Agriculture Canada

October 30, 1987 T-1-255

FOOD PRODUCTION AND INSPECTION BRANCH	DIRECTION GENERALE, PRODUCTION ET INSPECTION DES ALIMENTS	SECTION PESTICIDES

TRADE MEMORANDUM

Re: Guidelines for Determining Environmental Chemistry and Fate of Pesticides

Section 9 of the Pest Control Products Regulations requires that applicants for registration of pest control products shall provide data to support the registration of their products.

The purpose of this memorandum is to provide a guideline for the development of data to determine the environmental chemistry of pesticides and to predict their fate in the environment.

These guidelines have been prepared cooperatively by Agriculture Canada, Environment Canada and the Department of Fisheries and Oceans. Comments received on a proposed guideline, distributed earlier as a Memorandum to Registrants have been considered and incorporated where appropriate. Accordingly, this trade memorandum replaces Memorandum to Registrants R-1-222, dated November 19, 1984. Should you require a copy of the complete guidelines, please contact Information Secretariat, Pesticides Directorate Agriculture Canada, K1A 0C6.

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R&T\T-1-255E.JEH

PUBLIÉ AUSSI EN FRANÇAIS

ENVIRONMENTAL CHEMISTRY AND FATE

GUIDELINES FOR REGISTRATION

OF

PESTICIDES IN CANADA

Prepared cooperatively by:

- Agriculture Canada
- Environment Canada
- Department of Fisheries and Oceans

July 15, 1987

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6.1 OVERVIEW

A. GENERAL INFORMATION

1. Purpose and Description of Guidelines

The purpose of these guidelines is to:

1) outline the registration requirements for studies demonstrating environmental chemistry and fate;

2) suggest methods and approaches for generating and reporting the required data;

3) facilitate the prediction of exposure of man and non-target organisms to the active ingredient(s) of a pest control product and its environmental transformation product(s).

To achieve this objective, it is necessary:

1) to identify active ingredients and transformation products, pathways and rate determining factors of the transformation process;

2) to assess environmental persistence and mobility of the active ingredient and its major transformation products;

3) to predict the persistence and transport patterns, with respect to specific uses.

REGISTRANTS ARE REQUIRED TO PROVIDE DATA THAT WILL PERMIT THESE REQUIREMENTS TO BE SATISFIED.

The guidelines have been prepared taking into consideration those of the Environmental Protection Agency (EPA)(1), the United Nations Food and Agriculture Organization (FAO)(2) and the Organization of Economic Cooperation and Development (OECD)(3). Additional requirements are included relating to Canadian geography and climate. **FIELD STUDIES OF DISSIPATION AND ACCUMULATION CARRIED OUT IN CANADA RELATIVE TO DESIRED CANADIAN USE PATTERNS ARE MANDATORY.**

Because of the many variables involved for each pesticide, e.g., manufacture, method of application, potential distribution, chemistry, and toxicity, set protocols are not developed, particularly in the case of field trials. These

guidelines are meant to be flexible, yet indicate clearly the types of tests necessary for evaluation.

Omission of any "required" study must be justified on scientific grounds. Where the decision is made (on a case-by-case basis) to not conduct a laboratory study with a major transformation product, the applicant for registration must provide the rationale for not conducting the study.

The protocols presented here are recommended as a standard approach to specific tests. Tests conducted according to other scientifically supportable protocols are also acceptable. References included provide a basis for the development of other protocols, and are referenced to specific tests. References apply to an entire area or section of study in most cases, and not to an individual item. They are for guidance and are not a restricted list.

Use patterns are divided into two general categories -- "terrestrial" and "aquatic" with a separate section on "special situations". Data requirements are based on intended use pattern (See Section 6.1D).

As part of the requirement to demonstrate the environmental safety of a pesticide, the basic charge on an applicant is to demonstrate the extent to which an active ingredient or a major transformation product may:

a) persist in the areas of application, or

b) migrate out of the areas (or the soil layers) of application, under the range of environmental conditions (moisture, temperature, pH, soil type, climate) likely to be encountered in the intended regions of use in Canada.

Applicants for registration should present their conclusions in the summary (Section 6.1) of the data submission. Conclusions should be supported by discussion based on the evidence in the data submission. In particular, the findings in field studies (e.g., studies in small plots or ponds) must be adequately explained and supported by the results of the laboratory studies. Provided that such explanation and support are available, the field studies will normally constitute the section of the environmental fate data submission that carries the greatest weight in the regulatory decisions. The laboratory studies of physicochemical properties and simulated environmental behaviour of a

pesticide serve to refine the design of the field studies as well as to permit the interpretation or explanation of the findings from the field. Deviations from expected results in the field studies should be explained.

Results of environmental chemistry and fate studies will be evaluated by Environment Canada and the Department of Fisheries and Oceans and the evaluations will be relayed to Agriculture for use in risk management decisions. By following these guidelines, data provided should be sufficient to allow evaluation. The need for additional data will depend on the quality of data provided, properties of the pesticide and intended use pattern. Evaluation will follow the philosophy stated by OECD in discussing this aspect: **"SCIENTIFIC JUDGMENT RATHER THAN RIGID CRITERIA SHOULD BE EXERCISED IN ACCEPTING OR REJECTING [CERTAIN] TEST RESULTS"**.

2. Experimental Design

The following qualifications of experimental design apply to the different studies discussed in the guidelines. In general, reference can be made to EPA Guidelines (1) for guidance in experimental design: the comments included here are intended to stress important points and to provide additional guidance in problem areas.

a) In all studies, the OECD Principles of Good Laboratory Practice should be applied (3).

b) Test substances will consist of the analytical grade of the active ingredient for radioisotopic studies, technical or analytical grade of the active ingredient for other laboratory studies, and the formulated product for most field studies.

c) Although radioisotopic analytical methods are preferred when the material balance of the parent compound and transformation products is desired, other appropriate analytical methods for detecting compounds are acceptable in many cases.

d) Untreated controls should be included in each experiment, where necessary; i.e., untreated controls are not always needed in studies involving radioisotopic material.

e) Unless otherwise noted, all pesticide treatments should be done at least in duplicate.

f) Either 20NC or 25NC should be chosen as a standard test temperature in laboratory studies (except where otherwise noted). The consistent use of a single standard temperature will allow calculations of parameters such as distribution ratios [see Section 6.2 A.1(c)].

g) Field studies of dissipation and accumulation must be conducted in Canada. Types of soil selected for field studies should be representative of intended major use sites in Canada. Similar soil types should be used in laboratory tests, although laboratory studies may be conducted outside Canada.

3. Reporting

a) Experimental: Reports should contain descriptions of ALL experimental design parameters. The following check list and the "Reports" section included with each study protocol contain information concerning details to be considered for inclusion in reports. Because the necessity for certain details will depend on the intended use and particular properties of a pesticide, these lists should be considered as guidelines rather than rigid or exhaustive criteria. Justification for omission of any listed detail should be provided.

b) Results: Control values as well as values for test material and major transformation products should be included in reports. Uncorrected data must be submitted. If corrections are made to presented data, (e.g., for extraction efficiency), these should be clearly stated. Where appropriate, numerical results should be presented in terms of rate constants, half-lives (for first-order reactions) or DT_{50}'s (time for 50% decrease) and graphical or tabular displays. Sample calculations should be included where appropriate.

c) The inclusion of data obtained using reference material(s) [i.e., well-studied pesticides, or substances such as those suggested in reference (3)] is strongly recommended because they can provide an indication of the reproducibility and sensitivity of the test system as well as aid interpretation of the

probable environmental behaviour of the test material. Data which has been developed for purposes other than Canadian registration and which provides additional, relevant evidence of environmental chemistry and fate (e.g., sensitized photodegradation in natural water samples, field studies carried out in other countries) should be submitted for evaluation. Certain physicochemical properties of pesticides required under Part 2, Product Chemistry may be necessary for evaluation of environmental chemistry and fate studies (e.g., UV-VIS absorption spectra, dissociation constants) and applicants should ensure such properties are submitted.

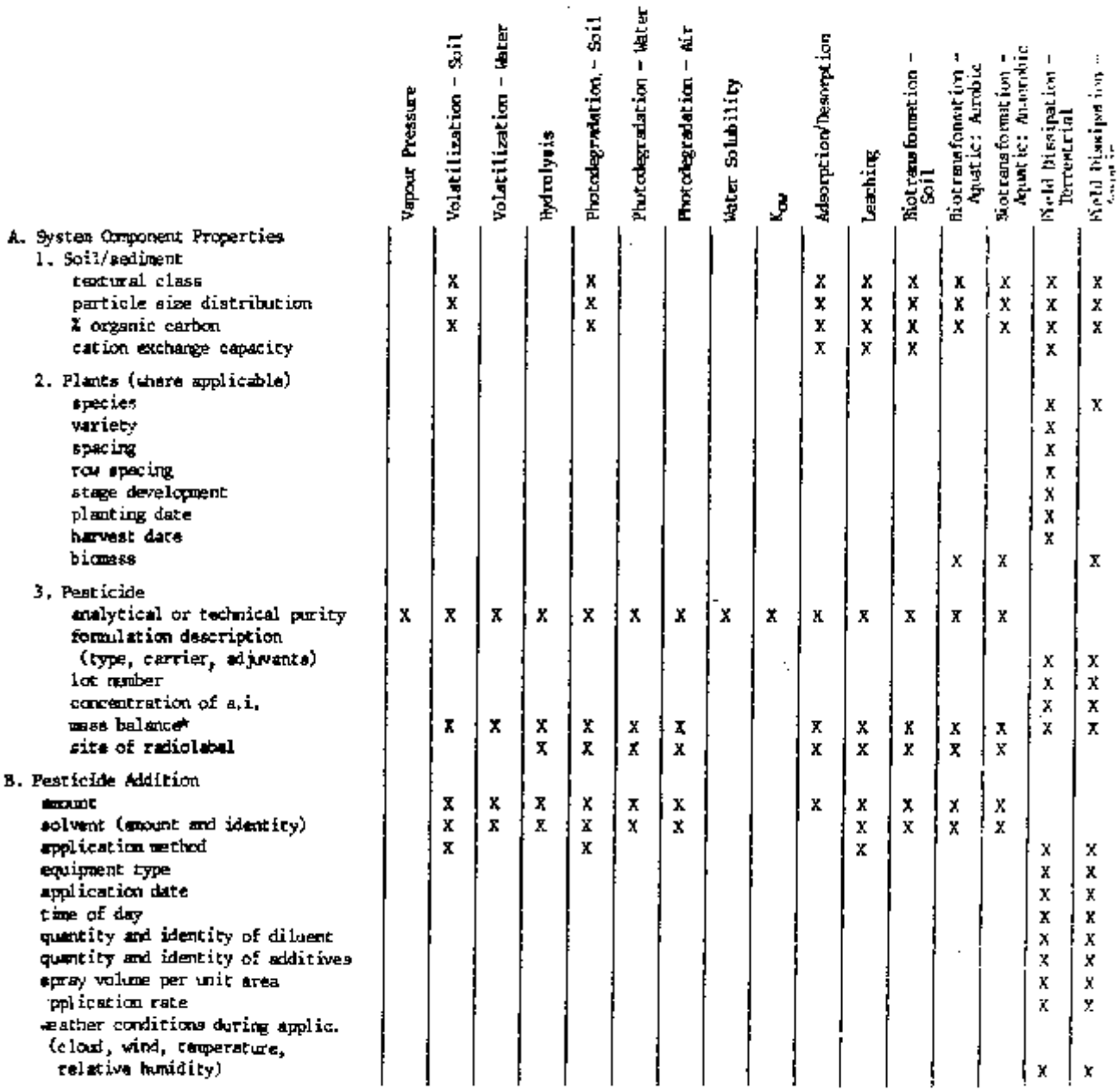
REFERENCES

1) EPA. 1982. Pesticide Assessment Guidelines. Subdivision D. Product Chemistry. EPA-540/9-82-018, and Subdivision N. Chemistry: Environmental Fate. EPA-540/9-82-021.

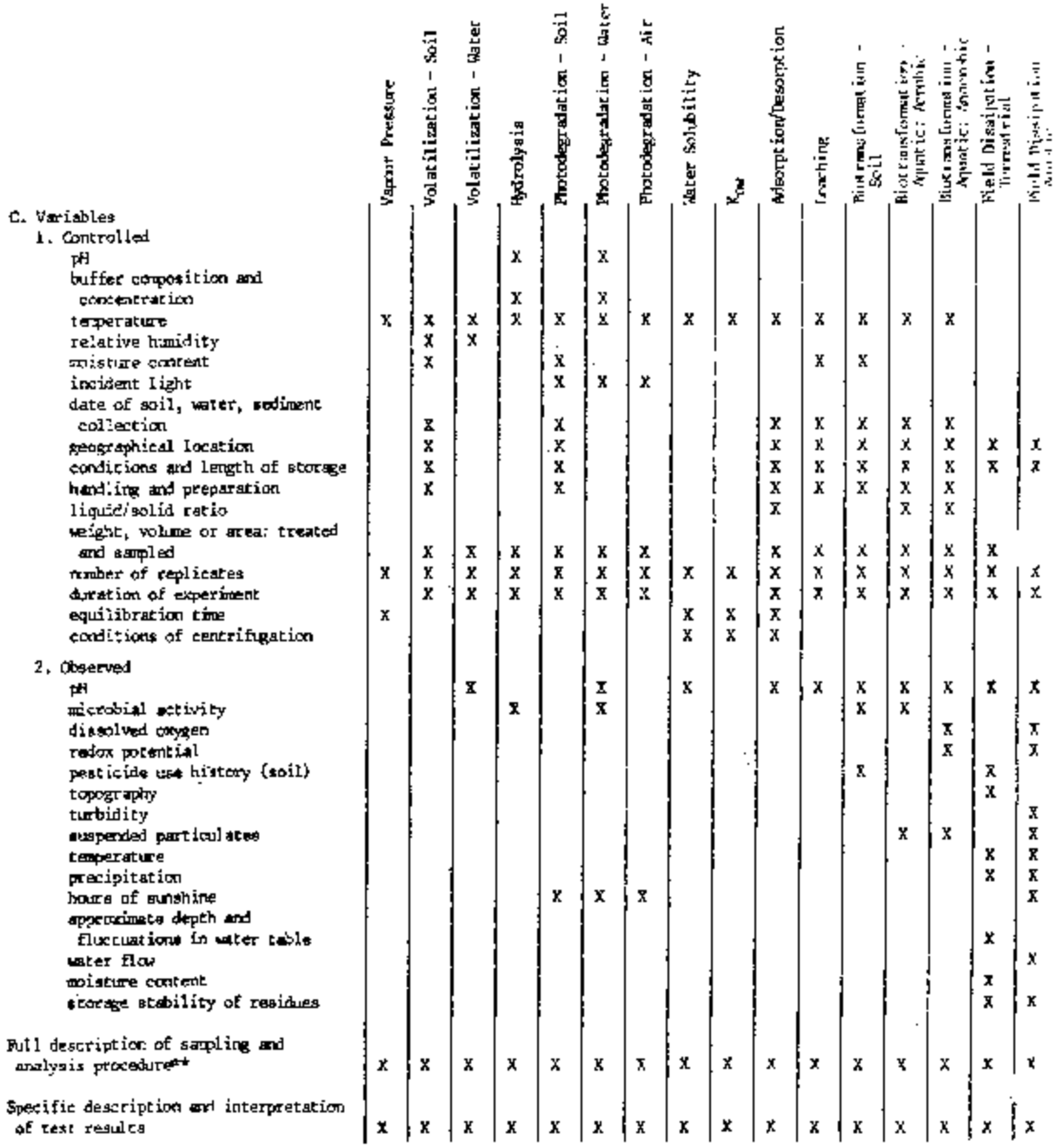
2) FAO. 1981. Second Expert Consultation on Environmental Criteria for Registration of Pesticides. Food and Agriculture Organization of the United Nations. Rome.

3) OECD Chemical Group. 1981. OECD Guidelines for Testing of Chemicals. Expert Group on Physical Chemistry.

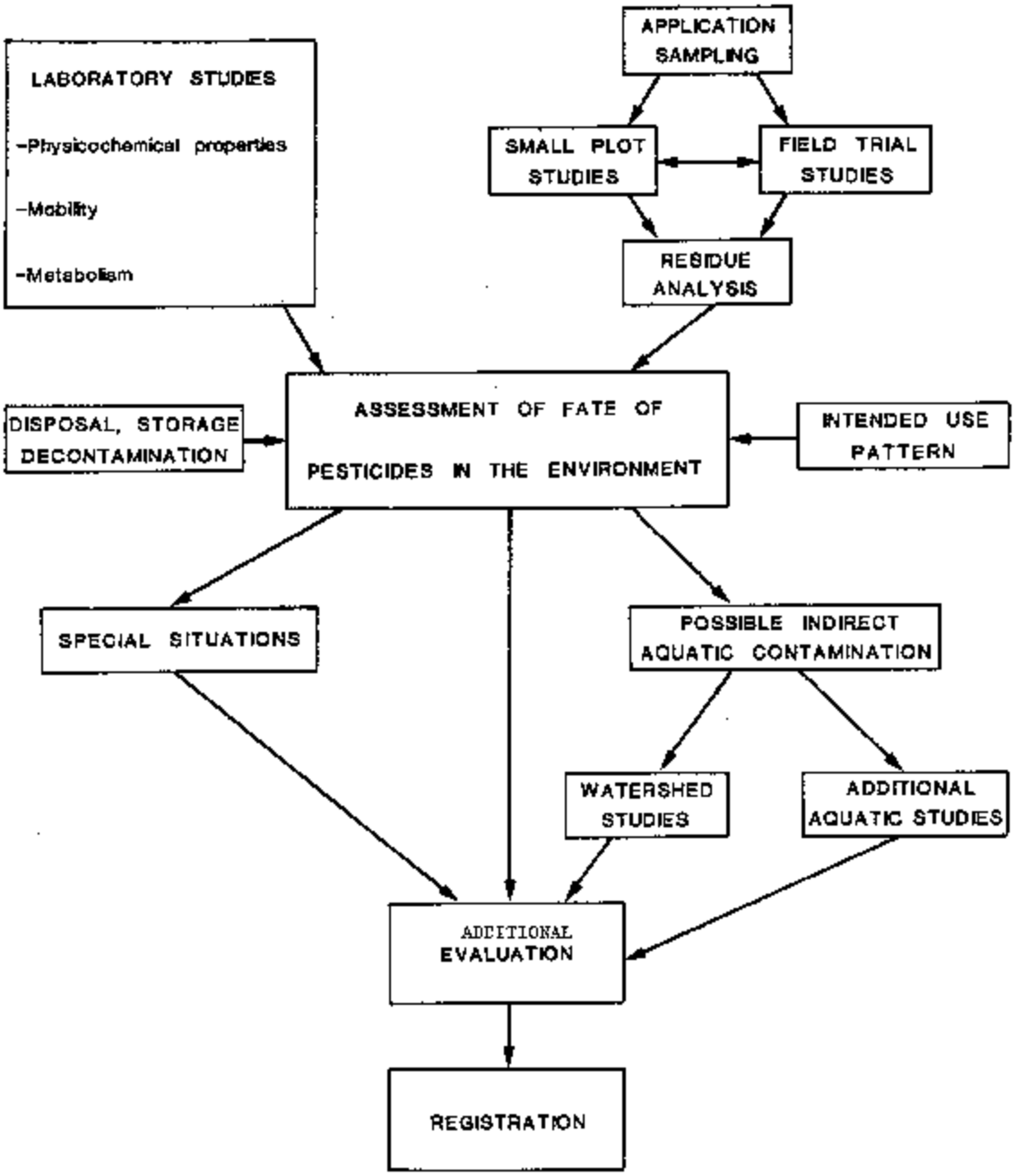
CHECKLIST FOR REPORTING DETAILS



* Some tests are not amenable to producing a mass balance. In such cases a rough estimate of the mass balance should be attempted.


C. Variables
1. Controlled
pH
buffer composition and concentration
temperature
relative humidity
moisture content
incident light
date of soil, water, sediment collection
geographical location
conditions and length of storage
handling and preparation
liquid/solid ratio
weight, volume or area treated and sampled
number of replicates
duration of experiment
equilibration time
conditions of centrifugation
2. Observed
pH
microbial activity
dissolved oxygen
redox potential
pesticide use history (soil)
topography
turbidity
suspended particulates
temperature
precipitation
hours of sunshine
approximate depth and fluctuations in water table
water flow
moisture content
storage stability of residues
Full description of sampling and analysis procedure**
Specific description and interpretation of test results
Vapor Pressure
Volatilization - Soil
Volatilization - Water
Hydrolysis
Photodegradation - Soil
Photodegradation - Water
Photodegradation - Air
Water Solubility
K_{ow}
Adsorption/Desorption
Leaching
Biotransformation - Soil
Biotransformation - Aquatic: Aerobic
Biotransformation - Aquatic: Anaerobic
Field Dissipation - Terrestrial
Field Dissipation - Aquatic

C. FLOW CHART OF ENVIRONMENTAL CHEMISTRY REQUIREMENTS FOR PESTICIDE REGISTRATION



D. DATA REQUIREMENTS FOR ENVIRONMENTAL FATE*

Special Study	Proposed Use Pattern: Terrestrial	Aquatic	Situations
LABORATORY STUDIES			
A. Physicochemical properties			
1. Vapour pressure and volatilization	+	+	+
2. Hydrolysis	+	+	+
3. Photodegradation - Soil	+		
- Water	+	+	
- Air			+
4. Solubility in water	+	+	+
5. Octanol/water partition co-efficient	+	+	+
B. Mobility			
1. Adsorption - desorption	+	+	
2. Leaching	+	+	
C. Biotransformation			
1. Soil] Aerobic	+		
] Anaerobic			+
2. Aquatic] Aerobic	+	+	+
] Anaerobic	+	+	+
FIELD STUDIES			
Dissipation and Accumulation			
1. Terrestrial Small plot/Large-scale**	+		+
2. Aquatic Small/Large-scale***	+	+	+

* Omission of any "required" study must be justified on scientific grounds.

** Either small plot or large-scale field trials must be carried out in Canada. Refer to page 39 for clarification.

*** Refer to page 49 for more details.

6.2 LABORATORY STUDIES

In all laboratory studies, all pesticide treatments should be done in at least duplicate (unless otherwise noted).

A. PHYSICOCHEMICAL PROPERTIES AND PROCESSES OF PARENT COMPOUND AND MAJOR TRANSFORMATION PRODUCT(S)

Accurate values for the laboratory studies on the physicochemical properties of a compound are essential for prediction of environmental behaviour. All major transformation products (products present at a level of greater than 10% of the initial concentration of pesticide at any time during the study) must be identified. The physicochemical properties and rates of transformation of major transformation products should also be determined, however, these matters may be decided on a case-by-case basis.

1. Vapour Pressure and Volatilization

The purpose of these studies is to determine the likelihood of pesticide dissipation by volatilization. The vapour pressure of a substance is the saturation pressure of the vapour above the solid or liquid phase of the substance at thermodynamic equilibrium. Vapour pressure is a key indicator of the potential of a compound to volatilize. Volatile pesticides may become widely distributed in the environment and are also of particular concern in confined areas.

a) There is no single procedure for measuring vapour pressure that is applicable to the entire range of potential vapour pressure values. The appropriate method for determining vapour pressure can be chosen from five recommended methods and will depend on an estimate of the range in which the vapour pressure of the pesticide in question lies (3,6):

   i) Gas saturation method (recommended range: <1 Pa)

   ii) Vapour pressure balance (recommended range: 10^{-3} to 1 Pa)

   iii) Static method (recommended range: 10 to 10^{5} Pa)

iv) Isoteniscope
(recommended range: 10^2 to 10^5 Pa)

v) Dynamic method
(recommended range: 10^3 to 10^5 Pa)

For vapour pressure determinations of most pesticides, the gas saturation method is recommended for the following reasons:

- the range of vapour pressures that can be accurately determined by this method would encompass the vapour pressures of most pesticides
- provided that the chemical component of interest is present as a discrete phase, the test material need not be pure as long as the detection method is specific for the test compound. Impurities in the test compound will produce erroneous results in the static, isoteniscope, and vapour pressure balance methods.

b) Vapour pressure should be determined at 20NC or 25NC (the chosen temperature should be used in all laboratory studies, except where otherwise noted). In general, extrapolation of vapour pressure beyond the temperature employed in a particular test should be avoided. In the dynamic method, however, vapour pressure is calculated by determining the boiling point of a sample as a function of reduced pressure. This determination will normally be done at temperatures considerably above ambient, and may require extrapolation to 20NC or 25NC. The vapour pressure balance method and other methods based on the effusion cell, also involve elevated temperatures and will require extrapolation from the experimental conditions. In any case, data must not be extrapolated if a change of state occurs between the temperature of interest and the temperature at which the vapour pressure is measured.

c) When low distribution ratios (1) or high Henry's Constant (5×10^{-7} atm m^3 mol^{-1}) indicate that a compound may be volatile, a laboratory study should be conducted to determine the potential contribution of volatilization to the dissipation of the pesticide in the field. Observations of volatilization made during biotransformation or photodegradation studies (conducted in flow through systems) may satisfy requirements for volatilization studies. Where

volatilization is going to be a major explanation of pesticide dissipation in the field, then specific laboratory data demonstrating volatilization (1, 2, 4, 5, 6) should be submitted. A highly volatile compound that also is moderately to highly toxic will require further testing in a confined area such as a greenhouse, particularly if significant inhalation exposure to workers would be liable to occur according to proposed use.

Reports

The following information should be included in reports:

i) Vapour Pressure

1. Analytical or technical purity of active ingredient.
2. Temperature of determination.
3. Number of replicates.
4. Equilibration time.
5. Full description of test methods and sampling and analysis procedure.
6. Specific description and interpretation of test results. Vapour pressure should be reported in pascals (1 mm Hg = 1 Torr = 133.32 Pa).

ii) Volatilization

1. Soil textural class, particle size distribution, organic carbon content and moisture content.
2. Analytical or technical purity of active ingredient.
3. Mass (materials) balance at end of study.
4. Amount of pesticide added and amount and identity of solvent.
5. Soil application technique.
6. Temperature of determination.
7. Relative humidity.
8. Soil collection date, geographical location of collection site, length and conditions of soil storage and soil handling and preparation.
9. Weight, volume or area, treated and sampled.
10. Number of replicates.
11. Duration of experiment.
12. Observed pH of test solution.
13. Full description of test methods and sampling and analysis procedures.
14. Specific description and interpretation of test results.

REFERENCES

1) Burkhard, N. and J.A. Guth. 1981. Rate of volatilization of pesticides from soil surfaces; Comparison of calculated results with those determined in a laboratory model system. Pestic. Sci. 12: 37-44.

2) Nash, R.G. 1983. Comparative volatilization and dissipation rates of several pesticides from soil. J. Agric. Food Chem. 31: 210-217.

3) OECD Chemical Group. 1981. Vapour pressure curve. OECD Test Guideline No. 104. Expert Group on Physical Chemistry. May 1981.

4) Sanders, P.F. and J.N. Seiber. 1983. A chamber for measuring volatilization of pesticides from model soil and water disposal systems. Chemosphere 12: 999-1012.

5) Smith, J.H., D. MacKay, and C.W.K. Ng. 1983. Volatilization of pesticides from water. Res. Rev. 85: 73-88.

6) Spencer, W.F., and M.M. Cliath. 1983. Measurement of pesticide vapour pressures. Res. Rev. 85: 57-71.

7) Spencer, W.F., W.J. Farmer, and W.A. Jury. 1982. Review: Behaviour of organic chemicals at soil, air, water interfaces as related to predicting the transport and volatilization of organic pollutants. Environmental Toxicology and Chemistry 1: 17-26.

Other details on experimental procedure and pertinent references can be found in:

EPA. 1982. Pesticide Assessment Guidelines.
Subdivision D. Product Chemistry.
EPA - 540/9-82-018
Subdivision N. Chemistry: Environmental Fate
EPA - 540/9-82-021

2. Hydrolysis

The purpose of this study is to determine the rate of pesticide degradation by hydrolytic reactions and to determine the identity of major transformation products; the rates of hydrolysis of major transformation products should also be determined, however, this may be decided on a case-by-case basis. In principle, a dilute solution of chemical is maintained at a constant pH and temperature while changes in concentration of parent chemical and major transformation products are followed over time. It should be noted that, besides hydrolysis, other possible reaction mechanisms of a compound in water, such as elimination and isomerization, are covered in this study. The information gained from hydrolysis studies is useful in estimating persistence of pesticide residues in the environment.

a) Identification of major hydrolysis products must be done using radiolabeled pesticide. Materials present at concentrations greater than 10% of the initial pesticide concentration at any time during the study should be identified. The rates of hydrolysis of the parent compound and major hydrolysis products can be determined using any suitable analytical technique.

b) Rates of hydrolysis should be determined at one pesticide concentration in distilled, buffered water, using sealed containers. Hydrolysis should be examined at three pH values (acidic, neutral, basic). Test temperature should be 20°C or 25°C (depending on which temperature is chosen as standard for laboratory studies). Temperature extrapolations are acceptable. Light should be excluded from test systems to prevent phototransformations.

c) The experiment must be conducted under sterile conditions. Samples should be taken and examined for contaminants at appropriate intervals to ensure that sterility is maintained for the duration of the test period.

d) Solutions should be largely aqueous, keeping the cosolvent, if used, under 1% (V/V) of the final concentration.

e) The buffer concentration should be relatively low (around 0.01 N) and the results of the hydrolysis study

should be evaluated with the possible effects of buffer catalysis being considered.

f) Samples for analysis should be taken at a minimum of 5 time intervals starting at zero time to provide at least 6 measurements to determine the rate constants. At least one observation should be made after half disappearance of the parent compound or, if the parent compound is slow to hydrolyze, the final observation can be taken at 30 days from initiation of the test.

Reports

The following information should be included in reports:

1. Analytical purity of active ingredient.
2. Mass (materials) balance at end of study.
3. Site of radiolabel.
4. Amount of pesticide added, and amount and identity of solvent.
5. Buffer composition and concentration, pH and possible buffer catalysis effects.
6. Temperature of determination.
7. Volumes treated and sampled.
8. Number of replicates.
9. Duration of experiment.
10. Results of sterility checks.
11. Full description of sampling and analysis procedures.
12. Specific description and interpretation of test results.

REFERENCES

1) Chapman, R.A., and C.M. Cole. 1982. Observations on the influence of water and soil pH on the persistence of insecticides. J. Environ. Sci. Health, B17: 487-504.

2) Faust, S.D., and H.M. Gomaa. 1972. Chemical hydrolysis of some organic phosphorus and carbamate pesticides in aquatic environments. Environmental Letters 3: 171-201.

Other details on experimental procedure and pertinent references can be found in:

EPA. 1982. Pesticide Assessment Guidelines.
Subdivision N. Chemistry: Environmental Fate
EPA - 540/9-82-021

3. Photodegradation

Photodegradation studies are required to permit the assessment of the significance of this mode of dissipation of a pesticide or its major transformation products.

Identification of major photodegradation products must be done with radiolabeled pesticide. The rates of degradation of the pesticide can be determined using any suitable analytical technique; the rates of degradation of major transformation products should also be determined on a case-by-case basis.

Studies using natural sunlight are acceptable, provided that the parameters are well defined and documented. If artificial light sources are employed in these studies, all light of wavelengths less than those in sunlight reaching the earth's surface (i.e., less than 290 nm) should be excluded by the use of selective filters or light sources which do not emit these wavelengths (1).

i) Soil

a) Soil photodegradation studies are required if the mode of pesticide application indicates that application will result in deposition and residence at the soil surface. Thus, studies would not be required for soil incorporated compounds.

b) One or more concentrations should be tested on one representative soil used in the soil biotransformation study. One test concentration is sufficient, but some studies routinely involve a range of concentrations.

c) Experimental controls shall consist of soil samples treated with the test pesticide at the same concentration(s) as the test samples. Controls should be held in darkness, but otherwise under the same conditions as treated samples (e.g., same apparatus, same temperature).

d) Samples for analysis should be taken at a minimum of 4 time intervals starting at zero time to provide at least 5 measurements over a period of up to 30 days. One observation after one half of the test substance has degraded is sufficient. Therefore, the full 30-day test period may not be necessary. A photoproduct

present at any time during the study at 10% or more of the initial concentration must be identified.

e) The use of flow through test systems (2), is recommended as a means of quantifying the formation of volatile photoproducts.

f) Soil used in photodegradation studies need not be sterilized. If soil is sterilized, do not do so by steam or heat (which could change the chemical nature of the soil components).

Reports

The following information should be included in reports:

1. Soil textural class, particle size distribution, % organic carbon and soil moisture content.
2. Analytical or technical purity of active ingredient.
3. Mass (materials) balance at end of study.
4. Site of radiolabel.
5. Amount of pesticide added, pesticide application method, and amount and identity of solvent.
6. Temperature of determination.
7. Details of incident light: duration, wavelength distribution, intensity and identity of source when artificial light is used, or, when sunlight is used, hours and intensity of sunshine, details of test location and study date.
8. Soil collection date, geographical location of collection site, length and conditions of soil storage and soil handling and preparation.
9. Weight or area; treated and sampled.
10. Number of replicates.
11. Duration of experiment.
12. Full description of test methods, sampling and analysis procedures.
13. Specific description and interpretation of test results.

REFERENCES

1) Choudhry, G.G. and G.R. Barrie Webster. 1985. Protocol guidelines for investigations of photochemical fate of pesticides in water, air, and soils. Res. Rev. 96: 79-136 (for description of artificial light sources available).

2) Klehr, M., J. Iwan, and J. Riemann. 1983. An experimental approach to the photolysis of pesticides adsorbed on soil: Thidiazuron. Pestic. Sci. 14: 359-366.

3) Nilles, G.P., and M.J. Zabik. 1975. Photochemistry of bioactive compounds. Multiphase photodegradation and mass spectral analysis of basagran. J. Agric. Food Chem. 23: 410-415.

ii) Water

a) Photodegradation studies in water are required for all compounds to determine the products and rates of degradation caused by this activity. The ultraviolet-visible (UV-VIS) absorption spectrum of a pesticide (1,5) can provide an indication of the wavelengths of light (in the range of 290 to 800 nm) at which photodegradation may occur. The UV-VIS absorption spectrum should be submitted with photodegradation studies in water (this is also required under Part 2, Product Chemistry). Since solvents may alter light absorption by a molecule, the UV-VIS absorption spectra will be most useful when measured in distilled water, or using a minimum of photochemically inert cosolvent.

b) One or more concentrations should be tested using sterile, distilled water buffered to a pH which minimizes hydrolysis. The study should be conducted at either 20NC or 25NC, depending on which of these temperatures was chosen as standard for other laboratory studies. With compounds of low solubility, a photochemically inert organic cosolvent (eg. spectrograde acetonitrile) may be used at a concentration not greater than 1% (V/V), if necessary.

c) Experimental controls should consist of sterile, distilled water, buffered to the same pH, and treated at the same concentration of pesticide, as the test samples. Controls should be held in darkness, but otherwise under the same conditions as treated samples, (e.g., same apparatus, same temperature).

d) Samples of control and test solutions should be examined for contaminants at appropriate intervals to ensure that sterility is maintained for the duration of the test period.

e) Samples for analysis should be taken at a minimum of 4 time intervals starting at zero time to provide at least 5 measurements over a period of up to 30 days. One observation after one-half of the test substance has degraded is sufficient. Therefore, the full 30-day test period may not be required. A photoproduct present at anytime during the study at 10% or more of the initial concentration must be identified.

f) The use of flow through test systems is recommended as a means of quantifying the formation of volatile photoproducts.

Reports

The following information should be included in reports:

1. Analytical or technical purity of active ingredient.
2. Mass (materials) balance at end of study.
3. Site of radiolabel.
4. Amount of pesticide added, and amount and identity of solvent.
5. Buffer composition and concentration.
6. Temperature of determination.
7. Details of incident light: duration, wavelength distribution, intensity and identity of source when artificial light is used, or, when sunlight is used, hours and intensity of sunshine, details of test location and study dates.
8. Volumes treated and sampled.
9. Number of replicates.
10. Duration of experiment.
11. Observed pH of test solution.
12. Results of sterility checks.
13. Full description of test methods, sampling and analysis procedures.
14. Specific description and interpretation of test results.

REFERENCES

1) EPA. 1979. Toxic substances control: Discussion of premanufacture testing policy and technical issues; request for comment. 44 FR 16267-8.

2) Lemaire, J., I. Campbell, H. Hulpke, J.A. Guth, W. Merz, J. Philp, and C. von Waldow. 1982. An assessment of test methods for photodegradation of chemicals in the environment. Chemosphere 11: 119-164.

3) Miller, G.C., and R.G. Zepp. 1983. Extrapolating photolysis rates from the laboratory to the environment. Res. Rev. 85: 89-110.

4) Nakagawa, M., and D.G. Crosby. 1974. Photodecomposition of nitrofen. J. Agri. Food Chem. 22: 849-853.

5) OECD Chemical Group. 1981. UV-VIS absorption spectra. OECD test guidelines no. 101. Expert group on physical chemistry. May 1981.

6) Wong, A.S., and D.G. Crosby. 1981. Photodecomposition of pentachlorophenol in water. J. Agric. Food Chem. 29: 125-130.

Other details on experimental procedure and pertinent references can be found in:

EPA. 1982. Pesticide Assessment Guidelines. Subdivision N. Chemistry: Environmental Fate EPA - 540/9-82-021

iii) Air

a) A photodegradation study in the vapour phase to determine products and rates of degradation of highly volatile pesticides and major transformation products is required on a case-by-case basis.

b) One concentration at 30± 2NC shall be tested.

c) Experimental controls should consist of air samples treated with the test pesticide at the same concentration as the test samples. Controls should be held in darkness, but otherwise under the same conditions as treated samples (e.g., same apparatus, same temperature).

d) Samples for analysis should be taken at a minimum of 4 time intervals starting at zero time to provide at least 5 measurements over a period of up to 30 days. One observation after one-half of the test substance has degraded is sufficient. Therefore, the full 30-day test period may not be required. A photoproduct present at any time during the study at 10% or more of the initial concentration must be identified.

Reports

The following information should be included in reports:

1. Analytical or technical purity of active ingredient.
2. Mass (materials) balance at end of study.
3. Site of radiolabel.
4. Amount of pesticide added, and amount and identity of solvent.
5. Temperature of determination.
6. Details of incident light: duration, wavelength distribution, intensity and identity of source when artificial light is used, or, when sunlight is used, hours and intensity of sunshine, details of test location and study dates.
7. Volume treated and sampled.
8. Number of replicates.
9. Duration of experiment.
10. Full description of test method, sampling and method analysis procedure.
11. Specific description and interpretation of test results.

REFERENCES

1) Crosby, D.G., and K.W. Moilanen. 1974. Vapour-phase photodecomposition of aldrin and dieldrin. Arch. Environ. Contam. Toxicol. 2: 62-74.

2) Woodrow, J.E., D.G. Crosby, T. Mast, K.W. Moilanen, and J.N. Seiber. 1978. Rates of transformation of trifluralin and parathion vapours in air. J. Agric. Food Chem. 26: 1312-1316.

3) Woodrow, J.E., D.G. Crosby, and J.N. Seiber. 1983. Vapour-phase photochemistry of pesticides. Res. Rev. 85: 111-125.

4. Solubility in Water

The solubility of a pesticide in water is its equilibrium concentration in a saturated solution at a stated temperature. This property is useful in predicting pesticide partitioning, mobility and fate in the environment.

a) Compounds should be as pure as possible since certain impurities (ie. solvents in technical formulations) can significantly affect the solubility of the pesticide.

b) The solubility of the pesticide should be determined at one specified temperature of either 20°C or 25°C (depending on the temperature chosen as standard for other laboratory studies). Solubility values should not be extrapolated from other temperatures (3). Temperature must be controlled for the duration of the test.

c) The column elution method for determining solubility may be used for compounds with solubility below 10^{-2} g L^{-1} (8). The flask method should be used for compounds with solubilities above 10^{-2} g L^{-1}(8). The following comments pertain to the flask method for determining solubility:

   i) Centrifugation is commonly used to separate excess solute from the saturated solution. Adequate centrifugal force (e.g., 3 hrs at 17,000-20,000 RPM, 35,000 x g) is required to achieve this. Care must be taken in withdrawing samples from the centrifuge tubes as some pesticidal material may float on the surface of the supernatant. Teflon or other plastic materials should not be in contact with the pesticidal compounds at any stage of the determination. Stainless steel and Pyrex glass are preferred.

   ii) Equilibration time. Samples should be taken periodically until two successive samples give the same value (within experimental error). The time interval between sampling will be compound dependent and will also depend on the amount of solute excess in the system, the temperature and the mode of equilibration

(eg. the intensity of shaking or tumbling).

iii) Sample preparation. When pesticide solubility is above approximately 0.1 ug mL^{-1}, sample preparation is straight forward - dispense the solute into a glass container (foil-lined screw cap) and add an appropriate amount of distilled water. At lower solubilities only very small amounts of test substance are involved and this sometimes retards the rate at which solubility equilibrium is reached. In these special situations, the solute is dissolved in a small volume of organic solvent (eg. acetone, hexane) and plated onto the glass surfaces of the equilibration vessel. The excess solvent is slowly evaporated off while rotating the glass vessel. Care must be taken to remove all the organic solvent before adding the distilled water.

d) Pesticide solubility in water may be a function of pH if the compound ionizes in aqueous solution. In such cases, it may be necessary to determine solubility at more than one pH.

Reports

The following information should be included in reports:

1. Analytical purity of active ingredient.
2. Temperature of determination.
3. Number of replicates.
4. Equilibration times and conditions of centrifugation.
5. Observed pH of test solution.
6. Full description of test method sampling and analysis procedure.
7. Specific description and interpretation of test results. Water solubility should be expressed in: ug mL^{-1} or ug L^{-1} and in moles L^{-1}.

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5. Octanol/Water Partitioning Coefficient (K_{ow})

The octanol/water partitioning coefficient is the ratio of the concentration of a pesticide in n-octanol to that in water at equilibrium in dilute solution. The K_{ow} of a pesticide indicates the likelihood of pesticide transfer from environmental media to organisms and the potential to

bioaccumulate. Radiolabelled compounds can be used for K_{ow} determinations. It is preferable, however, to use non-radiolabelled compounds because of problems with impurities and transformation products.

a) Reagent purity. Both solute and solvents should be of the highest obtainable purity. Deionized water should be distilled over $KMnO_4$ to remove organic impurities before use. Pesticide grade n-octanol is acceptable; however, reagent grade n-octanol must be further purified, preferably by extracting once with 0.1 N NaOH, twice with distilled water and then distilling the extracted octanol.

b) If radiolabeled pesticide is used for K_{ow} studies, some additional means of verifying the identity of the tagged material in solution must be presented (e.g., GLC or HPLC analysis). The presence of the isotope "tag" does not constitute verification of the presence of the pesticide, as it could just as well be on a remaining hydrolytic fragment. Stability data from the hydrolysis study may be submitted to satisfy this requirement.

c) Sample preparation. The purified reagents (n-octanol, water) should be mutually saturated. This is most easily accomplished in a 2 L separatory funnel which can then serve as a storage reservoir for both solvents. Equilibrate samples in 60 mL separatory funnels so that the water phases can be withdrawn after each equilibration. Relative volumes of the two solvents are theoretically not important, but if partitioning into the water phase is favoured (low K_{ow}), the volume ratio becomes important in practice, especially if using GLC analysis(1). If the K_{ow} is expected to be low, then the volume of octanol relative to that of water should be increased.

d) Phase partitioning. Samples in the 60 ml separatory funnels should be shaken carefully to avoid the formation of emulsions, and then allowed to stand for several hours before the water phase is withdrawn. Three extractions in triplicate would normally be sufficient to produce a reliable K_{ow} value. However, if substantial differences are obtained between the K_{ow} values calculated from the second and third extractions, which can be caused by impurities, the

extraction process must be continued until sequential K_{ow} values are the same (within experimental error).

e) Centrifugation. Centrifugation times for the water phase of AT LEAST one-half hour at sufficient g-forces (e.g., 34,000 x g) are recommended to assure complete separation of the two phases. Stainless steel centrifuge tubes should be used. Extreme care should be taken to avoid small droplets of octanol around the air-water interface when pipeting the water sample for analysis.

f) For accurate K_{ow} measurements, the solute concentration should not be allowed to approach its solubility limit in either phase. Initial solute concentrations of 2000 ug mL^{-1} or less in the octanol phase have proven satisfactory.

g) By convention $K_{ow} = \frac{[\text{solute}]\ \text{octanol}}{[\text{solute}]\ \text{water}}$

and, therefore, the K_{ow} is independent of the relative volumes of the two phases. It is important to analyse the solute concentration in both phases rather than calculate the concentration in one phase by difference from an initial concentration. Since it is only practical to analyse the solute concentration in the octanol phase after the last extraction, K_{ow} calculations should be made using this final solute concentration in octanol and then back calculating for the earlier extractions.

h) Although the K_{ow} determination is not greatly affected by temperature, it is recommended that temperature be controlled to $\pm$ 1NC. Test temperature should be either 20NC or 25NC depending on which of these temperatures was chosen as the standard for laboratory studies.

i) A reverse-phase HPLC method has been developed for estimating K_{ow} values from retention times (9,12). While this method has the advantage of being rapid and repeatable, its accuracy is highly dependent on the error associated with the direct determination of the reference K_{ow} values. Because of this limitation, the reverse-phase HPLC method is not recommended for obtaining primary K_{ow} values. However, when analytical problems are encountered (eg. with extraction

techniques), this method could be used for estimating the K_{ow}.

j) Direct generator column methods for producing primary K_{ow} values have been described (13,14), but have not, as yet, become widely used.

k) A discussion of special procedures for determining the K_{ow} of pesticides that ionize or exhibit other association/dissociation behaviour in solution can be found in reference (5).

Reports

The following information should be included in reports:

1. Analytical purity of active ingredient.
2. Temperature of determination.
3. Number of replicates.
4. Conditions of centrifugation, and equilibration time.
5. Full description of test methods, sampling and analysis procedures.
6. Specific description and interpretation of test results.

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B. MOBILITY

Mobility studies provide information concerning the ability of terrestrial-use pesticides and their major transformation products to move through soils and their potential to contaminate aquatic environments by leaching to groundwater, movement in surface runoff or with eroding soil. This

information is necessary in designing terrestrial field studies (i.e., selecting depth of soil core samples) and in determining the need for aquatic field studies with terrestrial-use pesticides. The adsorption/desorption properties of aquatic-use pesticides and their major transformation products will be considered in the design of aquatic field studies.

Mobility of pesticides intended for either terrestrial or aquatic use should be assessed by adsorption/desorption tests. The protocol for adsorption/desorption measurements presented in B.1 stresses major points that should not be overlooked in the generation of this type of data.

In addition, the leaching potential of pesticides (and their major transformation products) intended for terrestrial use should be assessed by **ONE** of the leaching test methods outlined in B.2. For pesticides intended for domestic outdoor, greenhouse, or aquatic use, testing of mobility by methods in B.2 is not necessary.

1. Adsorption/Desorption Measurements

a) Adsorbents

i) Type. Adsorption/desorption data should be obtained using <u>at least</u> <u>two</u> and preferably three or more soils, REPRESENTATIVE of the major areas of proposed use in Canada. Non-Canadian soils must be shown to be representative of the soils of the major areas of proposed use in Canada with respect to particle size distribution (% sand, % silt, % clay), % organic carbon, pH, cation exchange capacity and clay fraction mineralogy. It is suggested that the soils chosen for these studies be the same as those used in the biotransformation studies. Three typical soil types could include a sandy soil, a loam or sandy loam, and a clay or clay loam. A muck (organic) soil should be included if intended use warrants this. If a pesticide is intended for aquatic use, data for one representative aquatic sediment should be obtained.

ii) Preparation. Soil should be maintained at natural moisture levels. To produce a reasonably homogeneous substrate, soils are sieved through a screen of $\leq$ 4 mm mesh. Drying of soil should be avoided if possible, but may be necessary for sieving. If this is the case, soils may be partially air-dried to a workable moisture content.

b) Generation of isotherm data (batch method).

i) At least four concentrations of pesticide solution (made up by adding analytically pure pesticide to a solution of 0.01 N $CaCl_2$) should be added to the various soil types and shaken or tumbled in darkness for a specific period (12-18 hour period is usually sufficient) to reach equilibrium. Each treatment should be carried out in triplicate. After equilibration the soil-pesticide slurry should be centrifuged in stainless steel tubes at sufficient g-forces and for sufficient times, to allow separation (longer centrifugation times may be necessary if very fine, well-dispersed clays are present).

ii) Desorption studies should be conducted in sealable centrifuge tubes (e.g., Corex glass with foil-lined screw caps) so that both equilibration and centrifugation can be conducted in the same vessel. After the above-described adsorption step, a specific volume of supernatant is removed for analysis, and is replaced by the same volume of 0.01 N $CaCl_2$ solution to initiate the first desorption step. The sample is shaken and centrifuged to complete the first cycle. At least two further desorption cycles should be completed to generate the desorption isotherm. Only one initial starting concentration should be necessary for the desorption study.

It is not acceptable to present "single-point desorption" data, where single desorption cycles are performed on several different adsorption systems (at different equilibrium concentrations) and the resulting points joined to form an "isotherm". Slopes of "isotherms" formed with this approach are greater than the respective adsorption isotherms, which in reality is an impossibility. A proper desorption isotherm can have ONLY ONE adsorption point as its point of initiation.

Hysteresis, or irreversibility effects in desorption studies are, in part, a result of the methods used. The "consecutive desorption" method outlined above often exhibits more hysteresis than does the "dilution" method in which several identical adsorption systems are diluted to different extents. With the dilution method, it is important to use the same adsorbent weight and the same initial pesticide concentration in all samples.

c) Calculation of adsorption/desorption parameters. Most pesticide adsorption by soil slurries follows the empirical Freundlich Equation:

$$S = K\ C^N$$

where: S = amount adsorbed/unit weight adsorbent
C = equilibrium solution concentration of the adsorbate
K,N = constants

The "constant" K has long been employed as a measure of relative adsorption, but unfortunately, it really is NOT a constant, and actually has a complex set of units. If S is expressed in ug g^{-1} soil, and C in ug mL^{-1} solution the units of K are $ug^{1-N}\ g^{-1}\ mL^N$. Since pesticide adsorption varies over several orders of magnitude, K acquires many different units, WHICH ARE NOT COMPARABLE AND NOT ABLE TO BE DIRECTLY CONVERTED.

Consequently, there is only one universal way to report adsorption data, and the Freundlich Equation is rewritten as:

$$S = K_{MF}\ Z^N$$

where: S = moles g^{-1} adsorbent (oven-dry basis)
Z = mole fraction of pesticide in solution (for dilute solutions, moles pesticide/ moles water)
K_{MF}, N = regression constants

Most adsorption data falls in the mole fraction range 10^{-6} to 10^{-9}. Adsorption data usually yield a curved isotherm and, for statistical comparisons, must be linearized by taking logarithms.

i.e. $\log S = N \log Z + \log K_{MF}$
N = slope; $\log K_{MF}$ is the intercept at $\log Z = 0$ (pure pesticide in solution)

Instead of evaluating relative adsorption at log C = 0, where K was derived, a vertical transect is taken at an appropriate log Z value within the data range of the isotherm. For example, if the transect is taken at a mole fraction of 10^{-7}, then log Z = -7.0 and $\log S_{-7} = N(-7.0) + \log K_{MF}$, or, more generally: $\log S_Y = N(Y) + \log K_{MF}$ when log Z = Y.

From regression analysis both N and $\log K_{MF}$ will have numerical values, and therefore $\log S_Y$ acquires a certain value. The antilog, or S_Y value, is then used in the analogous fashion to the former "K", except that it now has units of mole g^{-1}, and can be used for comparisons between pesticides. S_Y values can be determined at any convenient point WITHIN the data range. Relative adsorption is dependent upon the point of comparison for non-linear adsorption isotherms with unequal values of N.

Actual isotherm data may still be reported in ug g^{-1} and ug mL^{-1} if desired, but must also be shown in mole g^{-1}, and mole fraction. All soil weights must be reported on an oven-dried (105°C) basis. Relative adsorption values must be reported using the S_Y designation. For further discussion see references 1, 2, 4.

d) Sample solid/liquid ratios from 1/1 to 1/200 are acceptable. For maximum precision it is important to adjust adsorbent concentration so that 20 to 80% of the solute will be adsorbed. For highly adsorbent soils use a lower solid/liquid ratio.

e) Be aware of pesticide decomposition or volatilization. Both of these factors can result in anomalously high adsorption values. Some knowledge of the hydrolytic stability of the pesticide in water should be obtained before generating adsorption/desorption data. This information is particularly important in desorption studies which may require several days to complete. USUALLY, pesticide stability is not less in a soil slurry than it is in distilled water over the short duration of an adsorption study.

f) If radiolabeled pesticide is used for adsorption studies, some additional means of verifying the identity of the tagged material in solution must be presented (e.g., GLC or HPLC analysis). The presence of the isotope "tag" does not constitute verification of the presence of the pesticide, as it could just as well be on a remaining hydrolytic fragment. Stability data from other soil and water tests may be submitted to satisfy this requirement.

g) There has been a trend in the literature to assume that the adsorption process is linear with respect to solution concentration, and thereby to simplify the Freundlich Equation to:

$$S = KC, \text{ where } N = 1$$

The linear equation may be a valid model at low, but environmentally realistic, pesticide concentrations.

h) Flow methods for determining adsorption/desorption parameters in soil-water systems have been described (6), however, these are more complicated than the batch methods.

Reports

The following information should be included in reports:

1. Soil textural class, particle size distribution, % organic carbon, cation exchange capacity and soil moisture content.
2. Analytical purity of active ingredient.
3. Mass (materials) balance at end of study.
4. Site of radiolabel.
5. Temperature of determination.
6. Soil collection date, geographical location of collection site, lengths and conditions of soil

storage and soil handling and preparation.
7. Liquid/solid ratio.
8. Number of replicates.
9. Duration of experiment.
10. Conditions of centrifugation.
11. Observed pH of initial soil-water slurries.
12. Full description of test methods, sampling and analysis procedures.
13. Specific description and interpretation of test results.

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2. Leaching

Pesticidal compounds of water solubility less than 0.5 ug mL^{-1} are relatively immobile in mineral soils (2) and those compounds of water solubility less than 50 ug mL^{-1} are relatively immobile in organic or muck soils (soils having organic carbon contents 17%). Consequently, it should not be necessary to determine their mobilities in the above stated situations UNLESS adsorption/desorption data suggest unusually low adsorption in comparison to water solubility.

The leaching potential of a pesticide and its major transformation products should be assessed by **ONE** of the following procedures. Each approach has strengths and weaknesses and the applicability of any one of these methods will depend on the particular situation.

As an alternative to individual investigations of leaching potential for each major transformation product, soil columns may be treated with "aged" soils or extracts of such soils and, similarly, soil thin-layer plates with extracts of such soils (4). Aged soils should be generated by incubating the parent chemical in soil for 30 days or one half life, whichever is shorter. The type of soil and conditions of incubation should be the same as used in soil biotransformation studies.

a) Soil thin-layer chromatography. The particular pesticides under examination are chromatographed on thin layer plates using the same representative, and characterized soils for adsorbents as were used in the adsorption/desorption studies. The method provides a rapid, quantitative means of assessing relative mobility using R_f values.

   It is recommended that coarse-textured soils be dry-sieved to <500 m in order to produce a uniform surface on the plate. The use of soils with larger particles may result in an underestimation of the mobility of the pesticide.

   Two possible major disadvantages:

   i) Soil TLC provides the quickest and best results when radiolabeled compounds are used since these can be easily visualized by autoradiograms. However, if non-radiolabeled material is used

this method is somewhat awkward since the soil layer on the TLC plate must be sectioned and each segment must be carefully scraped off, extracted and analysed by other means (e.g., GLC, HPLC).

ii) Soil texture compatibility. Soil TLC does not appear to be amenable to mobility studies with organic soils because of the water-repellent characteristics of these soils when air dry and because of possible problems with the binding of these soils to TLC plates. The use of organic soils for soil TLC studies should only be considered when the compound in question is very soluble (>50 ug mL^{-1}).

b) Soil column leaching. It is suggested that pesticides be eluted through a 30 cm soil column using a volume of distilled water equal to 20 inches (50.8 cm) times the cross sectional area of the column. The distributions of pesticide and major transformation products are determined by analysis of eluate fractions and 6 cm segments of the eluted soil column.

The soil column technique is practical only for medium to coarse-textured soils which retain a reasonable degree of permeability during the leaching process. The soil plug technique (3,7) circumvents many of the permeability problems experienced with longer soil columns when using finer-textured soils. Soils used in column or plug leaching studies should be the same as those used in adsorption/desorption studies.

c) Soil thick-layer chromatography combined with bioassay (1,5). This method may be useful for preliminary screening but should not be submitted as the sole leaching study for registration purposes.

SPECIAL NOTES:

1) It is important to do a mass (material) balance in the soil column and soil plug leaching studies. If not accounted for, volatility and degradation losses can produce significant errors in reporting the mobility of certain compounds.

2) All soil concentrations and soil weights must be based on oven-dry values (105°C).

3) Tests should be done at the highest proposed field application rate. Alternatively, a higher application rate may be used in order to cover a wide range of uses not envisaged at the time of original submission for registration.

Reports

The following information should be included in reports:

1. Soil textural class or bulk density, particle size distribution, % organic carbon, cation exchange capacity and soil moisture content.
2. Analytical purity of active ingredient.
3. Mass (materials) balance at end of study.
4. Site of radiolabel.
5. Amount of pesticide added, amount and identity of solvent application method.
6. Temperature of determination.
7. Soil collection date, geographical location of collection site, length and conditions of soil storage and soil handling and preparation.
8. Weight volume and area treated. Volume of eluate fractions.
9. Number of replicates.
10. Duration of experiment, equilibration time.
11. Observed soil pH.
12. Full description of test methods, sampling and analysis procedure (include description of methods used to generate aged soil).
13. Specific description and interpretation of test results.
14. For soil column (or plug) studies, the bulk density of the soil in the column (or plug).

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C. BIOTRANSFORMATION

The primary objective of biotransformation studies is to determine the nature and rates of formation of major pesticide transformation products in natural soil, sediment or water samples. These studies also permit determination of the microbial contribution to overall transformation processes. Controlled laboratory studies on transformation and persistence can be used in conjunction with physicochemical data and mobility studies to indicate probable fate in the environment, and aid in the design of field studies on dissipation and accumulation to substantiate such predictions.

1. Soil (Laboratory) - Degradation Pathways and Persistence.

a) Radioisotopic techniques must be used to identify the major transformation products. Suitable non-radioisotopic techniques may be used to determine rates of degradation (1). Major transformation products are

those that are present at more than 10% of the initial pesticide concentration.

b) It is important to specify whether laboratory persistence and degradation studies are done in "closed" or "open" systems. While conditions, eg. moisture, may be maintained better in a closed system, volatilization of the pesticide will be reduced. In an open system, while volatilization can occur, moisture is difficult to maintain - most pesticides will be more persistent in dry soil. Thus, any attempt to interpret laboratory persistence data in terms of what will happen under natural conditions should be done cautiously. Flow through systems or systems which permit the collection of volatile pesticide residues are recommended.

c) Rates of degradation of the parent compound and major transformation products should be determined under aerobic (both sterile and non-sterile) soil conditions at constant temperatures in darkness and at a constant moisture content. **RATE OF TRANSFORMATION MUST BE DETERMINED AT TWO SPECIFIED TEMPERATURES:** one in the lower temperature range of 3 - 8°C and one in a higher temperature range of 20° - 30°C.

d) It is recommended that soils used in biotransformation studies be freshly collected from the field and sieved wet through a screen of $\leq$ 4mm mesh. If the soil is too wet to sieve, partial air-drying to a workable moisture content may be necessary.

e) Non-sterile soils must be known to be microbially active and sterile soils must be known to be microbially inactive for the duration of biotransformation studies. This requirement could be satisfied by standard microbial plate counts (6).

f) One soil type, preferably a mineral soil, should be tested. If the use covers a crop that is grown in various soil types with wide geographic distribution, then more than one soil type should be included in biotransformation experiments. Further effects of varying environmental conditions on persistence can be determined by testing at two soil moistures (realistically high and low moisture contents). This information would aid in interpreting field results.

g) The pesticide is applied at one or two dosage rates. One concentration tested should be the maximum proposed field rate. If two rates are used, a 10-fold difference in concentration is the standard separation. Care must be taken in the method of introducing pesticide to soil, i.e., if pesticide is added with organic solvent, and the solvent is not completely removed, microbial activity may be affected. Soils should be treated when moist.

h) Suggested sampling times are at pretreatment, 0, 1, 3, 7, and 10 days, 2, 3, 4, 6, 8, and 10 weeks, 3, 4, 6, 9, and 12 months. Frequent initial sampling is necessary to determine the degradation pattern of compounds with rapid transformation. However, frequent initial sampling intervals will not be required when consideration of the structural properties of the pesticide molecule predicts that the compound will be persistent. The experiment can be terminated when the degradation pattern has been established or after one year.

i) Anaerobic biotransformation. Rates of degradation of the parent compound and major transformation products should be determined under anaerobic soil conditions when the pesticide is for use on flooded or poorly drained areas, or when assessment of physicochemical properties, mobility and degradation in soil indicate potential migration to subsoil. Anaerobic biotransformation studies are not required when anaerobic sediment/water studies are done. Soil from one-month samples of the aerobic soil biotransformation test can be waterlogged and/or purged with inert gases to create anaerobic conditions. In this way, the aerobic and anaerobic tests can run concurrently. Alternatively, anaerobic conditions can be imposed immediately after introduction of the pesticide to previously untreated soil (4). Samples should be taken at one and two months after the imposition of anoxic conditions.

Reports

The following information should be included in reports:

1. Soil textural class, particle size distribution, % organic carbon, cation exchange capacity, soil moisture content and soil moisture content at field capacity.
2. Analytical purity of active ingredient.

3. Mass (materials) balance at end of study.
4. Site of radiolabel.
5. Amount of pesticide added, application method and amount and identity of solvent.
6. Temperature of determination.
7. Soil collection date, geographical location of collection site, length and conditions of soil storage and soil handling and preparation.
8. Weight treated and sampled.
9. Number of replicates.
10. Duration of experiment.
11. Observed soil pH.
12. Results of microbial activity determinations.
13. Pesticide use history at collection site.
14. Full description of test methods, sampling and analysis procedure.
13. Specific description and interpretation of test results.

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2. Aquatic (Laboratory) - Anaerobic and Aerobic

Laboratory studies of aquatic biotransformation will be required for most pesticides that are intended for field use.

a) Radioisotopic methods must be used to identify the major products of transformation. Suitable non-radioisotopic techniques may be used to determine rates of degradation. Major transformation products are those that are present at more than 10% of the initial pesticide concentration.

b) Rates of degradation of the parent compound and major transformation products should be determined under anaerobic and aerobic conditions at constant temperature. **TWO TEMPERATURES (SPECIFIED) SHOULD BE TESTED:** one in the lower temperature range of 3° - 8°C and one in a higher temperature range of 20° - 30°C. There is no need to determine degradation products at the lower temperature, but a knowledge of the rate of degradation at this lower temperature would be useful. Aerobic incubations should be carried out under a standard lighting regime, e.g., 16 hrs light, 8 hrs dark, using fluorescent lights of the type recommended for plant cultivation. If the pesticide is photolabile, however, an additional aerobic incubation should be carried out in the dark, e.g., in foil-wrapped flasks. Light intensity, wavelength distribution and exposure time should be measured when aerobic incubations are not held in the dark. Anaerobic incubations should be held in the dark.

c) Aerobic degradation should be determined in unfiltered natural water held under static conditions (10), or aerated by shaking (7) or bubbling with air (3). Where physicochemical properties of the pesticide (e.g., adsorption/desorption parameters) suggest that sediment will be a major sink for the pesticide, degradation should be studied in sediment/water systems (1,2,6,11) rather than in unfiltered water. Aerobic conditions can be maintained in sediment/water systems by the use of open containers, or by shaking or bubbling with air. Water and sediment should be freshly obtained from a representative use site.

Anaerobic incubations in sediment/water systems should be carried out concurrently with aerobic incubations. Reducing conditions should first be established by holding samples under, or bubbling with, nitrogen.

d) Suggested sampling times include pretreatment, 0, 1, 3, 7, 10 days, 2, 3, 4, 6 weeks, 2, 3, 4, 6, 9, 12 months. Less frequent initial sampling may be permissible when consideration of the structural properties of the pesticide molecule predicts that the compound will be persistent. The study should continue until the patterns of degradation of the parent compound and its major transformation products are established, or for one year.

e) Sterile treated water or sediment/water samples, maintained under the same conditions as non-sterile samples, should serve as experimental controls. Standard plate counts should be performed to ensure that sterility is maintained for the test period.

f) Pesticide should be applied at one or two dosage rates. If two rates are used, a 10-fold difference in concentration is the standard separation. One pesticide concentration tested should be the maximum label-recommended rate or a concentration expected to occur in water runoff or as a result of spray drift (generally, < 1 ug mL^{-1}). The pesticide should be added to the water phase as a filter-sterilized aqueous solution (7) or, if this is not feasible owing to solubility limits, in a minimum volume of water miscible solvent, e.g., acetone or ethanol. The use of solvents may affect transformation through selection of types of microorganisms and effects on growth rates (9).

Reports

The following information should be included in reports:

1. Sediment textural class, particle size distribution, % organic carbon.
2. Microbial biomass.
3. Analytical or technical purity of active ingredient.
4. Mass (materials) balance at end of study.
5. Site of radiolabel.
6. Amount of pesticide used, and amount and identity of solvent.
7. Temperatures of determination.
8. Date of sediment and water collection, geographical location of collection site, handling and preparation (including sterilization method).
9. Liquid/solid ratio.

10. Weights and volumes treated and sampled.
11. Number of replicates.
12. Duration of experiment.
13. Observed pH.
14. Results of microbial activity determinations.
15. Dissolved oxygen (water) and redox potential (sediment) in anaerobic studies.
16. Suspended particulates.
17. Full description of test methods and sampling and analysis procedure.
18. Specific description and interpretation of test results.
19. Details of light intensity, wavelength distribution and exposure time should be reported, where appropriate (i.e., for aerobic incubations not held in the dark).

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Other details on experimental procedure and pertinent references can be found in:
EPA. 1982. Pesticide Assessment Guidelines.
Subdivision N. Chemistry: Environmental Fate
EPA - 540/9-82-021

6.3 FIELD STUDIES

Field studies are needed to demonstrate fate in the Canadian environment and to substantiate the physicochemical, mobility and biotransformation data from laboratory studies. Outdoor field studies are carried out under representative soil or aquatic conditions. It is mandatory that some field dissipation/accumulation studies be carried out under Canadian conditions to support final Canadian registration.

Definition of terms used:

1) Types of soil - the main differentiation is organic (muck) and mineral soil. Mineral soils may be further differentiated according to soil textural class (e.g., clay, silty clay loam, loamy sand, sand, etc.).

2) Plot - a single experimental unit, e.g., a control plot, a treated plot.

3) Replicate plot - one of two or more plots treated in an identical manner at one site.

4) Site - exact geographical location of a study.

5) Region - one of British Columbia, Prairies, Central Canada (Ontario/Quebec), Maritimes.

6) Area - a subset of a region characterized by similar climatic conditions or crops, e.g., the southern Ontario cornbelt is an area within the region of Central Canada (Ontario/Quebec).

7) Major tranformation products - degradation products/metabolites of the parent compound that are observed at any time in the laboratory studies (see section 6.2.C) at a level greater than 10 percent of the initial concentration of the parent compound.

8) Ideal application and planting techniques - the use of specially adapted application machinery to accurately apply pesticide in small plot field trials in a manner approximating field methods.

A. DISSIPATION AND ACCUMULATION - TERRESTRIAL

Terrestrial studies with pesticides under use conditions are necessary to substantiate laboratory findings, particularly with respect to dissipation/accumulation, leachability and potential carryover of residues.

Field studies to determine the behaviour of pesticides in soil can be performed in a variety of study systems, both small-scale and large-scale.

SMALL PLOT AND/OR LARGE-SCALE FIELD STUDIES MUST ADEQUATELY DEMONSTRATE THE BEHAVIOUR AND FATE OF THE TEST MATERIAL IN SOIL UNDER CANADIAN CONDITIONS.

1. The decision concerning the plot size in field studies rests with the applicant for pesticide registration.

2. The use of small plots in field studies is strongly recommended, but the use of small plots is not mandatory.

3. If the applicant for pesticide registration decides to conduct large-scale field studies and the generated pesticide dissipation data are not interpretable, then additional studies using small plots may be requested.

1. Study Systems

1.1 Small Plots

Small plots (2,6,7,8,12,13) are treated using ideal application techniques and, thus, minimize the difficulty in obtaining satisfactory pesticide dissipation curves. Small plots may range in size from microplots, typically 100 cm^2 - 2500 cm^2 (7,8,13) to 2-6 m^2.

Microplots - Microplots are amenable to the use of radiolabeled compounds, which may be necessary for pesticides that are applied at very low application rates. Microplots are most suitable for use with relatively immobile pesticides because of problems associated with removing deeper soil layers.

Small Plot Studies - When pesticide dispersion is uneven due to crop interference, dissipation curves may be difficult to generate or interpret. In such cases, the use of bare small plots (i.e., up to 2 x 2-6 m^2 not sown to intended crops) may be considered. Hand-weeding is the preferred method of maintaining plots plant-free. The bare plot study system is recognized by Environment Canada as an artificial system that is, nevertheless, useful to demonstrate an interpretable pesticide dissipation curve. It is recognized that bare soil plots have drawbacks, e.g., soil temperature and moisture regimes may not represent normal use conditions, and the contribution of plant uptake/retention to environmental fate will not be accounted for. Nevertheless, the factors involved in cropped plots are complex and variable, and their effects may be difficult to quantify and interpret. For these reasons, it is recommended that data from field studies using bare small plots be submitted. However, data from field studies using cropped plots will be acceptable, if these data are interpretable and dissipation curves can clearly be demonstrated.

1.2. Large Scale Studies

Large-scale studies (1,9,10) are conducted using normal agricultural practices (e.g., cultivation prior to planting, etc.) and equipment. The area of treated plots is typically 8 rows by 25 m, but may range to an entire field of several hectares, depending on the design of the experiment and the use for which the product is intended.

In large-scale field studies, the primary goal is to establish the dissipation of the pesticide and its major transformation products in soil. However, crop-residue data may, if environmental fate studies are not disrupted, be collected during these studies and used to meet requirements of part 5, Trade Memorandum T-1-237 for registration.

2. Experimental Design for Field Studies

2.1 Field Site Selection

Within each region in which a pesticide is intended for use, field study sites should be selected to take into account crop distribution and the associated range in soil texture (i.e., the finest versus the coarsest soils) and climate [i.e., low versus high precipitation and cool versus warm (soil) temperature].

The suggested number of mineral soil field-study sites, for a major crop grown across Canada, is outlined in Table 1. While the theoretical regional distribution of study sites (presented in Table 1) is designed to encompass the variation in soil and climatic conditions within each region, it may be modified by substitution** or addition in light of the target crop distribution or on the basis of well-substantiated scientific evidence concerning the environmental behaviour and fate of a particular pesticide. Applicants, however, are advised to seriously consider the guidance offered in Table 1, being mindful of their responsibility to demonstrate the fate of their pesticides in the Canadian environment.

Table 1

Suggested Regional Distribution for
Number of Mineral* Soil Sites for Field Studies

Region	Canadian	Appropriate American	Total
B.C.	1	1	2
Prairies	3	1	4
Central Canada (Ont., Que.)	2	2	4
Maritimes	1	1	2

* where a crop is grown on both mineral and organic soils, an additional Canadian site must be located in an organic soil.

** e.g., a registrant may request an exemption from doing a study upon provision of a rationale that an Ontario study should substitute for a Maritime study on the basis of similar conditions of soil, climate and water table, etc.

Canadian field studies are mandatory. However, the results of studies conducted at appropriate sites in northern states, under similar climatic conditions and with major types of soil as found in proposed use regions of Canada, may be submitted in lieu of some Canadian studies in accordance with the following (see also Table 1);

- one American site may be substituted for one of the two suggested for B.C.;
- one American site may be substituted for one of the four suggested for the Prairies;
- two American sites may be substituted for two of the four suggested for Central Canada;
- one American site may be substituted for one of the two suggested for the Maritimes.

For the region of Central Canada, in cases where a crop is grown only in southern Ontario, there should be a total of two study sites (one or more appropriate American sites plus at least one in southern Ontario).

Realistic situations are represented by the examples in Table 2.

Table 2

Number of Sites

Use	Region	Canadian	Appropriate American	Total
Major use in several regions e.g., cereals	Prairies	3	1	4
	Central Canada (Ontario)	1	1	2
	Maritimes	1	0	1
Use in 1 Region, e.g., field tomatoes	Central Canada (Ontario)	1	1	2
Orchard pesticide	B.C.	1	1	2
	Central Canada (Ontario)	1	1	2
	Maritimes	1	0	1

If the data generated by soil field studies do not adequately demonstrate the environmental fate of a pesticide under potential use conditions, then additional studies will be requested.

2.2 Number of Plots per Site

Replicate Plots - Two or more replicate plots are to be treated at each site.

Control Plots - Untreated control plots should always be included. Control plot samples are a source of uncontaminated soil for residue storage stability and recovery determinations in the laboratory and may not require as frequent sampling as treated plots (i.e., three times at maximum over a sampling season).

Buffer Zones - Plots are to be separated by buffer zones to prevent cross-contamination through drift during treatment.

2.3 Pesticide Application

Replicate plots should be treated at the maximum recommended application rate using the commercial formulation and following the proposed times of application for both single or multiple applications,

as appropriate. The method of pesticide application should follow, as closely as possible, normal commercial use application procedures.

2.4 Sampling Requirements

At each sampling time, care must be taken to obtain soil samples for residue analysis that are representative of the replicate plots; accurate and consistent sampling is vital for meaningful results.

a) Sampling Patterns

i) A random or systematic soil sampling pattern (11) may be followed, depending on the type of pesticide application. For example, the soil may be sampled in-row only (seed furrow or band treatment) or by a random pattern which covers the entire treatment area (broadcast application). Great difficulty may be encountered in obtaining interpretable results using an in-row sampling pattern; it is recommended that extreme care be taken in the application and sampling procedures.

ii) Outside rows of treated areas should be excluded from sampling in order to avoid variability resulting from possible undercoverage or drift; in the case of confined plots, edge effects may contribute to this variability.

iii) Soil core holes should be marked after sampling.

iv) Plugging holes may be useful in preventing the movement of treated soil to greater depths and subsequent anomalous results.

b) Depth of Soil Sampling

i) In order to fully demonstrate the fate of the pesticide under study, soil should be collected from a depth sufficient to encompass the vertical distribution of the pesticide and its major transformation products at each sampling time. Data from laboratory studies (physicochemical properties, mobility and transformation) can be used in conjunction with water recharge estimates, (e.g., average rainfall data, expected irrigation)

and soil permeability properties to establish appropriate core depths.

ii) Soil cores should be divided into (at least) an upper and lower section in order to determine the extent of leaching of the pesticide or its major transformation products. The lowest section of the sampled cores should not contain amounts of the active ingredient or major transformation products. Soil should be collected from depths adequate to ensure that this can be demonstrated.

iii) In the absence of rainfall or irrigation, initial or zero-time samples need be taken to only just past the depth of incorporation.

iv) At later sampling times, a sampling depth of 15 cm should be sufficient for compounds of low mobility.

v) When compounds of higher mobility are being studied, or with soil-incorporated compounds, deeper cores may be necessary, especially as the season progresses or if the season is wet.

c) Times of Soil Sampling

i) Soil sampling should be carried out prior to treatment, immediately after treatment (zero-time) and at increasing intervals (daily, weekly, monthly) between sampling times depending on prior estimates of pesticide dissipation.

ii) The dissipation of a product used in multiple applications over a season should be studied through a full cycle of applications (4).

iii) Residue data should be obtained until: (1) 90% of the pesticide and /or its major transformation products have disappeared from the soil profile or (2) the pattern of dissipation has been clearly established. It is necessary to determine more than the 50% decline time (DT_{50}) from the initial application because the dissipation rate constant often decreases with time (i.e., the half-life is not constant as in first-order kinetics).

iv) An overall plot sample should be taken at the end of the growing season to determine residue carryover to the next season (sampling in subsequent years may be necessary). Long-term studies are required if dissipation is slow to occur.

d) Number and Pooling of Samples

i) The number and diameter (typically 3 to 12 cm) of soil cores to be taken should be based on the size of the plot, the type of soil and the amount of soil required for analysis.

ii) Corresponding depths of soil cores from a single plot can be pooled and mixed thoroughly to give one representative composite sample from which an aliquot, (e.g., 300 g) can then be taken for analysis.

iii) Duplicate sets of cores should be taken at zero-time from each treated plot and analysed separately to firmly establish the initial residue value. The amount of pesticide in all subsequent soil samples will be evaluated in comparison to the zero-time value.

iv) Samples from replicate plots should not be pooled together.

v) An adequate number of cores per plot should be collected at each sampling time to ensure that the sample is representative of the plot. For example, a composite sample from a 2m x 1m small plot may consist of 10 to 15 soil cores (3 cm in diameter) per sampling time over a period of one year. For field studies of longer duration, small plots of larger area should be used to accommodate the collection of a greater total number of cores that results from the increase in number of sampling times. Owing to the increase in plot size, the number of cores collected per sampling time should be increased (e.g., if the plot dimensions are doubled, then the number of cores collected per sampling time should be increased by 50%).

vi) In large plots, soil cores of greater diameter are usual, but 20 or more cores should be collected; the variation present within large plots is greater than that in small plots because of less uniform pesticide application and greater natural variation in the soil.

vii) If, within a plot, there are areas of different types of soil, soil organic matter content, etc., or knolls/depressions, then representative cores from such areas should be pooled and analysed separately from other samples (i.e., all samples not pooled together).

e) Handling of Samples

Both soil and crop samples should be frozen if they cannot be extracted immediately. To check stability of pesticide residues during storage, untreated soil samples should be fortified with analytical standards (for parent chemical and major transformation products), stored and then extracted and analysed in the same manner as samples from treated field plots (5).

3. Special Problems

a) Low Application Rates:

Pesticides intended for use at very low application rates may present difficulties with respect to detection in soil soon after application. The preferred method for solving such problems is to lower detection limits by developing more sensitive analytical techniques. If this cannot be accomplished, field studies (small plot or large-scale) may be conducted at an elevated application rate of approximately 2-3 times the highest recommended rate. In support of studies at elevated application rates, a bioassay procedure and/or microplot dissipation study using radiolabelled pesticide may be acceptable on a case-by-case basis. Field studies using radiolabelled technical pesticide may be submitted as the sole field dissipation studies provided that they are supported by data from comparative (laboratory or field) studies conducted with technical and formulated pesticide. Such comparative studies would assess the similarity between the two test substances with respect to

pesticide dissipation (transformation and leaching) in soil.

When field studies are conducted with formulated product that contains radiolabelled pesticide, appropriate precautions must be taken to ensure that the radiolabelled pesticide behaves in exactly the same manner as non-labelled pesticide in the formulated product (i.e. simply "spiking" formulated product with radiolabelled pesticide may lead to unrepresentative and unacceptable results). It would be preferable to formulate (via lab scale process) with radiolabelled chemical.

b) Large Scale Orchard Studies

Small plot studies are recommended in orchard sites. However, if large scale studies are conducted, then the area to be treated should be large enough to allow the use of commercial application. Within this treated area, replicate plots should be established and should include five or more uniformly sized trees. Soil samples should be taken from two distinct areas within a replicate plot: (i) the dripline areas (in-row) and (ii) between rows. Samples should not be pooled between these two areas, but samples taken from within each of these areas may be pooled. In general, 10-15 soil cores/composite/sampling date should be sufficient. Pesticide residues in surface organic layers (thatch and/or plant litter) should be determined at appropriate intervals. Samples of the surface organic layer should not be mixed with samples of underlying soil. To obtain an adequate sample of the surface organic layer, it may be necessary to sample an area greater than that which would be sampled with a typical soil core sampler.

Reports

1. The following information should be considered for inclusion in reports:

   a) Soil textural class, particle size distribution and % organic carbon for each depth of soil to be sampled.

   b) Plant species, variety, spacing and row spacing, developmental stage (at sampling times), planting and

harvest dates, soil tillage, cultivation and fertilization, where applicable.

c) A general description of the formulation (e.g., type, carrier, adjuvants).

d) Formulation lot number and concentration of active ingredient.

e) A mass balance. For field studies conducted with non-labelled material, this can be satisfied by a direct comparison of the extractable amounts of parent chemical and major transformation products in the soil profile at each sampling time with the amount of pesticide initially applied. Calculation of residue concentrations on the basis of equivalent amounts of parent chemical per unit area of soil (e.g., kg ha -1) enables this direct comparison. For field studies conducted with radiolabelled material, the mass balance would include amounts of non-extractable or "bound" pesticide residues in the soil profile.

f) Site of radiolabel on the molecule, where applicable.

g) Application method, equipment type, application date and time of day applied. Quantity and identity of diluent and additives. Spray volume per unit area and application rate. Weather conditions during application, including cloud cover, wind, temperature and relative humidity.

h) Geographical location of test sites.

i) Conditions and length of sample storage.

j) Number of replicate plots. Duration of experiment.

k) Observed soil pH for each depth sampled.

l) Pesticide use history at site. Topography of site. Layout of plots. Temperature, precipitation, and plot irrigation data for the duration of the sampling season. Applicants for registration should note that where photodegradation is a major route of pesticide dissipation, the hours and intensity of sunlight should be documented.

m) Approximate depth and fluctuations in the water table (when pesticide residues are mobile and water tables shallow). The presence of a high water table (5 metres) may influence pesticide environmental behaviour and dissipation at the study site. If such water table data are not available from local surveys or sources, then they should be collected at the study site, if the study conditions of high water table and mobile pesticide occur together.

n) Sample moisture content and bulk density.

o) Storage stability of residues.

p) Full description of sampling and analysis procedure.

q) Specific description and interpretation of test results.

2. Additional information that may aid interpretation, such as soil porosity, soil moisture content at field capacity and at permanent wilting point, other weather conditions, historical weather data for the geographical area, soil taxonomic classification, soil series description, and general condition of the plots during the study, etc., could also be included.

3. Soil field sample residue data should be reported uncorrected for storage stability losses.

4. A comparison of the soil and climatic conditions between any American study sites and proposed Canadian use area(s) should be included in reports.

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Other details on experimental procedure and pertinent references can be found in:

EPA 1982. Pesticide Assessment Guidelines. Subdivision N. Chemistry: Environmental Fate EPA-540/9-82-021.

B. DISSIPATION AND ACCUMULATION - AQUATIC

A combination of laboratory and field studies is required for thorough assessment of fate and effects of pesticides in natural waters (2). Rates of dissipation and levels and types of transformation products in field studies may differ from those in laboratory studies. The aquatic field study is intended to confirm results of, and validate predictions from, laboratory studies and to indicate pesticide distribution in aquatic/sediment compartments.

Aquatic field studies will be required to clarify the fate of all pesticides applied directly to water. Aquatic field studies will normally be required for pesticides intended for large-scale forestry or agricultural use. In cases where field studies on pesticide dissipation in aquatic systems are not included in the submission, the applicant should provide a rationale for not conducting the studies. For example, mitigating factors to be considered in the rationale for not conducting aquatic field studies include:

1) low persistence
2) low mobility
3) low potential to bioaccumulate
4) low acute toxicity to aquatic organisms
5) a proposed use pattern with limited potential for environmental impact (e.g., speciality crops)

1. Study Systems

1.1 Small-scale aquatic field studies will normally be required. Natural or artificial small ponds or enclosures, 1-5 m^3, < 1 m in depth with < 50 m^2 surface area and with little or no inflow or outflow, are suitable for small-scale aquatic studies (4, 7, 8, 12). For small-scale field studies, only one Canadian location may be required if additional aquatic field dissipation studies from outside of Canada, but representative of Canadian conditions, are submitted for review. If the small-scale studies conducted in Canada are not satisfactory either because they do not

demonstrate the dissipation/accumulation of the pesticide, or because their results differ significantly from those of the non-Canadian studies, it may be necessary to conduct further studies in Canada. If no aquatic field studies from outside of Canada are submitted, Canadian studies must be conducted at a minimum of two locations. The locations selected should represent the extremes of aquatic and climatic conditions found in the regions of proposed major uses.

(2) Large-scale aquatic field studies. Although small-scale aquatic field studies are recommended, large-scale Canadian studies [including studies in limnocorrals (10), ponds, streams, etc.] may be substituted for them.

For pesticides intended for aquatic use and for pesticides with a high potential to impact (directly or indirectly) non-target aquatic organisms, large-scale aquatic field studies will be required so that the pesticide can be assessed under conditions of actual use. These studies may be combined with biological impact studies. Large-scale studies must be conducted in region(s) of proposed major use.

(3) Monitoring of pesticide residue levels in waters near treated areas (6,13) may be requested in addition to small- or large-scale aquatic studies to confirm that the stipulated operational use conditions do not lead to aquatic contamination through drift, runoff, erosion or leaching. In most cases, monitoring trials will be restricted to analysis of pesticide residues. Analytical sensitivity should be below expected effect levels for biological systems. In unusual circumstances where no-effect-levels below analytical sensitivity have been determined, biological indicators may be necessary. Monitoring trials will normally be done under temporary registration.

2. Experimental Design

Compartments of aquatic environments (e.g., water, sediment, biota) that are critical to the fate of a particular pesticide must be sampled thoroughly, (e.g., sediment and

suspended solids are important compartments for substances with water solubility less than 1 ug/mL^{-1}). Prediction of compartments likely to be critical requires a knowledge of the physicochemical properties, adsorption/desorption behaviour and transformation rates and products of the pesticide. Prediction can be facilitated by the use of multicompartment aquatic fate models such as EXAMS (1) and NRCC (9). These models can be used to predict fate and may be useful in designing field studies but, at present, cannot replace field studies.

(a) Aquatic study systems should simulate water chemistry and sediment characteristics likely to be encountered under proposed Canadian use conditions. For products used in water or on ditchbanks, the recommended method of application should be followed, and the product should be applied at the maximum proposed rate and number of applications. For other products, application should follow a "worst-case" scenario, e.g., inadvertent direct spray by aircraft or spray drift from adjacent field applications. Direct addition of the product to water is recommended in these cases. In those cases where the proposed label recommends multiapplication, the experimental design should include pesticide applications in accordance with the label instructions.

(b) The formulated (end-use) product should normally be used. Use of radiolabeled pesticide in small-scale studies may be considered as a means of estimating a mass balance.

(c) Aquatic dissipation field studies should be performed at least in duplicate. Sampling of control and treated sites prior to pesticide application is necessary to establish that similarity exists between these sites. Untreated controls should be sampled throughout studies to provide background data on water and sediment characteristics. The use of enclosures in ponds or shallow lakes facilitates replication.

(d) Pesticides should be applied at the time of year recommended for use. Sampling should be carried out prior to treatment, immediately after treatment and at increasing intervals between samplings (daily, weekly, monthly) depending on estimates of field dissipation from laboratory data.

(e) Samples from a single pond or enclosure can be pooled to yield a single representative sample per replicate for each sampling time. To encompass possible concentration gradients, the collection of surface (0-5 cm), and subsurface (20 cm) water samples is recommended. Sampler design should be such that contamination from a surface slick is minimized, (e.g., with an inlet that can be opened under water). Water from the different depths should be analysed separately.

(f) Sediment samples should consist of only the upper 5 cm or less. Deeper samples can result in decreased pesticide residue concentrations as a result of dilution. Devices which cause minimal disturbance to the sediment such as corers (5) are recommended. Alternatively, containers filled with sediment can be placed on the pond bottom before treatment and removed at intervals (7). The sediment sample should be drained of excess water, taking care to avoid loss of flocculent material at the sediment-water interface.

(g) If possible, the extraction of residues from water samples should be initiated immediately by the addition of a suitable solvent, when laboratory studies indicate rapid transformation of pesticide residues in natural waters. Otherwise, samples should be refrigerated immediately and extracted as soon as possible. Sediment samples should be frozen immediately. To check stability of pesticides during storage, water and sediment samples from untreated areas must be fortified with analytical standards, stored and analysed in the same manner as samples from treated areas.

(h) If plastic, especially polyethylene, is used to line artificial ponds or enclosures, sorption/desorption of the product to/from plastic liners should be monitored (4, 11).

(i) Studies may be combined with environmental toxicology studies on bioaccumulation and transformation in fish, non-target invertebrates and aquatic macrophytes.

Reports

The following information should be considered for inclusion in reports:

1. Sediment characteristics (textural class, particle size, distribution and % organic carbon).
2. Dissolved oxygen, pH, suspended solids, turbidity and temperature of water.
3. Redox potential, temperature, and pH of sediment.
4. Plant species, variety and biomass, where appropriate.
5. Geographical location and description of test site.
6. Weight, volume or area treated and sampled. Number of replicates.
7. Application method, equipment used, application date and time, quantity of diluent, spray volume per unit area, application rate and weather conditions during application (cloud, wind, relative humidity and air temperature).
8. Description of formulation used, (e.g., type, carriers), lot number and concentration of active ingredient.
9. Dates of sample collections and full description of sampling and analysis procedures.
10. Any correction(s) in the reported pesticide residue data should be clearly stated and supported with sample calculations.
11. Conditions and length of sample storage and storage stability data.
12. Precipitation, hours of sunlight and water flow rates during the study period.
13. A mass balance or rough estimate of mass balance where accuracy is not feasible.
14. Specific description and interpretation of test results.

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C. SPECIAL SITUATIONS RELATED TO INTENDED USE PATTERN

These situations have additional or extenuating requirements as they do not fit neatly into broad categories of "terrestrial" and "aquatic", or conversely, are more specific and thus require separate consideration.

1. Forestry

Forestry use of a pesticide is both terrestrial and aquatic (1,2,3). Therefore, the laboratory investigations should consist of those required for terrestrial use, while field studies would cover aquatic as well as terrestrial locations. Field studies of dissipation and accumulation will include residue data from the following strata: foliage, leaf litter, soil under litter, exposed soil, standing and moving water and sediment, fish and other non-target organisms. One site with replicated treatment is sufficient if the use pattern does not involve widely varying forest environments. Otherwise, at least two different sites must be tested. Some field persistence tests, terrestrial and aquatic, must be done under Canadian conditions, for registration in Canada for forestry use.

2. Tank Mixes

When two or more pesticides are to be applied together, dissipation and persistence are not normally influenced by the combination. Information on the individual components will be provided through the studies suggested in the guidelines. A laboratory or field persistence test may be required on a case-by-case basis.

3. Greenhouse

Greenhouse use involves a limited or restricted physical area. Tests on volatility and photodegradation in air have been mentioned (6.2 A 1, 3 (iii)). If adsorption/desorption studies are available, leaching studies will not be required. Field trials may consist of confined area tests or small plot studies; ie. large-scale field trials are not necessary as they are not applicable to intended use.

4. Domestic Indoor/Outdoor

Because use pattern involves a limited area and quantity, laboratory studies may be sufficient to determine environmental fate. If adsorption/desorption data are

available, leaching studies will not be necessary. Small-scale field trials may be required on a case-by-case basis depending on the extent of intended use. Large-scale field trials of dissipation and accumulation are inappropriate.

5. Miscellaneous

Most individual use patterns will fall into the terrestrial, aquatic, or special situation categories described. A case-by-case examination of unusual or new use patterns will determine the extent and type of study desired to assess environmental fate. Other related information may be required for a particular pesticide in addition to the studies outlined in the guidelines.

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6.4 STORAGE, DISPOSAL, AND DECONTAMINATION

A. LABEL AND PACKAGING INFORMATION

1. Storage life and stability under typical storage conditions:

   i) occurrence of deterioration or changes in pesticide;

   ii) corrosion test of containers; and

   iii) rinsability of container.

2. Proper disposal of excess material (1): unused pesticides and rinsates.

3. Disposal of empty containers (2).

4. Decontamination of personal or in-transit spills.

5. Safe re-entry period assessment eg. from photolysis in vapour phase (3).

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6.5 ASSESSMENT OF ENVIRONMENTAL FATE DATA

In order to register a pesticide, the fate of that pesticide when it enters biotic systems must be known. The purpose of environmental fate data is to determine if contamination is liable to occur and, if so, to what degree. The studies suggested in the guidelines will supply enough information to assess the impact that the intended use of a pesticide will have on the environment. This will be evaluated in conjunction with toxicological data to determine the safety or potential hazard of a compound. Analysis of the risks and benefits of a pesticide with particular emphasis on the distribution and extent of use, will lead to the decision to register the pesticide. This decision is the responsibility of the Pesticides Directorate, Agriculture Canada.

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