten and Van der Linden, 1991).

In case transformation was too high in the adsorption experiment, a reliable $K_{s/l}$ can only be calculated if besides the concentration in the liquid phase, also the amount of the substance adsorbed to the soil is determined.

Table 5: Soil, sorption and mobility, adsorption studies

	Items	Notes	Reliability lower?
Description	**1. test substance + position of label**		
	2. vehicle		
	3. soil[14]		
	3.1 soil type (US-class.)	3.1 soil must be relevant for the Dutch situation	
	3.2 pH		
	3.3 CEC and %clay		
	3.4 % o.m.		
	4. weight of soil sample		
	5. soil/water ratio (kg/dm^3)		
	6. temperature		
	7. number of concentrations	7. min. X concentrations [X= 4] should be used	7. Y
	8 number of replicas	8. test should be performed in duplo	8. Y
	9. shaking time	9. shaking time (in hours) should be shorter than the DT_{50} (in days)[15]; no longer than 48 hours	9. E
	10. extraction/analysis method	10. recovery should be >X% [X= 70]	10. E
Results	**1. distribution constants[16]**		
	2. Freundlich exponent	2. only $K_{s/l}$ with 1/n of X [X= 0.7 - 1.1] are used for the calculation of K_{om}[17]	2. Y
	3. the relative decrease should be ≥0.1	3. $K_{s/l}$ is not accurate if $K_{s/l}$ x (soil/water ratio) <0.1][18]	3. Y
	4. total recovery (maximum, and time)	4. recovery should be >X% [X= 80]	4. E
Pay attention	1. water solubility	1. initial and equilibrium concentrations should not exceed water solubility	1. Y
	2. transformation	2. there should be no major loss due to transformation (max. 3%), unless both the amount sorbed and the decline in concentration of the substance in the liquid phase has been determined	2. E
	3. pK_a of the substance	3. if $2<pK_a<6$: $K_{s/l}$ should be determined at pH 7-8	
	4. soil handling	4. no manipulation, with exception of sieving (2 mm) allowed. Sterilisation is not allowed	4. Y

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[14] For some pesticides (e.g. those containing a phosphate moiety) the amounts of sesqui-oxides/hydroxides in soil might explain the amounts sorbed. For some pesticides (e.g. those with a positive charge) the amounts of clay might explain the amounts sorbed.

[15] This requirement has to be met if only the decline in concentration of the substance in water is analysed and not the amount sorbed to the soil: if the shaking time (in hours) is longer than the DT_{50} (in days), the concentrations in both soil and water must be measured.

[16] Only the following $K_{s/l}$ values are selected for the risk assessment in the RIVM Conclusion: from a study performed in a laboratory; in a soil relevant for the Dutch situation (e.g. no paddy or volcanic soil); in a soil with a % o.m. ≥0.5 and ≤15; from a study with $K_{s/l}$ x ratio soil/water ≥0.1; from a study with $0.7 \leq 1/n \leq 1.1$.

[17] 1/n not within this range indicates poor accuracy or strange sorption behaviour.

[18] This requirement need not be met if the amount sorbed is also measured.

How to calculate the K_{om} from the $K_{s/l}$ is described in the previous section on column leaching experiments.

In case the $K_{s/l}$ is not reliable because [$K_{s/l}$ × (ratio soil/water)] <0.1, the result is not used for RIVM Conclusions, unless the substance is measured in both the water fraction and the soil fraction (Boesten, 1990).

For results not used for RIVM Conclusions the following standard sentence is used in Remarks:

- $K_{s/l}$ of soil ... is not used in RIVM Conclusion because ..... .

4.4.4 Transformation and sorption in saturated soil

The evaluation procedure for transformation and sorption studies in the saturated zone is under development.

4.4.5 Lysimeter and field leaching studies

Information on field studies can be found in the CTB Application Form (Anonymous, 1995).

Instructions for the lysimeter studies are presented in Table 6. In the TOXIS Header the soil properties of the A-horizon are recorded.

A tracer can be applied for a better interpretation of the water flow (after the growing season of the crop). A tracer is a compound not showing transformation and sorption, and not interfering with the compound under investigation (e.g. tritiated water, bromide, chloride).

Table 6: Soil, sorption and mobility, lysimeter studies

	Items	Notes	Reliability lower?	
Description	**1. substance**	1. as a.i. or as formulation		
	2. label	2. position of label should be reported, except when only the leachability of a single compound is investigated	2.	Y
	3. application rate	3. highest prescribed dosage; only problems with detection allow a higher dosage	3.	E
	4. tracer	4. use of a tracer is preferred (e.g. chloride, bromide)		
	5. number of replicas	5. at least two replicas	5.	Y
	6. soil type	6. give soil characteristics		
	6.1 sensitivity	6.1 soil relatively sensitive to leaching of compound is preferred		
	6.2 horizon/layers	6.2 soil horizon classifications and depths should be given	6.2	E
	7. soil history	7. no exposure to compound, nor metabolites, nor structural analogues in the past 5 years	7.	Y
	8. casing			
	8.1 material	8.1 inert material (e.g. stainless steel)	8.1	E
	8.2 dimensions	8.2 $\geq$0.1 m^2 surface area for cereals, $\geq$0.5 m^2 for other crops. Depth/Height c. 1 m.	8.2	Y
	9. soil column			
	9.1 profile	9.1 column should have an undisturbed soil profile.	9.1	Y
	9.2 hydrology	9.2 the hydrology within the lysimeter should reflect the field where it is placed.	9.2	E
	10. soil treatment	10. normal agricultural treatment, including the surroundings (at least one meter distance) of the lysimeter during the entire study	10	Y
	11. location of experiment			
	12. climatic conditions			
	12.1 air	12.1 air temperature (measured daily), potential evapotranspiration measured within 25 km (preferably on daily basis)		
	12.2 soil	12.2 soil temperature (different depths) in same soil type or in control lysimeter (give range),		
	12.3 precipitation	12.3 daily precipitation, measured within 100 m (give total)	12.3	E
	13. crop sampling	13. upon harvest tracer, r.a., and individual compounds in the crops are determined.	13.	Y
	14. soil sampling			
	14.1 during experiment	14.1 sampling of the plough layer is only allowed immediately before a soil tillage operation	14.1	Y
	14.2 upon termination	14.2 upon termination the soil is sampled in layers (max. 10 cm each) and analysed for tracer, total r.a., total extractable r.a., bound residues, individual compounds, bulk density and moisture content	14.2	Y
Results	**1. leachate**[19]			
	1.1 amount	1.1 in total >300 mm per year (give total) should be reached.	1.1	E
	1.2 frequency	1.2 measurements of leachate on a weekly basis.		
	1.3 composition	1.3 leachate is sampled and analysed for tracer, total r.a. and individual compounds		
	2. crop and soil	2. give results: see 'description' of items 13 and 14		
	3 total precipitation			
	3.1 precipitation	3.1 give amount of precipitation;		
	3.2 additional	3.2 additional water supply (artificial rain) max. 20 mm per day	3.2	Y
Pay att.	1. duration of test	1. until a reasonable certainty on the behaviour of the test substance is acquired	1.	E
	2. interpretation of results	2. laboratory experiments (soil degradation and column leaching) for the different horizons of the used soil (in case of unclear horizons, for the layers 0 - 30 cm, 30 - 60 cm, 60 - 100 cm) are necessary for proper interpretation (not necessary if only the leachability of non-identified metabolites is investigated)	2.	E

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[19] The soil should be relevant for Dutch situation; e.g. not a paddy soil or a volcanic soil. A %o.m. 0.5 - 2 is preferred.
The result of the lysimeter experiment has to be evaluated in view of the standard PESTLA scenario, before it can be used in the CTB decision tree for leaching to shallow groundwater (instructions in preparation, available c. May 1995).

4.5 Behaviour in surface water

4.5.1 General information

In case a compound reaches the water in a substantial amount, information on the behaviour of the pesticide in water is required. After local application of pesticides, main emission routes to the water are directl, via application in water and drift, or indirect, via run off, drainage (including leaching), and deposition from air of the pesticide, and via waste water of a sewage treatment plant (STP).

According to the Dutch pesticide registration policy (Anonymous, 1995), studies on hydrolysis are always requested. If a compound reaches the surface water in substantial amounts, studies on ready biodegradability are also requested. If a compound hydrolyses very rapidly in water (i.e. DT_{50} <1 day), the main metabolite(s) should be measured. For methods of research, OECD-guidelines 111, and 301 A - 301 F are suggested. If a compound proves to be "readily biodegradable", no further testing is required. If this is not the case, additional testing with natural waters and the accompanying sediment is required (G.2.1). For methods of research, OECD 302 or 303 are suggested. In case the compound is expected to reach the surface water via an STP, biotransformation tests according to OECD 302 (inherent biodegradability test with activated sludge) and 303 (simulation test with activated sludge) are required.

For pesticides that are applied directly in the water or that reach the water in large quantity (>50% of the applied dose), studies on photolysis in water are required.

Information on the flux of an active ingredient from a paint-layer or wood can be required. This applies for some wood preservatives and anti-foulings.

Concerning inorganic compounds, no instructions for testing are given in the registration procedures because of the complexicity of the (many) possible chemical reactions. It is preferred to indicate which important reactions take place in the water.

The instructions in Tables 7, 8, and 9, are based on the Appendices D.9 and G.2 of the CTB Application Form (Anonymous, 1995), and on OECD and EPA Guidelines.

A change in the requirements is under consideration. It is proposed to require the water/sediment study in all cases (if the compound reaches the surface water). The ready biodegradability test is not obligatory then.

Only *laboratory* experiments on hydrolysis, photolysis and degradation (in water/sediment systems) are used for the first step of the risk assessment.

4.5.2 Hydrolysis (see Table 7)

The hydrolysis test should be carried out at *three* different pHs: 4, 7, and 9. The hydrolysis at each pH has to be tested at 2 - 3 different temperatures, unless the substance is thermostable (see OECD 111). The hydrolysis rate can be influenced substantially by a small change in temperature, and the extent is dependent on the compound.

If raw data are available, the DT_{50} values always have to be recalculated. At least five time points including the value on t = 0. The DT_{50} value is calculated with a log-linear regression model using all selected time points, provided that first-order kinetics appear to be valid. If not, the DT_{50} is determined graphically (Linders et al., 1994). If $r^2 < 0.7$, the regression is not valid.

The recalculated DT_{50} value is recorded in the Header. The DT_{50} value calculated by the authors is mentioned in the Results. In the Remarks is stated

- DT_{50} in Header derived from data given by the author.

In the Remarks should always be stated, if the transformation followed first-order kinetics or not, and if the DT_{50} was calculated or determined graphically. In case the DT_{50} was extrapolated, this should also be mentioned in the Remarks.

DT_{50} values are converted to 20 °C (automatically by TOXIS) using the equation:

$$DT_{50}\ (20^\circ C) = DT50\ (T) \times e^{(0.08 \times (T-20))}$$

T = temperature (°C) at which the study was conducted

In the Remarks all deviations from the instructions (in this section and in the table) are stated.

If DT_{50} values are used in the RIVM Conclusion, in the Remarks is stated:

- DT_{50} value(s) used for the RIVM Conclusion is (are).. .
- Converted DT_{50} value(s) (20 °C) is (are) .. .

Table 7: Water, hydrolysis

	Items	Notes	Reliability lower?
Description	**1. test substance + position of label**		
	2. applied concentration(s)		
	3. use of vehicle		
	4. buffer system	4. buffer solution has to be used [20]	4. Y
	5. pH	5. preferably 4, 7, and 9	5. E
	6. duration test		
	7. temperature	7. temperature should be constant	7. Y
	8. light condition	8. test should be performed in darkness [21]	8. Y
	9. sampling frequency	9. sampling should be frequent [f= ≥5] enough to establish DT_{50} and kinetics	9. E
	10. extraction/analysis method	10. recovery should be >X% [X= 70]	10. E
Results	**1. DT_{50}[22]**		
	2. metabolites (maximum, and time)		
	3. total recovery (maximum, and time)	3. recovery should be substantial >X% [X= 80]	3. E
	4. kinetic order		
Pay attention	1. water solubility	1. test concentrations should not exceed water solubility	1. E
	2. the dissipation type	2. there should be no major loss due to biotransformation (*sterile conditions should be maintained*), photolysis, volatilisation, or adsorption to test vessel: is the test design adequate?	2. E

---

[20] Unless it has been proven by measurement that pH is not changing.

[21] Unless it has been proven that photodegradation is of no importance in the same buffer solutions.

[22] Only the following results are used for risk assessment in the RIVM Conclusion: DT_{50} determined at pH range 3 - 10.

4.5.3 Phototransformation (see Table 8)

The instructions in Table 8 are based on the EPA-540/9-85-014 Guideline.

If raw data are available, the DT_{50} values always have to be recalculated. At least five time points including the value on $t = 0$. The DT_{50} value is calculated with a log-linear regression model using all selected time points, provided that first-order kinetics appear to be valid. If not, the DT_{50} is determined graphically (Linders et al., 1994). If $r^2 < 0.7$, the regression is not valid.

The recalculated DT_{50} value is recorded in the Header. The DT_{50} value calculated by the authors is mentioned in the Results. In the Remarks is stated

- DT_{50} in Header derived from data given by the author.

In the Remarks should always be stated, if the transformation followed first-order kinetics or not, and if the DT_{50} was calculated or determined graphically. In case the DT_{50} was extrapolated, this should also be mentioned in the Remarks.

Table 8: Water, phototransformation

	Items	Notes	Reliability lower?
Description	**1. test substance + position of label**		
	2. applied concentrations		
	3. use of vehicle		
	4. buffer system	4. buffer solution has to be used [23]	4. Y
	5. pH		
	6. use of sensitiser		
	7. test duration		
	8. temperature		
	9. type of light; light intensity; light distance	9. artificial light should be comparable with sunlight (>290 nm)	9. E
	10. sampling frequency		
	11. extraction/analysis method	11. recovery should be >X% [X= 70]	11. E
	12. oxygen	12. excluded: yes or no	
	13. cosolvent	13. used: yes or no, if yes, % in Remarks	
	14. radical scavenger	14. used: yes or no	
Results	**1. DT_{50}**	1. in case of discontinuous exposure: it should be stated, if DT_{50} is calculated over the whole period, or only over sun hours	1. Y
	2. metabolites (maximum, and time)		
	3. total recovery (maximum, and time)	3. recovery should be substantial >X% [X= 80]	3. E
	4. kinetic order		
	5. quantum yield	5. if reported	
Pay attention	1. water solubility	1. test concentrations should not exceed water solubility	1. E
	2. the dissipation type	2. there should be no major loss due to e.g. biotransformation (*sterile conditions should be maintained)* volatilisation, and adsorption to the vessel: is the test design adequate?	2. E
	3. quantum yield	3. Φ is calculated as the amount of substance (in mol) transformed per amount of photons (in Einstein) and has no dimension.	
	4. optical density	4. if the optical density >1, the amount of light is the limiting factor in the transformation. Then the actual DT_{50} may be smaller.	4. E

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[23] Unless it has been proven by measurement that pH is not changing.

4.5.4 Water-sediment System, transformation (see Table 9)

The water-sediment system has to acclimate in order to recuperate from the disturbance and to allow the formation of gradient zones, especially from aerobic to anaerobic.

For water/sediment systems a distinction is made between the DT_{50} value for the pesticide in the aqueous phase ($DT_{50,wat}$), the DT_{50} value in the sediment phase ($DT_{50,sed}$), and the DT_{50} value for the whole water/sediment system ($DT_{50,sys}$). The first may not only include transformation but also volatilisation, and sorption onto e.g. the sediment.

If raw data are available, the DT_{50} values always have to be recalculated. The calculation of a reliable DT_{50} value has to meet the following conditions.

1. At least five time points including the value on $t = 0$ (to enable adequate regression analysis) within the first 100 days of the study (because after 100 days the biological activity of the system may have declined substantially. For the whole system only data showing mole fractions of $\geq 10\%$ are taken into account; at lower fractions e.g. diffusion may influence the transformation rate.

2. At least three (additional to point 1, see above) time points are needed to ensure that there is a lag phase. The lag phase is not included in the calculation of the DT_{50}.

3. To check whether the curve for the whole system shows first-order kinetics, or consists of a successive serie of two first-order processes, the 'hockey stick' model is used (see Appendix 3). All available time points are used for this check, including those outside the first 100 days (showing molar fractions $\geq 10\%$). In the case of a 'hockey stick' curve, a period with a higher transformation rate is followed by a period with a lower transformation rate, resulting in a hinge point in the transformation curve. With the 'hockey-stick' model a calculation is performed to estimate whether this model gives a better fit compared to the single log-linear model. In case of a significant hinge point ($p \leq 0.05$) within 50 - 100 days after application of the substance only the time points up to the hinge point will be used for the calculation of the DT_{50}. In case of a significant hinge point before 50 days, or in case the hinge point is after 100 days and the % residues are high (molar fraction >50%) after 100 days, both periods should be mentioned (hinge point and slopes), and expert judgement is required to establish the DT_{50}.

4. The DT_{50} value is calculated with a log-linear regression model using all selected time points, provided that first-order kinetics appear to be valid. If not, the DT_{50} is determined graphically. If $r^2 < 0.7$, the regression is not valid.

The recalculated DT_{50} value is recorded in the Header. The DT_{50} value calculated by the authors is mentioned in the Results. In the Remarks is stated

- ♦ DT_{50} in Header derived from data given by the author.

Table 9: Water, transformation in water/sediment systems

	Items	Notes	Reliability lower?
Description	**1. test substance + position of label**		
	2. applied concentration(s)		
	3. use of vehicle		
	4. test system:	4.	
	4.1 acclimatisation	4.1 at least 6 - 8 weeks	4.1 Y
	4.2 type and amount of water and sediment	4.2 the mass fraction of solids (dry weight) in the system should be X% based on total weight system [X= 1 or 10][25]	4.2 E
	4.3 aeration/volatile traps	4.3 water should be in aerobic condition[26]	4.3 Y
	5. characteristics of the system: CEC, pH, % o.m. of sediment, DO water, etc.[24]	5. an X cm sediment layer should be used: [X= 2] for small shallow water (agricultural pesticide) [X= 0.2] for large water (non-agricultural pesticide)	5. E
	6. temperature		
	7. light condition	7. 8 - 14 hours light requested by CTB	
	8. duration test	8. until 90% of a.i. is transformed, up to 100 days	8. E
	9. sampling frequency		
	10. extraction/analysis method	10. recovery should be >X% [X= 70]	10. E
Results	**1. $DT_{50,wat}$ / $DT_{50,sys}$ / $DT_{50,sed}$[27]**		
	2. major (≥5%) metabolites	2. maximum and time, amount after 100 days, amount at end	
	3. total recovery (maximum, and time)	3. recovery should be substantial >X% [X= 80]	3. E
	4. kinetic order		
	5. bound residue	5. maximum and time, amount after 100 days, amount at end	
	6. produced CO_2	6. maximum and time, amount after 100 days, amount at end	
Pay att.	1. water solubility	1. test concentrations should not exceed water solubility	1. E
	2. the dissipation type		
	3. inhibition		
	4. the dissociation of the compound	4. the pH may influence the partitioning between water and sediment	

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[24] Two types of sediment have to be tested for agricultural pesticides: one with a high, and one with a low %o.m.. For substances that will mainly reach larger surface waters (e.g. non-agricultural pesticides) only one system has to be tested (%o.m. <5).

[25] A higher percentage of sediment might reduce the biotransformation as there could be a high degree of sorption;
10%: in case the pesticide can reach mainly **small** surface waters after application;
1 %: in case the pesticide can reach mainly **larger** surface waters after application.

[26] If the water is aerated, the sediment should not be disturbed.

[27] Only the following DT_{50} values are selected for risk assessment in the RIVM Conclusion: at 15 - 25°C; in systems with 8 - 14 hours light per day.

In the Remarks should always be stated, if the transformation followed first-order kinetics or not, and if the DT_{50} was calculated or determined graphically. In case DT_{50} values are based on CO_2-production or dissipation, this is stated in the Header (Header field 'remarks') and in the Remarks. In case the DT_{50} was extrapolated, this should also be mentioned in the Remarks.

DT_{50} values, determined at a temperature ranging between 15 and 25°C, are converted to 20 °C (automatically by TOXIS) using the equation:

$$DT_{50}\ (20^\circ C) = DT50\ (T) \times e^{(0.08 \times (T-20))}$$

T = temperature (°C) at which the study was conducted

In the Remarks all deviations from the instructions (in this section and in the table) are stated.

If DT_{50} values are used in the RIVM Conclusion, in the Remarks is stated:

- DT_{50} value(s) used for the RIVM Conclusion is (are).. .
- Converted DT_{50} value(s) (20 °C) is (are) .. .

4.5.5 Ready biodegradability

See the OECD 301 and 306 Guidelines for details on testing of ready biodegradability and interpretation of results. Further instructions are under development.

4.6 Toxicity

4.6.1 General information

The hazard of a pesticide for organisms or ecosystems not only depends on its toxicity, but also on the emission and distribution (e.g. may the compound reach the water?). Therefore the kind of toxicity tests that are required according to Dutch pesticide registration policy, depends on this pattern of emission and distribution. Tests on the following organisms can be required:

- ***toxicity studies on birds;***
- ***toxicity studies on aquatic organisms, of three different trophic levels: micro-organisms (algae), invertebrates (crustaceans), and vertebrates (fish), if the compound is expected to reach the water;***
- ***studies on soil organisms (earthworms, nitrifying bacteria), in case the pesticide is expected to reach the soil;***
- ***bee studies, if the pesticide is applied on flowering crops where bees are used either for fertilisation or for the collection of honey;***
- ***studies on the respiration, or TOC-removal, or nitrification of activated sludge, if pesticides are discharged on an STP after application;***
- ***studies on the bioaccumulation in aquatic organisms, in case of non-organic compounds or compounds with a*** K_{ow} ***>1000;***
- ***studies on the side effects on beneficial (non target) arthropods, in case the pesticide can be used for integrated pest control.***

Primarily, short-term (acute) studies are requested; in case exposure to the compound appears to be long-term (depending on persistence and frequency of application), or the compound is resulting in medium risk after short-term exposure (applies for aquatic organisms only, conform USES 1.0), chronic tests are requested.

Tests have to be performed with the active ingredient(s) (a.i.). Sometimes tests performed with the formulation are preferred; this depends on the nature of the formulation or the way of application (e.g. a formulation containing more than one a.i.). If a test has been performed with a formulation containing one a.i., and this formulation is intended to be applied in the Netherlands, then no additional test with the a.i. is requested.

Additional data concerning metabolites may be required, depending on the nature of the compound and the metabolisation scheme (Anonymous, 1995).

Concentrations

In case of an acute or short-term test, the range of concentrations should be reported in the Description. In case of a chronic or long-term test, the individual tested concentrations should be reported.

The actual concentrations (if reported by the author) are reported in the Results. If the concentration in water is measured on more than one occasion, the measurements should be averaged to one mean concentration. When actual concentrations are reported, these should be used for recalculation of the results. Then in the Remarks is stated:

- ♦ The results in the header are based on actual concentrations.

Statistics

The L(E)C_{50}/LD_{50} value has to be checked for monotonically increasing data (this applies to birds, crustaceans, fish, and bees) if raw data are available (check with the Spearman-Karber computer program (Montana State University; Hamilton et al., 1977/1978). The recalculated value is given in the Header, and the corresponding 95% confidence limits in the Remarks. In the Remarks is stated:

- ♦ L(E)C_{50}/LD_{50} in the header was recalculated according to the Trimmed Spearman-Karber method, based on Hamilton et al. (1977/1978), using data from author.

4.6.2 Birds (see Table 10)

A test according to OECD 401 is required according to Dutch pesticide registration policy to determine the acute oral toxicity (for at least two species). Also a subacute oral toxicity test has to be performed, according to OECD 205 (two species); a semi-chronic oral toxicity test, if requested, should be done according to OECD 206.

The instructions in Table 10 are based on Appendix H.1 of the CTB Application Form (Anonymous, 1995) and on the OECD Guidelines 401 and 205.

Preferably, LD_{50} studies are summarised first, and then LC_{50} studies. The TOXIS database does not provide the functionality of changing the order of tests in the output generated for the reviewer. It is therefore wise to pay attention to the order of studies when recording information in TOXIS.

In case of orally acute (LD_{50}) studies the duration —Fixed Field in the Header— is not entered, because there is no real duration of exposure as in the dietary tests.

In acute study birds may vomit due to the high dose that is applied. This results in underestimation of the toxicity. In case no mortalities are observed and the birds have vomited, the LD_{50} is assumed to be higher than the highest concentration at which no vomiting occurs. The concentrations at which vomiting is observed are not used for calculations with the Spearman-Karber model. If the birds vomit, this should be reported under Results.

In case of a subacute 8 days-study, with an exposure time of 5 days and an observation period of 3 days, the following test durations have to be mentioned in the summary:

Header:	test duration 5 days; observation 3 days;
Remarks:	♦ For the RIVM Conclusion an 8-days LC_{50} of ... is used.

In chronic tests (according to OECD 206) the exposure time is at least 20 weeks, and the following effects are studied: mortality of the adults, egg production, cracked eggs, egg shell thickness, viability, hatchability and effects on young birds.

Table 10: Birds acute and subacute toxicity studies

	Items	**Notes**	**Reliability lower**
Description	**1. test substance**		
	2. test species		
	3. applied concentration(s) of toxicant (in feed or vehicle)	3. min. X concentrations [X= 5][28]	3. E
	4. use of vehicle		
	5. type of application	5. oral (by e.g. gavage), or dietary	
	6. sex, weight and age of the birds		
	7. feed type (LC_{50} study)		
	8. exposure time (LC_{50} study)	8. preferably X days [X= 5]	8. E
	9. observation time	9. LD_{50}: c. X days [X= 14] LC_{50}: after five days exposure X days of observation [X= 3]	9. E
	10. number of animals per group	10. min. X animals per concentration [X= 10]	10. E
	11. availability of feed and water [29]y		
	12. housing conditions		
	13. vehicle control		
Results	**1. LC_{50} or LD_{50}, and 95% confidence limits**	1. raw data should be available for recalculation	1. Y
	2. mortality data in all groups	2. the mortality in the controls should not exceed X% [X= 10] at the end of a test	2. Y
	3. sublethal effects (overt signs of toxicity and macroscopic effects)		
	4. feed consumption		
	5. body weight change		
	6. measured concentrations (LC_{50} study)	6. actual test concentrations should be at least 80% of nominal concentrations	6. E
	7. repellency[30]		
Pay attention	1. vomiting	1. if vomiting occurs the actual feed intake is unknown (lowered)	1. E
	2. repellency	2. if a substance is repellent actual feed intake is not known, and cause of death could be starvation rather than a toxic effect	
	3. LC_{50} test: stability/homogeneity of the substance in the diet	3. stability/homogeneity should be maintained throughout the test	3. Y
	4. LC_{50}: mortality at the lowest concentration	4. at the lowest concentration no toxic effects should appear	4. E

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[28] Unless the range finding test shows that LD_{50} >2000 mg/kg, or 8-day LC_{50} >5000 mg/kg feed.

[29] If applicable the following standard sentence is used: ♦ Feed and water were provided ad libitum.

[30] If repellency is claimed, or if feed consumption decreases with increasing test concentrations, see Luttik (1993).

4.6.3 Aquatic organisms (see Table 11)

According to the Dutch pesticide registration policy, short-term tests are required for three species from different trophic levels (usually algae, Daphnia, fish). Short-term tests with algae have to be performed according to OECD 201, NEN 6506, or Annex V.C3; short-term Daphnia tests according to OECD 202, NEN 6501, or Annex V.C2; and short-term fish tests according to OECD 203, NEN 6504, or Annex V.C1 (see Appendix 9).

Concerning long-term tests (if requested), Daphnia tests have to be performed according to EC Directive XI/681/86, and fish tests according to OECD 210.

The instructions in Table 11 are based on the Appendix H.2 of the CTB Application Form (Anonymous, 1995) and the OECD Guidelines 201, 202, and 203.

General information

The following order is used for summarising: first algae, then crustacea, fish, and other organisms. The TOXIS database does not provide the functionality of changing the order of tests in the output generated for the reviewer. It is therefore wise to pay attention to the order of studies when recording information in TOXIS.

If taxonomic revisions have resulted in changes in nomenclature, the new scientific names are used. If a report uses the old name, enter the new name in TOXIS, and mention the old name in the Remarks:
- 'new species name' previously named 'old species name'.

The latin name for rainbow trout has been changed since 1993: *Oncorhynchus mykiss* instead of *Salmo gairdnerii.*

The blue algae are classified as *Cyanophyta*, as to distinguish them from the green algae (which are classified as Chlorophyta).

Standard experiments with one-cell organisms (e.g. algae) lasting for 72 or 96 hours result in an EC_{50} and/or an NOEC. These sublethal endpoints are derived from short-term tests; however, they should be seen as endpoints for chronic toxicity due to the high turn-over rate of algae. All tests lasting $\leq$96 hours are recorded in TOXIS as short-term tests.

There are important differences between short-term and long-term tests. Naturally, the exposure time is longer in the latter (*Daphnia*, 14-21 days; fish, depending on species, e.g. 28 days). Further, the test system has to be either semi-static or flow through, never static. Also, feeding is allowed/required, in contrast with short-term tests. The studied effects are:

Daphnia:	effects on mortality, time of first production of young, number of young born, signs of intoxication;
fish:	effects on the stage of embryonic development, hatching and survival, abnormal appearance, abnormal behaviour, weight, and length (darkened skin is not considered relevant).

For more detailed information, the reader is referred to OECD 202 II (*Daphnia*, reproduction), and OECD 210 (fish, Early-Life Stage).

Table 11: Waterorganisms, short-term toxicity tests

	Items	Notes	Reliability lower?
Description	**1. test species**		
	2. test substance		
	3. concentrations (nominal)		
	3.1 number	3.1 ≥X concentrations [X= 5][31];	3.1 E
	3.2 range	3.2 test concentrations should not exceed water solubility >X times [X= 10]	3.2 Y
	4. use of vehicle	4. concentration vehicle <100 mg/l	4. E
	5. analysis method		
	6. age (crust.+fish); length+weight (fish)	6. *Daphnia*: maximum age 24h	6. Y
	7. number of animals	7 *Daphnia*: min. 20/conc.: preferably 4 replicas of 5; fish: min. 7/conc.	7. E
	8. test vessels	8. suitable for tested compound	
	9. loading		
	9.1 algae	9.1 algae: initial cell conc.= c. 10^4/ml	9.1 E
	9.2 Daphnia	9.2 *Daphnia*: max. 1 *Daphnia* per 2 ml	9.2 E
	9.3 fish	9.3 fish: max. 1 g/l (in flow-through tests the loading can be higher)	9.3 E
	10. control	10. vehicle: also solvent control should be tested	10. Y
	11. test system: static, renewal, flow through		
	12. exposure time	12. algae: 72-96h; *Daphnia*: 24-48h; fish: 96h	12. E
	13. test water/medium: temperature, pH, diss. O_2 (DO), hardness, salinity (seawater)	13. good quality natural water or reconstituted water; hardness 10-250 mg $CaCO_3$/l; pH 6.0-8.5; temperature: see OECD 201-203	13. Y
	14. feeding	14. no feeding	14. Y
	15. light conditions		
	15.1 algae	15.1 algae: source, continuous 120 $\mu E/m^2s \approx 8000$ lx	15.1 Y
	15.2 Daphnia	15.2 *Daphnia*: optional	15.2 E
	15.3 fish	15.3 fish: 12-16 h light per day	15.3 E
	16. effects studied	16. algae: biomass (b) or growth (r), *Daphnia*: immobility; fish: mortality and sublethal effects	16. E
	17. sampling frequency test concentrations	17. at least at the start and at the end of test	17. E
Results	**1. algae NOEC and EC_{50}, Daphnia EC_{50}, fish LC_{50}; 95% confidence limits**	1. preferably based on measured concentrations	1. E
	2. raw data	2. raw data should be available	2. Y
	3. mortality/effect	3. mortality/effect in the control should be <10% (or <1 fish if 7, 8, or 9 fish are used)	3. Y
	4. measured concentrations	4. measured test concentrations should be at least 80% of the nominal concentrations	4. Y
	5. pH, DO, temperature	5. pH and T should be constant; DO should be ≥60% of air saturation value; see Appendix 1.	5. E
	6. incipient LC50	6. incipient value should preferably be reached[32]	6. E
Pay attention.	1. the dissipation type	1. there should be no major loss due to hydrolysis, photolysis, volatilisation, or adsorption to glass. Is the test design adequate?	1. E
	2. log K_{ow}	2. bioconcentration, adsorption to glass and particle in solution may occur for lipophilic compounds	2. E
	3. effects at lowest and highest test concentration	3. lowest: no toxic effect should appear; highest: algae: at least 50% inhibition; *Daphnia*: 100% immobilisation is preferred; fish: no percentage mortality is mentioned.	3. E

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[31] The minimum number of test concentrations can be smaller if the range finding test shows that $L(E)C_{50}$ will probably be >100 mg/l (no mortalities at this concentration, else a full test should be performed), or if it is very likely that no mortalities will occur below the water solubility.

[32] If this is not the case, it can be required to repeat the experiment (Anonymous, 1995).

The actual averaged concentrations have to be used (and mentioned in the Results), if possible.

The Spearman-Karber model cannot be applied on algae because the measured effect is not monotonically increasing. Use the log-logistic model in GENSTAT for algae. For calculation of the NOEC see the instructions in the OECD201 Guideline.

In the Remarks it has to be reported whether the incipient L(E)C_{50} was reached or not. The incipient L(E)C_{50} is the L(E)C_{50} value that does not decrease in time any more. If an incipient L(E)C_{50} has not been reached, this indicates that the organisms tested may be more sensitive to the pesticide after a longer period of exposure.

If the L(E)C_{50} values of the last two (three) time points are the same, it is assumed that the incipient value is reached. If not, all the L(E)C_{50} values are put into a graph, and it is estimated whether the curve reaches a plateau; a minimum of three points is necessary to reach a conclusion, so for 48 hours tests (often with only two time points), the incipient value cannot be determined.

Dissipation of the substance

There should be no major loss due to photolysis, volatilisation, hydrolysis, adsorption to glass, etc. The test design should be adequate as to maintain >80% of the nominal concentration. In case photolysis occurs, tests with *Daphnia* and fish may be performed in the dark. In case of photolysis, volatilisation, hydrolysis, or adsorption to glass, a flow-through system might be adequate.

For tests with slightly soluble substances or rapidly hydrolysing substances some additional instructions are mentioned below.

Tests with slightly soluble substances[33]

In case very slightly soluble compounds (water solubility S <0.1 mg/l) are tested at concentrations up to the water solubility, and e.g. no effects are observed, the test in principle is reliable. However, the compound cannot be classified; only the 'bare' toxicity result is mentioned then (e.g. NOEC >0.05 mg/l). Toxicity values from a test in which the slightly soluble compound was tested at nominal concentrations that are larger than 10 times the solubility, should be regarded as unreliable (RI is 3).

The reader is referred to Vaal et al. (1992) for more information on the evaluation of slightly soluble substances.

Tests with rapidly hydrolysing pesticides [31]

The EC and OECD Test Guidelines have been devised for stable compounds. However, one can be confronted with an unstable pesticide.

According to the official Guidelines, the loss of test substance in an ecotoxicological test should be less than 20% to consider the compound stable enough for the purposes of toxicity testing. If this is not the case there is serious doubt if the test has been performed adequately, technically speaking. However, the high loss may be caused by fast hydrolysis of the compound; in this case the test has

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[33] Other difficult substances (e.g. volatile, strongly to glassware sorbing substances) are not dealt with in this Manual

been performed in a technically adequate way, in other words, the high hydrolysis rate is an intrinsic property of the compound and could not not have been avoided.[34]

If the loss of test substance is higher than 20%, first it should be checked if the loss is caused by bad performance of the test (in which case the RI is lowered), or by fast hydrolysis. The latter is done by inspecting the results of the hydrolysis test, and if these are not available (or in case of doubt), by consulting the specialist.

Once it is established that hydrolysis is the (main) cause for the high loss, the second question is if the metabolite(s) should have been tested instead of the parent compound (of course it can also be concluded that testing of both parent compound and metabolite(s) is necessary). The following limits[35]y are used:

$DT_{50} \geq 24$ h : the test is started with the parent substance
$DT_{50} < 4$ h : (n) tests are started with (n) metabolites
DT_{50} 4-24 h : expert judgement

In case the DT_{50} (hydrolysis) <4 h, and the toxicity test has been started with the parent compound, the result is considered as unreliable (RI is 3) because the (major) metabolite(s) should have been tested.

In all other cases (i.e. loss of test substance >20%, metabolites neither qualified nor quantified, DT_{50} (hydrolysis) ≥4 h), the test results are considered as less reliable (RI is 2), as it remains unclear which compound causes the observed effect(s) and at what concentration. The toxicity is expressed in terms of the nominal (initial) concentration (Whitehouse and Mallet (1993) use the term loading rate). Therefore the following standard sentences should be used in the Remarks:

- The toxicity is determined by a mixture of the parent compound and one or more transformation products because of rapid hydrolysis of the parent compound. Because these transformation products are not identified and quantified, the $L(E)C_{50}$ value in the Header is expressed in terms of the nominal concentration. The test is considered less reliable / unreliable.

In the RIVM Conclusion (see FEF, Appendix 8):

- A mixture of ...(parent compound) and unidentified and unquantified metabolite(s) is acute ...(classification) toxic for...(aquatic organism). This mixture was the result from rapid hydrolysis of the parent compound.

In the RIVM Conclusion —the subsection with the title **Aquatic organisms:**— the following should be stated (see FEF, Appendix 8):

- The $L(E)C_{50}$ values are expressed as nominal concentrations, due to rapid hydrolysis.

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[34] In some cases a different test system can avoid a high loss of the parent compound, for example applying a flow-through system instead of a (semi-)static system.

[35] The limits are derived from Whitehouse and Mallet (1993). The 12 hours limit is changed to 24 hours, because of the compatibility with the water/sediment transformation test (See Behaviour in surface water, General information, subsection 4.5.1).

Long-term exposure

For instructions on summarising and evaluating long-term toxicity tests the reviewer is referred to the OECD 202, the EC Directive XI/681/86, and the OECD 210 Guidelines.

Male daphnids may indicate bad culture conditions, and influence the number of offspring as well as the statistical analysis. Check the effect of the number of males on the calculated NOEC, as the latter should be based on females.

4.6.4 Bees, and other beneficial insects, mites, and spiders (see Tables 12a and 12b)

According to Dutch pesticide registration policy (Anonymous, 1995), arthropod tests have to be performed according to EPPO Guidelines on test methods for evaluating the side-effects of plant production products: EPPO Guideline no. 170 (for Honeybees); 142 (for Encarsia formosa), 151 (for Phytoseiulus persimilis); or EPPO Bulletin 15 (in case of other useful insects and mites). In 1994 the EPPO Guideline no. 180 (for Trichogramma cacoeciae) was published. Tests performed according to the latter four Guidelines are required, if the substance is claimed to be suitable for use in integrated pest control.

Three different types of arthropod tests can be distinguished, each corresponding to a different level in the sequential decision-making scheme (see Appendix 8, decision tree for bees, E. formosa and P. persimilis). At the first level, laboratory tests for oral (LD_{50}) and contact toxicity (LC_{50}) serve to classify the hazard of pesticides.

At the second level, a semi-field test is recommended to establish the duration of harmful activity, or to establish the effect of a dry pesticide film on plant or soil. Cage or tunnel trial tests fulfill this purpose by determining the effects on colony survival and development. In cage tests bees from small colonies are made to forage on a flowering crop in field cages. In tunnel tests the hazard of a pesticide to bees foraging the honey-dew secreted by cereal aphids is determined; for this purpose honey-dew is simulated by applying sucrose solution as a high volume spray, and bees from a small colony are made to forage on the sucrose.

At the third level a field test is recommended to show the effect of a direct spray of pesticide on plant or soil, inhabited by beneficial arthropods. Bee colonies are placed in or on the edge of large test fields, and bees are made to forage on a flowering crop in the adjacent test field only. Effects on colony survival and development are determined.

In case the product is an insect growth regulator, the toxicity to bee brood should be determined, and based on the resulting NOEL advancement to laboratory or cage tests is established; a suitable bee brood test method is described in EPPO Bulletin 22 (1992).

The instructions in Table 12a and 12b are based on the EPPO Guideline 170 (Honeybees) and EPPO Bulletin 15 (other arthropods).

Only oral LD_{50} values are considered relevant. Contact toxicity studies (topical application) are considered less relevant and are not used for RIVM Conclusions.

For the recording in TOXIS: give spiders and mites the code SPE in the TOXIS Fixed Field 'tests'. These Summaries are then printed in the AR under Special Ecological Studies.

Table 12a: Bees: LD_{50} studies

	Items	Notes	Reliability lower?
Description	**1. test substance**		
	2. species	2. preferably young adult worker bees; age and race should be known	2. Y
	3. oral exposure		
	3.1 test solution	3.1 10 or 20 µl test solution, containing 20-50% sucrose solution	3.1 E
	3.2 starvation	3.2 maximally two hours before test	3.2 Y
	4. applied concentration(s)	4. minimal five for correct statistical interpretation	4. E
	5. vehicle	5. acetone is preferred	5. E
	6. duration of test		
	6.1 treatment time (LD_{50} studies)		
	6.2 observation time		
	7. feed type		
	8. number of animals	8. min. 3 groups of 10 bees/concentration	8. E
	9. cage	9. cages should not cause control mortality; cages should be ventilated well	9. Y
	10. temperature	10. 25°C ± 2°C	10. Y
	11. light condition		
	12. humidity	12. 60 - 70% relative humidity	12. E
	13. method of anaesthesia (LD_{50} study)	13. e.g. CO_2: application should not lower temperature in cage	13. Y
	14. (vehicle) control	14. vehicle used: solvent control should be tested	14. Y
	15. reference substance (positive control)	15. e.g. parathion, dimethoate	15. Y
Results	**1. LD_{50} and 95% confidence limits**	1. only oral LD_{50}'s are relevant	1. Y
	2. mortality in control groups	2. should not exceed X% [X = 15]	2. Y
	3. overt signs of toxicity		
Pay att.	1. method of collection of bees	1. collection in early spring or late autumn should be avoided	1. E
	2. correction for control mortality		
	3. housing	3. bees should not be confined for more than one hour	3. E

Studies according to EPPO Guidelines

Instructions on how to evaluate laboratory tests are given in Tables 12a and 12b. In case of laboratory tests with other insects, mites, spiders, always check with the EPPO Guidelines (142, 151, 180) or EPPO Bulletin 15, for species-specific instructions on testing and evaluating. For instructions on semi-field and field tests with bees or other insects, mites, and spiders, see EPPO Bulletin 170 or 15.

After correction for control mortality, the LD_{50} can be calculated with an appropriate statistical method. If the control mortality >15%, the test should be repeated.

Instructions on how to evaluate tunnel experiments with bees, field experiments with bees, and bee brood tests, are under development.

Studies with insects, mites, and spiders, not according to EPPO-Guidelines

Other bee studies are evaluated according to the instructions in Table 12a. Other studies with beneficial insects, mites, and spiders are evaluated according to the instructions in Table 12b.

Bee studies according to BBA 23-1 (1980) Guidelines

Only the *oral* toxicity test of BBA 23-1 is comparable to the EPPO Guidelines. However, the temperature can be too low, the solvent control may be lacking, and the maximum number of concentrations (three) is too low for a proper dose-response relation. For these reasons this test might have a limited value.

The test results of testing the toxicity of exposure by contact, inhalation, and to spray aerosoles, or dispersed powder are not comparable to the EPPO Guideline. These tests should nevertheless be summarised (see BBA 23-1 guideline), but they are not used for the RIVM Conclusion.

Reviews

Bee studies reviewed in Beran (1970), Kupetz et al. (1979), and Stevenson (1978) get the RI of 1. Bee studies reviewed in Atkins et al. (1981) get the RI of 2. In TOXIS these studies have the reference numbers 714, 715, 716, and 717, respectively. The *Encarsia* studies of Oomen et al. (1994) (TOXIS reference 1646) get the RI of 1. In the Description the next standard sentence is included:

- Further information in referred literature.

Table 12b: Insects (bees excluded), mites and spiders, L(E)C_{50}/NOEC studies

	Items	Notes	Reliability lower?
Description	**1. test substance**		
	2. test species	2. species must be relevant for the crop; preferably laboratory-reared, uniform in age[36]	2. Y
	3. route of exposure	3. expose to fresh dry pesticide film; depending on behaviour of species use as exposure target: glass plates, plant leaves, or soil	3. Y
	4. test concentration(s)		
	4.1 nominal dose	4.1 recommended (field) concentrations	4.1 E
	4.2 actual dose	4.2 dose measured by weighing the target	4.2 E
	4.3 control dose	4.3 control groups with water application	4.3 Y
	5. vehicle	5. standard amount of fluid: 1-2 mg/cm^2 (glass and leaf), 6 mg/cm^2 (soil)	5. Y
	6. duration of test	6. adequate exposure period	6. E
	7. feed type		
	8. number of animals	8. number per test and per vessel depends on species; see Guideline	8. Y
	9. ventilation	9. adequate ventilation	9. Y
	10. housing conditions	10. see EPPO Guidelines	
	11. test conditions	11. see EPPO Guidelines	
Results	**1. reduction in beneficial capacity/mortality compared to controls**	1. see EPPO Guidelines (e.g. mortality, egg laying, feeding)	
	2. beneficial capacity/mortality controls		
	3. overt signs of toxicity		
Pay att.			

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[36] *Pardosa* species (wolf spiders): a breeding method has not been developed yet. Spiders collected in the field are allowed for testing.

4.6.5 Soil organisms (see Tables 13, 14)

According to the Dutch pesticide registration policy, nitrification tests have to be performed according to NEN 5795. Nitrification tests are always required, independently whether the pesticide is incorporated into the soil or not. A test on the effects on soil respiration is not obligatory, according to the Dutch registration policy.

According to the Dutch pesticide registration policy, acute tests on earthworms have to be conducted according to OECD 207, ISO 11268-1, or NEN 5794. Effects on reproduction of earthworms can be conducted according to NEN 5797.

The instructions in Table 13 and Table 14 are based on NEN 5795 Guideline, and Appendix H.4 of the CTB Application Form (Anonymous, 1995) and OECD Guideline 207, respectively.

Micro-organisms and enzymes in soil and manure

A specific feature of tests with micro-organisms is that the uptake of substances is in general very quick. Therefore the effect can be apparent after half an hour up to two hours after application. Also the adaptation can be very quick.

Four types of tests can be distinguished:

1. single species test (e.g. Microtox);
2. test on the activity of enzyms (e.g. dehydrogenase, phosphatase, arylsulphatase);
3. test on soil processes (e.g. respiration, nitrification);
4. test on microbial diversity.

Nitrification tests have to be performed with at least two soil types, relevant for the Dutch situation. Nitrification is a process in which several species of micro-organisms are involved. The process consists of the oxidation of ammonium to nitrite and the subsequent oxidation of nitrite to nitrate. The proces of nitrification is relatively susceptible to disturbance.

Sometimes tests have been carried out in which the effects on ammonification (organic-N to NH_4^+) or on denitrification (NO_3^- to N_2) were measured.

The Microtox test is carried out with e.g. *Photobacterium phosphoreum*, a salt water bacteria. The inhibition of light production is measured.

Storage conditions of sampled soil, that is not immediately used, are preferably as follows: in the laboratory at 4 °C for at most three months (to avoid anaerobic conditions); in the open or in a glasshouse under well-drained conditions (to avoid desiccation)

Table 13: Micro-organisms and enzymes in soil and manure

	Items	Notes	Reliability lower?
Description	1. **test substance**		
	2. **vehicle**		
	3. **applied concentrations**	3. min. X concentrations [X= 2]: the recommended dose, and ten times the recommended dose	3. E
	4. **soil**[37] 4.1 soil type (US-class.) 4.2 pH 4.3 CEC 4.4 % o.m.		
	5. analysis method		
	6. sampling frequency	6. min. X samples [X= 2]: after 7 or 14 days, and after 21 days	6. E
	7. **additives (lucerne meal, ammonia)**	7. micro-organisms can be influenced negatively by too high concentrations of some additives (e.g. ammonium sulphate)	
	8. ***light condition***	8. dark conditions are preferred	8. E
	9. ***temperature***	9. temperature should be X °C [X= 15 - 25]	9. Y
	10. ***moisture content***	10. pF 2 - 3	10. Y
	11. ***vehicle control (if applicable)***		
Results	1. **% reduction of level of enzymatic or other biochemical processes (with/without additives)**	1. relative to control	1. Y
	2. **time of recovery of activity**		
Pay atten.	1. the agricultural history soil	1. no manipulation with fertiliser, no (prior) use of pesticides that may have lead to adapted micro-organisms (in the previous five years). Special attention should be paid to compounds interfering with the N-cycle in the soil.	1. E
	2. storage	2. if there is no immediate use, storage in the lab or in the open should be appropriate (see text)	2. Y

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[37] Nitrification tests should be performed with two soil types.

Earthworms, LD_{50}/LC_{50} studies

In TOXIS the scientific name of worms can only consist of two words:
Eisenia foetida andrei becomes *Eisenia andrei*;
Eisenia foetida foetida becomes *Eisenia foetida.*

The risk assessment for earthworms is based on an acute test in soil. The soil can be a natural soil, or an artificial (laboratory composed) soil. An example of artificial soil test substrate (OECD 207): 10% sphagnum peat, 20% kaolin clay; 70% industrial sand; calcium carbonate is added to adjust the pH to 6.0 ± 0.5.

An LC_{50} value from a test with filter paper, or from an Artisol test (a medium of silica gel) is less useful for risk assessment. A description of an Artisol test is found in Reinecke (1992).

Earthworms, reproduction studies

Tests performed according to the ISO 11268-2 (draft) Guideline on reproduction of earthworms are not yet accepted officially.

Table 14: Earthworms, LD$_{50}$/LC$_{50}$ studies

	Items	Notes	Reliability lower?
Description	**1. test substance**		
	2. test species		
	3. applied concentration(s)	3. X treatment levels (geometric series) [X= 5], unless range finding test shows that LC50 >1000 mg/kg soil	3. E
	4. vehicle		
	5. analysis method		
	6. way of exposure (filter paper contact test or artificial soil test)	6. filter paper contact test or Artisol test are considered less relevant	6. Y
	7. medium:	7.	
	7.1 artificial soil	7.1 pH 6.0 ± 0.5 and moisture content should be c. 35% of dry weight	7.1 Y
	7.2 natural soil: soil type (US-class.); pH, CEC, % o.m.		
	8. duration of exposure and observation period	8.	
	8.1 artificial soil test	8.1 mortality is assessed 7 and 14 days after application	8.1 E
	9. number of worms per concentration	9.	
	9.1 artificial soil test	9.1 Four replicas/treatment level and 10 worms/replicate	9.1 E
	9.2 natural soil test		
	10. age and weight		
	10.1 age	10.	
		10.1 worms should be adult (min. 2 months old with clitellum)	10.1 Y
	10.2 weight	10.2 individual wet weight should be 300-600 mg	10.2 E
	11. temperature	11. 20 ± 2°C	11. E
	12. light condition	12.	
	12.1 artificial soil test	12.1 test should be performed under continuous light: illuminated cabinet or chamber controllable to 20 ± 2°C with a light intensity of 400-800 lx	12.1 E
	12.2 natural soil test		
	13. vehicle control (if applicable)		
Results	**1. LC$_{50}$ or LD$_{50}$ value, and 95% confidence limits**		
	2. mortality in control groups	2. the mortality in the controls should not exceed X% at the end of either test [X= 10]	2. Y
	3. overt signs of toxicity		
	4. bodyweight change		
Pay att.	1. moisture content	1. pay attention: e.g. 35% of dry weight is not the same as 35% of WHC	

4.6.6 Activated sludge (see Table 15)

According to the Dutch pesticide registration policy (Anonymous, 1995), tests have to be performed according to OECD 209, NEN 6511, or NEN 6512, if the substance is expected to be discharged to sewage treatment plants. Information on the effect on the respiration or TOC-removal, and on the effect on nitrification, of adapted activated sludge is required.

The instructions in Table 15 are based on OECD Guideline 209.

Instructions for the evaluation of tests on the effect on the nitrification of adapted activated sludge are under development.

Summaries are recorded in TOXIS under Toxicity tests. TOXIS prints this information in the ARs in Chapter 4.1.2 Aquatic Organisms.

Table 15: Influence on activated sludge (respiration)

	Items	Notes	Reliability lower?
Description	**1. test substance**		
	2. applied concentration(s)	2. at least X concentrations [X= 5] should be tested; difference between concentrations should not exceed factor 3.2	2. E
	3. test system (e.g. BOD-flask)		
	4. duration test	4. 30 minutes or 3 hours 'contact'	4. E
	5. type of microbial inoculum	5. usually activated sludge from a sewage treatment plant (STP)	
	6. source of sludge	6. e.g. a municipal or an industrial STP	
	7. temperature		
	8. air supply	8. aeration should take place	8. Y
	9. nutrient solution		
	10. controls	10. the two control respiration rates are within 15% of each other	10. Y
	11. reference substances	11. at least three concentrations of 3,5-dichlorophenol: the EC_{50} (3 hours) of 3,5-dichlorophenol must be in the range 5 - 30 mg/l	11. Y
Results.	**1. EC_{50} and 95% confidence limits**		
Pay attent.	1. water solubility	1. test concentrations should not exceed water solubility	1. E
	2. the dissipation type	2. there should be no major loss due to hydrolysis, photolysis, or volatilisation: is the test design adequate?	2. E

4.7 Bioconcentration in waterorganisms (see Table 16)

According to the Dutch pesticide registration policy (Anonymous, 1995), bioconcentration tests have to be conducted according to OECD 305 A - 305 E, with a clear preference for E.

The bioconcentration should be investigated in a study with fish or shell-fish, in case of non-organic compounds, or organic compounds with a log K_{ow} >3.

The instructions in Table 16 are based on OECD Guideline 305 E.

Bioconcentration in water organisms, calculation of BCF from log K_{ow}

The BCF can be calculated from the log K_{ow} with the next equation, used in USES 1.0 (RIVM, VROM, WVC, 1994):

$$\text{BCF(fish)} = 0.05 \times K_{ow}$$

This equation is based on the assumption that the partitioning of the compound between the lipids of the organism and water equals the partitioning between octanol and water. The factor 0.05 is introduced as it is assumed that 5% of the total weight of the organism consists of lipids. Hence when it is known that the organism has a substantial higher or lower lipid content, this equation cannot be used. Furthermore, this equation is valid for many neutral organic compounds, but for other compounds deviations have been found.

The equation is only used for organic compounds meeting the following conditions:
- log K_{ow} <6;
- molar mass <500;
- water solubility <1000 mg/l.

Bioconcentration in waterorganisms, studies with organisms

For compounds with a water solubility >1000 mg/l, or a logK_{ow} <4.3, it may be sufficient to calculate the BCF from the logK_{ow}.

The BCF should be based on fat weight (BCF_{fw}) (for organics with the exception of organo-metals), or on total wet weight (BCF_{wo}) (for e.g. dissociating compounds).

Studies based on OECD 305 A - E Guidelines should be checked with these Guidelines for evaluation. These Guidelines differ from each other with respect to the test system and the mathematical interpretation of the results.

The OECD Guidelines 305 B-E use the model mentioned below. Guideline 305 A uses a different model, therefore the equations mentioned below cannot be used for

Table 16: Bioconcentration in waterorganisms, studies with organisms

	Items	Notes	Reliability lower?
Description	**1. test substance**		
	2. test species		
	3. applied concentration(s)	3.1 min. X concentrations [X= 2]	3.1 Y
		3.2 highest concentration $<(0.1 \times LC_{50})$; lowest concentration $>(10 \times$ detection limit)	3.2 E
	4. vehicle	4. should not exceed 0.1 ml/l	4. E
	5. analysis method		
	6. test system		
	7. exposure time and depuration time	7. uptake phase: 3 hours - 30 days; depuration phase: 6 hours - 60 days, or 3 × DT_{50}	7. E
	8. age, length, weight of the organisms		
	9. number of animals per group		
	10. loading		
	11. (solvent) control		
	12. type of water (pH, DO, etc.)	12. DO should not vary more than ± 3 mg/l	12. E
	13. temperature	13. should not vary more than ± 1°C	13. E
	14. feeding		
	15. light condition		
	16. sampling frequency		
	16.1 samples of test water		
	16.2 samples of organisms	16.2 uptake phase: min. X [X= 4]; depuration phase: min. X [X= 5]	16.2 E
Results	**1. BCF**	1.1 preferably based on whole body wet weight or lipid content	1.1 E
		1.2 based on a.i., not on r.a.; give C_{water} and C_{fish}	1.2 Y
	2. steady state	2. steady state reached: yes/no, time point	2. E
	3. rate constants	3. k_1 (uptake) and k_2 (depuration); duration of phases should be sufficient	3. E
	4. k_2 and steady state	4. the time to reach 50% of the equilibrium concentration (steady state C_f) should equal the half-life for depuration[38]	4. E
	5. measured concentrations in water	5. should be (in water) at least 80% of the nominal concentrations	5. E
	6. signs of toxicity	6. no toxic effects should occur	6. Y
Pay atten.	1. test concentrations	1. test concentrations should not exceed water solubility, and should be <1 mg/l	1. E
	2. loss of test substance	2. there should be no major loss due to hydrolysis, photolysis, biotransformation in water, volatilisation, and adsorption to vessel or particles: is the test design adequate?	2. E

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[38] A difference of a factor X [X= 2] is acceptable.

305 A. The Guidelines 305 B-D consider only the calculation of the bioconcentration factor. When in these tests no steady state is reached, no bioconcentration factor can be determined, which means that the result is quite useless for conclusions. This does not apply for the 305 E Guideline, because the rate constants that are determined in this test give insight in the behaviour of the chemical in the environment.

The OECD 305 B - E Guidelines are based on the next model. The mass balance in the system consisting of water, organism (fish, or other) and test compound is:

$dC_f / dt = k_1 \times C_w - k_2 \times C_f$ [equation 1]

in which C_f is the concentration of test compound in the organism [µg/kg], C_w is the concentration in water [µg/l], t is time [d], k_1 is the uptake rate constant [$l.kg^{-1}.d^{-1}$], and k_2 is the elimination rate constant [d^{-1}]. The elimination rate constant k_2 describes every elimination process of the test compound from the organism, hence it includes physico-chemical elimination and biotransformation.

Integration of equation 1 is only possible when C_w is constant. When C_w declines it must be checked if the authors included this in their calculations of the rate constants. If not, the rate constants can be recalculated using BIOFIT (Gobas & Zhang, 1992). BIOFIT can also be used if the C_w is constant.

Assuming C_w is constant, the next calculations should be performed. Consider equation 1 during the phase of initial uptake, C_f will be then negligible:

$dC_f/dt = k_1 \times C_w$, or $C_f = k_1 \times C_w \times t$

However, it is difficult to determine when C_f ceases to be negligible, so it is hard to use this formula in a good way. Another way to determine the uptake rate constant is described in OECD 305 E. For this method the k_2 is nessecary.

Consider then equation 1 during period of elimination instead of uptake, C_w is then negligible:

$dC_f/dt = -k_2 \times C_f$ or $C_f = e^{(-k2 \times t)}$

When lnC_f is plotted against time, the slope of the straight line is k_2; k_2 can then be used to determine k_1. Integration of equation 1 gives:

$C_f = k_1/k_2 \times C_w \times [1 - e^{(-k2 \times t)}]$

k_1 is then calculated as (t means time point at which the C_w and C_f are determined):

$k_1 = C_f \times k_2 / C_w \times [1 - e^{(-k2 \times t)}]$

Finally, for the bioconcentration factor, consider equation 1 at equilibrium:

$k_1 \times C_w = k_2 \times C_f$

From this equation it is clear that the bioconcentration factor (K_c or BCF (l/kg)) is defined by:

$$BCF = k_1 / k_2 = C_f / C_w$$

The first part of this equation ($BCF = k_1 / k_2$) can be used to determine the BCF also when no equilibrium in the test has been reached. The second part ($BCF = C_f / C_w$) can never be used to calculate BCF when equilibrium has not been reached.

The k_2 or half-life (DT_{50}) determines when equilibrium will be reached. The time required to reach 50% of equilibrium concentration in the organism equals the half-life in the depuration part of the experiment. Whether the time to reach 50% equilibrium and the halflife are comparable or not, should be checked in the original uptake curve. If this is not the case, the RI of the test is lowered.

Equation 1 considers first-order, one-fish compartment kinetics. However, more-fish compartments are possible. When more compartments are present, more rate constants are needed to describe the kinetic behaviour of the compound.

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[39] The ministry of WVC has been renamed in Aug. 1994 into VWS (Volksgezondheid, Welzijn en Sport)

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Appendix 1: Useful formulas, units, and air saturation values

General

deci	(d)	$= 10^{-1}$	deca	(da)	$= 10^{1}$
centi	(c)	$= 10^{-2}$	hecto	(h)	$= 10^{2}$
milli	(m)	$= 10^{-3}$	kilo	(k)	$= 10^{3}$
micro	(µ or u)	$= 10^{-6}$	mega	(M)	$= 10^{6}$
nano	(n)	$= 10^{-9}$	giga	(G)	$= 10^{9}$
pico	(p)	$= 10^{-12}$	tera	(T)	$= 10^{12}$

Temperature

$$x = \frac{y - 32}{1.8}$$

$x : {}^{\circ}C$
$y : {}^{\circ}F$

Pressure

The vapour pressure is calculated with:

$$\log P = \frac{0.05233 \times a}{T} + b$$

P = vapour pressure [mm Hg]
T = temperature in Kelvin [K]
a and b are constants, that can be calculated,
if the vapour pressure is known for two or more temperatures

- 1 mm Hg = 133 Pa [N/m^2]
- 1 mm Hg = 1 Torr
- 760 mm Hg = 101300 Pa
- 101300 Pa = 1 atm
- 1 atm = 1 bar
- 1 psi (pounds per square inch) = 6860 Pa

Soil

- CEC: 1 meq/100 g = 10 mmol/kg

The pF is expressed in [log $cm_{water\ column}$]. Alternative units are [bar] and [Pa]. When reading the pF scale in [bar] (see Appendix 2), be aware that the bar scale is logarithmic.

pF	cm_{H20}	bar	kPa
1	10	0.01	1.0
2	100	0.1	10
3	1000	1	100
4	10000	10	1000

Water hardness

- German degree (dH): 1° hardness = 17 mg/l as $CaCO_3$, 10 mg/l as CaO
- French degree: 1° hardness = 10 mg/l as $CaCO_3$
- English degree: 1° hardness = 1 grain/gallon as $CaCO_3$
  1 grain/gallon as $CaCO_3$ = 10 mg/l as $CaCO_3$
- American degree: 1° hardness = 1 mg/l as $CaCO_3$

Recalculation from English/American units

Length	**Volume**
1 inch = 2.54 cm 1 foot= 12 inches = 30.48 cm 1 yard = 3 feet = 0.9144 m 1 mile = 1.60934 km	1 cubic inch = 16.3871 cm^3 1 cubic foot = 28.3168 dm^3 1 cubic yard = 0.76455 m^3 1 pint = 1/8 engl. gallon = 0.568261 dm^3 1 quart = 1/4 engl. gallon = 1.13652 dm^3 1 engl. gallon = 4.54609 dm^3 1 amer. gallon = 3.785 dm^3 1 fluid pint = 1/8 amer. gallon 1 fluid quart = 1/4 amer. gallon
Area	**Weight**
1 sq. inch = 6.4516 cm^2 1 sq. foot = 9.290304 dm^2 1 sq. yard = 0.8361 m^2 1 sq. mile = 2.59 km^2 1 acre = 4047 m^2	1 grain = 64.7989 mg 1 ounce (oz) = 28.35 g 1 pound (lb) = 0.453592 kg

Toxicity

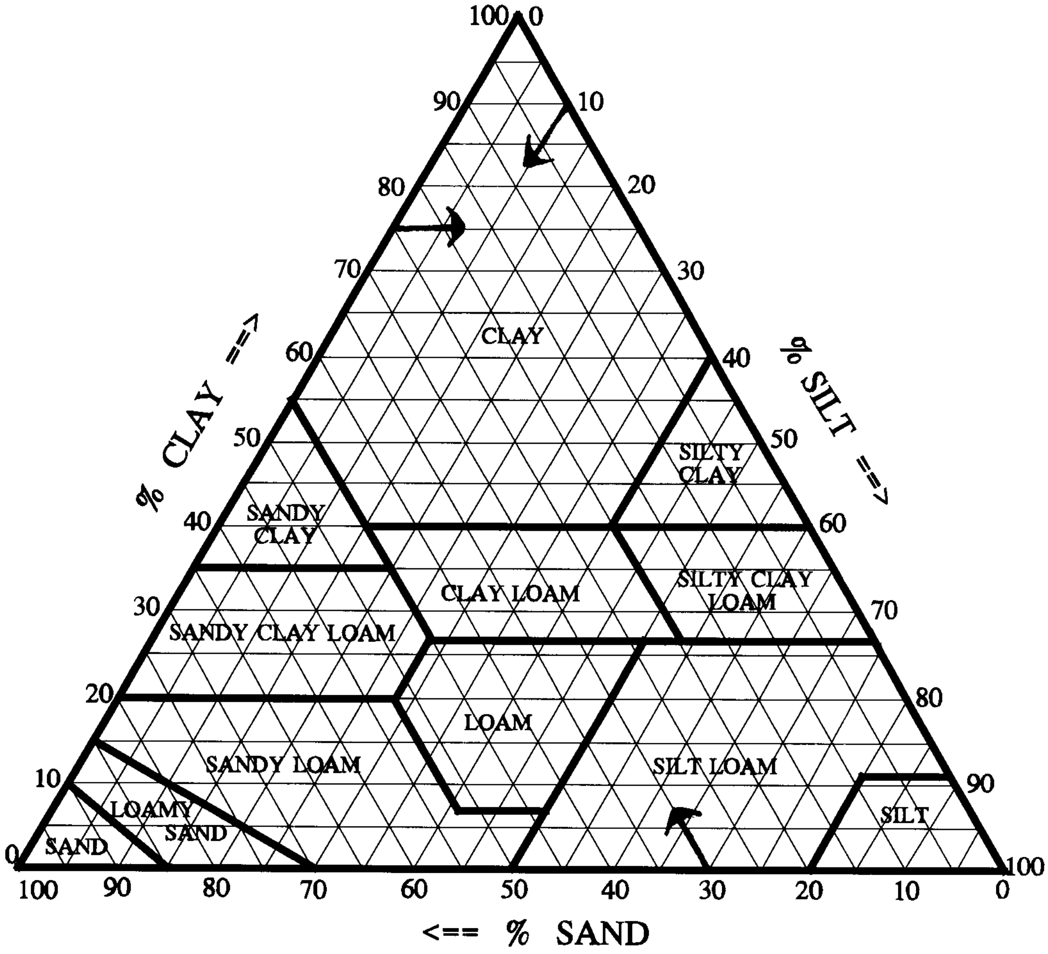
TLm (median tolerance limit) is comparable to the LC_{50}.
MATC is the mean of the NOEC and LOEC.

O_2 saturation in water

temperature °C	Solubility of oxygen in mg/l (100% saturation at 1 atm)				
	fresh water	seawater 5 g Cl/l	seawater 10 g Cl/l	seawater 15 g Cl/l	seawater 20 g Cl/l
10	11.3	10.7	10.1	9.6	9.0
11	11.1	10.5	9.9	9.4	8.8
12	10.8	10.3	9.7	9.2	8.6
13	10.6	10.1	9.5	9.0	8.5
14	10.4	9.9	9.3	8.8	8.3
15	10.2	9.7	9.1	8.6	8.1
16	10.0	9.5	9.0	8.5	8.0
17	9.7	9.3	8.8	8.3	7.8
18	9.5	9.1	8.6	8.2	7.7
19	9.4	8.9	8.5	8.0	7.6
20	9.2	8.7	8.3	7.9	7.4
21	9.0	8.6	8.1	7.7	7.3
22	8.8	8.4	8.0	7.6	7.1
23	8.7	8.3	7.9	7.4	7.0
24	8.5	8.1	7.7	7.3	6.9
25	8.4	8.0	7.6	7.2	6.7
26	8.2	7.8	7.4	7.0	6.6
27	8.1	7.7	7.3	6.9	6.5
28	7.9	7.5	7.1	6.8	6.4
29	7.8	7.4	7.0	6.6	6.3
30	7.6	7.3	6.9	6.5	6.1

Appendix 2: Soil classification, pF graphs, and concentrations in soil after multiple applications

Soil classification

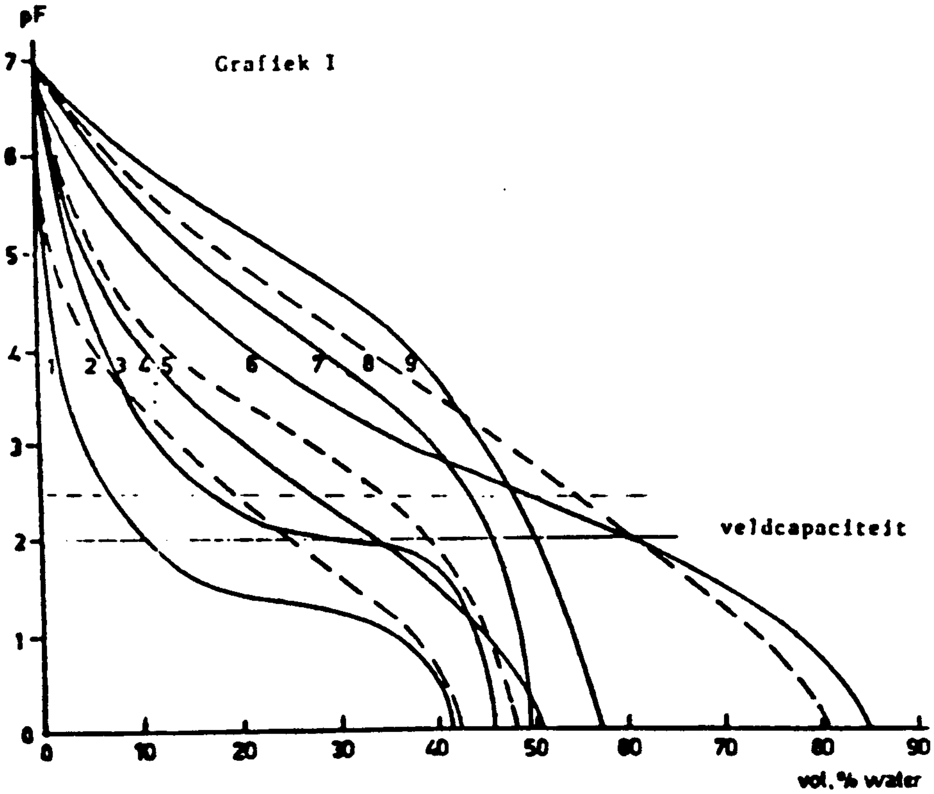


Textural classes of mineral soils according to the US soil classification. Particle sizes: clay <2 µm, silt 2 - 50 µm, sand >50 µm. USDA (1951).

pF-graphs

Different soil classes and their pF graphs (= 'curve')

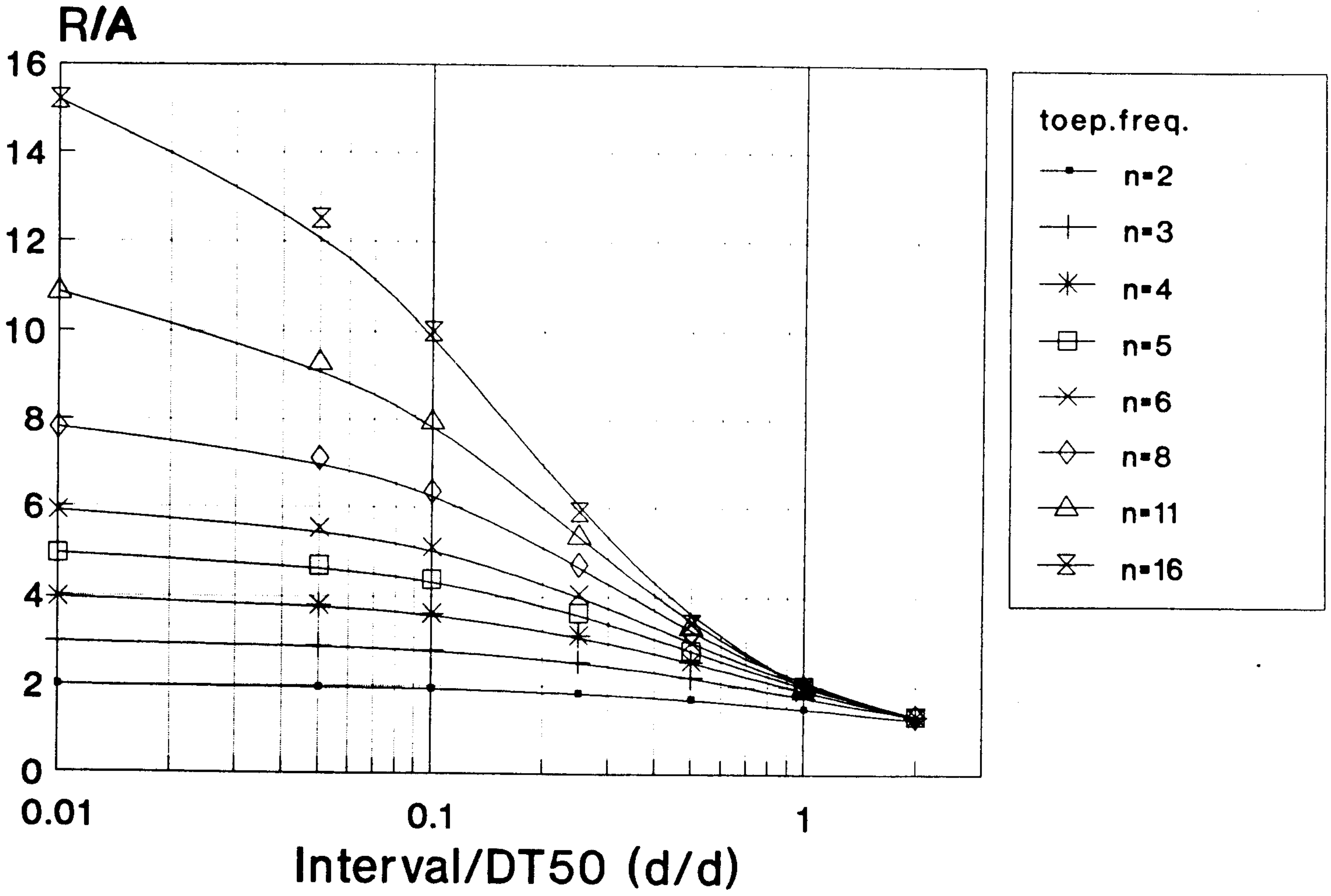
clay	zware klei	curve 9
sandy clay	klei	curve 7
silty clay	klei	curve 7
clay loam	klei	curve 7
silty clay loam	klei	curve 7
sandy clay loam	zavel	curve 4
loam	zavel	curve 4
sandy loam	lichte zavel	curve 3
silt	löss	curve 5
silty loam	löss	curve 5
loamy sand	lemig zand	curve 2
sand	duinzand	curve 1
peat	laagveen	curve 8
muck	veen	curve 6 or 8



Graph: pF graphs of some Dutch soils:
1. dune sand (Westland); 2. loamy sand (Venlo); 3. sandy loam (15% $CaCO_3$)(Polder "Het Grootslag", N-Holland); 4. loam (15% $CaCO_3$)(Polder "Het Grootslag", N-Holland); 5. silt/silty loam (Maastricht); 6. "Bolster" muck (Drenthe); 7. sea clay (Hoog Karpsel, N-Holland); 8. muck (Boskoop); 9. clay (Bommelerwaard)

Assessment of the moisture content of the soils of curves 1 t/m 7			
curve	soil type	40% MWHC	75% FC
1	sand	too wet	correct
2	loamy sand	correct	correct
3	sandy loam	correct	correct
4	sandy clay loam loam	correct	correct
5	silt silty loam	too dry	correct
7	sandy clay silty clay clay loam silty clay loam	too dry	too dry

Concentrations in soil after multiple applications



Application rates in soil experiments should be within the range of half to two times the recommended dosage. In case of multiple applications, this should be checked with the graph above. R is the (theoretical) dose after n applications [kg/ha], A is the dose per application [kg/ha], Interval is the interval between the dosages [days], DT_{50} is the biotransformation half-life [days].

Example: (single) dose is 3 kg a.i./ha; DT_{50} is 50 days; number of applications is 5; interval is 10 days. Then Interval/DT_{50} value on the X axis is 0.2. The corresponding R/A value on the Y axis (in this case determined via the line representing n = 5) is c. 3.8. The residu after the fifth application (R) equals 3.8 × 3 = 11.4 kg/ha. Therefore dosages in soil experiments should be within the range of 5.7 - 23 kg a.i./ha.

Appendix 3: Hockey-stick model on determining the transformation kinetics in soil

This Appendix gives a short description on how to determine a DT_{50} if during the transformation different phases can be distinguished. It could concern a succession of two first-order processes (with different transformation rates), resulting in a hinge point in the transformation curve.
To determine if a significant hinge point is present in the curve, the 'Hockey-stick' model in GENSTAT is used. The hinge point should be observed within the first 100 days after application of the substance. The 'Hockey-stick' model is available on the computer in the ACT library.

What to do?

Keys that should be pressed are given in brackets <>.

1. Feed the data into a WP file, that should be made under c:\genstat\jandata; **a zero (0)** cannot be included, but instead (e.g.) a **0.0001** can be used. Per line, **one** time point and corresponding concentration can be included. End the file by typing < :> behind the last concentration. The file should be saved as a DOS text: successively **<ctrl F5>, <1>, and <1>**. This file should be saved with the extension **.dat** (e.g. cynmethyl.dat). Then leave WP.
2. Start the 'Hockey-stick' model by typing **<ne hockey.gen>**. The data file (e.g. cynmethyl.dat) can be connected by typing the name of the desired data file at: **2. open name='...'**. Then leave by **<F3><e>**. The output file is generated by **<genstat hockey.gen cynmethyl.out><rtrn> (the output file has the extension .out)**. The output file can be examined by the DOS command **<type cynmethyl.out ¦ more>**. The file can also be printed (directly from the library computer or - if copied on a disc - on another printer). Print from WP.

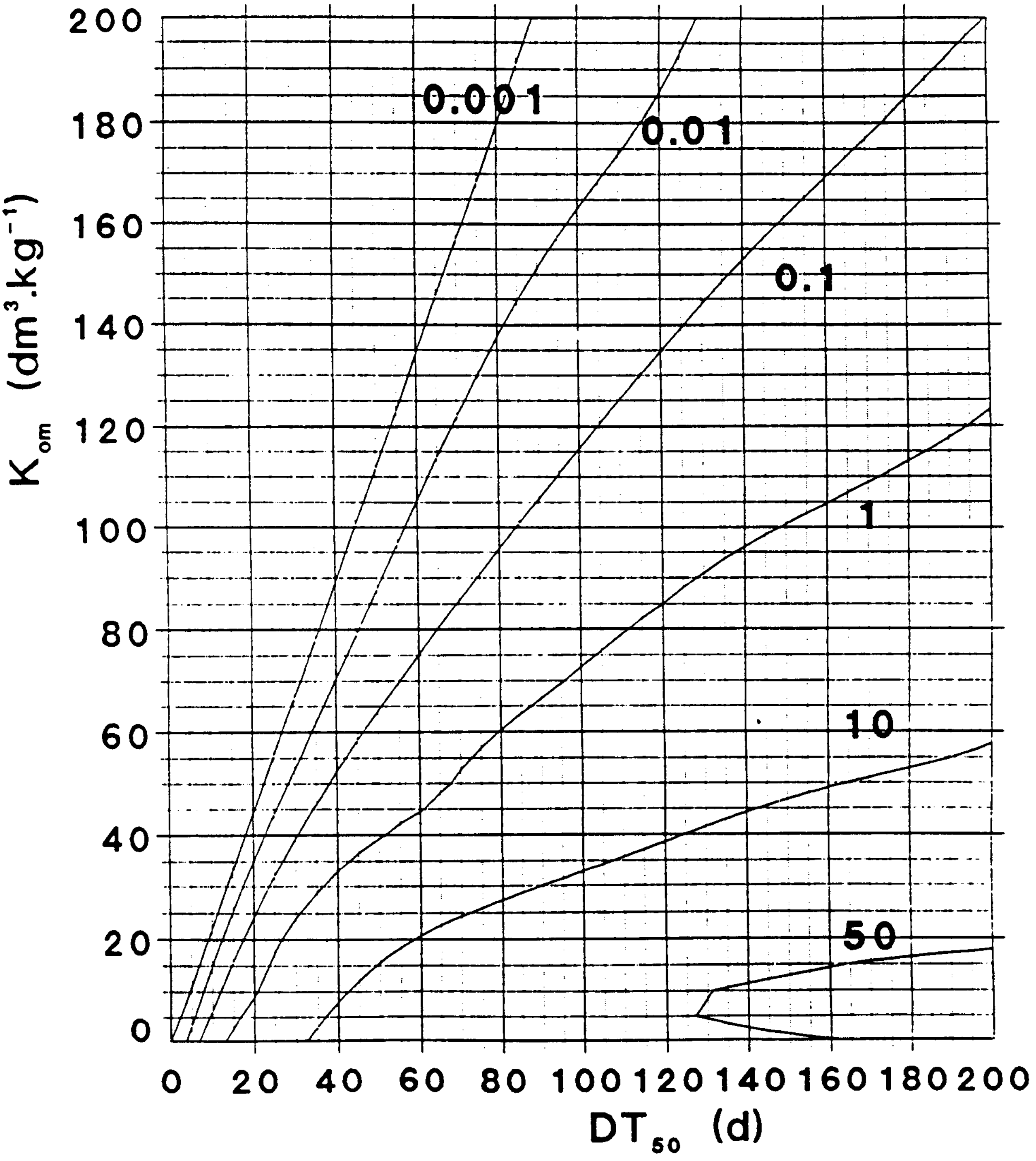
Interpretation

The place of the hinge point is indicated with δ under 'Estimates of parameters'.

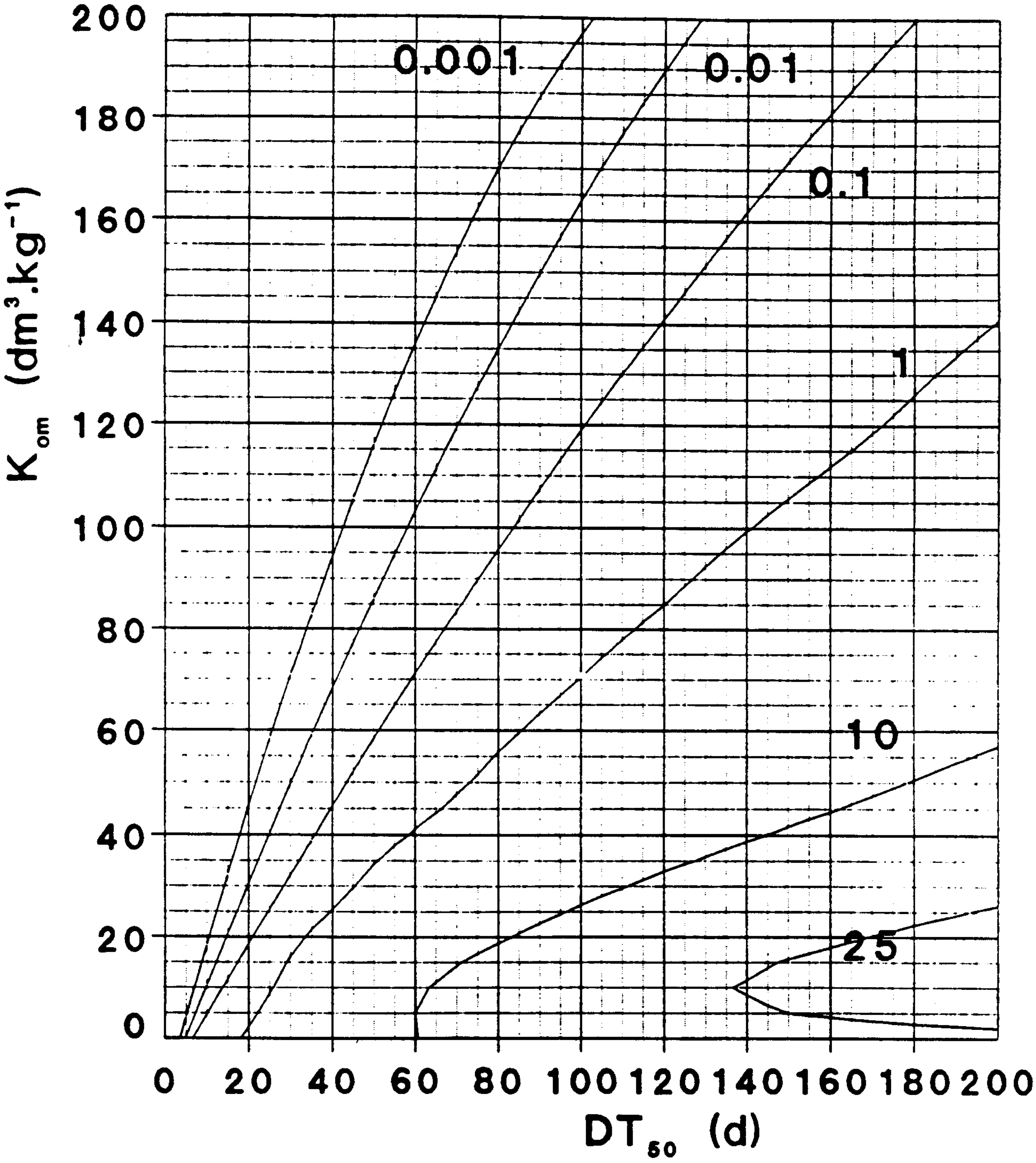
The 'p value' refers to the chance that both samples are from the same population. The p value is printed at the end of the output file. The significant level α is 5%, or 0.05. In case p ≤ 0.05 the zero hypothesis is rejected (there is a significant hinge point in the curve), and in case p >0.05 the zero hypothesis is accepted (there is no significant hinge point in the curve). If a * is printed, this means that there is no significant hinge point in the curve.

Appendix 4: PESTLA model on accumulation and leaching (graphical)

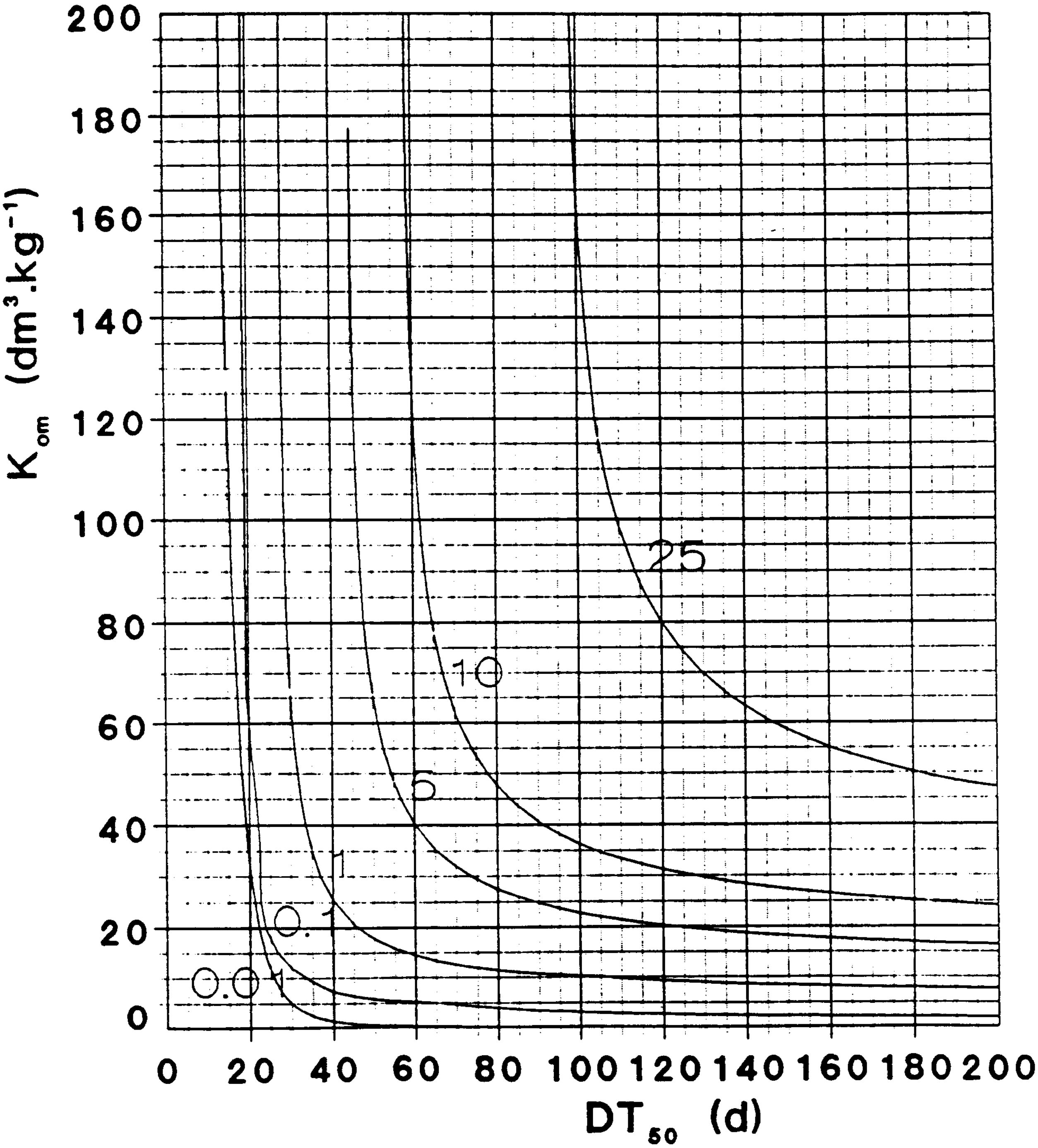
Graphical representation of PESTLA, spring application: maximum concentrations of the pesticide (metabolite) in the upper groundwater (1 - 2 m), as a function of K_{om} and DT_{50}; the graphs are contour lines and the numbers are the concentrations belonging to them, in µg/l. The lines are based on an application rate of 1 kg a.i./ha.



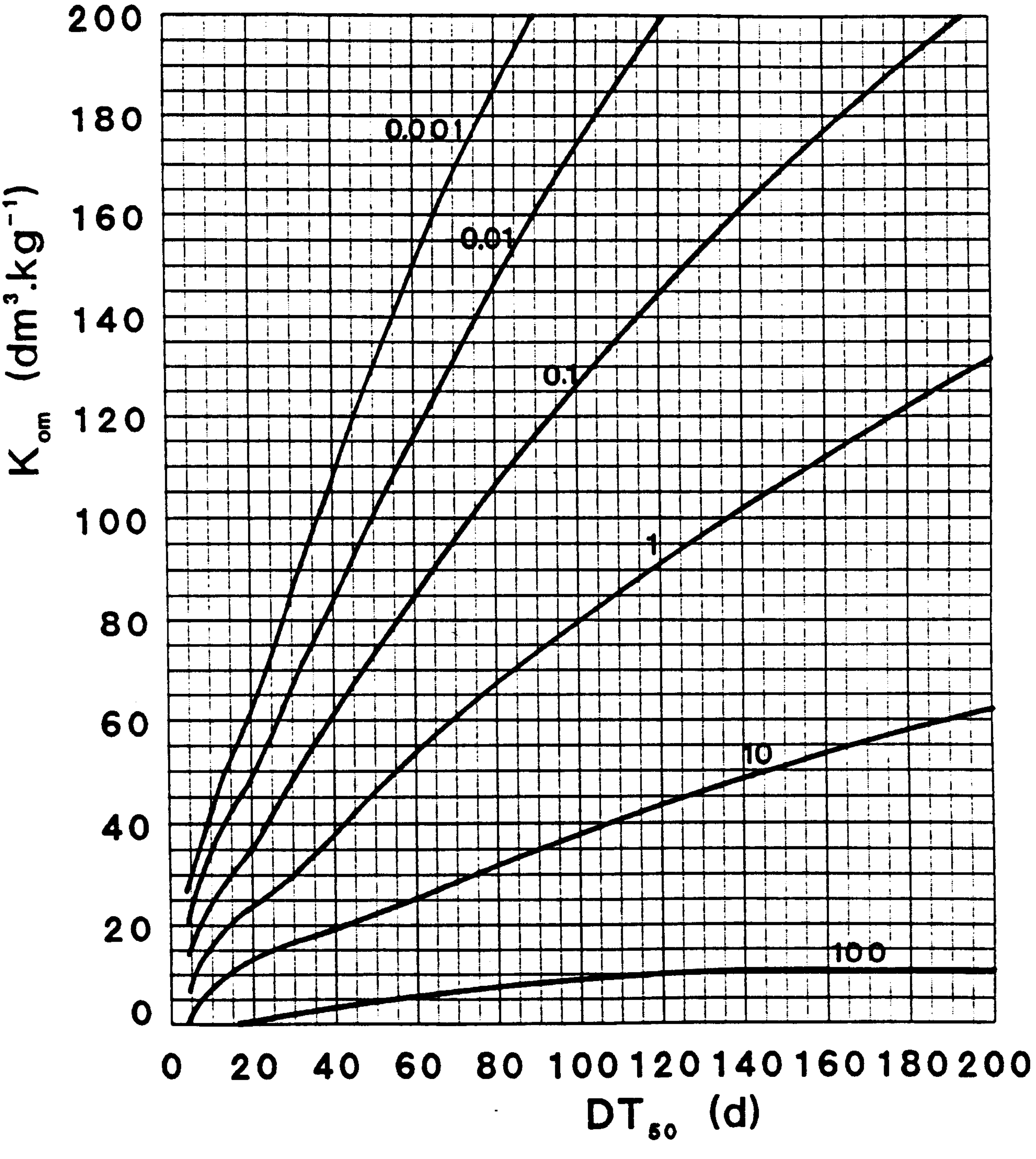
Graphical representation of PESTLA, spring application: the leaching of the pesticide (metabolite) from the upper meter of the soil, as a function of K_{om} and DT_{50}; the graphs are contour lines and the numbers belonging to them give the leaching as percentage of the applied dose.



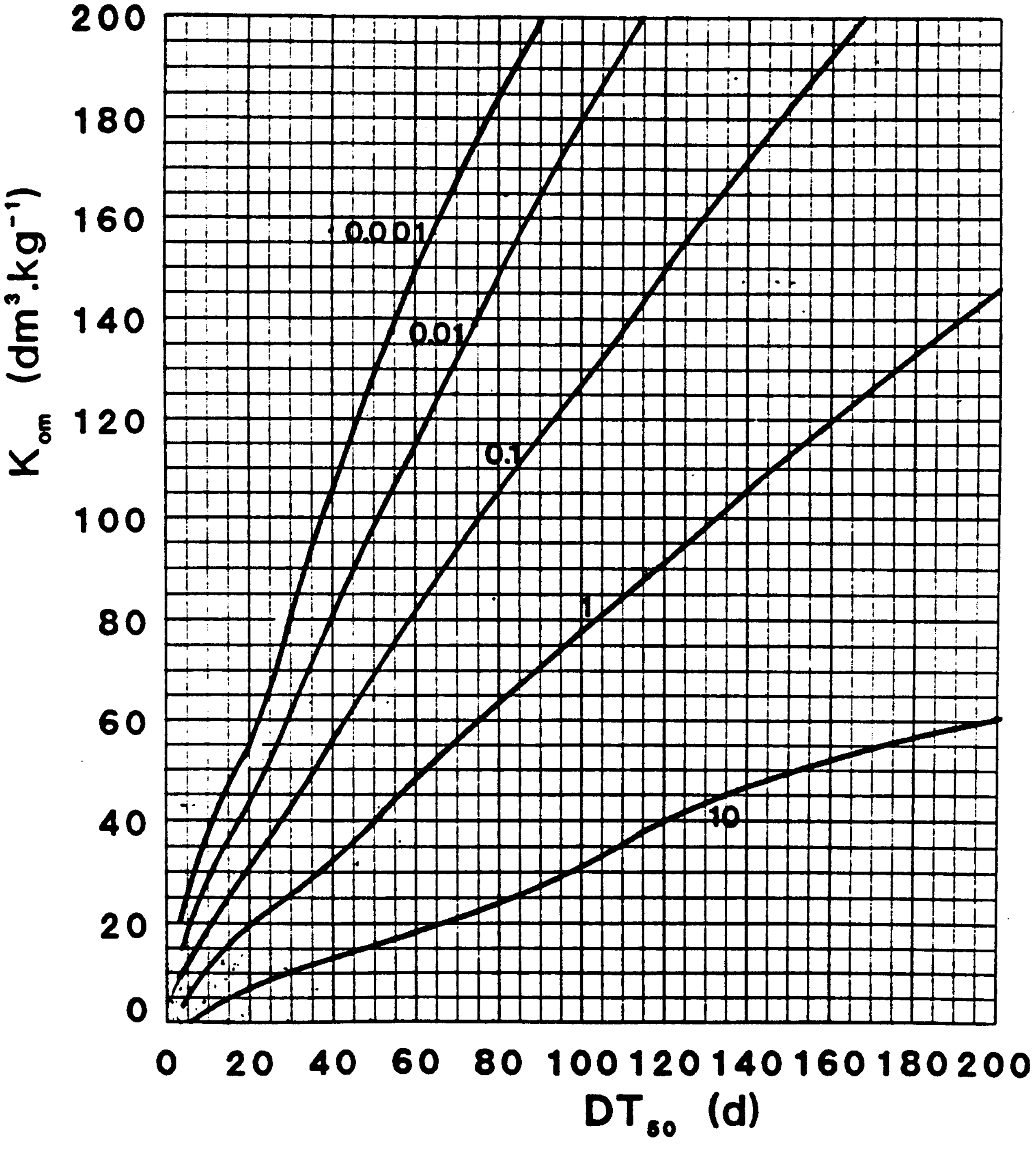
Graphical representation of PESTLA results, spring application: the amount of pesticide (metabolite) that is still present in the plough layer 1 year after application; the graphs are contour lines and the numbers belonging to them give the percentage of the applied dose.



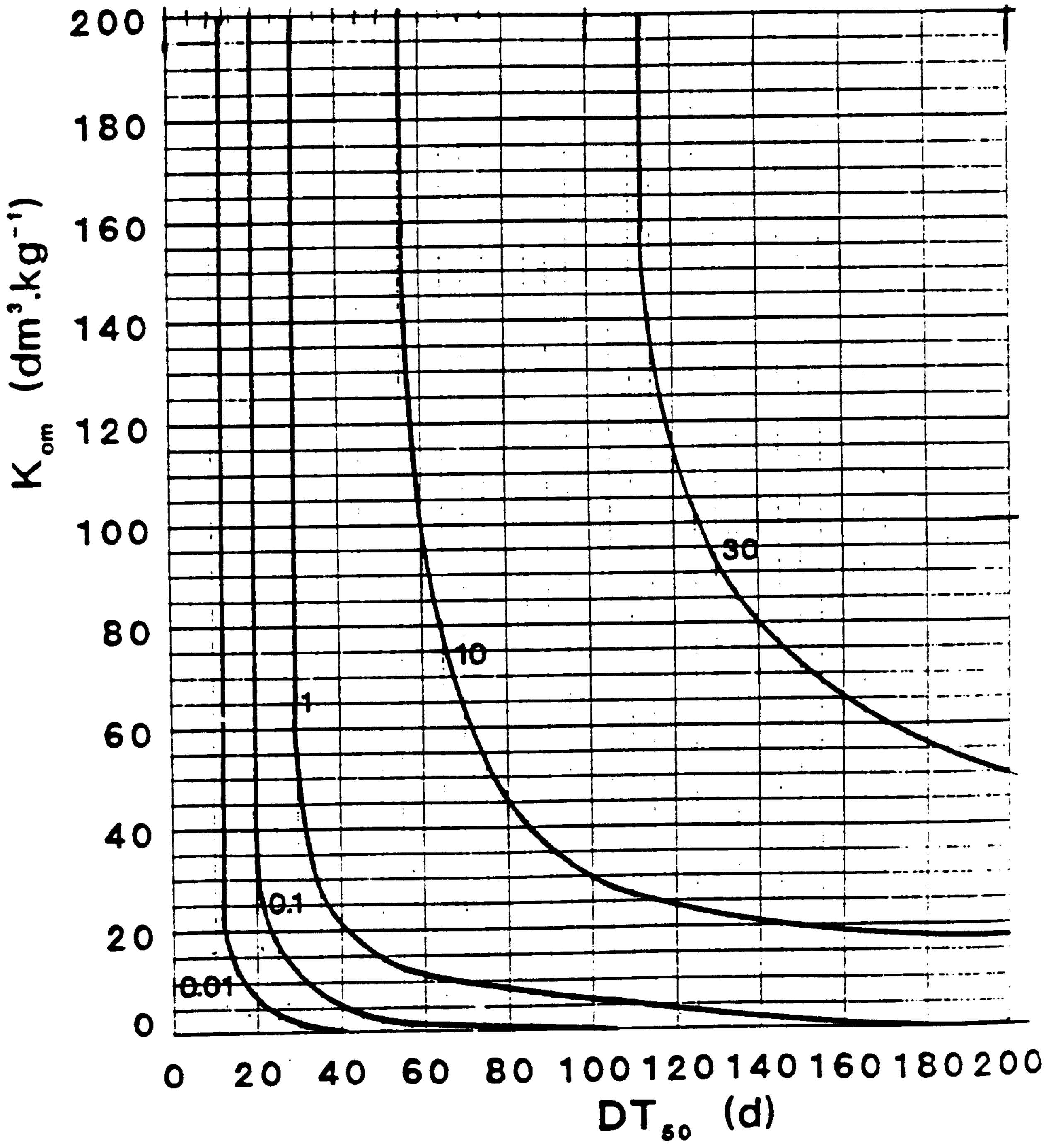
Graphical representation of PESTLA results, autumn application: maximum concentrations of the pesticide (metabolite) in the upper groundwater (1 - 2 m), as a function of K_{om} and DT_{50}; the graphs are contour lines and the numbers are the concentrations belonging to them, in µg/l. The lines are based on an application rate of 1 kg a.i./ha.



Graphical representation of PESTLA results, autumn application: the leaching of the pesticide (metabolite) from the upper meter of the soil, as a function of K_{om} and DT_{50}; the graphs are contour lines and the numbers belonging to them give the leaching as percentage of the applied dose.



Graphical representation of PESTLA results, autumn application: the amount of pesticide (metabolite) that is still present in the plough layer 1 year after application; the graphs are contour lines and the numbers belonging to them give the percentage of the applied dose.



Appendix 5: Classification of data on physico-chemical properties, environmental behaviour and ecotoxicology

Remarks:

1. The classifications that are given below are BBM classifications; only for aquatic organisms an EU-classification is available. For aquatic organisms both the BBM and EU-classification are given.
2. The quantitative result should always be given in the RIVM Conclusion.

Physical properties

* **Solubility (S) at 20-25 °C**

Classification		S [mg/l]
zeer slecht oplosbaar	very slightly soluble	< 0,1
slecht oplosbaar	slightly soluble	0,1 - 10
matig oplosbaar	moderately soluble	10 - 1000
goed oplosbaar	readily soluble	≥ 1000

* **Vapour pressure (P) at 20-25 °C**

Classification		P [Pa]
weinig vluchtig	very slightly volatile	< 0,0001
enigszins vluchtig	slightly volatile	0,0001 - 0,01
matig vluchtig	moderately volatile	0,01 - 1
vluchtig	volatile	1 - 100
zeer vluchtig	highly volatile	≥ 100

* **Volatility from water (Henry's Law Constant)**

Classification (Lyman, 1982)		H [-]
weinig vluchtig	very slightly volatile	< 1E-5
matig vluchtig	moderately volatile	1E-5 - 0,03
zeer vluchtig	highly volatile	> 0,03

Transformation and mobility in soil

* **DT_{50} at 20 °C, pF = 2, top soil**

Classification		DT_{50} [d]
zeer slecht afbreekbaar	very slightly degradable	> 180
slecht afbreekbaar	slightly degradable	60 - 180
redelijk afbreekbaar	fairly degradable	20 - 60
goed afbreekbaar	readily degradable	< 20

* Mobility at 20 °C

Classification		R_f	$K_{s/l}$ [dm^3/kg]	K_{om} [dm^3/kg]
- zeer weinig mobiel	immobile	0 - 0,09	> 2,6	> 100
- weinig mobiel	slightly mobile	0,10 - 0,34	0,53 - 2,6	20 - 100
- matig mobiel	moderatly mobile	0,35 - 0,64	0,15 - 0,53	5 - 20
- mobiel	mobile	0,65 - 0,89	0,03 - 0,15	1 - 5
- zeer mobiel	highly mobile	0,90 - 1,00	< 0,03	< 1

Transformation in water

* Transformation water/sediment system $DT_{50,sys}$ at 20 °C (whole system)

Classification		DT_{50} [d]
- zeer slecht afbreekbaar	very slightly degradable	> 180
- slecht afbreekbaar	slightly degradable	60 - 180
- redelijk afbreekbaar	fairly degradable	20 - 60
- goed afbreekbaar	readily degradable	< 20

* Hydrolysis DT_{50} at 20 °C, pH 7

Classification		DT_{50} [d]
- slecht hydrolyserend	slightly hydrolysing	> 30[40]
- matig hydrolyserend	moderately hydrolysing	10 - 30
- redelijk hydrolyserend	fairly hydrolysing	4 - 10
- goed hydrolyserend	readily hydrolysing	1 - 4
- zeer goed hydrolyserend	very rapidly hydrolysing	< 1

* Phototransformation in water DT_{50} (continuous light regime)

Classification		DT_{50} [d]
- weinig afbreekbaar	slighty degradable	> 30[41]
- matig afbreekbaar	moderately degradable	10 - 30
- redelijk afbreekbaar	fairly degradable	4 - 10
- goed afbreekbaar	readily degradable	1 - 4
- zeer goed afbreekbaar	very rapidly degradable	< 1

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[40] When a preliminary test was performed, in which < 10% of the pesticide was hydrolysed at 50 °C within five days, then the pesticide is considered hydrolytically stable. No main test need to be performed, and the extrapolated DT_{50} (20 °C) is > 500 days

[41] When a preliminary test was performed, in which < 10% of the pesticide was photolysed at 20 - 25 °C within 30 days, then the pesticide is considered photolytically stable

Toxicity

* **Aquatic organisms, acute: algae (96-h EC50), *Daphnia* (48-h LC_{50}) and fish (96-h LC_{50})**

RIVM/ACT-classification		EU-classification	$E(L)C_{50}$ [mg/l]
zeer weinig giftig	very slightly toxic		> 100
weinig giftig	slightly toxic	harmful	10 - 100
matig giftig	moderately toxic	toxic	1 - 10
zeer giftig	highly toxic	very toxic	< 1

Remark: the EU-classification is given between brackets after the RIVM/ACT-classification:
- ♦ Dutch: (volgens EU-classificatie:...)
- ♦ English: (according to EU-classification:...)

* **Aquatic organisms, chronic**

Classification		NOEC [mg/l]
zeer weinig giftig	very slightly toxic	> 1
weinig giftig	slightly toxic	0,1 - 1
matig giftig	moderately toxic	0,01 - 0,1
zeer giftig	highly toxic	< 0,01

* **Birds, acute oral**

Classification		LD_{50} [mg/kg bw]
weinig giftig	slightly toxic	> 500
matig giftig	moderately toxic	50 - 500
giftig	toxic	5 - 50
zeer giftig	highly toxic	< 5

* **Earthworms, soil test**

Classification		LC_{50} [mg/kg dry soil]
zeer weinig giftig	very slightly toxic	> 1000
weinig giftig	slightly toxic	100 - 1000
matig giftig	moderately toxic	10 - 100
giftig	toxic	1 - 10
zeer giftig	highly toxic	< 1

* **Bees, contact and oral**

Classification		LD_{50} [µg/bee]
zeer weinig giftig	very slightly toxic	> 100
weinig giftig	slightly toxic	10 - 100
matig giftig	moderately toxic	1 - 10
giftig	toxic	0,1 - 1
zeer giftig	highly toxic	< 0,1

* **Other beneficial insects, mites, and spiders, laboratory testing (bees excluded)**

Classification		Reduction in beneficial capacity [%]
- onschadelijk	harmless	< 30
- weinig schadelijk	slightly harmful	30 - 79
- matig schadelijk	moderately harmful	80 - 99
- schadelijk	harmful	> 99

Bioconcentration

Classification		BCF_{wo}
- weinig concentrerend	slightly concentrating	< 100
- matig concentrerend	moderately concentrating	100 - 1000
- sterk concentrerend	highly concentrating	> 1000

Appendix 6: Additional Questions (Dutch)

AANVULLENDE VRAGEN betreffende:

Auteurs:

Maand + jaar:

Fysisch-chemische gegevens

0 Gegevens omtrent de K_{ow} van ...... worden noodzakelijk geacht (D.8).
0 Gegevens omtrent de pK_a van ...... worden noodzakelijk geacht (D.8).
0 Gegevens omtrent de wateroplosbaarheid van ...... worden noodzakelijk geacht (D.8).
0 Gegevens omtrent de dampspanning van ...... worden noodzakelijk geacht (D. 8).
0 (andere vraag)

Gedrag in grond

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de omzetting van ...... in grond. Uitvoering van een omzettingssnelheidstudie in tenminste 3 / 2 extra / 1 extra grondsoort(en) volgens G.1.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de omzettingsroute van ...... in grond. Uitvoering van een proef ter bepaling van de omzettingsroute van ...... in 1 grondsoort volgens G.1.1 wordt noodzakelijk geacht.
Indien een metaboliet in een omzettingsstudie in de bodem wordt gevormd in een gehalte van meer dan 10% (stoffractie) van de hoeveelheid toegevoegd ...... , dient met die metaboliet een omzettingssnelheidstudie in tenminste 3 grondsoorten volgens G.1.1 en een schudproef of een kolomproef met tenminste 3 grondsoorten ter bepaling van $K_{s/l}$ volgens G.1.2 uitgevoerd te worden.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de mobiliteit van ...... in grond. Uitvoering van een schudproef of een kolomproef met tenminste 3 / 2 extra / 1 extra grondsoort(en) ter bepaling van $K_{s/l}$ volgens G.1.2 wordt noodzakelijk geacht.
Indien de pK_a ligt tussen 2 en 6 dienen sorptiegegevens geleverd te worden in tenminste 3 grondsoorten met pH 7 - 8.

0 De pK_a van ...... ligt tussen 2 en 6. Uitvoering van een schudproef met tenminste 3 / 2 extra / 1 extra grondsoort(en) met pH 7 - 8 ter bepaling van $K_{s/l}$ volgens G.1.2 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de mobiliteit van de metabolieten van ...... Uitvoering van een kolomstudie met verouderd residu in tenminste 1 grondsoort volgens G.1.2 wordt noodzakelijk geacht[42].

0 (andere vraag)

Gedrag in water

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[42] Op termijn zullen studies met drie grondsoorten vereist zijn

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de omzetting van ...... in een water/slib systeem. Uitvoering van een studie met tenminste 2 / 1 extra slootbodem-materia(a)l(en) volgens G.2.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de adsorptie aan slibdeeltjes van ...... Uitvoering van een studie met 2 (kleine ondiepe wateren) / 1 (grote oppervlaktewateren) slootbodem-materia(a)l(en) volgens G.2.2 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de fotochemische afbraak van ...... Uitvoering van een studie volgens G.2.3 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de hydrolyse van ...... Uitvoering van een studie volgens D.9 wordt noodzakelijk geacht.

0 (andere vraag)

Organismen in het milieu

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de acute orale toxiciteit van ...... voor vogels. Uitvoering van een studie met tenminste 2 / 1 extra vogelsoort(en) volgens H.1.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de subacute orale toxiciteit van ...... voor vogels. Uitvoering van een studie met tenminste 2 / 1 extra vogelsoort(en) volgens H.1.2 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de semi-chronische orale toxiciteit voor vogels. Uitvoering van een studie volgens H.1.3 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de vorm, de oppervlaktestruktuur en de grootteverdeling van het granulaat. Verstrekking van deze gegevens wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de acute toxiciteit van ...... voor algen. Uitvoering van een studie volgens H.2.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de acute toxiciteit van ...... voor kreeftachtigen. Een studie volgens H.2.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de acute toxiciteit van ...... voor vissen. Uitvoering van een studie volgens H.2.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de chronische toxiciteit van ...... voor kreeftachtigen. Uitvoering van een studie volgens H.2.2 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de chronische toxiciteit van ...... voor vissen. Uitvoering van een studie volgens H.2.2 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de toxiciteit van ...... voor bijen. Uitvoering van een studie ter bepaling van de orale en contact / orale / contact LD_{50} volgens H.3.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de toxiciteit van ...... voor nuttige insekten en mijten. Uitvoering van een studie volgens H.3.2 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de toxiciteit van ...... voor regenwormen. Uitvoering van een studie volgens H.4.2 wordt

noodzakelijk geacht.

0 (andere vraag)

Overige gegevens

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent het effekt van ......op de bodemademhaling. Uitvoering van een studie volgens H.4.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent het effekt van ...... op de nitrificatie. Uitvoering van een studie volgens H.4.1 wordt noodzakelijk geacht.

0 Op grond van de Kow wordt voor ...... een BCF van > 1000 berekend. Uitvoering van een bioconcentratiestudie met organismen (bij voorkeur met vis) ter bepaling van de BCF volgens H.6 wordt noodzakelijk geacht.

0 Lozing van ...... op het riool is te verwachten. Uitvoering van een studie naar het effect van ...... op de respiratie of de TOC-verwijdering (OECD 305A-E) en op de nitrificatie door geadapteerd slib volgens H.5 (OECD 209 en OECD 305A-E) wordt noodzakelijk geacht.

0 Een volledige beschrijving van een methode voor de kwalitatieve en kwantitatieve bepaling van residuen van ...... in water, volgens G.2.5, dient geleverd te worden.

0 (andere vraag)

Metabolieten

De metaboliet(en) m......, m......, m...... enz. en m...... wordt/worden in de bodem gevormd in een gehalte van meer dan 10% van de toegevoegde hoeveelheid ......

m... :
m... : etc.

0 Gegevens omtrent de pK_a van de metaboliet(en) m......, m......, m...... en m...... worden noodzakelijk geacht (D.8).

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de omzetting van de metaboliet(en) m......, m......, m...... en m...... in grond. Uitvoering van een omzettingssnelheidstudie in tenminste 3 / 2 extra / 1 extra grondsoort(en) volgens G.1.1 wordt noodzakelijk geacht.

0 Er zijn geen / onvoldoende / onvoldoende betrouwbare gegevens beschikbaar omtrent de mobiliteit van de metaboliet(en) m......, m......, m...... en m...... in grond. Uitvoering van een schudproef of een kolomproef met tenminste 3 / 2 extra / 1 extra grondsoort(en) ter bepaling van $K_{s/l}$ volgens G.1.2 wordt noodzakelijk geacht.
Indien de pK_a van deze metaboliet(en) ligt tussen 2 en 6, dienen sorptiegegevens geleverd te worden in tenminste 3 grondsoorten met pH 7 - 8.

0 De pK_a van metaboliet m...... ligt tussen 2 en 6. Uitvoering van een schudproef met tenminste 3 / 2 extra / 1 extra grondsoort(en) met pH 7 - 8 ter bepaling van $K_{s/l}$ volgens G.1.2 wordt noodzakelijk geacht.

0 (andere vraag)

Appendix 7: List of Guidelines

OECD Guidelines

Guideline Code	Guideline Description
OECD101	UV-VIS absorption spectra
OECD102	Melting Point/Melting Range
OECD103	Boiling Point/Boiling Range
OECD104	Vapour Pressure Curve
OECD105	Water Solubility
OECD106	Absorption/Desorption
OECD107	Partition Coefficient (n-octanol/water)
OECD108	Complex Formation Ability in Water
OECD109	Density of Liquids and Solids
OECD110	Particle size Distribution/Fibre Length and Diameter Distributions
OECD111	Hydrolysis as a Function of pH
OECD112	Dissociation Constants in Water
OECD113	Screening Test for Thermal Stability and Stability in Air
OECD114	Viscosity of Liquids
OECD115	Surface tension of Aqueous Solutions
OECD116	Fat Solubility of Solid and Liquid Substances
OECD117	Partition Coefficient (n-octanol/water), HPLC Method
OECD201	Algae, Growth Inhibition Test
OECD202	*Daphnia* spp. Acute Immobilisation test and Reproduction Test
OECD203	Fish, Acute Toxicity Test
OECD204	Fish, Prolonged Toxicity Test: 14-day Study
OECD205	Avian Dietary Toxicity Test
OECD206	Avian Reproduction Test
OECD207	Earthworm, Acute Toxicity Tests
OECD208	Terrestrial Plants, Growth Test
OECD209	Activated Sludge, Respiration Inhibition Test
OECD210	Fish, Early-Life Stage Toxicity Test

OECD301	Ready biodegradability:
OECD301A	DOC Die-away Test
OECD301B	CO2 Evolution Test
OECD301C	Modified MITI Test (I)
OECD301D	Closed Bottle Test
OECD301E	Modified OECD Screening Test
OECD301F	Manometric Respirometry Test
OECD302	Inherent biodegradation:
OECD302A	Modified SCAS Test
OECD302B	Modified Zahn-Wellens Test
OECD302C	Modified MITI Test (II)
OECD303A	Aerobic Sewage Treatment: Coupled Units Test
OECD304A	Inherent Biodegradability in Soil
OECD305	Bioconcentration:
OECD305A	Sequential Static Fish Test
OECD305B	Semi-Static Fish Test
OECD305C	Degree of Bioconcentration in Fish
OECD305D	Static Fish Test
OECD305E	Flow-Through Fish Test
OECD306	Biodegradability in Seawater
OECD401	Acute Oral Toxicity

EPPO Guidelines

Guideline Code (EPPO Bulletin)	Guideline Description
(15)	Standard methods to test the side-effects of pesticides on natural enemies of insects and mites developed by the IOBC/WPRS Working Group 'Pesticides and Beneficial Organisms'. Eppo Bulletin 15 (1985) 214-255.
(22)	Method for honeybee brood feeding test with insect growth-regulating insecticides. Oomen, P.A., A. de Ruijter & J. van der Steen. Eppo Bulletin 22 (1992), p. 613-616.
142	Guidelines for the evaluation of side-effects of plant protection products. No 142. *Encarsia formosa*. Eppo Bulletin 19 (1989), 355-372.
151	Guidelines for the evaluation of side-effects of plant protection products. No 151. *Phytoseiulus persimilis*. Eppo Bulletin 20 (1990), 531-550.
170	Guideline on test methods for evaluating the side-effects of plant protection products on honeybees. Eppo Bulletin 22 (1992) p. 203-215.
180	Guidelines for the evaluation of side-effects of plant protection products. No 180. *Trichogramma cacoeciae*. Eppo Bulletin 23 (1994), 329-352.

EPA Guidelines

Guideline Code	Guideline Description
EPA-540/9-85-002	Honey bee - acute contact LD_{50} test
EPA-540/9-85-003	Honey bee - toxicity of residues on foliage
EPA-540/9-85-005	Acute toxicity test for freshwater invertebrates
EPA-540/9-85-006	Acute toxicity test for freshwater fish
EPA-540/9-85-007	Avian single-dose LD_{50}
EPA-540/9-85-008	Avian dietary LC_{50} test
EPA-540/9-85-009	Acute toxicity test for estuarine and marine organisms (estuarine fish 96-hour acute toxicity test)
EPA-540/9-85-010	Acute toxicity test for estuarine and marine organisms (shrimp 96-hours acute toxicity test)
EPA-540/9-85-011	Acute toxicity test for estuarine and marine organisms (mollusc 96-hour flow-through shell deposition study)
EPA-540/9-85-012	Acute toxicity test for estuarine and marine organisms (mollusc 48-hour embryo larvae study)
EPA-540/9-85-013	Hydrolysis studies
EPA-540/9-85-014	Aqueous photolysis studies
EPA-540/9-85-015	Aerobic soil metabolism study
EPA-540/9-85-016	Soil photolysis study
EPA-540/9-85-017	Soil column leaching study
EPA-540/9-85-130	Non-target plants: target area testing
EPA-540/9-85-130	Growth and reproduction of aquatic plants
EPA-540/9-85-135	Non-target plants: terrestrial field testing
EPA-540/9-85-136	Aquatic field testing
EPA-540/9-85-137	Fish life-cycle toxicity tests
EPA-540/9-86-138	Fish early life-stage
EPA-540/9-86-139	Avian reproduction test
EPA-540/9-86-141	*Daphnia magna* life-cycle (21 day renewal) chronic toxicity test
EPA-540/9-86-152	Acute dietary LC_{50} test for waterfowl and upland gamebirds (addendum)
EPA-540/9-87-198	Aquatic testing for marine/estuarine and freshwater fish and invertebrates (addendum)
EPA-540/9/88-006	Wild mammal toxicity test (addendum)

ISO Guidelines

Guideline Code	Guideline Description
ISO 11268-1	Soil quality — Effects of pollutants on earthworms (*Eisenia fetida*) — Part 1: Determination of acute toxicity using artificial soil substrate
ISO 11268-2 (draft)	Soil quality — Effects of pollutants on earthworms (*Eisenia fetida*) — Part 2: Method for the determination of effects on reproduction

EU Guidelines

Guideline Code	Guideline Description
Annex V.A1[43]	Melting/Freezing temperature
Annex V.A2	Boiling temperature
Annex V.A3	Relative density
Annex V.A4	Vapour pressure
Annex V.A5	Surface tension
Annex V.A6	Water solubility
Annex V.A7	Fat solubility
Annex V.A8	Partition coefficient
Annex V.A9	Flash point
Annex V.A10	Flammability (solids)
Annex V.A11	Flammability (gases)
Annex V.A12	Flammability (substances and preparations which, in contact with water or damp air, evolve highly flammable gases in dangerous quantities)
Annex V.A13	Flammability (solids and liquids)
Annex V.A14	Explosive properties
Annex V.A15	Auto-flammability (determination of the temperature of self-ignition of volatile liquids and of gases)
Annex V.A16	Auto-flammability (solids - determination of relative self-ignition temperature)
Annex V.A17	Oxidising properties (solids)
Annex V.C1	Acute toxicity for fish
Annex V.C2	Acute toxicity for *Daphnia*
Annex V.C3	Growth inhibition test with algae
Annex V.C4A	DOC - die away test
Annex V.C4B	Modified OECD screening test
Annex V.C4C	Carbon dioxide CO2-development test
Annex V.C4D	Manometric respiration test
Annex V.C4E	Closed Bottle test
Annex V.C4F	MITI test
Annex V.C5	Degradation - biochemical oxygen demand
Annex V.C6	Degradation - chemical oxygen demand
Annex V.C7	Degradation - abiotic degradation: hydrolysis as a function of pH

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[43] This Annex V is implemented in the EC Council Directive of 25 April 1984 containing technical adaptations of Directive 67/548/EEC on the approximation of the laws, regulations and administrative provisions relating to the classification, packaging, and labelling of dangerous substances (84/449/EEC)

BBA Guidelines

Guideline Code	Guideline Description
BBA IV/4-1	Persistence of plant protection products in the soil; degradation, transformation, and metabolism
BBA IV/4-2	Seepage behaviour of plant protection products
BBA VI/1-1	Auswirkungen auf die Aktivität der Bodenmikroflora
BBA 23-1	Richtlinien für die Prüfung von Pflanzenbehandlungsmitteln auf Bienengefährlichkeit
BBA 23-2.3.3	Richtlinie zur Prüfung der Wirkung von Pflanzenbehandlungsmitteln auf Nutzarthropoden der Baumschicht im Freiland
BBA 23-2.3.4	Richtlinie für die Prüfung der Auswirkung von Pflanzenbehandlungsmitteln auf Raubmilben im Weinbau
BBA 25-1	Richtlinie zur Prüfung von Pflanzenbehandlungsmitteln auf Vogelgefährdung
BBA 36	Unterlagen zum Verhalten von Pflanzenbehandlungsmitteln im Boden im Rahmen des Zulassungsverfahrens.
BBA 37	Seepage behaviour of plant protection products
BBA 55	Prüfung des Verhaltens von Pflanzenbehandlungsmitteln im Wasser

NEN Guidelines

Guideline Code	Guideline Description
NEN 6501	Determination of acute toxicity with *Daphnia magna*
NEN 6502	Determination of chronic toxicity with *Daphnia magna*
NEN 6504	Determination of acute toxicity with *Poecilia reticulata*
NEN 6506	Determination of toxicity with algae
NEN 6511	Water - Determination of acute toxicity in nitrificating active sludge by measurement of ammonia degradation.
NEN 6512	Water - Determination of acute toxicity in aerobic active sludge by measurement of the respiration rate.
NEN 5794	Determination of the acute toxicity of chemical substances to earthworms
NEN 5795	Determination of the influence of chemical substances on the nitrification in soil
NEN 5797	Determination of the effect of chemical substances to reproduction of earthworms

Appendix 8: Final Environmental File (FEF)

(authors)
(date)

RISK ASSESSMENT ACCORDING TO ENVIRONMENTAL DECISION TREES

(COMMON NAME)

In applying different formulations containing (name) the following hazards, based on the environmental decision trees, are expected:

application	number form.	leaching to groundwater	aquatic organisms	birds	earth-worms	bees	beneficial insects	nitrification

N = no, L = low, M = medium, H = high, U = unknown

General remark

This monograph represents a complete summary of all delivered data.

General information

Common name :
Chemical name :
Structural formula :

Goal of application : (herbicide, insecticide etc.)
Formulations : 1. (kind), (amount a.i. in % or g/l)
2. etc.

Overview of application :

application	no. form.	dose kg a.i./ha	frequency	interval (days)	time of application

Physico-chemical properties

- Solubility:

(name) is (classification) soluble in water: the solubility is ... mg/l at ... °C.

- Volatility:

(name) is (classification) volatile: the vapour pressure is ... Pa at ... °C.

- Other properties:

The molar mass of (name) is (value) g/mol.
The log K_{ow} of (name) is (value/is not available/is not available but is not considered relevant in view of the high solubility).
The pK_a for (name) is (value/unknown/unknown but not considered relevant).
Henry's law constant (calculated) for (name) is (value).

(name) is (classification) bioconcentrating.
Based on the log K_{ow} (value) a BCF of (value) can be calculated for (name).
In a study on fish/crustaceans (or other organism) a .. -day BCF of (value) was calculated for the whole organism.

Ecotoxicology

Behaviour in soil

- Transformation in the upper soil:

(name) is (classification) degradable in soil.
(may have to be repeated for metabolites formed in amounts of > 10% of mass fraction)

In laboratory studies the following DT_{50} values (20 °C) were found for the aerobic transformation of (name): (values) days.

In field studies the following DT_{50} values were found: (values) days.

In the aerobic transformation of (name), (amount) metabolite(s) was/were found. The main metabolite(s) (> 5% of the applied amount of (name)) was/were:
mI : (chemical name) (max. ...% after ... days; ...% after ... days (end)),
mII : (chemical name) (max. ...% after ... days; ...% after ... days (end)),
etc.
m.. : (chemical name) (max. ...% after ... days; ...% after ... days (end)).

For metabolite m.. DT_{50} values of (values) days were found.
(may have to be repeated for other metabolites)

Bound residues (place of label) reached a maximum of (value) % after .. days under aerobic conditions. (if the maximum is not reached at the end of the study add the final percentage and time).
(may have to be repeated for another label)

The amount of $^{14}CO_2$ (place of label) reached a maximum of (value) % after .. days under aerobic conditions. (if the maximum is not reached at the end of the add the final percentage and time)
(may have to be repeated for other label)

In laboratory studies the following DT_{50} values (20 °C) were found for the anaerobic transformation of (name): (values) days.

In the anaerobic transformation (name) metabolites m.., m.. etc. were found (for a new metabolite:)
m.. : (chemical name) (max. ...% after ... days; ...% after ... days).

- Transformation in the saturated zone:
In laboratory studies the following DT_{50} values (20 °C) were found for the (an)aerobic transformation of (name): (values) days.

In the (an)aerobic transformation of (name) metabolites m.., m.. etc. were found. (for a new metabolite:)
m.. : (chemical name) (max. ...% after ... days; ...% after ... days).

For the/a preliminary calculation of accumulation and leaching of (name), the following/no DT_{50} values (20 °C) are available: (values) days (mean/median (value) days, standard deviation (value)/range (values)).
(may have to be repeated for metabolites formed in an amount of > 10%)

- Mobility:
(name) is (classification based on K_{om}) mobile in soil.

In a column study/column studies the following Rf values were found for (name): (values). Calculated K_{om} values are: (values) dm^3/kg.

In an adsorption study/adsorption studies the following $K_{s/l}$ values were found for (name): (values) dm^3/kg. Calculated K_{om} values are: (values) dm^3/kg.

(text classification/column study/adsorption study for metabolites formed in amounts of > 10% may have to be repeated)

In a column study with .. days aged residue (composition after ageing) ..% of the applied radioactivity (possibly a specification of the amount of a.i. or metabolite(s)) recovered from the leachate.

For the/a preliminary calculation of accumulation and leaching of (name), the following/no K_{om} values are available: (values) dm^3/kg (mean/median (value) dm^3/kg, standard deviation (value)/range (values)).
(may have to be repeated for metabolites formed in amounts of > 10%)

Calculation of accumulation and leaching[44]

On the basis of the standard calculation with the "Steungroep-M" model the next (preliminary) expectations can be obtained for the application of (name) in spring/autumn (using a mean/median DT_{50} of ... days with a minimum of ... and a maximum of ... days, and a mean/median K_{om} of ... dm^3/kg with a minimum of ... and a maximum of ... dm^3/kg)*:

- a concentration in the shallow groundwater of ... mg/m^3 (minimum ... and maximum ... mg/m^3);
- leaching of ...% (minimum ... and maximum ...%) of the dose from the upper metre of the soil;
- a residue of ...% (minimum ... and maximum ...%) of the dose in the plough layer after 1 year, which corresponds with a concentration of ... µg/kg (minimum ... and maximum ... µg/kg).

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[44] In the future data concerning the mobility in the saturated zone (aerobic and anaerobic), and a calculation of the concentration in deeper groundwater might be included

(may have to be repeated for application in autumn and/or for metabolites formed in amounts of > 10%)
* (if the calculation concerns a metabolite also mention: , a relative molecular mass of (percentage) and a formation percentage of ..%)

If using (name) (no/an unknown/a low/a medium/a high) hazard of leaching of (the a.i./metabolite m..) to the shallow groundwater is expected.

If using (name) (no/an unknown/a low/a medum/a high) hazard of leaching of (indication of metabolite formed in amounts of <10%) to the shallow groundwater in a concentration of > 0.1 µg/l is expected.
(state chemical name if not mentioned before)

Behaviour in water

- Transformation in water/sediment systems:

(name) is (classification based on DT_{50} system) degradable in a water/sediment system.

In water/sediment systems DT_{50} values of (values) days were found for (name) in the water phase and of (values) days for the system were found.
With the transformation of (name) in a water/sediment system metabolites m.. (max. ..% after .. days, water phase/sediment), m.. etc. were found in the water phase.
With the transformation of (name) in a water/sediment system metabolites m.. (max. ..% after .. days, water phase/sediment), m.. etc. were found in the sediment.

- Hydrolysis:

For the hydrolysis of (name) at (temperature) °C and a pH of .., .. etc., DT_{50} values of .., .. etc. days are found, respectively.
With the hydrolysis of (name) metabolites m.. (max. ..% after .. days), m.. etc. were found.

- Photolysis:

For the photolysis of (name) in (natural/artificial sunlight) DT_{50} values of (values) days were found.
(possibly: In these experiments (naam) may have served as a photosensitizer.)
With the photolysis of (name), metabolites m.. (max. ..% after .. days), m.. etc. were found.

For the calculation of the expected concentration of (name) in surface water according to slootbox, no/a/a mean DT_{50} of .. days is available.

Toxicity for organisms in the environment

- Occurrence in the environment:

(measured concentrations in different environmental compartments)

- Birds:

(name) is orally acute (classification) toxic for birds: LD_{50} (value or range) mg/kg body weight (.. species)
(possibly: a ..% formulation of (name) is etc.)

In (a) diet study/studies (a) (period)-day LC_{50} value(s) according to slootbox of (value or range) mg/kg feed were found for (name) (.. species).
(may have to be repeated with "were found for a ..% formulation of (name)")

- Risks for birds:

Upon single application of a/different formulation(s) containing (name) the following hazards are expected for birds, based on an LD_{50} of .. mg/kg body weight and an LC_{50} of .. mg/kg feed:

Table: Risk assessment of the chance of acute death in birds

application	no. form.	dose kg a.i./ha	exposure route	concentration mg/l or mg/kg	hazard

N = no, L = low, M = medium, H = high, U = unknown

- Combination toxicity for birds:
Upon single application of a/different formulation(s) containing, besides (name) one or more other active ingredients, the following hazards are expected for birds, based on the toxicity values below:

number formulation	LD50 in mg/kg body weight		LC50 in mg/kg feed	
	calculated*	measured	calculated*	measured

* based on additivity

Table: Risk assessment of the chance of acute death in birds

application	no. form.	dose kg a.i./ha	exposure route	concentration mg/l or mg/kg	hazard#

N = no, L = low, M = medium, H = high, U = unknown
# if available, based on the measured toxicity value

- Aquatic organisms:

* Algae:

(name) is (classification) toxic (according to EC classification: (classification) toxic) for algae: ..-hour NOEC (possibly: NOE_rC or NOE_bC) (value or range) mg/l (.. species); .. -hour EC_{50} (possibly: E_rC_{50} or E_bC_{50}) (value or range) mg/l (.. species)
(possibly: a ..% formulation of (name) is etc.)

* Crustaceans:

(name) is acutely (classification) toxic (according to EC classification: (classification) toxic) for crustaceans: .. -hour LC_{50} (value or range) of mg/l (.. species).
(evt. a ..% formulation of ... is etc.)

(possibly chronic data)

* Fish:

(name) is acute (classification) toxic (according to EC classification: (classification) toxic) for fish: .. -hour LC_{50} (value or range) of mg/l (.. species).
(possibly: a ..% formulation of ... is etc.)

(possibly chronic data)

- Risks for aquatic organisms:

* Acute:

Upon application according to agricultural requirements for a/different formulation(s) containing (name) and assuming a ditch depth of 25 cm, the following hazards for algae, crustaceans and fish caused by drift to the surfacewater are expected, based on an EC_{50} of .. mg/l for algae, an EC_{50}/LC_{50} of .. mg/l for crustaceans and an LC_{50} of .. mg/l for fish:

Table: Risk assessment of the chance of **acute** death in or effects on crustaceans and fish, and the chance of growth inhibition in algae

application	no. form.	dose kg a.i./ha	emission %	concentration* mg/l or µg/l	risk assessment algae	*Daphnia*	fish
mushrooms			most critical				
			least critical				

N = no, L = low, M = medium, H = high, U = unknown
* calculated according to slootbox

* Chronic:

Upon application according to agricultural requirements for a/different formulation(s) containing (name) and assuming a ditch depth of 25 cm, the following hazards for algae, crustaceans and fish caused by drift to the surface water are expected, based on an NOEC of .. mg/l for algae, an NOEC of .. mg/l for crustaceans and an NOEC of .. mg/l for fish:

Table: Risk assessment of the chance of **chronic** effects on algae, crustaceans and fish #

application	no. form.	concentration* in µg/l or mg/l			risk assessment		
		algae (4d)	*Daphnia* (21d)	fish (28 d)	algae	*Daphnia*	fish
mushrooms (most critical)							
mushrooms (least critical)							

N = no, L = low, M = medium, H = high, U = unknown
* calculated according to slootbox
\# for dose and emission see risk assessment acute effects

If using (name) (no/an unknown/a low/a medium/a high) hazard for aquatic organisms is expected.

- Combination toxicity for aquatic organisms:

* Acute:

Upon application according to agricultural requirements for a/different formulation(s) containing, besides (name), one or more other active ingredients, and assuming a ditch with a depth of 25 cm, the following hazards for algae, crustaceans and fish caused by drift to the surface water, are expected, based on the toxicity values below:

number formulation	algae EC50 in mg/l		*Daphnia* E(L)C50 in mg/l		fish LC50 in mg/l	
	calculated*	measured	calculated*	measured	calculated	measured

* based on additivity

Table: Combination toxicity. Risk assessment of the chance of **acute** death or effects on crustaceans and fish and the chance of growth inhibition in algae

field of application	no.	dose	emission	concentration*	risk assessment#		
	form.	kg a.i./ha	%	mg/l or µg/l	algae	*Daphnia*	fish
mushrooms			most critical				
			least critical				

N = no, L = low, M = medium, H = high, U = unknown
* calculated according to slootbox and based on the combination product
\# if available, based on the measured toxicity value

* Chronic:
Upon application according to agricultural requirements for a/different formulation(s) containing, besides (name), one or more other active ingredients and assuming a ditch depth of 25 cm, the following hazards for algae, crustaceans and fish, caused by drift to the surface water, are expected, based on the toxicity values below:

number formulation	algae NOEC in mg/l		*Daphnia* NOEC in mg/l		fish NOEC in mg/l	
	calculated*	measured	calculated*	measured	calculated*	measured

* based on additivity

Table: Combination toxicity. Risk assessment of the chance of **chronic** effects on algae, crustaceans and fish

application	no.	concentration* in µg/l or mg/l			risk assessment#		
	form.	algae (4d)	*Daphnia* (21d)	fish (28d)	algae	*Daphnia*	fish
mushrooms (most critical)							
mushrooms (least critical)							

N = no, L = low, M = medium, H = high, U = unknown
* calculated according to slootbox and based on the combination product
\# if available, based on the measured toxicity value
@ for dose and emission see risk assessment acute effects

- Earthworms:
(name) is (classification) toxic for earthworms: .. -day LC_{50} (value or range) mg/kg (.. species).
(possibly: a ..% formulation of ... is etc.)

- Risks for earthworms:
Upon application according to agricultural requirements for a/different formulation(s) containing (name) the following hazards for earthworms are expected, based on an LC_{50} of .. mg/kg soil:

Table: Risk assessment of the chance of acute death in earthworms

application	number formulation	dose kg a.i./ha	concentration mg/kg	hazard

N = no, L = low, M = medium, H = high, U = unknown

- Combination toxicity for earthworms:
Upon application according to agricultural requirements for a/different formulation(s) containing, besides (name), one or more other active ingredients, the following hazards for earthworms are expected, based on the toxicity values below:

number formulation	LC50 in mg/kg soil	
	calculated	measured

Table: Combination toxicity. Risk assessment of the chance of acute death in earthworms

application	number formulation	dose kg a.i./ha	concentration mg/kg	hazard#

N = no, L = low, M = medium, H = high, U = unknown
# if available, based on the measured toxicity value

- Bees:
(name) is (classification) toxic for bees: oral LD_{50} (value or range) µg/bee (.. species); contact LD_{50} (value or range) µg/bee (.. species).
(possibly: a ..% formulation is etc.)

- Beneficial insects and mites:
(short description of the test and the results)

- Bees, beneficial insects and mites:
Upon application according to agricultural requirements for a/different formulation(s) containing (name), the following hazards for bees, beneficial insects and mites are expected, based on an oral LD_{50} of .. µg/bee and a contact LD_{50} of .. µg/bee for bees:

Table: Risk assessment of the chance of acute death in bees, beneficial insects and mites

application	no. form.	dose kg a.i./ha	hazard bees	name insect/mite	reduction %	hazard insect/mite

N = no, L = low, M = medium, H = high, U = unknown

- Combination toxicity for bees, beneficial insects and mites:
Upon application according to agricultural requirements for a/different formulation(s) containing, besides (name), one or more other active ingredients, the following hazards for bees, beneficial insects and mites are expected, based on the toxicity values below:

number formulation	LD50 oral in µg/bee		LD50 contact in µg/l	
	calculated	measured	calculated	measured

if available, based on the measured toxicity value

Table: Combination toxicity. Risk assessment of the chance of acute death in bees, beneficial insects and mites

application	no. form.	dose kg a.i./ha	hazard bees	name insect/mite	reduction %	hazard insect/mite

N = no, L = low, M = medium, H = high, U = unknown

- Soil respiration:
Upon application of (name) in normal and .. fold doses, no significant effect on the soil respiration was observed after .. days at ..°C.
(Or: ....(description of effect) was observed).

- Nitrification:
Upon application of (name) in normal and .. fold doses, no significant effect on the nitrification (and ammonification) was observed after .. days at .. °C.
(Or: .... (description of effect) was observed).

Upon application according to agricultural requirements for a/different formulation(s) containing (name), the following hazards for nitrification are expected, based on an EC_{50} of .. mg/kg:

Table: Risk assessment of the chance of influence on the nitrification

application	no. form.	dose kg a.i./ha	concentration mg/kg	hazard

N = no, L = low, M = medium, H = high, U = unknown

- Influence on activated sludge:
() has an IC_{50} of ... mg/l / NOEC of ... mg/l for activated sludge.

- Risk for activated sludge:
Upon application of () the next concentrations in the STP and the next hazards for activated sludge are calculated with USES, using an NOEC of ... mg/l (which equals a NEC of ... mg/l) / IC_{50} of ... mg/l (which yields a NEC of ... mg/l):

Table: Risk assessment of the chance of influence on activated sludge

application	no. form.	dose kg a.i./ha	concentration mg/kg	hazard

N = no, L = low, M = medium, H = high, U = unknown

Appendix 9: Glossary

ACT	Adviescentrum Toxicologie (Dutch), Toxicology Advisory Centre (English)
acute toxicity test	toxicity test serving to study the effects occurring in a short time following the administration of a single dose or multiple doses given within this short time period
Additional Questions	a list with the questions that should be answered by a company that claims admission of a pesticide on the Dutch market. This list is made after accordation of the Advisory Report by the Toxicology Advisory Group and reflects the uncompleteness of the data supplied by a company
adsorption	enrichment of one or more components in an interfacial layer
advection	intercompartmental transfer of a chemical by a carrier that physically flows from one compartment to the other; examples are atmospheric deposition, sedimentation, and resuspension
adverse effect	change in morphology, physiology, growth, development or lifespan of an organism
Advisory Report	a report on a pesticide that consists of the Summaries of the supplied tests and the RIVM Conclusion with the concluding remarks on the physicochemical properties, the fate in the environment, the effects, and the hazards
AR	see Advisory Report
AQ	see Additional Question(s)
BAV	Beoordeling Algemeen Volksgezondheid (Dutch), Department of Evaluating Public Health Aspects of Chemicals (English)
BBM	Beoordeling van Bestrijdingsmiddelen in het Milieu (Dutch), Department of Evaluating Environmental Aspects of Pesticides (English)
BCF	BioConcentration Factor: the ratio of the test substance concentration in (part of) an organism (e.g. fish, plant) to the concentration in a medium (e.g. water, soil) at steady state
bioaccumulation	the net result of the uptake, distribution, and elimination of a substance due to all routes of exposure
bioconcentration	the net result of the uptake, distribution and elimination of a substance due to water-borne exposure
biodegradation	see degradation

biotransformation	see transformation
BOD	Biological Oxygen Demand
bound residue	residu that cannot be extracted from soil or sediment after several subsequent extractions, applying methods that do not alter the chemical structure of these residues substantially
CEC	Cation Exchange Capacity
Chemical Identity	menu in TOXIS to registrate identifying data of a substance (e.g. CAS number)
Chemobiokinetics	menu in TOXIS to registrate the metabolism routes of a substance
chronic toxicity test	toxicity test in which organisms are observed during the whole life-span and in which exposure to a substance takes place over the whole observation time or a substantial part thereof
CTB	College voor de Toelating van Bestrijdingsmiddelen (Dutch), Board for the Authorisation of Pesticides (English). Up to 1994 the name was Committee for Registration of Pesticides
degradation	conversion of a molecule to smaller molecules by (micro)biological or chemical action
degradation rate	the rate at which a pesticide can be degraded. This is often expressed as a DT_{50}
degradation route	route along which a substance is degraded to metabolites
Description	unstructured part of a Summary consisting of the Methodology, the Results, and the Remarks (see Summary, Results, and Remarks). A Description and a Header form a Summary. One Description per test
Detection	menu in TOXIS to registrate detection methods
DGM	Directoraat-Generaal Milieubeheer (Dutch); Directorate General for Environmental Protection (English)
dissipation	disappearance of the parent compound from a compartment (such as soil or water) in which various processes such as conversion, evaporation, leaching, etc. can play a role.
DO	Dissolved Oxygen
DOC	Dissolved Organic Carbon
dose-response assessment	the estimation of the relationship between dose or concentration and the incidence and severity of an effect

drift	the part of the applied dose of a sprayed pesticide that reaches the surface water directly, expressed as a fraction of the nominal dose per m^2
drift percentage	see emission percentage
DT_{50}	time in which 50% of the parent compound has disappeared from soil or water by transformation or degradation (under standard conditions). See degradation and transformation
$DT_{50,sys}$	DT_{50} in the whole system (water + sediment). This DT_{50} pertains to a biodegradation test in water with the accompanying sediment
$DT_{50,wat}$	DT_{50} in the water column. This DT_{50} pertains to a biodegradation test in water with the accompanying sediment. This DT_{50} is often considered as relevant for the actual exposure of algae, waterfleas and fish (rather than the $DT_{50,sys}$
EC_{50}	median Effective Concentration: 1. the concentration resulting in a 50% change in a parameter (e.g. algal growth) relative to the control 2. the concentration at which a particular effect (e.g. daphnia immobilization) is observed in 50% of the organism population relative to the control
ECO	Laboratorium voor Ecotoxicologie (Dutch), Laboratory of Ecotoxicology (English)
effect	the extent of biological changes
effect assessment	(more precisely, dose-response assessment) the estimation of the relationship between dose or level of exposure to a substance, and the incidence and severity of an effect
emission percentage	loss from the compartment the product is applied, depending on the method of application and calculated as a fraction of the dosage used
EPPO	European and Mediterranean Plant Protection Organisation
exposure assessment	the determination of the emissions, pathways and rates of movement of a substance and its transformation or degradation in order to estimate the concentrations/doses to which environmental compartments, human populations are or may be exposed
FC	field capacity
FEF	see Final Environmental File
field capacity	the moisture content of the soil at pF= 2 - 2.5
Final Environmental File	the RIVM Conclusion in a specific format to be used as a crucial document for registration policy

Fixed Field	see Header
formulation	form and composition in which a pesticide is marketed (such as a dust, granules, a liquid); beside the active ingredient the formulation contains ingredients which make it more manageable, or improve its application potential, efficacy or safety
Formulations	menu in TOXIS to registrate formulations
Free Text	unstructured part of a Summary to be stored in TOXIS in which any kind of information can be included
GLP	Good Laboratory Practice: a set of rules describing how a laboratory should work, how it should be organised and how it can produce valid data; GLP principles are described by e.g. OECD
Guideline	an official Guideline (i.e. authorized by national or international institutions, e.g. EPA, NEN, BBA, OECD) for the protocol and the report of a test
H	see Henry's Law Constant
hardness (of water)	property of water indicating the total amount of calcium, magnesium and barium
hazard	the potential of a substance to cause adverse effects at a particular degree of exposure (Dutch: gevaar, risico *in algemene zin*)
hazard identification	the identification of the adverse effects which a substance has an inherent capacity to cause
Header	structured part of a Summary to be stored in TOXIS. A Header contains the most relevant items of a test and forms in this way the "head" of a summarised test. The Header contains Fixed Fields (i.e. specifically meant for including a particular item, e.g. one for the pH and one for the DT_{50} in a soil degradation test)
Henry's law constant	air-water partition coefficient; the ratio between the partial pressure in the gas phase of a compound and its concentration of a substance in water. Henry's law constant can be with ($Pa \times m^3 \times mol^{-1}$, synonym is H') or without dimension (synonym is H)
hydrolysis	a chemical reaction of a substance with water in which a part of the molecule of the reacting substance is replaced by an OH group
IC_{50}	median Inhibitory Concentration: the concentration resulting in a 50% inhibition of growth relative to the control
index	value used as a measure for e.g. reliability (see Reliability Index). The plural, for reasons of convenience, is indicators

indicator	plural of index (see index)
Instructions	the instructions comprise both the official Guidelines on testing and reporting and the BBM-directives on summarising and evaluating test reports. Instructions refer to all guidance giving statements in this report: standards, cut-off values, useful formulas, standard sentences, selection criteria, etc; they point out what items should be included in the summary and how to handle the abundant information
IUPAC	International Union of Pure and Applied Chemistry
K_{aw}	air-water partition coefficient. See Henry's law constant
K_F	Freundlich coefficient: a soil-water partition coefficient —or sorption coefficient— *dependent* on the ratio 1/n (n is an empirical entity which describes the non-linearity of an adsorption isotherm)
K_{om}	sorption coefficient normalised to the fraction of organic matter in soil
K_{ow}	octanol-water partition coefficient
$K_{s/l}$	a soil-water partition coefficient —or sorption coefficient— *independent* on the ratio 1/n (n is an empirical entity which describes the non-linearity of an adsorption isotherm)
LAC	Laboratorium voor Anorganische Chemie (Dutch), Laboratory of Inorganic Analytical Chemistry (English)
LBG	Laboratorium voor Bodem en Grondwater Onderzoek (Dutch), Laboratory for Soil and Groundwater Research (English)
leachate	water leached out from a soil (column)
leaching	transfer of a chemical from the top layer of soil to the subsoil (and subsequently to the groundwater)
LC_{50}	median Lethal Concentration: a statistically derived concentration that can be expected to cause death in 50% of animals exposed for a specified time
LD_{50}	median Lethal Dose: statistically derived single dose that can be expected to cause death in 50% of dosed animals
LNV	Ministerie van Landbouw, Natuurbeheer en Visserij (Dutch), Ministry of Agriculture, Nature Management and Fisheries
LOC	Laboratorium voor Organische Chemie (Dutch), Laboratory of Organic Analytical Chemistry (English)
long term	duration of exposure ≥96 hours (aquatic organisms); duration of exposure

	≥5 days feeding (birds)
metabolite	substance formed from the parent compound by transformation, synonym for transformation product
maximum water holding capacity	the moisture content at pF = 0 (saturation)
mineralisation	degradation of a substance into inorganic end products; it is usually estimated in terms of CO_2 production.
MWHC	see maximum water holding capacity
NOEC	No-Observed-Effect-Concentration: the highest concentration without adverse effects
o.c.	organic carbon
o.m.	organic matter
P	vapour pressure
partition coefficient	ratio of the distribution of a substance between two phases when the heterogeneous system (of two phases) is in equilibrium; the ratio of concentrations (or, strictly speaking, activities) of the same molecular species in the two phases is constant at constant temperature
PD	Plantenziektenkundige Dienst (Dutch), Plant Protection Service (English)
PEC	Predicted Environmental Concentration; the expected concentration in an environmental compartment
persistence	residence time of a substance in a compartment; the disappearance rate being dependent on one or more dissipation processes
PESTLA	PESTicides Leaching and Accumulation model: calculates concentrations in soil and groundwater
photochemical transformation	the breakdown of a compound as a result of irradiation by light
photolysis	see photochemical transformation
phototransformation	the reaction of a compound with (hydroxyl, ozone, nitrate) radicals produced by the action of light
PIEC	Predicted Initial Environmental Concentration
pK_a	$-\log K_a$. K_a is the dissociation constant of an acid or base at equilibrium,

	in other words, the pH at which 50% of the molecules of is dissociated—an acid—or protonated—a base
quality	the degree of excellence of a test as determined by both its reliability and usefulness (see reliability and usefulness)
Quality Assurance	internal laboratory control system to ascertain that tests are in compliance with GLP principles
reliability	the intrinsic reliability of a test with respect to methodology and description
Reliability Index	value —1,2,3,or 4— indicating the reliability of a test
Reliability Indicators	plural of Reliability Index
Remark(s)	unstructured part of a Summary to enter critical statements on e.g. the reliability of a test, and on the usefulness of the test for the hazard assessment
Result(s)	unstructured part of a Summary to enter the results of a test and the comments of the reviewer
Rf	retardation factor: the distance moved by a substance relative to the distance moved by the water front
RI	see Reliability Index
risk	the probability of a substance to cause adverse effects (Dutch: risico, in statistische zin)
risk assessment	determination of the relation between the predicted exposure and adverse effects in four major steps: hazard identification, dose-response assessment, exposure assessment and risk characterisation
risk characterisation	the estimation of the incidence and severity of the adverse effects likely to occur in a human population or environmental compartment due to actual or predicted exposure to a substance; it may include *risk estimation* (i.e. the quantification of that likelihood using probability functions)
RIVM Conclusion	the RIVM Conclusion contains the concluding remarks on classification of the physico-chemical data, the environmental fate data, and the (eco)toxicological data (i.e. the effect and the exposure assessment) and on the hazard assessment. Together with the Summaries, the RIVM Conclusion forms an Advisory Report
RIZA	Rijksinstituut voor Integraal Zoetwaterbeheer en Afvalwaterbehandeling (Dutch), Institute for Inland Water Management and Waste Water Treatment (English)

S	water solubility
short-term	duration of exposure ≤96 hours (aquatic organisms); duration of exposure ≤5 days feeding (birds)
SLOOT.BOX	a model which calculates the concentration in a fictitious ditch as a result of drift
STP	Sewage Treatment Plant (synonym for WWTP)
Substances	menu in TOXIS to registrate substances
Summary	a Summary is a concise text, consisting of a Header and a Description (see Header and Description) including the most relevant aspects of a test.
summary table	a table in this report with a concise overview of the items in a particular test that can influence the reliability
TAG	see Toxicology Advisory Group
TLC	Soil Thin or Thick Layer Chromatography
TOC	Total Organic Carbon
Toxicology Advisory Group	a panel of specialists from both ACT and other laboratories of RIVM. Each Advisory Report has to be accorded by such a panel (synonym for beoordelingsgroep, Dutch)
transformation	conversion of a molecule to larger or smaller molecules by (micro)biological or chemical action
transformation rate	the rate at which a pesticide can be transformed. This is often expressed as a DT_{50}
transformation route	route along which a substance is transformed to metabolites
Uniform Principles	EU guidance on the evaluation of plant protection products
usefulness	the extent to which a test is appropriate for a particular purpose (e.g. standard setting procedures, hazard or risk assessment) Synonyms: relevance, bruikbaarheid (Dutch)
USES	Uniform System for the Evaluation of Substances, a decision-support system, including models for calculation of exposure and hazard in environmental compartments
water holding capacity	the moisture content at field capacity (pF = 2 - 2.5)

wo	whole organism
WWTP	Waste Water Treatment Plant (synonym for STP)