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Guidance Document on Persistence in Soil

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This document has been conceived as a working document of the Commission Services which was elaborated in co-operation with the Member States. It does not intend to produce legally binding effects and by its nature does not prejudice any measure taken by a Member State within the implementation prerogatives under Annex II, III and VI of Directive 91/414/EC, nor any case law developed with regard to this provision. This document also does not preclude the possibility that the European Court of Justice may give one or another provision direct effect in Member States.

1 Introduction

Annexes II and III of Directive 91/414/EEC set out the data requirements for the inclusion of an active substance onto Annex I of the Directive and for the authorisation of a plant protection product at Member State level. Annex VI of the Directive includes the decision making criteria for the authorisation of plant protection products at Member State level. Given that no other harmonised criteria are currently available, Annex VI should also be used in an appropriate way for the decision on whether or not Annex I listing of an active substance can be recommended. Over the last few years, at several meetings in the frame of Directive 91/414/EEC, issues related to environmental fate and behaviour (persistence in soil) have been discussed. Several points in environmental fate and behaviour (persistence in soil) of Annexes II, III and VI were identified during these discussions where expert judgement is required or where there is scope of different interpretation.

The purpose of this document is to provide guidance to regulatory authorities in Member States and to Notifiers on the use of the environmental fate and behaviour (persistence in soil) sections of Annexes II, III and VI, allowing flexibility in decision making where necessary. This document should also serve as a bridge to the assessment of the ecotoxicological effects in the terrestric and aquatic ecosystems (Draft Guidance Document on Aquatic Ecotoxicology, Document 8075/VI/97 and Draft Guidance Document on Terrestrial Ecotoxicology, Document 2021/VI/98). The primary aim is to promote consistency and transparency in decision making for the benefit of the public, industry and policy makers. To enhance efficiency of the review process is a further objective as repetitive discussions on general items in meetings dedicated to individual substances lead to a prolongation of the process of Annex I listing.

The environmental fate and behaviour (persistence in soil) data requirements for active substances and plant protection products are set out in Annex II, section 7 and Annex III section 9 of Directive 91/414/EC, respectively. It should be noted that the introduction to both these sections provide useful information on the purpose and use of data submitted. It is clearly stated that the data submitted must be sufficient to permit an assessment of the environmental fate and behaviour (persistence in soil). The environmental and agricultural significance of persistent compounds should be assessed according to defined procedures. In order to fulfil this objective, tests additional to those outlined in Annex II and III may be needed in individual cases if there is an established concern and specific justification.

2 Persistence

This document deals with persistence in soil of active substances in plant protection products as well as, where they are relevant from the toxicological, ecotoxicological or environmental point of view, metabolites, breakdown or reaction products resulting from these active substances.

The aim of this guidance document is to provide applicants with advice concerning the information which should be submitted in order to allow an evaluation of persistent active substances and with elaborations on how to assess accumulation levels. Persistence is defined as the residence time of an active substance in a defined compartment of the environment as outlined in Annex VI of Council Directive 91/414/EEC. Persistent active substances are those which exceed the limits defined in this Annex VI, point 2.5.1.1. The reason for focusing on the soil compartment is that this is the compartment of concern with regard to persistence.

The persistence of an active substance is the integrated effect or weighted summation of transformation and loss processes like

- processes which result in transformation, degradation and eventually mineralisation of a substance, including microbial degradation, chemical hydrolysis, and photochemical reactions,
- other processes, such as leaching, volatilisation and uptake by plants.

Factors influencing persistence or, conversely, the disappearance of an active substance include crop factors (morphology, cuticle characteristics, stage of growth at treatment, and rate of growth), characteristics of the active substance (Henry's law constant, water solubility, susceptibility to physical, chemical, and biological transformation processes), and environmental conditions (rainfall, wind, humidity, temperature, light-intensity) (Himel et al., 1990).

Assessment of the intrinsic biodegradability is fundamental in the assessment of the fate and behaviour of all chemical substances, including active substances in plant protection products. However, for the interpretation of disappearance observed in the field studies on active substances also other processes (photolysis, leaching, volatilisation) as well as specific environmental conditions (e.g. climate) must be taken into account. Further, in the assessment of persistence, the proposed use of products should not only be considered for one year (e.g. application rate) but also over a longer time period (e.g. differentiation between repeated application in one year, in succeeding years or in non-consecutive years).

Annex VI to Council Directive 91/414/EEC concerning the placing of plant protection products on the market lays down Uniform Principles aiming to ensure that evaluations and decisions with regard to authorization of plant protection products, provided they are chemical preparations, result in the implementation of the requirements of Article 4 (1) (b), (c), (d) and (e) of the Directive by all the Member States at the high level of protection of human and animal health and the environment. In the evaluation of the fate, the distribution and the effects of the plant protection product in the environment, Member States shall have regard to all aspects of the environment. With respect to **persistence of active substances** the following is provided:

2.5.1.1 Fate and behaviour in the environment (Annex VI, part C, decision-making):

No authorization shall be granted if the active substance and, where they are of significance from the toxicological, ecotoxicological or environmental point of view, metabolites and breakdown or reaction products, after use of the plant protection product under the proposed conditions of use:

- during tests in the field, persist in soil for more than one year (i.e. DT90 > 1 year and DT50 > 3 months), or
- during laboratory tests, form non-extractable residues in amounts exceeding 70 % of the initial dose after 100 days with a mineralization rate of less than 5 % in 100 days,

unless it is scientifically demonstrated that under field conditions there is no accumulation in soil at such levels that unacceptable residues in succeeding crops occur and/or that unacceptable phytotoxic effects on succeeding

crops occur and/or that there is an unacceptable impact on the environment, according to the relevant requirements provided for in points 2.5.1.2, 2.5.1.3, 2.5.1.4 and 2.5.2.

In the following chapters guidance is given on how to deal in detail with persistence of active substances including non-extractable residues in the evaluation and decision-making process according to the Uniform Principles.

3 Determination of DT50/DT90 and influential factors

In context of Annex VI of Council Directive 91/414/EEC (part C, Decision-making, point 2.5.1.1) soil persistence is related to the DT50 and DT90 values, respectively from field dissipation studies. Evaluation starts, however, earlier - with consideration of laboratory soil degradation studies.

Criteria for conducting field dissipation studies

According to Annexes II and III of the Council Directive 91/414/EEC, points 7.1.1.2.2 and 9.1.1.2, respectively, soil dissipation studies should provide (best-possible) estimates of the time needed for dissipation of 50 % and 90 % of the active substance under field conditions. Where relevant, information on relevant metabolites, degradation and reaction products must also be reported.

Generally, field studies have to be conducted in those cases where the $DT50_{lab}$ (for calculation see below) determined in the laboratory at 20 °C and at a moisture content of the soil related to a pF value of -10 to -32 kPa (suction pressure) is greater than 60 days. The decision for the conductance of field dissipation studies should be triggered by the results of realistic worst case laboratory degradation studies. In general, to assess whether an active substance is persistent or not, the worst case degradation value from laboratory studies should be used, unless there is convincing justification to disregard individual results due to non representative extreme conditions (e.g. due to methodological problems). A detailed consideration should be carried out if there is any special correlation to the soil type, e.g. pH dependence. Test conditions as specified in the Annexes II and III, points 7.1.1.2.1 and 9.1.1.1, respectively, for conductance of these studies should be followed as close as possible.

If plant protection products containing the active substance are intended to be used in cold climatic conditions, where the soil temperature over the time period following the application is $\leq 10^{\circ}$ C, field tests have to be conducted where in the laboratory DT50 at 10 °C is greater than 90 days (Annex II, point 7.1.1.2.2).

The persistence of formulated plant protection products in soil must be investigated unless it is possible to extrapolate from data obtained on the active substance and relevant metabolites, degradation and reaction products in accordance to the requirements of Annex II, point 7.1.1.2. This extrapolation is, for example, not possible for slow-release formulations (Annex III, points 9.1.1.1 and 9.1.1.2).

Guidance for conducting laboratory and field dissipation studies

Guidance for conducting laboratory and field dissipation studies is given in the Annexes II (chapter 7) and III (chapter 9) of the Council Directive 91/414/EEC.

Methods for calculation of DT50/90-values

General recommendations

For the calculations of DT50 and DT90 values in laboratory as well as field dissipation studies the following recommendations are given:

- For a sound regression analysis in calculating DT50, at least 5 sampling times are required including zero time.
- Care should be taken when using time points from near the end of soil degradation/field dissipation studies for the calculation of DT50/90 values, when the concentration of the remaining active substance is low (< 2-5 % initial concentration), especially when concentrations are approaching the limit of quantification for the method of analysis for non-radiolabelled studies.
- Experience shows that DT50 can usually be calculated from first-order kinetics, and this is the preferred method. The determination coefficient r^2 should be in a range between 0.85 and 1.0. In a practice there will be many cases where r^2 will be lower than 0.85. In such situations is advisable to distinguish if a DT50 is needed for modelling purposes or as a trigger value for further (field) studies. Since most models can handle only 1st order kinetics, for pragmatic reasons the determination coefficient $r^2 \ge 0.7$ can still be accepted. In order to trigger further studies a DT50 value can be calculated according to the best fit. If the use of first order kinetics to calculate degradation rates results in a determination coefficient of $r^2 < 0.7$, then other methods can be tested and used:
 - -- As a first option, approximation of two degradation/dissipation rates to first order kinetics (one for the initial part and one for the later part of the degradation/dissipation process) should be tested which may be shown up by a hinge point in the curve. Rather simple statistical methods are available in standard statistical software to show this. A hinge point can arise as a result of a change in the contribution to degradation of various processes over a period of time. For example, a hinge point may be caused by a significant decline of microbial activity or bioavailability in the soil or by adaptation. Therefore the hinge point does not represent an instantaneous change in the degradation process but is the product of the limitations of sampling intensity and does not reflect gradual changes in processes and possible bioavailability.
 - -- The results of the fit give the first order rate coefficient as one of the two regression coefficients. The DT50 and DT90 are calculated using the formula:

DT50 =
$$\frac{\ln 2}{k}$$
 and DT90 = $\frac{\ln 10}{k}$

When there is a hinge point in the degradation curve, the calculation of the DT90 is less simple, the complication may be taken into account on a case-by-case basis.

- -- Mathematical models which fit the data can be used (e.g., Gustafson and Holden (1990), ModelMaker, TopFit).
- From the shape of the curve of concentration against time, one can decide whether a lag phase has to be taken into account. A lag phase may be assumed where at least three measurement points are more or less on a horizontal line. The length of the lag phase has to be reported. The DT50 is then calculated by leaving out the experimental results within the lag phase. At least five sampling times including zero time must be available after excluding the 3 points of the lag phase.

Special aspects of laboratory studies

The following aspects should be taken into account when considering soil degradation studies in the laboratory:

- Often DT90_{lab} is difficult or impossible to obtain for persistent compounds because of the obvious problems with extrapolation beyond the end of study periods and the general problem with extremely long study durations making statistical analysis of the data very inaccurate. When first order kinetics is applicable, then mathematically the DT90_{lab} can be estimated as 3 times DT50_{lab}. In addition to these points, loss of microbial activity of the soil might result in a decrease in the rate of degradation after approximately 2 to 4 months of incubation.

- Effect of temperature on the degradation rate, where relevant.
- The Arrhenius equation is a validated relationship which can be used to describe temperature effects on transformation. A Q_{10} -value of 2.20 could reasonably be used to extrapolate DT50 data derived at 20 °C to expected values at 10 °C (FOCUS, Doc. 7617/VI/96). A Q_{10} -value can also be calculated if degradation studies have been carried out at different temperatures. In every case, the method used for calculating the compound-specific Q_{10} -value should be clearly described.
- The methods described are also used for metabolites, breakdown or reaction products, where they are relevant from the toxicological, ecotoxicological or environmental point of view, if separate studies with these substances are available.

Special aspects of field dissipation studies

If the trigger values of $DT50_{lab} > 60 d (20 °C)$ or $DT50_{lab} > 90 d (10 °C)$ are exceeded, then individual field dissipation studies on a range of representative soils (normally four different types) must be conducted according to the relevant guidance e.g. SETAC or BBA (Annexes II and III, points 7.1.1.2.2 and 9.1.1.2, respectively). The soil types should be representative for the regions of the intended use of the product. The following aspects should be taken into account when $DT50/90_{field}$ values are to be determined:

- In laboratory experiments mainly (bio)degradation is considered whereas in field studies in addition to (bio)degradation other dissipation routes like volatilisation, leaching or photolysis do occur.

Consequently, a clear assessment concerning the routes of dissipation in addition to (bio)degradation must be given. The following recommendations should be considered:

- -- For most compounds, volatilisation is not an important factor (however it may be a dominant factor for some active substances).
- -- The dissipation rates should be calculated from the soil residues using only those soil core depth segments where quantifiable residues occur, so that the influence of leaching on the dissipation rate is minimised, and taking into account the possibility that the highest residue level in soil may not occur on the day of treatment but several days after.
- -- Photolysis may be an important degradation mechanism for some compounds, especially when applied to bare soil and not incorporated.
- For further interpretation of the results from field studies and their use in exposure assessment, the range of all observed $DT50_{field}$ and $DT90_{field}$ values should be reported, together with a full consideration of any possible correlation with the soil/climatic conditions present in the field studies. Differentiation according to soil/climatic conditions can be done taking into account the FOCUS scenario regions in Europe. Further guidance can be obtained from FOCUS documents currently being developed.

Because the standard number of field dissipation studies required is four there is little likelihood that the particular contribution of each variable (i.e. temperature, soil water content, soil type, soil pH, soil microbial biomass etc.) can be determined since there are more potential variables than results. When interpreting field dissipation study results, important parameters such as soil temperature and moisture (and for some compounds pH, texture, and organic matter/carbon content) should be considered. Although originally developed for laboratory studies, one way to consider these effects is to use the Walker corrections to adjust field rate to standard temperature or moisture conditions (Hurle and Walker 1980, Walker and Allen 1984, FOCUS Report Soil Persistence Models and EU Registration, Document 7617/VI/96). A model for such calculations is available (Walker 1974).

- Field dissipation data should be seen as more definitive than modelling predictions according to the final report of the work of the Soil Modelling Workgroup of FOCUS (Doc. 7617/VI/96). Nevertheless, simulation models may be used to preliminarily extrapolate the field study results to other regions of use. Extrapolation should only be undertaken if the behaviour of the active substance at the original field site is well understood and can be adequately simulated by a model using laboratory derived input parameters (i.e. the knowledge of active substance properties obtained from laboratory degradation studies, laboratory sorption studies, vapour pressure measurements etc. can be shown to be reflected in the field behaviour).

Use of data from other geographical areas

For many existing active substances field dissipation studies conducted in, for example, the United States under conditions comparable to Europe have normally been taken into consideration for the environmental exposure assessment. However for new active substances it is anticipated that appropriate field dissipation studies will always be conducted in European countries and in areas of intended use unless it can be demonstrated that dissipation data generated outside of Europe is comparable to soil/climatic and agricultural conditions within Europe.

4 Determination of the soil accumulation potential

If the trigger values of DT50 of 3 months and DT90 of one year in the field or non-extractable residues greater than 70 % with a mineralization rate of less than 5 % in 100 days are not met, no authorisation shall be granted (Annex VI, part C, point 2.5.1.1). However, this provision is not intended to set a cut-off point to the evaluation process, because its "unless" clause allows further consideration:

It is to be scientifically demonstrated that there is no accumulation in soil under field conditions at such levels that unacceptable residues in succeeding crops occur and/or unacceptable phytotoxic effects on succeeding crops occur and/or that there is an unacceptable impact on the environment (Annex VI, part C, point 2.5.1.1). The assessment procedure is described in the Draft Guidance Document on Terrestrial Ecotoxicology, Document 2021/VI/98.

Possible ways to deal with the "unless"-clause on persistence are outlined below in more detail. Regarding further consideration of the "unless" statement for non-extractable residues see chapter 6.

If on the basis of soil dissipation studies it is established that $DT90_{field} > one$ year and repeated applications are envisaged, whether in the same growing season or in succeeding years, the possibility of accumulation of residues in soil and the level at which a plateau concentration is achieved must be investigated unless reliable information can be provided by a model calculation or another appropriate assessment (Annexes II and III, points 7.1.1.2.2 and 9.1.1.2, respectively). One of the following approaches can be followed:

- A soil accumulation study generally takes several years to determine a plateau level after multiple applications. Multiple applications may be made during one growing season and/or during more years. As a rule, soil cores should be taken from control and treated plots prior to application and following the last application in one growing season or prior to and following applications during more years. The plateau level just before the last application should be determined, which means in the resulting "saw-teeth"-curve extrapolating the lowest points in order to obtain the plateau concentration to be used for risk assessment. It may show that before a plateau level has been reached already effects on succeeding crops or phytotoxic effects are noticeable. In such cases these effects have to be taken into account. With regard to the assessment of effects of persistent substances on terrestrial organisms, the extrapolation of the upper limits of the resulting "saw-teeth"-curve is of interest. The peak represents the exposure pattern that is relevant for toxicity testing as well as for the TER-calculations.
- The FOCUS Soil Modelling Workgroup (Doc. 7617/VI/96) recommends a simple model for **calculation** of the plateau concentration assuming dissipation to be first order. If dissipation cannot be assumed to be first order then equivalent procedures can be used, or more detailed simulations models and/or field data can be used.
- It is also possible to conduct **integrated studies** in which both, the determination of plateau level as well as further ecotoxicological testing is combined.

5 Plateau concentration versus unacceptable residues, effects and impact

Persistent substances may give rise to long-term exposure of organisms in the environment and may spread to other compartments of the environment from the one they are applied to. Therefore, extra precaution needs to be taken when evaluating substances with DT50 and DT90 respectively greater than the limits set out in the Uniform Principles. According to the Uniform Principles the determined plateau concentration in soil must not lead to unacceptable residues in succeeding crops and/or unacceptable phytotoxic effects on the succeeding crop and/or unacceptable impact on the environment. The assessment of these effects for non-target organisms or succeeding crops considers the bioavailibility of the persistent substances and is outlined in the Draft Guidance Document on Terrestrial Ecotoxicology, Document 2021/VI/98.

Unacceptable residues in succeeding crops

For data requirements refer to Annexes II and III, sections 6 and 8 of Council Directive 91/414/EEC. With regard to Uniform Principles refer to Annex VI, part C, point 2.4 of Council Directive 91/414/EEC. Further guidance is currently being developed (Draft Guidelines for the generation of data concerning residues (Doc. 1607/VI/97)).

Unacceptable phytotoxic effects on succeeding crops

Phytotoxic effects are discussed in the context of efficacy data which are considered at Member State level and should not be further discussed here. For data requirements refer to Annex III, section 6, point 6.1.1 of Council Directive 91/414/EEC. With regard to Uniform Principles refer to Annex VI, part C, point 2.2.5 of Council Directive 91/414/EEC. Further guidance is currently being developed by EPPO (Draft Guideline for the Efficacy Evaluation of Plant Protection Products).

Unacceptable impact on the environment

Groundwater

For data requirements refer to Annexes II/III, sections 7 and 9 of Council Directive 91/414/EEC. With regard to Uniform Principles refer to Annex VI, part C, 2.5.1.2 of Council Directive 91/414/EEC. The evaluation of the possibility of an active substance to reach groundwater has to take into account realistic worst case scenarios, e.g. range of DT50 values, of K_{oc} values, different climates. Further guidance is currently developed by FOCUS. A first draft of a report "FOCUS ground water scenarios in the EU registration process" has been prepared by Boesten et al (December 1999).

Surface water

For data requirements refer to Annexes II/III, sections 7 and 9 of Council Directive 91/414/EEC. With regard to Uniform Principles refer to Annex VI, part C, 2.5.1.3 of Council Directive 91/414/EEC. Further guidance is currently developed by FOCUS.

Air

For data requirements refer to Annexes II/III, sections 7 and 9 of Council Directive 91/414/EEC. With regard to Uniform Principles refer to Annex VI, part C, 2.5.1.4 of Council Directive 91/414/EEC. Further guidance is

currently being developed by EPPO (Decision Making Scheme for the Environmental Risk Assessment of Plant Protection Products – Air).

Impact on non-target species

For data requirements refer to Annexes II/III, sections 8 and 10 of Council Directive 91/414/EEC. With regard to Uniform Principles refer to Annex VI, part C, 2.5.2 of Council Directive 91/414/EEC. Further guidance is currently being developed (Draft Guidance Paper on Aquatic Ecotoxicology (Doc. 8075/VI/97) and Draft Guidance Paper on Terrestrial Ecotoxicology (Doc. 2021/VI/98).

6 Non-extractable residues

According to Annex VI of Council Directive 91/414/EEC (part C, Decision-making, point 2.5.1.1) no authorization shall be granted if non-extractable residues during soil laboratory studies are formed in amounts exceeding 70 % of the initial dose after 100 days with a mineralization rate of less than 5 % in 100 days.

Definition

Non-extractable residues (sometimes referred to as 'bound' or 'non-extracted' residues) in plants and soil are defined as chemical species originating from pesticides used according to good agricultural practice that cannot be extracted by methods which do not significantly change the chemical nature of these residues. These non-extractable residues are not considered to include fragments through metabolic pathways leading to natural products.

(Annex VI, part B, Evaluation, point 2.5.1.1 and Annex II, point 7.1.1.1).

Guidance for conducting studies

Appropriate studies to determine the extent of the formation of non-extractable residues and the consequent mineralization rate are those that are designed to determine the route of degradation in soil (Annex II, point 7.1.1.1).

The "unless"-statement for non-extractable residues

If non-extractable residues are formed in amounts exceeding 70 % of the initial dose after 100 days with a mineralization rate of less than 5 % in 100 days no authorisation shall be granted unless it is scientifically demonstrated that under field conditions there is no accumulation in soil at such levels that unacceptable residues in succeeding crops occur and/or that unacceptable phytotoxic effects on succeeding crops occur and/or that there is an unacceptable impact on the environment.

A comprehensive evaluation of the significance of non-extractable residues is still very difficult. Numerous studies and reviews over the past 30 years have concluded that "bound" or "non-extractable" residues are of little concern (Kahn 1982, Anonymous 1986, Calderbank 1989). Other scientific literature indicates the possibility of a release or substitution of substances bound to the soil by e.g. changes in pH, cation exchange capacity (IUPAC 1984, Cheng and Koskinen 1986, Ebing 1987). Studies on retention mechanisms, bioavailability and on possible change in bioavailability due to changes of soil parameters (e.g. changes in land use) could provide results which enable an assessment of whether or not the non-extractable residues may have an unacceptable impact on the

environment. Regarding studies on bioavailability refer to Boesten (1993), Belfroid (1994), Van Straalen (1996), Kelsey et al. (1997) and Torslov and Larsen (1997). With regard to the effect of sorption and/or desorption on bioavailability and biodegradation refer to Beck et al. (1995), Hatinger and Alexander (1995), Knaebel et al. (1996), Pignatello and Xing (1996) and Bosma et al. (1997). The effect of earthworms on bioavailability and biodegradation is considered particularly by Ma et al. (1995) and Meharg (1996). The significance of identifying different forms of adsorbed and bound contaminants in soil with a brief review of techniques is described by Jones et al. (1996). For several topics e.g. methods to determine the nature of the bond, new extraction methods, stimulation of formation of bound residues, the role of dissolved organic matter on turnover and translocation, bioavailability and uptake of bound residues by plants and soil organisms refer to the report of the Deutsche Forschungsgemeinschaft (1998).

The small fractions likely to be released from bound residues are likely to have no additional significance from the regulatory view point (SCP 1999).

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8 Glossary

DT50 _{lab} /DT90 _{lab}	disappearance time of 50/90 $\%$ of the substance in laboratory studies
$DT50_{field} / DT90_{field}$	disappearance time of 50/90 % of the substance in field studies
EEC	European Economic Community
EPPO	European and Mediterranean Plant Protection Organization
FOCUS	Forum for the Co-ordination of pesticide fate models and their Use
IUPAC	International Union of Pure and Applied Chemistry
k	first-order degradation rate constant
K _{oc}	linear partition coefficient related to organic carbon content
Q ₁₀	change in rate for a 10 °C change in temperature
r^2	determination coefficient
TER	toxicity exposure ratio



