

ETHOPROPHOS (149)

*First draft prepared by Trijntje van der Velde-Koerts and Bernadette C. Ossendorp,
National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands*

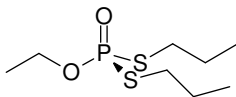
EXPLANATION

Ethoprophos, a nematicide and soil-insecticide, was evaluated for residues in 1984 and in 1987. The toxicology of ethoprophos was periodically reviewed by the 1999 JMPR. Ethoprophos was listed as a priority compound under the periodic re-evaluation programme of the 30th Session of the CCPR (ALINORM 99/24 App VII) for residue review by the 2001 JMPR. The manufacturer requested the postponement of the residue evaluation.

The basic manufacturer supplied information on identity, metabolism and environmental fate, residue analysis, use pattern, residues resulting from supervised trials on strawberry, banana, cucumber, melon, pepper, tomato, potato, sweet potato and sugar cane, fate of residues during storage and in processing, residues in food in commerce or at consumption and national maximum residue limits.

IDENTITY

ISO common name: ethoprophos
 Chemical name
 IUPAC: *O*-ethyl *S,S*-dipropyl phosphorodithioate
 CAS: *O*-ethyl *S,S*-dipropyl phosphorodithioate
 CAS Registry No: 13194-48-4
 CIPAC No: 218
 Synonyms and trade names: *S,S*-dipropyl *O*-ethyl phosphorodithioate;
 prophos; ethoprop;
 Mocap; VC9-104; ENT-27318; AE F034142
 Structural formula: established by IR, ¹H-NMR, ¹³C-NMR, ³¹P-NMR, APCI-MS (+/-)
 and UV-VIS (according to OECD 101) (Boeuf *et al.*, 2000)



Molecular formula: C₈H₁₉O₂PS₂ (Boeuf *et al.*, 2000)

Molecular weight: 242.34 (Boeuf *et al.*, 2000)

Physical and chemical properties

Pure active ingredient

Physical and chemical properties were determined with the pure active ingredient, unless specified otherwise.

Property	Description or result	Method(s) (References)
Minimum purity	980 g/kg	(Barriere, 2004a)
Appearance	colourless clear liquid, odour not assessed	(Ristorcelli, 2001a)

Property	Description or result	Method(s) (References)
Vapour pressure	78 mPa at 20°C 123 mPa at 25°C calculated from Clausius-Clapeyron relationship from measurements at 16.0-20.4-24.2-31.4°C	OPPTS 830.7950 92/69/EC A4 OECD 104 Knudsen effusion method (Ristorcelli, 2001b)
Melting/freezing point	below -70°C (203.2 K)	OPPTS 830.7200 92/69/EC A1 differential scanning calorimetry (Ristorcelli, 2001a)
Octanol/water partition coefficient	Log K_{ow} = 2.99 (95% confidence interval 2.9-3.1), temperature not stated K_{ow} is not pH-dependent	OPPTS 830.7570 92/69/EC A8 OECD 117 HPLC method (Ristorcelli, 2001d)
Water solubility	1.3-1.4 g/L (RSD 1.9%-3.8%), 20°C; solubility not pH-dependent, determined from pH 4 to 9	OPPTS 830.7840 92/69/EC A6 OECD 105 flask method (Ristorcelli, 2001c)
Solubility in organic solvents	purity 94.4% (technical grade) > 500 g/L at 20°C in acetone; acetonitrile; 1,2 dichloroethane; ethyl acetate; n-hexane; methanol; n-octanol; toluene	OPPTS 830.7840 92/69/EC A6 OECD 105 flask method (Ristorcelli, 2001c)
Relative density (D_{4}^{20})	1.096, measured at 20.7°C instead of 20°C	OPPTS 830.700 92/69/EC A3 oscillating densitometer (Ristorcelli, 2001a)
Hydrolysis in aqueous solution, study 1	- Chemical and radiochem. purity >98% - Stability tested over 6 week period at 2 and 200 mg/l at pH 3, 6, 9, in the dark with radio-label on C-1 carbon of the propyl group. Half-life 28-36 weeks at pH3, 20°C 33-39 weeks at pH 6, 20°C 39-44 days at pH 9, 20°C 16-21 weeks at pH 3, 35°C 14-16 weeks at pH 6, 35°C 10-14 days at pH 9, 35°C - Identified degradation product <i>O</i> -ethyl <i>S</i> -propyl phosphorothioic acid (mP). - Radioactivity was partly lost (up to 40% at 35°C at pH 9 at 7-42 days) as unknown volatile compounds.	Fed. Reg. 43, (132) 29717 (Norris, 1983), non-GLP
Hydrolysis in aqueous solution, study 2	-Radiochem. purity >98%, chemical purity not stated - Stability tested over 30 day period at 10 mg/l at pH 5, 7, 9, in the dark, with 1-ethyl- ^{14} C radiolabel at $25 \pm 1^{\circ}\text{C}$. -Stable at pH5 and 7, 25°C; -Half-life 83 days at pH =9, 25°C; -Identified degradation products ethyl alcohol and <i>S,S</i> -dipropyl phosphorodithioic acid (mK) -Total ^{14}C recovery >99%	US EPA 161-1 (Das, 1989)
Hydrolysis in aqueous solution, study 3	-Radiochem. purity >98%, chemical purity not stated -Stability tested over a 20 day period at 10 mg/l at pH 4, in the dark, with radio-label on C-1 carbon of the propyl group at 60, 70 and 80°C $\pm 1^{\circ}\text{C}$. - Half-life 10 days at 60°C; Half-life 3.5-4.0 days at 70°C Half-life 1.4 days at 80°C Stable at pH 4 at 20°C, half-life >365 days, calculated from Arrhenius plot. - Identified degradation product <i>S,S</i> -dipropyl phosphorodithioic acid (mK) - Total ^{14}C recovery >93%.	OECD 111 (Maurer, 2002)

Property	Description or result	Method(s) (References)
Photolysis in aqueous solution; study 1	-Radiochem. purity 98.6%, chemical purity >97.7% - Stability tested over 30 day period at 22 mg/l at pH 7, with 1-ethyl- ¹⁴ C radiolabel at 25 ± 1°C, continuous radiation with xenon-arc lamp at one-half the intensity of sun light. - Stable with or without sensitizer (1% v/v acetone); half-life could not be calculated - Total ¹⁴ C recovery > 98%.	US EPA 161-2 (Carpenter, 1989)
Photolysis in aqueous solution; study 2	-Radiochem. purity 99.5%, chemical purity not stated - Stability tested over 30 day period at 15 mg/l at pH 7, with 1-ethyl- ¹⁴ C radiolabel at 25 ± 1°C, continuous radiation with xenon-arc lamp at one-half the intensity of sun light. - Half-life 122 days without sensitizer Half-life 104 days with sensitizer (1% v/v acetone). - Total ¹⁴ C recovery > 98%.	US EPA 161-2 (Gorman, 1995)
Dissociation constant	purity 94.4% (technical grade); pH of a 1% (w/v) suspension in water, at 23°C =3.45 Ethoprophos does not dissociate.	OPPTS 830.7000; (Bascou, 2001)

Technical material

Property	Result	Method(s) (references)
Minimum purity	940 g/kg	(Barriere, 2004a)
Main impurities	no data provided	
Appearance	colourless, clear liquid, odour not assessed	(Ristorcelli, 2001a)
Relative density (D ₄ ²⁰)	1.093, measured at 20.7°C in stead of 20°C	OPPTS 830.700 92/69/EC A3 oscillating densitometer; (Ristorcelli, 2001a)
Freezing point	below -70°C (203.2 K)	OPPTS 830.7200 92/69/EC A1 differential scanning calorimetry (Ristorcelli, 2001a)
Stability	Technical material was placed in 3 different kind of packaging containers. No significant decrease was observed after a 1 year storage period at ambient temperature (23-25°C) and 50% relative humidity.	EPA 40 CFR 158.175, D 63-17 (Eubanks, 1991)

Formulations

Ethoprophos end-use products are formulated mainly as granulates (GR 50, 100, 150, 200 g ai/kg), or as emulsifiable concentrates (EC, 69.6, 172.9, 200, 720 g ai/l). Ethoprophos can also be formulated as microgranulate (MG, 100, 200 g ai/kg) or as an emulsifiable gel packaged in a water-soluble bag (gel, 720 g ai/l).

FAO specifications for technical and formulated ethoprophos have not been published.

Abbreviations and code names

Table 1. Metabolite and degradation product codes used in the present review.

Code used here	Code used in study reports	Name	Found in
mA	M1 AE0592496	<i>O</i> -ethyl <i>S</i> -propyl hydrogen phosphorothioate (<i>O</i> -ethyl <i>S</i> -propyl phosphorothioate)	rat/rabbit; goat?; hen?;

Code used here	Code used in study reports	Name	Found in
			plant; aerobic soil
mB		propanethiol	volatile
mC		dipropyl disulfide	plant; not found in rat; not tested in livestock not found in aerobic soil
mD		ethyl propyl sulfide	plant, not found in rat; not tested in livestock not found in aerobic soil
mE		ethyl propyl sulfoxide	plant, aerobic soil not found in rat, not tested in livestock
mF		ethyl propyl sulfone	plant, aerobic soil not found in rat, not tested in livestock
mG		methyl propyl sulfide	plant?, rat, not tested in livestock
mH		methyl propyl sulfoxide	plant?, rat, not tested in livestock
mI		methyl propyl sulfone	plant?, rat, not tested in livestock
mJ		ethyl dihydrogen phosphate (ethyl phosphate)	plant, rat, goat?; hen?
mK	AE 0712739 RPA 112748	<i>S,S</i> -dipropyl hydrogen phosphorodithioate (desethyl ethioprophos)	hydrolysis product in water rat, rabbit; not found in plant, not tested in livestock
mL		<i>S</i> -propyl dihydrogen phosphorothioate	plant?; rat; rabbit; not tested in livestock
mM		<i>S</i> -ethyl glutathione	rat; rabbit not tested in plant, not tested in livestock
mN	OME	<i>O</i> -ethyl <i>O</i> -methyl <i>S</i> -propyl phosphorothioate	aerobic soil?; plant?; hen? not found in rat; not found in goat
mO	SME	<i>O</i> -ethyl <i>S</i> -methyl <i>S</i> -propyl phosphorodithioate	aerobic soil?; plant?; hen? not found in rat; not found in goat
mP	SH	<i>O</i> -ethyl <i>S</i> -propyl <i>S</i> -hydrogen phosphorodithioate <i>O</i> -ethyl <i>S</i> -propyl phosphorodithioate	hydrolysis product in water; rat; not tested in plant, livestock or soil

Table 2. Other abbreviations used in the present review.

Code	Abbreviation for:
DAT	days after (last) treatment
ACN	acetonitrile
ai	active ingredient or active substance
CEC	cation exchange capacity
DCM	dichloromethane or methylene chloride
kg dw	kilogram dry weight (feed or soil)
EC	emulsifiable concentrate
EI +/-	electron impact with positive/negative ionisation (for MS)
eq	ethioprophos equivalents
GBq	giga Becquerel
GC-AFID	gas chromatography with alkaline flame ionisation detection
GC-ECD	gas chromatography with electron capture detection

Code	Abbreviation for:
GC-FPD	gas chromatography with flame photometric detection
GC-MC	gas chromatography with microcoulometric detection
GC-MS	gas chromatography with mass spectrometric detection
GC-MS-MS	gas chromatography with tandem mass spectrometric detection
GC-NPD	gas chromatography with nitrogen phosphorus detection
GC-PFPD	gas chromatography with pulsed flame photometric detection
GC-TSD	gas chromatography with thermionic specific detection = GC-NPD
GI tract	gastro intestinal tract
GPC	gel permeation chromatography
HPLC	high performance liquid chromatography
ILV	Independent laboratory validation
LSC	liquid scintillation counting
m/z	mass to charge ratio (mass spectrometry)
om	organic matter
PTVLV injector	programmable temperature vaporizing large volume injector
SIM	single ion monitoring (mass spectrometry)
TLC	thin layer chromatography
TAR	total applied radioactivity or total administered radioactivity
TRR	total recovered radioactivity

METABOLISM AND ENVIRONMENTAL FATE

Animal metabolism

The Meeting received information on the fate of orally dosed ethoprophos in lactating goats and laying hens. Ethoprophos was labelled at the 1-ethyl position. Metabolism in laboratory rats was summarized and evaluated by the WHO Core Assessment Group of the JMPR in 1999.

Ruminants: lactating goats

[1-ethyl-¹⁴C]ethoprophos was administered orally once daily for seven consecutive days by capsule to two lactating goats (Alpine; 46-54 kg; two years old) at an actual (mean) dietary concentration of 32 mg ai/kg dry feed (Byrd, 1993, 1994). The radiochemical purity after repurification was 99.0%, with a specific activity after dilution of 0.11 GBq/mmol. One control goat received capsules containing 100 mg cellulose. Mean feed intake was 632 g/goat/day. Immediately after the sixth dose one goat was enclosed for 24 hours in a plastic gas collection tent with an NaOH trapping solution. The tent was removed immediately before the seventh dose. Urine and faeces were collected once daily; milk was collected twice daily and was pooled for each 24 h period after dosing. Whole blood was collected before termination. The goats were slaughtered 20-21 hours after the final dose and liver, bile, kidney, muscle (from rear leg and lumber spine), fat (omental and peripheral), GI tract tissue and contents were sampled. Samples were homogenised and stored frozen at -15°C for 1 month. Radioactivity was determined in urine, faeces, milk, blood, tissues, cage rinse and expired volatiles by LSC and combustion-LSC. Tissues and faeces were freeze-dried and sequentially (Soxhlet) extracted with hexane, chloroform and acidic MeOH. Solid residues from muscle were acid- and base-hydrolysed (1 M HCl, 1 M NaOH, each for 1 h at 98°C), solid residues from liver and kidney were acid- and base-extracted (0.1 M HCl, 0.001 M NaOH) and digested with pronase E and protease (37°C, overnight) and 6 M HCl. Milk samples were extracted with chloroform/MeOH/water; urine was extracted with ACN/MeOH. Extracts and digests were chromatographed on normal and reverse-phase TLC plates (3 different solvent systems) with detection by LSC. Reference compounds were ethoprophos, mA, mJ, mN, mO, amino-acids (Ala, Arg, Asn, Asp, Cys, Gln, Glu, Gly, His, H-Pro, Ile, Leu, Lys, Met, Phe, Pro, Ser, Thr, Trp, Tyr, Val) and fatty acids (oleic, stearic, myristic, palmitic).

Results. Of the total radioactivity administered 92% and 84% (mean 88%) was recovered from the two goats: 76% in urine, 2.4% in faeces, 2.0% in cage rinse, 0.27% in expired air (24 h period;

extrapolates to 2% for 7 day period), 1.7% in milk, 3.6% in liver, 1.2% in GI tract and contents, 0.27% in the remaining tissues, blood and bile samples.

Residues in milk reached a plateau on the first day of treatment. The following average concentrations were found in edible tissues of the two goats: 0.49 mg/kg eq in milk (average over day 0-7; maximum 0.68 mg/kg eq); 8.8 mg/kg eq in liver; 0.93 mg/kg eq in kidney; 0.095 mg/kg eq in muscle; 0.051 mg/kg eq in fat.

Radioactivity in liver, kidney, muscle, fat and milk was fractionated into extractable, hydrolysable (acid/base) and solid residues (Table 3); the solid residues could be solubilised using protease/acid digestion. A limited set of extracts was characterized. The total extracted radioactivity in liver, kidney, muscle and fat was more than 100%. According to the study author high recoveries in muscle and fat were explained by the low levels of radioactivity.

In tissues almost all the radioactivity in liver and kidney remained in the post-extracted solids. Enzyme/acid digests of these solids co-chromatographed with amino acid standards. According to the study authors the formation of radioactive amino acids occurred via hydrolysis of ethioprophos to ethanol with subsequent conversion to acetaldehyde, acetate, acetyl-coenzyme A, (acetyl-Co A) and amino acids (tricarboxylic acid cycle). The acidic MeOH extract of liver contained three radioactive spots (1.1%, 1.4%, 0.45% of the TRR in liver), the first spot contained mA and/or mJ, but the other two spots did not co-chromatograph with any of the reference compounds used.

In milk 55% was extractable with chloroform. When the radioactivity in the chloroform extract was saponified, the major saponified fraction co-chromatographed with badly-resolved fatty acid standards (palmitic acid, oleic acid and stearic acid). According to the study authors the formation of radioactive fatty acids occurred via hydrolysis of ethioprophos to ethanol with subsequent conversion to acetyl-CoA and fatty acids.

Conclusions. After 7 daily doses of 32 mg ai/kg in the dry feed of dairy goats, the administered radioactivity was mainly excreted in the urine (76% of the TAR). Levels in milk attained a steady state 1 day after the first dose. Radiolabel concentrations were highest in tissues responsible for metabolism and excretion (liver and kidney). The metabolism of ethioprophos was shown to be extensive with most of the radioactivity apparently incorporated into natural products such as fatty acids and amino acids. The parent compound was not found. Primary metabolites tentatively identified were mA and/or mJ.

Table 3. Fractionation of radioactivity in edible tissues of dairy goats, treated for 7 days with 32 mg ai/kg dry feed.

Sample	Mean residue mg/kg eq	extractable		not extractable						Total %
		hexane, chloroform, acidic MeOH		released by acid/base		released by enzyme or 6 M HCl		solids		
		%	mg/kg eq	%	mg/kg eq	%	mg/kg eq	%	mg/kg eq	
Liver	8.8	4.9	0.43	3.6	0.32	103	9.1			111
Kidney	0.93	16	0.15	7.3	0.068	88	0.82			111
Muscle	0.095	24	0.022	132d	0.13	1.6	0.002			158
Fat	0.051	84	0.043	na		73	0.037			157
Milk	0.49	55b 16c	0.27b 0.076c					10a	0.05	81

na: not applicable.

a not treated by enzymes or strong acids.

b chloroform phase.

c MeOH/water phase.

d acid/base hydrolysis.

Poultry: laying hens

Radiolabelled [1-ethyl-¹⁴C]ethoprophos was administered orally once daily for seven consecutive days by capsule to two groups of laying hens (3 and 6 hens; Leghorn; weight 1.3-1.8 kg; 102-104 weeks old) (Bates and Byrd, 1993). The radiochemical purity after repurification was 99.0% with a specific activity after dilution of 0.16 GBq/mmol. The hens were originally dosed with 10 mg ai/kg dry feed (2 mg ai per hen per day). Owing to observed toxicity, the study was terminated and restarted with a lower dose. In the final experiment hens were dosed at an actual (mean) dietary concentration of 2.1 mg ai/kg dry feed. Mean feed intake was 96 g/hen/day. One group of three hens was placed in a plastic gas collection tent with an NaOH trapping solution during the whole study. One control group of three hens received gelatine capsules with 100 mg cellulose. Expired volatiles and excreta were collected once daily. Eggs were collected twice daily and pooled for each 24 h period after dosing. Eggs were separated into whites and yolks. Whole blood was collected before termination and separated into red blood cells and plasma. The hens were killed 16-20 hours after the final dose and liver, kidney, muscle (thigh and breast), fat (mesenteric and peripheral), skin with adhering fat, and GI tract tissue and contents were sampled. Samples were homogenised and stored frozen at -15°C for 1 month. Radioactivity was determined in excreta, eggs, blood, tissues, cage rinse and expired volatiles by LSC and combustion-LSC. Tissues, eggs and excreta from the two groups were pooled, freeze-dried and sequentially (Soxhlet) extracted with hexane, chloroform and acidic MeOH. Solid residues from muscle and egg whites were acid- and base-hydrolysed (1 M HCl, 1 M NaOH, each for 1 h at 98°C), solid residues from liver and kidney were acid and base extracted (0.1 M HCl, 0.001 M NaOH) and digested with pronase E and protease (37°C, overnight) and 6 M HCl. Extracts and digests were chromatographed on normal and reverse-phase TLC plates (3 different solvent systems) with detection by LSC. Reference compounds were ethoprophos, mA, mJ, mN, mO, amino-acids (Ala, Arg, Asn, Asp, Cys, Gln, Glu, Gly, His, H-Pro, Ile, Leu, Lys, Met, Phe, Pro, Ser, Thr, Trp, Tyr, Val) and fatty acids (oleic, stearic, palmitic, myristic).

Results. Of the total radioactivity administered 63% (3 hens in gas tent) and 64% (6 hens) was recovered: 44% in excreta, 0.31% in cage rinse, 3.6% in expired volatiles, 1.0% in egg whites, 9.3% in egg yolks, 2.6% in liver, 3.6% in GI tract tissue and contents, 0.62% in the remaining tissues and blood samples.

A plateau was reached in egg whites on the 3rd day of treatment, but not in egg yolks during the treatment period of 7 days. The following average concentrations were found in the edible tissues of the two groups of hens: 0.021 mg/kg eq in egg whites (average over days 3-7; maximum 0.029 mg/kg eq); 0.30 mg/kg eq in egg yolks (average over days 0-7; maximum 0.64 mg/kg eq); 1.2 mg/kg eq in liver; 0.40 mg/kg eq in kidney; 0.010 mg/kg eq in muscle; 0.076 mg/kg eq in fat; 0.021 mg/kg eq in skin with adhering fat.

Radioactivity in liver, kidney, muscle and fat was fractionated into extractable, hydrolysable (acid/base) and solid residues (Table 4); the solid residues could be solubilised using protease/acid digestion. A limited set of extracts was characterized.

Almost all the radioactivity in liver and kidney again remained in the post-extracted solids, and enzyme/acid digests of these solids co-chromatographed with amino acid standards. The study authors concluded that the radioactive amino acids had been formed as in goats. The organic (acidic MeOH) extract of liver contained three radioactive zones (1.9%, 0.95%, 2.0% of the TRR in liver): the first contained mA and/or mJ, the second did not co-chromatograph with any of the reference compounds used and the third co-chromatographed with mN and/or mO.

In egg yolks 84% of the TRR was extractable in hexane and 11% in chloroform. The hexane fraction was saponified, and the saponified fraction co-chromatographed with badly resolved palmitic, myristic, oleic and stearic acids. The study authors concluded that radioactive fatty acids had been formed as before.

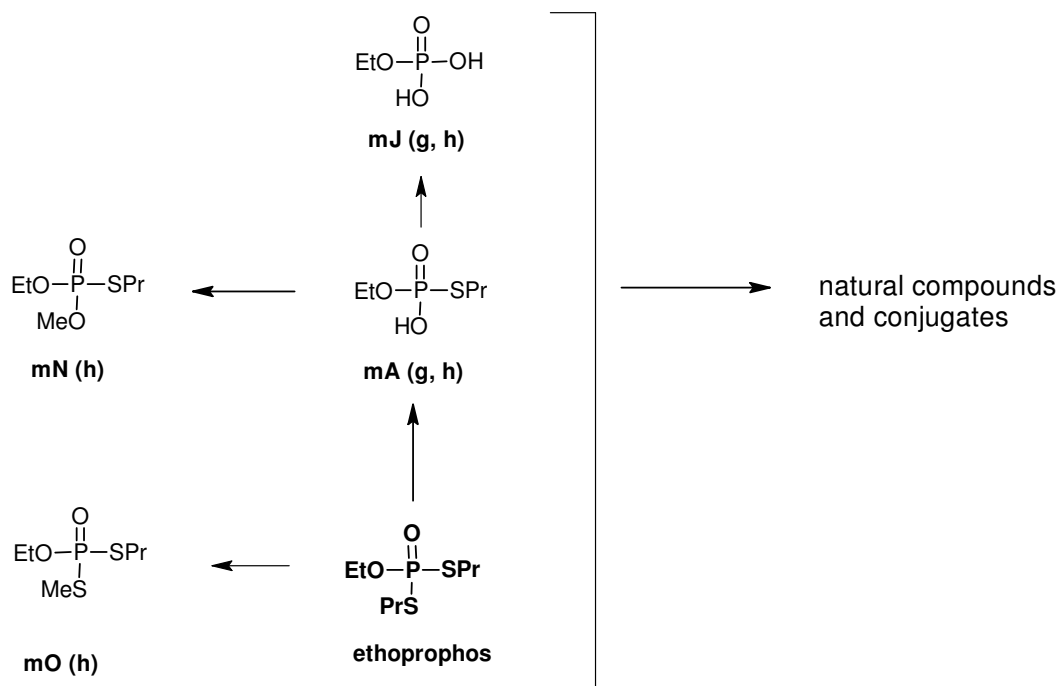
Conclusion. After 7 daily doses of 2.1 mg ai/kg in the dry feed of laying hens, the administered radioactivity was mainly found in the excreta (44% of the TAR). Radiolabel concentrations were again highest in the liver and kidney. The metabolism of ethoprophos was similar to that in goats, and the parent compound was not found. Metabolites tentatively identified were mA and/or mJ and mN and/or mO.

Table 4. Fractionation of radioactivity in edible tissues of laying hens treated with 2.1 mg ai/kg dry feed.

Sample	Mean residue * mg/kg eq	extractable		not extractable				Total %
		hexane, chloroform, acidic MeOH		released by acid/base		released by enzyme or 6 M HCl		
		%	mg/kg eq	%	mg/kg eq	%	mg/kg eq	
Liver	1.2	14	0.16	3.7	0.043	99	1.2	116
Kidney	0.42	33	0.14	6.4	0.027	102	0.43	142
Muscle	0.010	28	0.003	a23 b53	0.008	1.5	0.000	105
Fat	0.069	97	0.067	na		36	0.024	133
Egg white (day 6)	0.016	12		a14 b95				121
Egg yolk (day 6)	0.30	h84 c11						95

* combined samples from the two groups of hens.

na: not applicable. a: acid hydrolysis b: base hydrolysis h: hexane-extractable c: chloroform-extractable



(Me: methyl; Et: ethyl; Pr: propyl, g: goat, h: hen).

Figure 1. Proposed metabolic pathways of ethoprophos in livestock.

The main route in livestock is incorporation into natural compounds. Metabolites mA, mJ, mN and mO were tentatively identified¹ and mP was inferred from end products.

Plant metabolism

The Meeting received information on the fate of ethoprophos after soil treatment before planting of pulses/oilseeds (French beans), cereals (maize), root and tuber vegetables (potatoes) and leafy crops (cabbage). The ethoprophos was ¹⁴C-labelled in the ethyl or propyl group of the molecule.

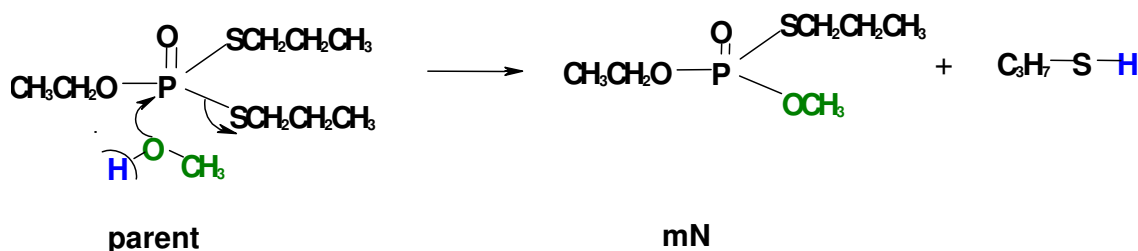
Crop category: pulses/oilseeds

French beans (snap beans/green beans, *Phaseolus vulgaris* L.)

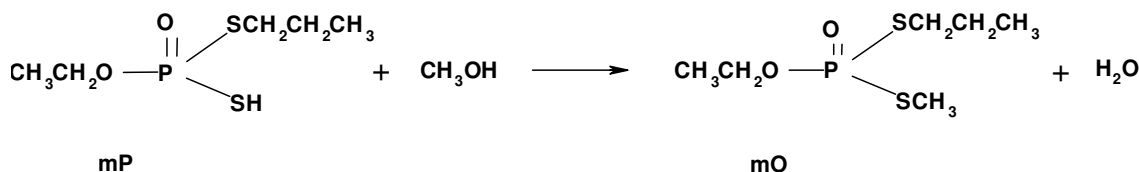
French bean bedding plants (variety Contender) were planted in a greenhouse (Maryland, USA, 1968) in clay pots, filled with steam-sterilized soil treated with [1-¹⁴C-ethyl]ethoprophos or [1-¹⁴C-propyl]ethoprophos (Menzer *et al.*, 1971; non-GLP). The radiochemical purity was not stated; the specific activity (undiluted) was 0.046 GBq/mmol (ethyl label) or 0.10 GBq/mmol (propyl label). Degradation of ethoprophos in steam-sterilized soil was slower than in unsterilized soil because the adsorptive capacity of the soil and the microbial activity was reduced. Hence a maximum amount of ethoprophos was available for uptake by the plants. Ethoprophos was applied as a GR formulation at 14.3 mg ai/kg soil (dosage in kg ai/ha not stated).

¹ Formation of metabolites mN and mO

Trace degradation products mN and mO may be artefacts formed during the extraction with MeOH (Barriere, 2004c). Formation of mN (*O*-ethyl *O*-methyl *S*-propyl phosphorothioate) may be explained by the fact that thiolphosphorous esters can be trans-esterified by alcohols in acidic medium or in the presence of catalysts according to the following reaction scheme:



The degradation product mO (*O*-ethyl *S*-methyl *S*-propyl phosphorodithioate) may be formed by esterification of degradation product mP (*O*-ethyl *S*-propyl phosphorodithioate) with MeOH according to the following reaction scheme:



Although most extractions were done with MeOH without addition of acid, the sample matrix can be acidic itself. Further experiments to confirm this hypothesis are not available.

French bean plants were grown for 63 days and sampled at weekly intervals from day 7 onwards. Storage conditions were not stated. Plant samples were homogenised and successively extracted with MeOH/water (1:1, v/v) and DCM (or vice versa). Extracts were combined and allowed to separate. Soil plus plant roots were homogenised and extracted with MeOH (Soxlet extraction for 16 h). Radioactivity in plant solids and solvent extracts was determined by combustion-LSC and LSC; the soil remaining after extraction was not analysed. Metabolites in the DCM extracts were characterized or identified by silica gel column chromatography with LSC detection, normal-phase TLC (four different solvent systems) with autoradiographic detection, GC-FPD with phosphorus and sulfur detection and IR spectrometry. Metabolites in the MeOH/water extracts were characterized or identified by anion exchange chromatography with identification by co-chromatography with the reference compounds parent, mA, mB, mC, mD, mE, mF, mG, mH, mI, mJ, mK and S,S,S,S-tetrapropyl tetrathio pyrophosphate, and dipropyl sulfide.

Results. The total recovered radioactivity in soil extracts and in plants is shown in Table 5. Residues in soil extracts decreased with time; bound residues in soil were not determined. Total residues in bean plants increased with time: 2.2-13% of TAR (ethyl label) and 0.58-8.3% of the TAR (propyl label) at days 7 to 63. Residue concentrations in mg/kg were not given. Extractable residues predominated at first but unextractable residues exceeded them later, >57% from day 21 onwards.

In the DCM extracts of bean plants the major compounds (Table 6) were the parent (maximum 13%) and mD (maximum 9.2%). The parent decreased with time and was less than 10% from day 28 onwards. Minor amounts of mC, mE(+mH), mF(+mI) were present at some sampling points. The identity of mE/mH and mF/mI could not be established with certainty because these metabolites could not be separated on silica gel or TLC. Because both spots were present in ethyl- and propyl-labelled plants, the spots are either mE+mH and mF+mI or the spots consist of mE and mF alone. Metabolite mK was not found, but according to the study authors is not stable and loses mB to form mL, which was found in the plants (not shown in Table 6). Although the presence of mG was suspected in some plant extracts, amounts were too small for confirmatory analyses.

In the MeOH extracts of soil, the major compound was the parent; in the MeOH/water extracts of bean plants the major compounds were mA and mJ (the data were not reported).

Table 5. Percentage recovery of total applied ^{14}C and distribution of radioactivity in extracts of soil and bean plants.

DAT	Percentage recovery in bean and soil						Distribution of radioactivity in bean plant extracts ³					
	whole bean plant ¹		MeOH extracts of soil		Total recovered ²		MeOH-water		DCM		Solids	
	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr
	% of TAR						% of TRR					
7	2.2	0.58	111	89	113	89	27	31	54	29	19	40
21	4.2	2.2	73	76	77	78	13	23	23	20	64	57
28		5.5	58	60		65	lost	16	lost	9.2	---	75
35	9.0	6.1	62	57	71	63	7.6	8.4	17	11	76	81
42	9.0	5.9	53	61	62	67	2.6	3.2	18	19	79	78
49	11	8.1	45	39	56	47	10	6.7	23	9.8	67	84
56	14	8.3	37	32	51	40	6.8	3.9	13	11	80	85
63	13	8.3	24	26	36	34	12	6.3	15	10	73	84

DAT: days after treatment (bean bedding plants planted on day of treatment).

Et: ethyl label

Pr: propyl label.

¹ calculated by reviewer (sum of % residues recovered in MeOH/water, DCM and plant solids).

² calculated by reviewer (sum of % residues in soil extracts and whole plants (extracts + solids)).

³ calculated by reviewer from % residues recovered in extracts divided by % residues recovered in plants.

Table 6. Characterisation of radioactive compounds in DCM extracts of French bean plants.¹

DAT	parent		mC		mD		mE(+mH)		mF(+mI)		unknown ²		missing ³	
	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr
	% of TRR													
7	8.3	13.4	na	ND	1.4	ND	ND	ND	ND	ND	4.2	3.9	40.4	12.0
21	8.8	12.3	na	ND	3.2	ND	1.3	0.9	ND	ND	5.0	3.1	4.4	3.3
28	lost	4.7	na	ND	lost	0.2	lost	ND	lost	0.5	lost	3.7	-	0.0
35	6.1	5.4	na	ND	2.6	ND	1.2	0.2	0.6	0.7	3.9	3.1	2.3	1.7
42	12.6 ⁴	10.4	na	ND	d	1.1	0.5	ND	4.2	ND	1.1	3.3	0.1	3.8
49	1.0	1.5	na	0.5	6.5	1.6	ND	ND	ND	ND	9.5	4.5	5.8	1.6
56	2.8	3.0	na	0.4	5.6	1.2 ^e	ND	ND	ND	ND	2.4	4.1	2.2	2.0
63	3.1	3.8	na	0.2	9.2	1.2	ND	ND	0.9	ND	1.7	3.8	0.4	1.1

ND: not detected

DAT: days after treatment (bean bedding plants were planted on the day of treatment)

Et: ethyl label

Pr: propyl label

na: not applicable (mC does not contain the specified label)

¹ calculated by reviewer from % of compound in DCM extract times total % recovered in DCM extract (Table 5).

² eluted with MeOH from silica gel column; major compound with ethyl label was mJ (percentage not stated).

³ calculated by reviewer from % recovered in DCM extract minus the sum of parent and metabolites.

⁴ sum of parent and mD.

⁵ sum of mD and mG.

In a related study, bean samples from the same experiment were extracted first by DCM then by MeOH/water (Menzer and Iqbal, 1968). The recovered radioactivity was about the same as described above. In the DCM extracts the parent and metabolites mC, mD and mE were identified together with unidentified compound and one unknown metabolite was found in the MeOH extract. In an addendum details of the metabolite identification were explained (Mobil, 1968a). In a second addendum the unknown compound in the DCM extract was identified as the parent and the unknown metabolite in the MeOH extract was identified as mJ (Mobil, 1968b).

In another related study French bean seedlings were planted in soil treated with 100 mg/kg ethoprophos, most likely unlabelled (Mobil, 1968a). When the plants were 2 weeks old, the shoots were harvested. Samples were homogenised with hexane and the filtrate was fractionated on a silica gel column using gradient elution with hexane/ether. Three ethoprophos related compounds were found in the fractions: 56 mg/kg mD, 72 mg/kg mC and 3000 mg/kg parent.

Crop category: cereals

Maize (corn, *Zea mays* L.), study 1

Maize seeds (variety not stated) were planted in a greenhouse (Menzer *et al.*, 1971; Menzer and Iqbal, 1968, non-GLP). Experimental methods as in the French bean study (see above). Maize plants were grown for 100 days and sampled at 10-day intervals from day 18 onwards.

Results. The total recovered radioactivity in plant and soil extracts is shown in Table 7. Residues in soil extracts were broadly constant; bound residues in soil were not determined. The total recovered residues exceeded 100% for the ethyl label from day 58 onwards. According to the study authors this could be partially explained by the difficulty of obtaining a homogeneous sample for analysis and the large volume of pulp resulting after extraction of the maize plants, whilst the recovery of the propyl label was generally lower because of the probable release of the extremely volatile propyl-labelled mB.

Residues in maize plants increased with time (Table 7): 0.96-74% of TAR (ethyl label) and 0.26-35% of TAR (propyl label) at days 18 to 100. Residue concentrations in mg/kg were not given.

Residues were mainly extracted at the early time-points but mainly unextractable later: >67% from day 28 onwards.

In the DCM extracts of maize plants the major compounds (Table 8) were the parent (maximum 40% of the TRR) and mD (maximum 7.6% of the TRR). The parent decreased with time and was less than 10% from day 48 onwards. Minor amounts of mC, mE(+mH) and mF(+mI) were present at some sampling periods; the unknown residues from the ethyl label were mainly mJ (percentage not given). Metabolite mK was not found, but is apparently not stable and loses mB to form mL, which was found (not shown in Table 8). Although the presence of mG was suspected in some plant extracts, amounts were too small for confirmatory analyses.

In the MeOH extracts of the soil, the major compound was the parent; in the MeOH/water extracts of maize plants the major compounds were mA and mJ (the data were not reported).

Table 7. Percentage recovery of total applied ^{14}C and distribution of radioactivity in extracts of soil and maize plants.

DAT	% recovery in maize plants and soil						Distribution of radioactivity in maize plant extracts ³					
	whole maize plants ¹		MeOH extracts of soil		Total recovered ²		MeOH-water		DCM		solids	
	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr
	% of TAR						% of TRR					
18	0.96	0.26	65	44	66	45	46	31	48	42	6.3	27
28	2.4	1.6	75	55	77	57	7.5	42	38	53	55	5.0
38	8.4	2.0	59	43	67	45	3.5	9.9	11	23	85	67
48	5.6	2.9	90	41	96	44	---	9.0	22	17	78	74
58	11	8.3	109	59	120	67	3.0	2.1	21	5.3	76	93
68	21	10	104	49	125	59	6.3	5.3	12	5.8	82	89
78	41	12	64	46	106	57	2.2	1.5	4.2	3.9	94	95
88	59	29	70	37	129	66	0.8	1.4	2.6	1.4	97	97
100	74	34	49	27	123	62	1.5	1.1	2.1	1.2	96	98

DAT: days after treatment (maize seeds were planted on the day of treatment).

Et: ethyl label

Pr: propyl label.

¹ calculated by the reviewer (sum of % residues recovered in MeOH/water, DCM and plant solids).

² calculated by the reviewer (sum of % residues in soil extracts and whole plants (extracts + solids)).

³ calculated by the reviewer from given % residues recovered in extracts divided by % residues recovered in plants.

Table 8. Characterisation of radioactive compounds in DCM extracts of maize plants.¹

DAT	parent		mC		mD		mE(+mH)		mF(+mI)		unknown ²		missing ³	
	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr	Et	Pr
	% of TRR													
18	39.7	13.4	na	ND	1.4	ND	ND	ND	ND	ND	5.3	28.9	1.1	0
28	27.8	41.2	na	1.0	2.8	0.9	ND	2.4	ND	ND	6.1	7.4	1.1	0
38	3.1	18.9	na	ND	3.8	ND	0.3	ND	ND	ND	3.7	4.4	0.6	0
48	9.6	9.6	na	ND	1.9	0.8 ⁴	0.4	2.8	ND	ND	9.7	3.7	0.4	0
58	4.0	3.3	na	ND	7.6	ND	ND	ND	1.6	ND	7.6	2.0	0	0
68	4.3	3.1	na	ND	2.1	0.2	ND	ND	ND	ND	5.5	2.5	0	0
78	0.9	1.9	na	ND	1.3	0.0	ND	ND	0.8	0.4	1.1	1.6	0	0
88	0.5	0.3	na	0.1	0.9	0.1	ND	ND	0.2	ND	1.0	0.9	0	0
100	1.2	0.3	na	0.1	0.1	0.3 ⁴	ND	ND	0.3	0.1	0.5	0.4	0	0

ND: not detected;

DAT: days after treatment (maize seeds planted on day of treatment).

Et: ethyl label

Pr: propyl label

na: not applicable (mC does not contain specified label).

¹ calculated by reviewer from % of compound in DCM extract times total % recovered in DCM extract.

² eluted with MeOH from silica gel column; major compound in ethyl-label was mJ (percentage not stated).

³ calculated by reviewer from % recovered in DCM extract (from Table 7) minus sum of parent and metabolites.

⁴ sum of mD and mG.

In a related study, maize samples from the same experiment were extracted first by DCM then by MeOH/water (Menzer and Iqbal, 1968). The same compounds were found as in the corresponding study with French beans (see above for details).

Maize, study 2

Silt loam (pH 5.6, 3.7% om, CEC 21 meq/100g, 7% clay particles) was treated with [1-ethyl-¹⁴C]ethoprophos (EC formulation; radiochemical purity 98.6%; specific activity after dilution 0.070 GBq/mmol) at a rate of 13 kg ai/ha in plastic-lined wooden boxes in a field in Kentucky, USA (Johnson, 1991a, GLP). The actual concentration in the soil was 10 mg ai/kg. The application mixture was incorporated to a depth of 10 cm. Sweet corn seeds (variety Early extra sweet) were planted 3 days after soil treatment and sampled at the green forage stage (soil, whole plant), at maturity (shanks, husks, silks, grain, empty cobs) and at the fodder stage (soil, senescent stalks without cobs). Plant and soil samples were stored frozen at – 20°C (storage time not stated). Plant samples were successively extracted with MeOH/water, MeOH and DCM. The remaining solids were extracted first with 0.1 M HCl and then with 0.1 M NaOH. ¹⁴C in plants and solvent extracts was determined by (combustion)-LSC. The LOQ was 0.05 mg/kg eq in maize and 0.01 mg/kg eq in solvent extracts. Metabolites in MeOH/water and DCM extracts were identified or characterized by reverse-phase HPLC (one solvent system; detection by UV (220, 230 nm) or beta radioactivity or LSC of collected fractions). The extracts were also subjected to normal-phase TLC-autoradiography (solvent system I for DCM extracts and solvent system II for MeOH/water extracts). Reference compounds used in TLC and HPLC were the parent, mA, mJ, mN and mO.

Results: The total [¹⁴C]ethoprophos residue in plants and soil and the distribution of the ¹⁴C residue in plants in the various extraction solvents is shown in Table 9. The distribution of metabolites in the MeOH/water and DCM extracts is shown in Table 10.

The total radioactive residue in the samples was very low: in corn forage a TRR of 2.2 mg/kg was detected, in corn cobs 0.27 mg/kg, in grain 0.25 mg/kg, husks 0.79 mg/kg and fodder 1.4 mg/kg. Most of the TRR was solvent-extractable in all samples. Acid or base hydrolysis released a further 6%-14% of the TRR in the forage, grain, cobs and fodder, but in forage 13% and in grain, cobs and fodder more than 40% of the TRR was still bound. During extraction and characterisation 5%-26% of the radioactivity was lost, for which the study author does not give an explanation.

Ethyl phosphate (mJ) was the major metabolite detected in all three plant parts (10%, 35% and 8.9%). The parent ethoprophos and *O*-ethyl *S*-propyl phosphorothioate (mA) were also present in small amounts in forage and fodder. The extracts of forage and fodder also contained small amounts of unidentified components (<6% each). Several of these unidentified components were less polar than ethoprophos.

Conclusions. The proposed metabolic pathways indicate that the primary degradation proceeds from parent ethoprophos to ethyl phosphate through hydrolysis of the two thiopropyl esters. Ethyl phosphate was the predominant metabolite (8.9%-35%). The parent, metabolites mA, mN, mO and several unidentified metabolites were found at minor quantities (<10%).

Table 9. Total-¹⁴C residue in maize and soil and distribution of radioactivity (% of the TRR) in maize extracts.

Sample	DAT	Total ¹⁴ C (mg/kg eq)	Extractable residues, %			Unextractable residues, %			Missing ¹
			MeOH- water	MeOH	DCM	0.1 M HCl	0.1 M NaOH	Solids	
green forage	27	2.2 ²	52	6.2	2.6	3.2	11	13	11%

Sample	DAT	Total ^{14}C (mg/kg eq)	Extractable residues, %			Unextractable residues, %			Missing ¹
			MeOH- water	MeOH	DCM	0.1 M HCl	0.1 M NaOH	Solids	
husks, shanks, silks	69	0.79							
mature grain	69	0.25	26	3.9	9.2	1.8	9.7	44	5.1%
empty cobs	69	0.27	34	12	ND	1.2	7.8	60	none
fodder (stalks)	94	1.4	9.8	14	3.1	1.1	5.1	41	26%
soil	27	4.1 (5.0) ³							
soil	94	2.1 (2.7) ³							

DAT: days after treatment (corn planted at DAT 3)

ND: not detected.

¹ calculated by reviewer from 100% minus sum of extractable and unextractable residues (Table 8).

² study text and Table 2 say 18 mg/kg, figure in the study summary and appendix 2.02 mg/kg.

³ mg/kg dry weight

Table 10. Characterisation of radioactive compounds in MeOH, MeOH/water and DCM extracts of maize (% total recovered radioactivity and mg/kg ethrophosphos equivalents) at DAT 27 (green forage), 69 (mature grain), 94 (fodder).

Sample	parent		mA		mJ		mN (f)		mO (f)		unknown		total	missing (c)
	%	mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%	%
green forage	7.8	0.17	2.3	0.05	10	0.23	0.8	0.02	0.3	0.01	20a	0.42	41	20
mature grain	ND	ND	ND	ND	35	0.09	ND	ND	ND	ND	ND	ND	35	4.6
fodder	0.5	0.01	0.8	0.01	8.9	0.13	1.8	0.03	1.1	0.02d	3.1b	0.06e	16	10

ND: not detected.

a represents at least 9 separate components, each less than 5.5%.

b represents at least 6 separate components, each less than 0.8%.

c calculated by the reviewer:

% total: sum of % metabolites + % parent (Table 9)

% missing: sum of % extractable residues (Table 9) minus % total (Table 10).

d in the study table 0.02 mg/kg; in the study summary 0.01 mg/kg.

e in the study table the summation is 0.06 mg/kg; in the study summary 0.04 mg/kg.

f reference compounds mN and mO could be detected by HPLC but mN could not be detected by TLC solvent system II and retention times for mN and mO in TLC solvent system I were the same. According to the study author confirmation of the identity of extractable components by TLC was marginal owing to sample interferences. Raw TLC data were not reported. Further confirmatory analysis is therefore desirable.

Crop category: root and tuber vegetables

Potatoes, study 1

Silt loam (pH 5.6, 3.7% om, CEC 21 meq/100g, 7% clay particles) was treated with [1-ethyl- ^{14}C]ethrophosphos (EC formulation; radiochemical purity 98.6%; specific activity after dilution 0.070 GBq/mmol) at a rate of 13 kg ai/ha in plastic-lined, wooden boxes in a field in Kentucky, USA (Johnson, 1991b, GLP). The actual concentration in the soil was 15 mg ai/kg. The application mixture was incorporated to a depth of 10 cm. Potatoes (variety Kenebeck) were planted 3 days after soil treatment. Soil and plants were sampled at the “new potato” stage and at maturity. Potato tubers, vines and soil samples were stored frozen at -20°C (storage time not stated). Potato tubers were subdivided into three groups: a) soil removed with dry cloth, b) soil removed by thorough washing; c) tubers washed and peeled, and pulp and peel analysed separately. Tubers from group b and vines were successively extracted with MeOH/water, MeOH and DCM. The remaining potato sample was extracted first with 0.1 M HCl and then with 0.1 M NaOH. ^{14}C in potato and solvent extracts was determined by (combustion) LSC. The LOQ was 0.012 mg/kg eq in potato vines and tubers and 0.01 mg/kg eq in solvent extracts. Metabolites in MeOH/water and DCM extracts were identified or characterized by reversed-phase HPLC (one solvent system; UV at 220, 230 nm or beta radioactivity or LSC of collected fractions). The extracts were also subjected to normal-phase TLC with

autoradiography (solvent system I for DCM extracts and solvent system II for MeOH/water extracts). Reference compounds used in TLC and HPLC were the parent, mA, mJ, mN and mO.

Results: The total ^{14}C residue in potatoes and soil and the distribution in the analytical fractions are shown in Tables 11 and 12. The compounds identified in the MeOH/water and DCM extracts are shown in Table 13.

Total radioactive residues were 0.24-0.54 mg/kg eq in tubers and 1.1-3.8 mg/kg eq in vines. Most of the TRR was extracted with aqueous MeOH. Acid and base hydrolysis solubilized a further 17% of the radioactivity in the vines, while 31% of the TRR and 23% of the TRR remained fibre-bound in vines and tubers respectively.

Analysis of aqueous and organic extracts of the vines and tubers showed that the most abundant radioactive component in both was mJ (ethyl phosphate 12% and 38% respectively). Parent ethoprophos and mA (*O*-ethyl *S*-propyl phosphorothioate) were also present in small amounts in the vines but were not detected in tubers. Several unidentified radiolabelled compounds were detected (<3% each). Most of these were less polar than the parent compound.

Conclusion. The primary degradation proceeds from ethoprophos to ethyl phosphate (mJ) through loss of the two *S*-propyl groups, as in maize. Ethyl phosphate was the predominant metabolite in potato tubers (38%). The parent was not found in the tubers, while unidentified metabolites were found in minor quantities (<10%).

Table 11. Total ^{14}C residue in potatoes and soil (expressed as mg/kg ethoprophos).

DAT	growth stage	Vines	Tubers-A	Tubers-B	Tubers-C (pulp)	Tubers-C (peel)	Soil
62	immature	1.1	0.24	0.26	0.24	0.40	2.4
93	mature	3.8	0.33	0.54	0.35	0.51	2.2

DAT: days after treatment (potato tubers were planted at DAT 3)

Tubers-A: soil removed with dry cloth.

Tubers-B: soil removed by thorough washing.

Tubers-C: washed and peeled, pulp and peel were analysed separately.

Table 12. Total ^{14}C residue (expressed as mg/kg ethoprophos) in potato tubers and vines and distribution of radioactivity (% of the TRR) in extracts of potato tubers and vines.

Sample	DAT	Total ^{14}C mg/kg eq	Extractable residues, %			Unextractable residues, %			Missing ¹
			MeOH-water	MeOH	DCM	0.1 M HCl	0.1 M NaOH	solids	
Vines	62	1.1	29	12	2.6	2.9	14	31	8.5%
Tubers-B	93	0.54	43	12	0.5	0.0	0.0 ²	23	22%

DAT: days after treatment (potato tubers planted at DAT 3).

tubers-B: soil removed by thorough washing.

¹ calculated by reviewer from 100% minus sum of extractable and unextractable residues.

² base hydrolysis produced a thick gel of semi-solubilised starch; this activity was classified as solids.

Table 13. Characterisation of radioactive compounds in MeOH/water and DCM extracts of potato tubers and vines.

Sample	parent		mA		mJ		mN ¹		unknown		total	missing ⁴
	% of TRR	mg/kg	% of TRR	mg/kg	% of TRR	mg/kg	% of TRR	mg/kg	% of TRR	mg/kg	% of TRR	% of TRR
Vines	2.7	0.03	1.5	0.02	12	0.14	1.0	0.01	13 ²	0.14	31	13
Tubers-B	ND	ND	ND	ND	38	0.21	ND	ND	1.2 ³	0.01	39	17

ND: not detected; Tubers-B: soil removed by thorough washing.

¹ mN was only detected by HPLC, not by TLC, so identity uncertain.

² represents at least 9 separate components, each less than 3%.

³ represents at least 3 separate components, each less than 0.6%.

⁴ calculated by reviewer: % total: sum of % metabolites + % parent (Table 13);
 % missing: sum of % extractable residues (Table 12) minus % total (Table 13).

Potatoes, study 2

This supporting study was conducted to determine the nature of the bound residues in potatoes (O'Neal and Johnson, 1995, GLP). Soil was treated with [1-ethyl-¹⁴C]ethoprophos (EC formulation; radiochemical purity 97%; specific activity after dilution 1.84 mCi/mmol) at a dose rate of 13 kg ai/ha in plastic-lined, wooden boxes in a field in Kentucky, USA (1995). The application mixture was incorporated to a depth of 10 cm. The soil was sandy loam (pH 7.0, 2.8% om, CEC 5.8 meq/100g, 8.3% clay particles). The actual concentration in the soil was 5.9 mg ai/kg. Potatoes (minituber variety Kennebec) were planted 3 days after soil treatment and harvested 118 days after treatment (new potato tubers) and 167 days after treatment (mature potatoes). Potato tubers, vines and soil samples were stored frozen at -20°C (storage time not stated). New potato tubers were extracted with MeOH/water, MeOH and DCM. The total radioactive residue was determined by (combustion) LSC. The LOQ was 0.01 mg/kg eq in potato vines, tubers and solvent extracts. A separate extraction was carried out for characterization of the unextractable residues. New potato tubers were subjected to a sequential extraction scheme using 0.05 M phosphate buffer for isolation of extractable residues (DCM had extracted very little residue), beta-amylase for starch digestion (20 h, 30°C, pH 7), pronase E (20 h, 25°C, pH 7.2) for protein digestion, 50 mM EDTA/acetate (pH 4.6, 6 h, 80°C) for pectin extraction, acetic acid/sodium chlorite (1 h, 70°C) for lignin extraction, KOH (24 h, 27°C) plus acetic acid (1 h, ambient) for hemicellulose extraction, sulfuric acid (4 h, ambient) neutralized with KOH to pH 7 for cellulose hydrolysis. The starch isolated from potato tubers by amylase digestion was analysed using reversed and normal-phase radio-HPLC to show whether ¹⁴C-labelled glucose was present.

Results: The total ¹⁴C residue in potatoes is shown in Table 14. The total radioactive residue was 0.51 mg/kg in new potato tubers harvested 118 days after treatment. This result correlates closely with the TRR determined in the first metabolism study with potatoes (0.54 mg/kg at 90 DAT). The extractability of the TRR was the same as reported in the previous metabolism study: 37% of the TRR was extracted with MeOH/water (80/20), 7% of the TRR with MeOH and a further 1% with DCM. The distribution of the bound radioactive residue in the cell wall fractions is given in Table 15. Normal and reversed-phase radio-HPLC showed the presence of [¹⁴C]glucose in the combined phosphate buffer - amylase extract of the starch (47% of the extract).

Conclusion: Supplementary characterisation and fractionation demonstrated that most of the fibre-bound ¹⁴C was incorporated into plant structural components. The fibre-bound radioactive residue associated with starch was identified as [¹⁴C]glucose.

Table 14. Total ¹⁴C residue in potatoes (expressed as mg/kg ethoprophos).

DAT	Growth stage	Vines	Tubers
118	new potato	na	0.51
167	mature potato	2.2	0.97

DAT: days after treatment (potato tubers were planted at DAT 3)

Table 15. Distribution of the fibre-bound residue in potato tubers (as % of the TRR).

TRR mg/kg eq	Phosphate buffer extract	Starch	Protein	Pectin	Lignin	Hemicellulose	Cellulose	Remaining solids	Missing
0.51	41	11	8.5	4.4	3.7	8.2	8.8	6.9	7.8

Crop category: leafy cropsCabbage

Silt loam (pH 5.6, 3.7% om, CEC 21 meq/100g, 7% clay particles) was treated with [1-ethyl-¹⁴C]ethrophophos (EC formulation; radiochemical purity 98.5%; specific activity after dilution 0.067 GBq/mmol) at a rate of 11 kg ai/ha in plastic-lined, wooden boxes in a field in Kentucky, USA (Johnson, 1990, GLP). The actual concentration in the soil was 7.6 mg ai/kg. The application mixture was incorporated to a depth of 7.6 cm. Cabbage bedding plants (variety Stonehead) were planted 2 days after soil treatment. Soil and plants were sampled at the leafy stage and at maturity, and stored at -20°C (storage time not stated). Cabbage was successively extracted with MeOH/water, MeOH and DCM. The remaining solid was extracted first with 0.1 M HCl (30 min) and then with 0.1 M NaOH (30 min). ¹⁴C in cabbage and solvent extracts was determined by combustion-LSC and LSC respectively. The LOQ was 0.05 mg/kg eq in cabbage and 0.01 mg/kg eq in solvent extracts. Metabolites in MeOH/water and DCM extracts were identified or characterized by normal-phase TLC with autoradiography (solvent system I for DCM extracts and solvent system II for aqueous extracts). Radioactive bands were scraped from the plates and further analysed by reverse-phase HPLC (one solvent system; detection by UV at 230 nm) or beta radioactivity or LSC of collected fractions). Reference compounds used in TLC and HPLC were the parent, mA, mN and mO.

Results: The total ¹⁴C residue in cabbage and its distribution in the various extraction solvents are shown in Table 16. The compounds identified in the MeOH/water and DCM extracts are shown in Table 17.

The total radioactive residue in leafy cabbage was 15.6 mg/kg eq and in heads 3.1 mg/kg eq. Most of the TRR was extracted with aqueous MeOH. Part of the fibre-bound residue was solubilized by either acid or base hydrolysis; 11% of the TRR or less remained fibre-bound. Ethyl phosphate (mJ) was the major metabolite found in both leafy and head cabbage (21 and 24% respectively). Ethrophos and mA were also present in small amounts in both. The MeOH/water extracts of immature cabbage also contained significant amounts of three unidentified components (9.3%, 7.6%, 6.4%). These components were present in reduced amounts in mature cabbage heads (4.5%, 0.4%, 1.0%). One of these unidentified components showed relatively polar chromatographic behaviour and accounted for about 9.3% of the TRR in leafy cabbage.

Conclusions: The metabolism in cabbage is essentially the same as in maize and potatoes. The parent, mA, mN and mO and unidentified metabolites were found in minor quantities (<10%).

Table 16. Total-¹⁴C residue in cabbage and soil and distribution of radioactivity in extracts of cabbage (% of the TRR).

Sample	DAT	Total ¹⁴ C mg/kg eq	Extractable residues			Unextractable residues			Missing ¹
			MeOH- water	MeOH	DCM	0.1 M HCl	0.1 M NaOH	solids	
Leaves + heads	33	16	65%	6.8%	3.8%	4.5%	8.6%	11%	1.4%
Mature heads	87	3.1	57%	8.1%	1.2%	3.4%	13%	5.6%	12%
Wrapper leaves	87	8.8							
Soil	33	5.0							
Soil	87	3.3							

DAT: days after treatment (cabbage planted at DAT 2).

¹ calculated by reviewer from 100% minus sum of extractable and unextractable residues.

Table 17. Characterisation of radioactive compounds in MeOH, MeOH/water and DCM extracts of cabbage (expressed as % total recovered radioactivity and mg/kg ethoprophos equivalents) at DAT 33 (immature), 87 (mature).

Sample	parent		mA		mJ		mN ¹		mO ¹		unknown		total	missing ²
	%	mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%	%
Immature leaves + heads	4.0	0.60	2.5	0.5	21	3.3	1.7	0.3 ⁴	0.6	0.09 ⁴	26 ³	4.4	55	20
Mature heads	0.8	0.03	0.3	<0.03	24	0.7	1.7	0.05 ⁴	0.4	0.01 ⁴	9.6 ³	0.4	37	30

ND: not detected.

¹ only detected by HPLC, not confirmed by TLC, so identity is not certain.

² calculated by reviewer % total: sum of % metabolites + % parent;

% missing: sum of % extractable residues (Table 16) minus % total (Table 17).

³ represents at least 6 separate components; 3 components were less than 1.6%; the other components were 9.3%-7.6%-6.4% in immature leaves/heads and 4.5%, 0.4% and 1.0% in mature heads.

⁴ calculated by reviewer from % mN or % mO times 15.6 mg/kg (immature) or 3.1 mg/kg (mature).

Further characterisation of the unextractable and hydrolysable residues is described in an addendum (Wootton and Johnson, 1991). Mature cabbage head from the previous study was successively extracted with DCM and 0.05 M potassium phosphate buffer (pH 7.0) to remove extractable residues. The unextractable residues were subjected to a sequential extraction scheme using beta-amylase for starch digestion (20 h, 30°C), pronase E (16 h, 30°C) for protein digestion, 50 mM EDTA/acetate (pH 4.5, 6hr, 80°C) for pectin extraction, acetic acid/sodium chlorite (4 h, 70°C) for lignin extraction, KOH (24 h, 27°C) plus acetic acid (1hr, ambient) for hemicellulose extraction, sulfuric acid (4 h, ambient) and KOH (pH 6.5-7.5) for cellulose hydrolysis.

Results: ¹⁴C distribution in the various extracts is shown in Table 18. The extractable residue in the phosphate buffer (34%) was lower than in MeOH/water + MeOH (65%, see previous study). Most of the unextractable ¹⁴C in cabbage was incorporated into lignin. According to the study authors, *O*-dealkylation of organophosphate pesticides esters in plants is not uncommon. The subsequent utilisation by a plant of the liberated ethanol as a carbon source is a normal physiological response.

Conclusion: Supplementary characterisation and fractionation demonstrated that most of the fibre-bound residues in cabbage were incorporated into plant structural components, but mainly into lignin.

Table 18. Total ¹⁴C residue in cabbage and distribution of radioactivity in extracts of cabbage head (% of the TRR).

Sample	DAT	¹⁴ C total mg/kg eq	Extractable		Unextractable residues						
			DCM	phosphate buffer	starch	protein	pectin	lignin	hemi- cellulose	cellulose	solids
Mature heads	87	2.7	4.4%	34%	4.9%	5.9%	3.7%	38%	5.5%	8.5%	0.2%

DAT: days after treatment (cabbage planted at DAT 2).

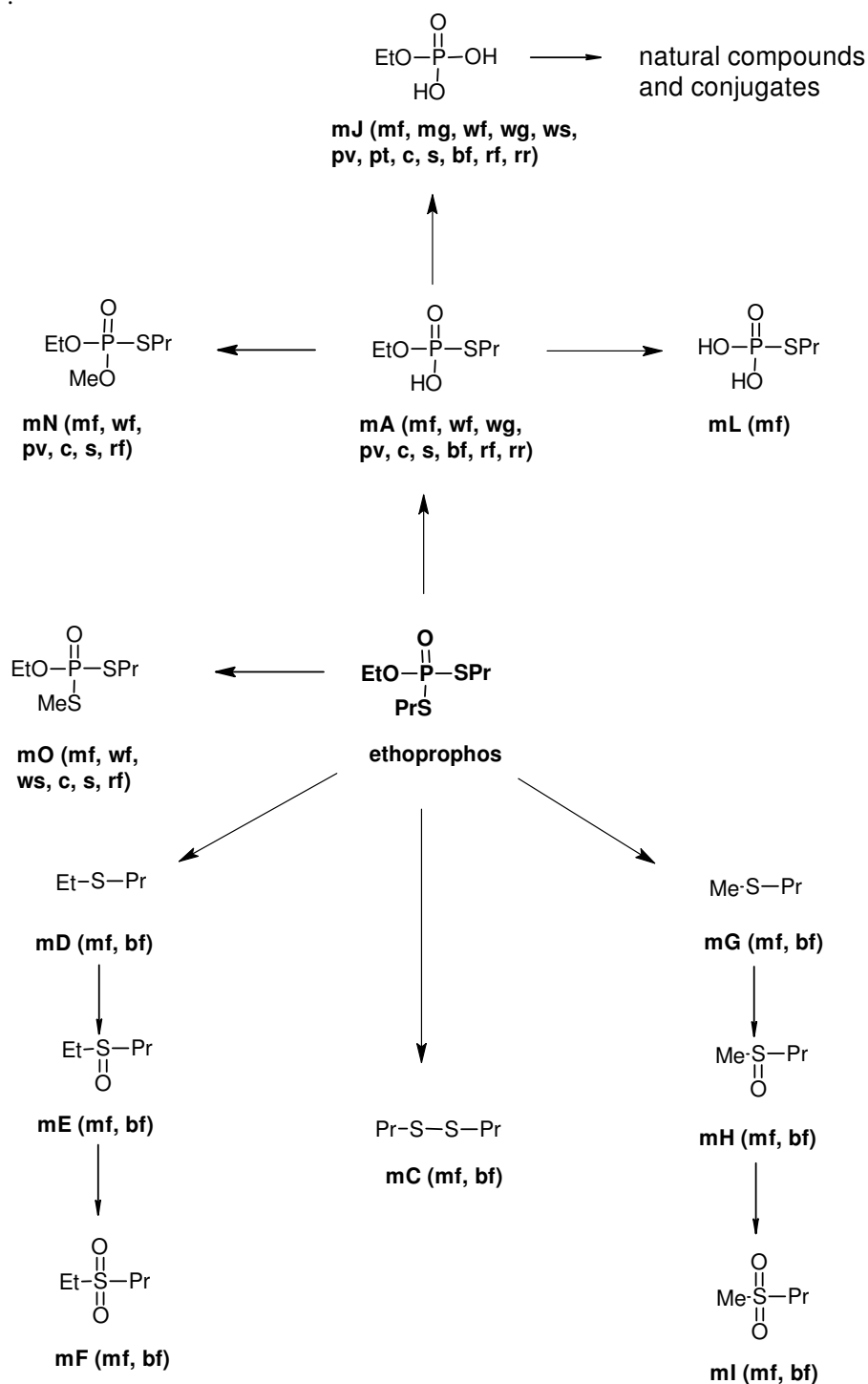


Figure 2 Proposed metabolic pathways of ethrophophos in plants (Me=methyl; Et=ethyl; Pr=propyl, mf=maize forage, mg= maize grain, wf=wheat forage, wg=wheat grain, ws=wheat straw, pv= potato vines, pt= potato tubers, c=cabbage, s=spinach, bf=bean forage, rf=radish forage, rr=radish roots). The major route in plants is ethrophophos – mA - mJ. Metabolites mG, mH, mI, mL, mN and mO were tentatively identified See also footnote to Figure 1.

Environmental fate in soil

The Meeting received information on aerobic degradation in soil and studies on rotational crops (confined and field). Information on anaerobic degradation in soil, photodegradation in or on soil, adsorption-desorption in or on soil, soil leaching, and field dissipation was submitted but was not relevant for this evaluation and was therefore not evaluated.

Degradation in soil under aerobic conditions

Study 1

The route and rate of degradation of propyl-labelled ethoprophos was investigated in a sandy clay loam (SCL) and a sandy loam (SL) under aerobic conditions in the dark at $10 \pm 1.5^\circ\text{C}$ and $22 \pm 2^\circ\text{C}$ (Greenslade *et al.*, 1984, non-GLP). The specific activity (undiluted) was 22.2 mCi/mmol and radiochemical purity 99.6%. The soils were adjusted to 50% of their maximum water holding capacity (62% and 70% field capacity for SCL and SL respectively). The incubation flasks, containing 100 g fresh soil each, were linked in parallel to a common glass manifold and air was drawn through this system to collect $^{14}\text{CO}_2$ in NaOH and other volatile products in a Tenax 15 tube. Based on an application rate of 10.5 kg/ha in the field, the test substance was applied at a nominal concentration of 14 mg ai/kg dw soil. The soils characteristics are summarised in Table 20. Duplicate samples were taken after 0, 1, 3, 7, 14, 27, 41, 60 and 90 days of incubation at 22°C and 0, 1, 3, 7, 14, 27, 41, 70 and 110 days at 10°C . After extraction at ambient temperature with MeOH, diethyl ether, hexane and 0.1N ammonia solution, the MeOH and selected ether extracts was analysed by normal-phase TLC with detection by autoradiography followed by LSC. Reference standards chromatographed with the extracts were the parent, mC, mD, mE and mF. The concentration of ethoprophos was also determined by GC-FPD method 5.

Degradation at 22°C . Overall recoveries of the applied radioactivity at each sampling were similar for both soils at 98%-103% at days 0-3 and 86%-90% at days 14-90. The radiolabelled material applied was almost completely extractable with MeOH, diethyl ether, hexane and ammonia on day 0 (100% and 102% of the TAR for SCL and SL respectively), but only 18% and 14% were extractable on day 90. At day 90, 56% and 60% of the TAR were mineralised as carbon dioxide. Other volatile radioactivity was not detected in the study ($<0.007\%$ of the TAR). The unextractable radioactivity amounted to 11% and 14% of the TAR at day 90 and was associated with humic acid, fulvic acid and humin fractions (0.5%-7.9% of the TAR each).

In MeOH and diethyl ether extracts of both soils, most of the radioactivity was associated with unchanged ethoprophos and accounted to 90%/94% (day 0) and 9.0%/7.2% (day 90), see Table 19. Radioactivity associated with polar products retained at the origin of the TLC plates could not be identified but was only in the range of 0.6%-1.5% / 0.5%-1.1% of the TAR. Trace products (0.1%-0.5% of the TAR) corresponding to R_F values of mE and mF were detected. Some very minor less polar products were found, the sum of which accounted for 0.3%-2.1% and 0.2%-1.2% of the TAR.

Degradation at 10°C . The radiolabelled material applied was almost completely extractable by MeOH on day 0 (93% of the TAR for both SCL and SL), while 20% of the TAR was MeOH-extractable at day 110. Contents in other extracts were not reported. At day 110, 50% and 43% of the TAR were found as ^{14}C -carbon dioxide. Other volatile radioactivity was not monitored. Concentrations of ethoprophos fell from 12 and 13 mg/kg (day 0) to 2.1 and 1.8 mg/kg at day 110 (Table 19). Results as % of the TAR were not reported.

Rate of degradation. The soil moisture content was in line with the level which is generally considered adequate for assessment of degradation rate (40% of the maximum water holding

capacity or 75% of field capacity is equivalent to ca. pF 3 for the soil types under investigation).

Semi-log plots of ethoprophos concentrations versus time demonstrated that the degradation was first order. Calculated DT₅₀ and DT₉₀ values are shown in Table 20.

Conclusion: The main degradation product in soil under aerobic conditions at 10°C and 22°C was ¹⁴CO₂ which accounted for 56%-60% of the TAR (90 days, 22°C) or 43%-50% of the TAR (110 days, 10°C). No other major product could be identified. Most of the radioactivity in the extracts was due to unchanged ethoprophos 90%-94% (day 0, 22°C) and 7.2%-9.0% (day 90, 22°C). The half-life at 22°C was 24-25 days and nearly twice as long at 10°C.

Table 19. Degradation of ethoprophos in soils (% of the TAR and mg/kg dw soil).

Days	sandy clay loam (22°C)		sandy loam (22°C)		sandy clay loam (10°C)		sandy loam (10°C)	
	% of TAR	mg/kg eq ¹	% of TAR	mg/kg eq ¹	% of TAR	mg eq/kg ¹	% of TAR	mg/kg eq ¹
0	90	14	94	14	-	12	-	13
1	90	14	95	15	-	13	-	12
3	82	13	89	14	-	11	-	9.7
7	72	12	77	12	-	11	-	7.3
14	58	8.4	59	9.2	-	8.8	-	8.0
27	38	5.7	40	6.4	-	8.2	-	7.0
41	29	4.3	26	4.1	-	6.7	-	7.0
60	18	2.6	14	2.4	-	-	-	-
70	-	-	-	-	-	4.0	-	4.0
90	9.0	1.2	7.2	1.2	-	-	-	-
110	-	-	-	-	-	2.1	-	1.8

- not sampled or not reported.

¹ determined by GC-NPD method 2.

Table 20. Soil characteristics and degradation times of ethoprophos.

Soil	Temp. (°C)	om (%)	pH (water)	CEC (meq/ 100 g dw soil)	Clay (%)	Moisture pF ¹	DT ₅₀ (days)	DT ₉₀ (days)
Sandy clay loam	22	3.2	5.7	19.6	28	3.0	25	82
Sandy loam	22	6.4	7.0	25.1	16	3.0	24	80
Sandy clay loam	10	3.2	5.7	19.6	28	3.0	43	144
Sandy loam	10	6.4	7.0	25.1	16	3.0	42	139

¹ pF values estimated by reviewer, based on pF-curves of representative soils

Study 2

The route and rate of degradation of ethyl-labelled ethoprophos was investigated in a loamy sand soil under aerobic conditions in the dark at 25°C and 80% of the field capacity (1/3 bar moisture) equivalent to a moisture content of 6.1% (Jordan *et al.*, 1986, non-GLP). The specific activity was 3.32 mCi/mmol; the radiochemical purity was >99%. The incubation flasks, containing 50 g fresh soil each, were linked in parallel to a common glass manifold and air was drawn through to collect to collect volatile metabolites in tubes containing XAD-4 resin and active charcoal and ¹⁴CO₂ in a 0.1 M NaOH trap. The test substance was applied at a concentration of 11.9 mg/kg soil. The soil characteristics are summarised in Table 21. Duplicate samples were taken after 0, 1, 3, 7, 14, 28, 56, 84, 112, 168 and 252 days of incubation. After extraction at ambient temperature with MeOH, diethyl ether and ammonium carbonate solution (2%), the combined MeOH and ether extracts were analysed by normal-phase TLC (four different solvent systems) with detection by autoradiography followed by LSC. Reference standards chromatographed with the extracts were parent, mA, mN and mO. Extracts from day 3 and day 112 were also analysed by GC-FPD. Unextractable residues were refluxed with 2% HCl - MeOH for 2 h and extracted with ethyl acetate for TLC analysis.

Table 21. Characteristics of soil.

Texture	Loamy sand
% Clay	7.6
pH	5.3
% om	1.7
CEC (meq/100 g soil)	5.9
% Field capacity (moisture content at 1/3 bar)	7.72

Results: Overall recoveries of the applied radioactivity at each sampling were in a range of 94% to 101% . The radiolabelled material applied was completely extractable with MeOH, diethyl ether and ammonium carbonate on day 0 (100% of the TAR), while 29% of the TAR were extractable on day 252. On day 252, 54% of the TAR was found as $^{14}\text{CO}_2$, while other volatile components accounted for up to 2.5% of the TAR. The unextractable radioactivity amounted to 10% of the TAR.

In combined MeOH and ether extracts, most of the radioactivity was associated with unchanged ethioprophos and accounted for 97%-99% of the TAR on day 0 and 24%-25% of the TAR on day 252. One major product was identified as mA (max. 3.6%-7.9% of the TAR) in addition to mO (max. 0.7% of the TAR) and mN (max. 0.3% of the TAR). Unknown compounds of medium polarity were detected amounting to 2.2% and 4.8%, the latter only in two selected samples. Radioactive polar products retained at the origin of the TLC plates could not be identified but were only in the range of 0.1-1.0% of the TAR. The presence of the parent, mA, mO, mN and one unknown compound was confirmed by GC-FPD.

In the ethyl acetate extracts of the HCl-MeOH-hydrolysed soil bound residues, the parent was the major compound identified at 2.8%-3.5% of the TAR on day 252.

Owing to the very low soil moisture of 6.1% degradation was delayed so no information on degradation kinetics could be obtained.

Conclusion. The major degradation product in soil under aerobic conditions at 25°C was $^{14}\text{CO}_2$ which accounted for 54% of the TAR after 252 days. Most of the radioactivity in the extracts was from unchanged ethioprophos. The product which accumulated to the greatest extent in the soil was mA.

Study 3

The rate of degradation of unlabelled ethioprophos was investigated in a humic sand (Speyer 2.2), a sandy loam (Speyer 2.3) and a loamy silt soil under aerobic conditions in the dark at 20°C and 40% of the maximum water holding capacity (Fuchsbichler, 1992, **non-GLP**). The incubation flasks, containing 100 g fresh soil each, were closed with a cotton wool plug. The test substance was applied at a concentration of 10 mg/kg soil. The soil characteristics are summarised in Table 23. Samples were taken after 0, 2, 4, 8, 16, 32, 64, 100 and 115 days of incubation and analysed using method 172, version 1991. Results are shown in Table 22. They were not corrected for concurrent recoveries (88%-94%), nor for interferences (<0.01 mg/kg).

Rate of degradation. Half-lives were calculated using Timme's method. The moisture content of the humic sand may have been too high, whereas the loamy silt soil may have been incubated in relatively dry conditions (based on pF curves of representative soils, 40% of maximum water holding capacity is equivalent to pF <2 for sand and pF 4 for loamy silt). The DT_{50} values, however, do not indicate that degradation has been adversely influenced by the conditions, the value of the humic sand is consistent with DT_{50} values found in other studies. Calculated DT_{50} and DT_{90} values are shown in Table 23.

Conclusion: The half-life at ambient temperature was 10-25 days.

Table 22. Degradation of ethoprophos in soils (expressed as mg/kg).

Days	humic sand Speyer 2.2	sandy loam Speyer 2.3	loamy silt
0	8.6	9.4	8.1
2	8.2	6.9	5.6
4	7.9	6.8	6.2
8	8.0	6.1	3.0
16	4.4	4.8	2.4
32	2.0	2.3	0.62
64	0.42	1.1	0.10
100	0.61	0.60	
115	0.27	0.35	

Table 23. Soil characteristics and degradation times of ethoprophos.

Soil	Temp. (°C)	% org. C	pH	CEC (meq/100 g)	% clay ($<2\ \mu\text{m}$)	Moisture pF ¹	DT ₅₀ (days)	DT ₉₀ (days)	Remark
Humic sand 2.2	20	2.3	5.5	9.7	5.1	1.5	23	76	too dry
Sandy loam 2.3	20	1.3	6.5	9.5	8.3	3.0	25	85	-
Loamy silt	20	1.4	6.8	-	17	4.0	10	34	-

¹ pF values estimated by reviewer, based on pF-curves of representative soils

Confined rotational crop study

A sandy loam soil was sprayed with [1-ethyl-¹⁴C]ethoprophos as an EC formulation at a rate equivalent to 13.4 kg ai/ha (Wootton and Johnson, 1992). The specific activity of the test substance after dilution was 1.06 mCi/mmol with a radiochemical purity of 96.3% at application. Soil characteristics were pH 7.0, 0.41% om, CEC 17 meq/100 g; 6% clay, moisture holding capacity at 0.33 bar 32%. The test substance was incorporated into the top 10 cm of soil. The soil was placed outside in boxes inside a screened enclosure (Watsonville, CA, USA, 1989-1991), which was heated and covered with plastic during winter months, and left fallow for 30 to 365 days after treatment. Wheat (Anza), spinach (Polka) and radish (Cherry Belle) were planted at 30, 120 and 365 DAT for each crop. Immature and mature crops were harvested from each planting interval. Soil samples were collected at application, at each planting and at each harvest. The total radioactive residue in crops and soil was determined by combustion LSC (LOQ 0.01-0.13 mg/kg). Crop and soil samples were stored frozen for 1-648 days, then extracted successively with MeOH/water, MeOH and DCM. Remaining solids were hydrolysed sequentially with 0.1 M HCl and 0.1 M NaOH (reflux 30 min each). Extracts were analysed by reversed-phase HPLC with LSC detection (LOQ 0.01-0.11 mg/kg). Retention times were compared with those of known reference standards: the parent, mA, mJ, mN, and mO. HPLC results were confirmed by normal-phase TLC and/or GC-MS.

Crop development was normal for wheat and radish, but spinach planted at DAT 30, 120 and 365 was stunted as a result of the phytotoxicity of ethoprophos.

The total radioactive residue in soil is shown in Table 24 and ranged from 14 mg/kg at application to 7.8 mg/kg at 30 DAT, 1.4 mg/kg at 120 DAT, 0.88 mg/kg at 365 DAT and 0.77 mg/kg at 484 DAT. Dissipation of the TRR in soil was biphasic. An initial rapid decrease occurred in the first 90 days after treatment, so only little radioactivity remained in soil for uptake by rotated crops.

The total radioactive residue in crops is shown in Table 25 and was relatively high in all species in the 30 DAT rotational planting. Crops in the 120 DAT rotation generally showed TRR levels at about 10-25% of the same species at 30 DAT, except wheat straw and immature spinach. At 365 DAT, the TRR in rotational crops was generally an order of magnitude lower than in the 30 DAT crops. In all crops there was less total extractability over time. All crops harvested at an immature stage showed similar extractability, while mature crops showed different patterns of extractability.

Total extractability in mature wheat was generally lower than in mature spinach or mature radish. Part of the remaining solids could be hydrolysed by acid or alkaline treatment, but 1.9%–42% of the TRR remained fibre-bound, with the highest proportion in wheat chaff at 365 DAT.

Table 24. Distribution of radioactivity in extracts of soil.

Rotational Interval (days)	Sampling (DAT)	TRR (mg/kg eq)	MeOH/H ₂ O ¹	DCM	0.1 N HCl	0.1 N NaOH	Solids	Total
			(% of TRR)					
30 (application)	0	13	110	0.1	1.0	0.9	0.2	112
30 (planting)	30	7.8	72	0.1	13	8.5	1.5	95
30	84	1.1	50	0.8	6.7	22	9.5	89
30	132	0.54	14	0.2	11	39	22	86
30	169	0.96	23	0.7	6.6	29	14	73
120 (application)	0	15	33	0.2	5.9	5.2	1.2	46
120 (planting)	120	1.4	38	0.8	5.1	18	17	80
120	182	1.1	30	1.2	3.6	3.4	2.9	41
120	202	0.98	30	2.2	10	30	23	95
120	268	0.92	16	1.9	8.3	32	26	85
365 (application)	0	13	101	0.3	1.5	0.8	0.2	104
365 (planting)	365	0.88	10	2.1	5.1	21	22	61
365	399	0.66	11	1.0	17	28	31	88
365	406	0.63	10	0.6	9.0	24	4.5	49
365	426	0.78	8.6	2.3	6.0	16	13	46
365	428	0.67	8.4	1.5	9.9	49	22	92
365	484	0.77	8.0	0.0	9.3	48	27	93

¹ Combined MeOH and MeOH water extracts

Table 25. Distribution of radioactivity in extracts of rotational crops.

Sample	Planted (DAT)	Harvest (DAT)	TRR (mg/kg)	MeOH/H ₂ O ¹	DCM	0.1 N HCl	0.1 N NaOH	Solids	Total
(% of TRR)									
Imm radish, foliage	30	65	15	72	1.2	1.7	10	4.1	89
Imm spinach, foliage	30	84	23	84	0.3	3.4	2.7	2.3	93
Imm wheat, forage	30	84	28	64	1.7	8.0	18	7.2	99
Mat radish, whole plant	30	84	4.3	85	1.2	3.1	6.4	6.1	102
Mat spinach, foliage	30	132	19	60	2.7	9.8	17	10	99
Mat wheat, straw	30	169	47	52	0.9	2.6	17	16	88
Mat wheat, grain	30	169	14	20	2.1	33	17	6.5	78
Mat wheat, chaff	30	169	41	48	1.6	7.5	22	20	99
Imm radish, foliage	120	182	1.6	59	1.7	13	9.4	6.0	89
Imm spinach, foliage	120	202	10	90	1.2	7.7	10	1.9	111
Imm wheat, forage	120	182	5.0	53	3.7	9.6	14	8.6	88
Mat radish, foliage	120	202	3.0	65	1.3	5.7	7.8	7.3	87
Mat radish, root	120	202	1.3	72	0.6	7.3	57	5.1	142
Mat spinach, foliage	120	268	3.0	70	1.6	3.3	8.3	9.6	93
Mat wheat, straw	120	268	38	56	2.2	2.8	1.5	19	82
Mat wheat, grain	120	268	5.0	26	2.1	0.7	2.2	25	56
Mat wheat, chaff	120	268	17	46	1.1	2.6	3.6	11	64
Imm radish, foliage	365	399	0.61	68	6.9	4.7	8.0	24	111
Imm spinach, foliage	365	426	0.87	68	3.1	6.1	9.8	6.2	93
Imm wheat, forage	365	406	0.61	60	2.6	3.2	22	11	99
Mat radish, foliage	365	406	1.2	36	1.5	2.7	11	1.5	53
Mat radish, root	365	406	0.19	41	5.3	7.8	24	11	89
Mat spinach, foliage	365	428	0.92	61	0.6	5.2	11	17	95
Mat wheat, straw	365	484	0.65	58	3.9	5.1	11	35	113
Mat wheat, grain	365	484	0.29	24	5.3	9.3	21	29	88
Mat wheat, chaff	365	484	0.50	54	2.9	13	14	42	125

imm: immature; mat: mature

¹ Combined MeOH and MeOH water extracts

Parent ethoprophos was the major compound identified in extracts of soil at all sampling times, except at 426 DAT where mJ was predominant (Table 26). Ethyl phosphate (mJ) was the primary product (0.14 mg/kg at 0 DAT) and remained relatively constant at 0.01 to 0.06 mg/kg throughout the study. The concentration of mA was very low in soil and was confirmed at only two samplings, 30 and 84 DAT.

Parent ethoprophos was present in extracts of immature and early maturing crops (radishes) at the 120 day as well as the 30 day rotational intervals (Table 27). No ethoprophos was found in the mature wheat or spinach planted at 120 days nor in any crops at 365 days. The major component of every crop sample was mJ, but mA was also found. A large number of unidentified compounds was found, some with levels above 10% of the TRR or 0.05 mg/kg eq. Hydrolysis of immature spinach extracts from the 120 day rotational interval (0.2 M HCl, 30 min) demonstrated that unknown compound 2 (12% of the TRR) and unknown compound 4 (8.5% of the TRR) were conjugates of ethyl phosphate (mJ). Isolated levels of unknown compounds 1, 3, and 5-18 were not sufficient to establish identity.

The major component in acid and base hydrolysates of crops and soil was the parent: 0.01 mg/kg in soil at DAT 132 and 0.13 mg/kg in mature wheat straw from the 120 day rotational interval. Most of the remainder was mN (0.02 mg/kg eq).

Mature wheat straw was selected for characterisation of bound residues. Plant material was sequentially treated to isolate extractable residues (50 mM phosphate buffer, pH 7, 10 min, ambient), starch (α -amylase, pH 7, 20 h, 30°C), protein (pronase E, pH 7.2, 16 h, 30°C), pectin (50 mM EDTA buffer, pH 4.5, 6 h, 80°C), lignin (glacial acetic acid/sodium chlorite, 4 h, 70°C), hemicellulose (24% potassium hydroxide, 24 h, 27°C), and cellulose (72% sulfuric acid, 4 h, ambient). Results showed general incorporation into cellular components: extractable 40% of the TRR, starch 7.7% of the TRR, protein 1.5% of the TRR, pectin 1.9% of the TRR, lignin 11% of the TRR, hemicellulose 14% of the TRR, cellulose 10% of the TRR and insoluble residue 22% of the TRR (overall recovery 105%).

Storage stability: Mature spiked radish showed no degradation of [^{14}C]ethoprophos after 648 days of storage, but results for other samples were not reported and degradation patterns from beginning and end of the study were not compared.

Conclusions: The total radioactive residue in soil decreased from 14 mg/kg to approximately 1 mg/kg in the first 90 days after treatment, so only little radioactivity remained in the soil for uptake by rotated crops. However, radioactive residues were found in all crop samples at all samplings, even though ethoprophos was not detected in plants after the 120-day rotational interval. The main product found in both soil and plants was ethyl phosphate (mJ).

The metabolism of ethoprophos in rotational crops appears to be, as before, by loss of an S-propyl group to give mA, then loss of the second propyl group to give mJ. The ethyl group can then be incorporated into plant cellular components.

Table 26. Metabolites in combined extracts¹ of soil.

Rotational interval (days)	DAT ¹	TRR (mg/kg eq)	parent (% of TRR)	mJ (% of TRR)	mA (% of TRR)	mO (% of TRR)	mN (% of TRR)	Other ² (% of TRR)
30 (application)	0	13	96	1.1	nd	nd	nd	13
30 (planting)	30	7.8	40	nd	32	nd	nd	0.6
30	84	1.1	45	2.0	0.9	nd	nd	2.9
30	132	0.54	12	2.8	nd	nd	nd	nd
30	169	0.96	21	2.3	nd	nd	nd	0.3
120 (application)	0	15	16	17	0.1	0.1	0.1	0.4
120 (planting)	120	1.4	38	1.0	nd	0.3	nd	nd

Rotational interval (days)	DAT ¹	TRR (mg/kg eq)	parent (% of TRR)	mJ (% of TRR)	mA (% of TRR)	mO (% of TRR)	mN (% of TRR)	Other ² (% of TRR)
120	182	1.1	28	1.8	nd	1.2	nd	nd
120	202	0.98	29	3.0	nd	0.5	nd	nd
120	268	0.92	14	4.3	nd	nd	nd	nd
365 (application)	0	13	88	12	0.1	0.5	0.1	0.5
365 (planting)	365	0.88	7.4	4.3	nd	0.8	nd	nd
365	399	0.66	8.4	3.8	nd	nd	nd	nd
365	426	0.78	1.8	7.3	nd	nd	nd	1.8

nd: <LOQ (0.001-0.005 mg/kg)

¹ combined MeOH/water, MeOH and DCM extracts.

² extractable radioactivity found in endogenous materials and /or minor unidentified products, 1-4 different compounds, each 0.1%-13% of the TRR or 0.01-1.7 mg/kg eq.

Table 27. Metabolites in combined extracts¹ of rotational crops.

Sample	Planted (DAT)	Harvest (DAT)	TRR (mg/kg eq)	parent (% of TRR)	mJ (% of TRR)	mA (% of TRR)	mO (% of TRR)	mN (% of TRR)	Other ² (% of TRR)
Immature radish, foliage	30	65	15	18	22	14	nd	nd	19
Immature spinach, foliage	30	84	23	1.9	51	4.1	0.2	nd	27
Immature wheat, forage	30	84	28	6.5	31	nd	0.3	0.8	27
Mature radish, whole plant	30	84	4.3	7.6	24	21	0.2	0.3	32
Mature spinach, foliage	30	132	19	0.4	28	nd	nd	1.8	33
Mature wheat, straw	30	169	47	1.3	23	nd	0.6	nd	27
Mature wheat, grain	30	169	14	nd	21	nd	nd	nd	1.2
Mature wheat, chaff	30	169	41	0.4	36	nd	0.2	0.4	12
Immature radish, foliage	120	182	1.6	7.8	21	9.8	nd	nd	22
Immature spinach, foliage	120	202	10	5.2	34	nd	nd	2.5	50
Immature wheat, forage	120	182	5.0	4.9	33	10	nd	nd	8.6
Mature radish, foliage	120	202	3.0	3.7	24	18	nd	nd	21
Mature radish, root	120	202	1.3	5.1	29	nd	nd	nd	39
Mature spinach, foliage	120	268	3.0	nd	21	nd	nd	nd	50
Mature wheat, straw	120	268	38	nd	42	nd	0.4	nd	15
Mature wheat, grain	120	268	5.0	nd	25	0.7	nd	nd	2.5
Mature wheat, chaff	120	268	17	nd	40	nd	nd	nd	7.0
Immature radish, foliage	365	399	0.61	nd	6.6	nd	nd	nd	68
Immature spinach, foliage	365	426	0.87	nd	46	nd	nd	nd	23
Immature wheat, forage	365	406	0.61	nd	40	5.4	nd	nd	18
Mature radish, foliage	365	406	1.2	nd	18	6.2	0.8	1.3	12
Mature radish, root	365	406	0.19	nd	31	nd	nd	nd	14
Mature spinach, foliage	365	428	0.92	nd	42	nd	nd	nd	19
Mature wheat, straw	365	484	0.65	nd	31	nd	nd	nd	31
Mature wheat, grain	365	484	0.29	nd	18	nd	nd	nd	4.6
Mature wheat, chaff	365	484	0.50	nd	39	nd	nd	nd	18

nd: <LOQ (0.01-0.11 mg/kg, depending on sample)

¹ combined MeOH/water, MeOH and DCM extracts.

² extractable radioactivity found in endogenous materials and /or minor unidentified products, 2-8 different compounds per sample, each 0.1%-24% of the TRR or 0.05-2.8 mg/kg eq.

Field rotational crop study

Unlabelled ethrophosphos was applied once as EC formulation before planting at an actual rate of 13.5 kg ai/ha in a volume of 224-279 l/ha, using a tractor-mounted broadcast sprayer (R008901, Norris,

1997). The formulation was incorporated into a sandy loam soil (pH 6.8-8.3; 1.5-2.0% om; 5-9% clay; CEC 8.7-9.7 meq/100g; 10%-16% moisture at 0.33 bar). Rotational crops included root vegetables (radish roots), leafy crops (radish leaves, red leaf lettuce, collards), cereals (winter wheat, spring wheat and sorghum), pulses/oilseeds (cow peas, wando peas, green peas, soya beans and mustard forage). Crops were planted in 1995-1996 at two US sites, California (CA) and North Carolina (NC), at 1, 4, 8 and 12 months after application. Rainfall was less than normal in California and greater than normal in North Carolina. In North Carolina, the cold and wet weather delayed the planting at the 8 months rotational interval to 10 months and mature wando peas were lost owing to the very cold winter. The crops were harvested at maturity or the specified growth stage. Spring wheat did not mature sufficiently for grain and straw to be sampled. Samples were stored at -10°C for 90-412 days until extraction. Extracts were stored for 0-11 days and analysed for ethoprophos and mA, using GC-FPD method 4, version 13. Samples containing the highest residues were analysed by GC-MS to confirm the presence of ethoprophos and mA. Results were not corrected for concurrent recoveries (63%-112% for the parent and 58%-99% for mA), nor for interferences (<0.01 mg/kg, except ethoprophos in radish roots 0.013 mg/kg).

Results: Residues of ethoprophos and mA in the rotational crop samples were below the LOQ of 0.01 mg/kg in all treated samples from both test sites except radish root and radish leaves (tops). Table 28 shows the average residues of ethoprophos and mA found in the radish samples. Because of matrix interferences in radish roots, the LOQ for ethoprophos should be increased to at least $0.013 \times 10/3 = 0.05$ mg/kg in radish roots. The presence of ethoprophos and mA in radish root and top was confirmed by GC-MS, but the levels measured by GC-MS were at least an order of magnitude lower than measured by GC-FPD. The crops were not washed before analysis so treated soil could adhere to the radishes. This soil may account for the residues found in the root crop.

Table 28. Average residues¹ of ethoprophos and mA in radish grown on ethoprophos treated soil in the USA.

Sample	Planting (DAT; date)	Harvest (DAT)	California		Planting (DAT; date)	Harvest (DAT)	North Carolina	
			Parent	mA			Parent	mA
Radish, root	31 17 July 95	32	0.018; 0.028; mean 0.023	0.030; 0.048; mean 0.039	31 23 June 95	34	0.015; 0.016; mean 0.016	<0.01 (2); mean 0.039
	119 13 Oct 95	35	<0.01; 0.011; mean 0.010	<0.01; 0.012; mean 0.011	120 20 Sept 95	57	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01
	241 12 Feb 96	50	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01	296 ² 14 Mar 96	60	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01
	362 12 June 96	89	NA	NA	365 22 May 96	40	0.012; 0.014; mean 0.013	<0.01 (2); mean <0.01
Radish top	31 17 July 95	32	< 0.01 (2); mean <0.01	0.066; 0.13; mean 0.096	31 23 June 95	34	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01
	119 13 Oct 95	35	< 0.01 (2); mean <0.01	0.022; 0.029; mean 0.026	120 20 Sept 95	57	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01
	241 12 Feb 96	50	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01	296 ² 14 Mar 96	60	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01
	362 12 June 96	89	NA	NA	365 22 May 96	40	<0.01 (2); mean <0.01	<0.01 (2); mean <0.01

NA: not analysed

¹ residues were from duplicate field samples.

² Owing to unusually wet weather in North Carolina, crops had to be planted 10 instead of 8 months after treatment.

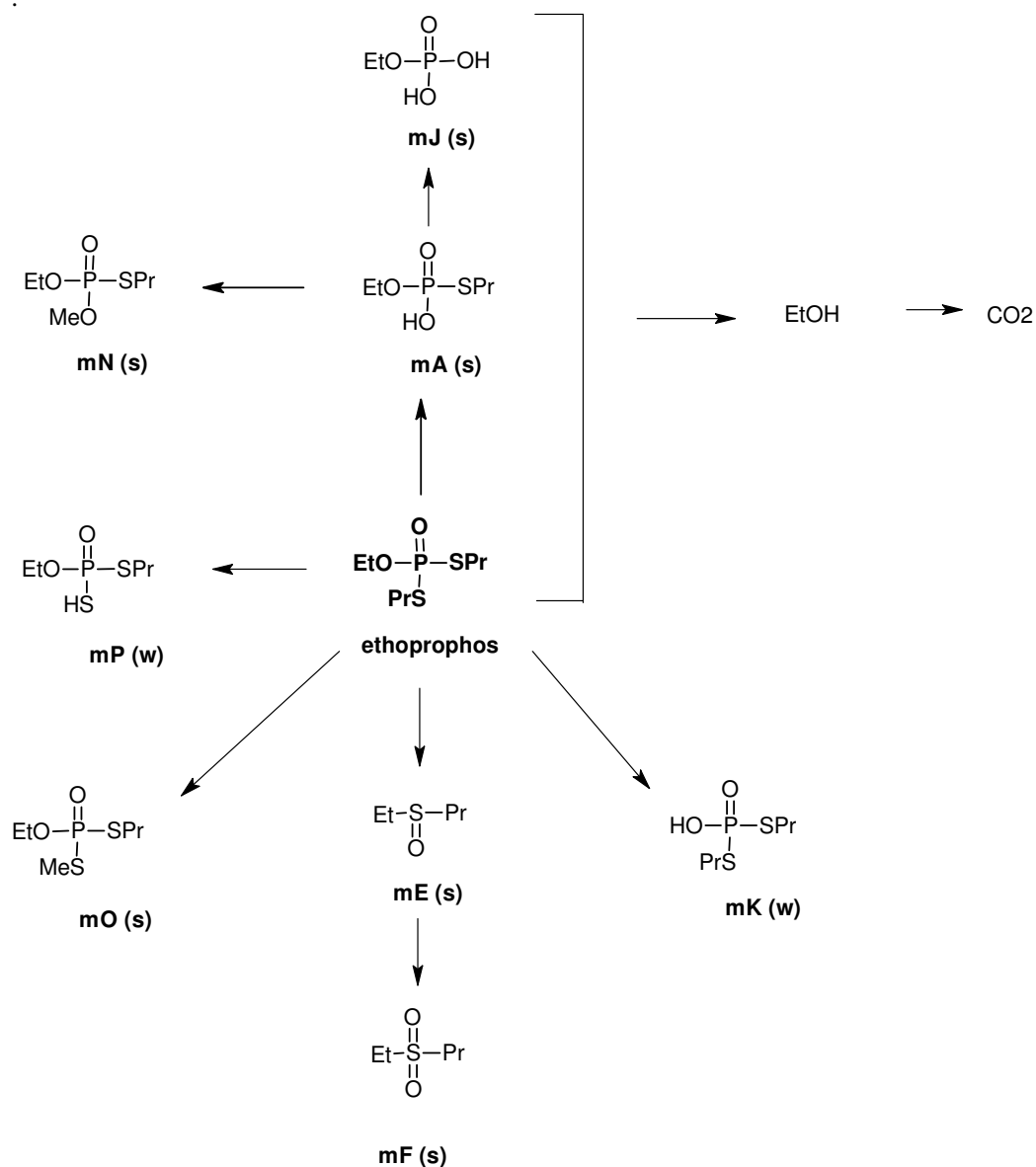


Figure 3. Proposed degradation pathways of ethoprophos in soil and water (Me = methyl; Et=ethyl; Pr=propyl, s=soil, w=water). The major route in soil is degradation to CO₂. Metabolites (mN and mO) were tentatively identified. See also footnote to Figure 1.

Environmental fate in water/sediment systems

The Meeting received information on hydrolysis and photolysis in water. These studies are summarized in physical and chemical properties. Information on biodegradability in water/sediment systems was submitted but was not relevant for this evaluation and was therefore not evaluated.

RESIDUE ANALYSIS

Enforcement methods for foodstuffs of plant and animal origin

The Meeting received information on enforcement and monitoring methods for the determination of parent ethoprophos in foodstuffs of plant and animal origin.

Dutch multi-residue method MRM-1

Ethoprophos was included in the list of 448 organochlorine, organophosphorus, pyrethroid and nitrogen-containing compounds that can be analysed using the Dutch multi-residue method MRM-1 (RIVM, 1996). This employs various extraction and clean-up modules and is suitable for foodstuffs of plant and animal origin. Quantification of ethoprophos was validated for non-fatty samples (<5% fat), which were extracted with ethyl acetate or acetone. Ethoprophos can be quantified by GC-NPD or GC-MS. For lettuce, the recovery was 110% (RSD 9.7%, n=10) at 0.02 mg/kg and 102% (RSD 3.6%, n=10) at 0.12 mg/kg, using GC-MS (ion trap) as quantification technique. The LOQ for organophosphorus compounds is 0.01-0.05 mg/kg depending on the modules used.

German multi-residue method DFG-S8

In addition, ethoprophos was also included in the list of 121 organohalogen, organophosphorus or triazine compounds that can be analysed using the German multi-residue method DFG-S8 (DFG, 1987). The method is suitable for fruits, vegetables, herbs and honey. Samples were macerated with acetone followed by clean-up. Ethoprophos could be quantified by GC-ECD or GC-AFID. The reported LOQ for ethoprophos was 0.02 mg/kg. Recoveries above 70% are reported for ethoprophos in lettuce at 0.2 mg/kg and in green beans and mushrooms at 0.1 mg/kg.

German, multi-residue method DFG-S19

In the original version of DFG-S19 ethoprophos was not listed (DFG, 1987b). In the updated version of DFG-S19 ethoprophos was included (DFG, 1992). The method is suitable for foodstuffs of plant and animal origin. Samples were macerated with acetone/water followed by clean-up. Ethoprophos was quantified by GC-FPD. No validation results are published.

In addition, ethoprophos was also included in the list of organochlorine, organophosphorus, nitrogen containing and other pesticides that can be analysed using the extended revision of the German multi-residue method DFG-S19 (DFG, 1999). The method consists of different modules for extraction (E1-E9), clean-up (GPC, C1, C2) and detection (D1-D4). The method is suitable for foodstuffs of plant and animal origin. Ethoprophos quantification was validated for crops with high water content (water >70%, fat <2.5%, w/w), crops with low water content (water <70%, fat <2.5%, w/w) and °C with high fat content (fat >2.5%, ww).

For crops with high water content or °C with high fat content samples are extracted with acetone/water (2+1) followed by clean-up (E1, GPC). Ethoprophos is quantified by GC-FPD (D2) or GC-MS (D4). The mean recovery for unspecified °C and detection techniques was 80% (RSD 19%, n=20) at 0.05-0.30 mg/kg (2 laboratories).

For °C with low water content (samples were first soaked in water). The mean recovery for unspecified °C and unspecified detection techniques was 79% (RSD 12%, n=5) at 0.20 mg/kg (1 laboratory).

The extended revision of DFG-S19 (with slight modification) was independently validated for potatoes and tomatoes by another laboratory (Table 29) and was renamed method AR 274-01 (Dorn,

2001). Crops (50 g) were extracted and purified according to extraction module E1 and GPC clean-up. Further procedures were modified. The GPC eluate was fractionated on silica gel. Fractions containing ethoprophos were concentrated, diluted with toluene and analysed by GC-PFPD (Chrompack Sil-8 capillary column) using external standardisation. A confirmatory method using GC-MS (same column) was provided. The fragment ion m/z 158 was used for quantification and the fragment ions m/z 199 and m/z 139 for identification.

Table 29. Validation results for the determination of ethoprophos using GC-PFPD method AR 274-01.

Sample	LOQ reported mg/kg	Spike mg/kg	Confirm. ratio PFPD/MS	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
potato	0.01	0.01 0.10	0.87 0.90	87 77-97 84 73-91	10% 10%	5 5	<0.3LOQ (n=3)	6 single points; 0.01-0.75 ug/L, in toluene; $r^2 > 0.99$	Dorn, 2001 (ILV)
tomato	0.01	0.01 0.1	0.87 0.80	85 77-90 84 70-102	6% 16%	5 5	<0.3LOQ (n=3)	6 single points; 0.01-0.75 ug/L, in toluene; $r^2 > 0.99$	idem

USA, FDA multi-residue protocols PAM 1

Ethoprophos was tested through the USA FDA multiresidue protocols A, B, C, D and/or E as described in Pesticide Analytical Manual Volume 1 (PAM 1, 1989) (Ver Hey, 1991). Because all of the protocols were unsuccessful, Ethoprophos could not be determined by the USA FDA multi-residue protocols.

Method AR 271-01

Method AR 271-01 was proposed as an enforcement method for the determination of ethoprophos in milk, eggs, meat, fat and liver (Barbier, 2001).

Samples (25 g) were macerated with MeOH and filtered. For fatty °C such as fat and liver, lipids were removed by low-temperature precipitation (1.5 h at -20°C) followed by filtration. After concentration and addition of 10% NaCl, residues were partitioned into hexane. The hexane-phase was filtered through sodium sulfate and concentrated, before clean-up using a Florisil cartridge. The eluate was evaporated to dryness and redissolved in hexane. For fat this step was not necessary. Ethoprophos was quantified by GC-PFPD (semi-capillary column Rtx-1701 for milk, egg and beef meat, CP SIL 24 CB for fat and liver) using external standardisation. For milk, egg and beef meat GC-MS-MS (capillary CP Sil 8 CB, EI, isolation ion $m/z=158$, quantification m/z 94+114+30), and for fat and liver GC-PFPD with a different polarity column (VA-5) were used as confirmatory methods.

The method was independently validated by a different laboratory (Class, 2001). The following modifications were introduced: the CP SIL 24 CB column was used for all °C tested (milk, beef meat and fat) and the calibration was performed by external standardization using matrix-matched standards for beef meat and liver. The results are shown in Table 30.

Conclusion: The method is considered valid as an enforcement method for the determination of ethoprophos in the range 0.01-0.1 mg/kg in foodstuffs of animal origin (milk, eggs, meat, fat and edible offal).

Table 30. Validation results for the determination of ethoprophos using GC-MS method AR 271-01.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
whole milk	0.01	0.01 0.10	87 71-112 89 81-101	19% 9%	5 5	<0.3LOQ (2)	6 single points; 0.02-0.1 mg/l; in hexane $r^2 > 0.99$	Barbier, 2001 (method validation)
whole egg	0.01	0.01 0.10	96 79-111 95 75-105	13% 13%	5 5	<0.3LOQ (2)	idem	idem
beef meat	0.01	0.01 0.10	84 66-107 88 68-110	19% 18%	5 5	<0.3LOQ (2)	idem	idem
pork fat	0.01	0.01 0.10	96 79-111 110 93-119	15% 10%	5 5	<0.3LOQ (2)	idem	idem
beef liver	0.01	0.01 0.10	84 81-87 79 70-92	3% 11%	5 5	<0.3LOQ (2)	idem	idem
milk	0.01	0.01 0.10	82 70-90 84 70-91	9% 10%	5 5	<0.3LOQ (2)	8 single points; 0.005-1.0 ug/L; in hexane; $r^2 > 0.99$	Class, 2001 (ILV)
beef meat	0.01	0.01 0.10	96 75-110 96 89-102	16% 6%	5 5	<0.3LOQ (2)	5 single points; 0.01-1.0 ug/L; matrix matched; $r^2 > 0.99$	idem
liver	0.01	0.01 0.10	77 50-96 82 73-93	26% 9%	5 5	<0.3LOQ (2)	5 single points; 0.01-1.0 ug/L; matrix matched; $r^2 > 0.99$	idem

Analytical methods for plant materials used in trials and studies

The Meeting received information on analytical methods for the determination of ethoprophos and mA in foodstuffs of plant origin as used in various studies (rotational crop, supervised residue trials, storage stability, processing and monitoring studies).

Method R-89-A (1966-1974)

Method R-89-A was used in trials on sugar cane (C032664), potato (R007982/C034085), cucumbers (C034085) and bananas (C034087). The method is based on extraction with hexane, clean-up on a Florisil column with MeOH as final eluent and determination of the parent by GC-MC. Table 31 shows the validation results.

Table 31. Validation results for the determination of ethoprophos using GC-MC method R-89-A.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
Sugar cane stalks	0.02	0.02 0.03	94 85-100 98 90-106	8% -	4 2	<0.3LOQ (3)	not verified	C032664 (trial/processing) 1967 USA Anal. 1966-1970
Banana, whole	0.02	0.02 0.03	95 90-100 96 -	4.3% -	4 1	<0.3LOQ (3)	not verified	C034087 (trial) 1968-1969 Costa Rica; Côte d'Ivoire Anal. 1969
Cucumber, whole	0.02	0.02	98 90-100	5.1%	4	<0.02 (4)	not verified	C034085 (trial) 1969-1973 USA Anal 1969-73
Potato, whole	0.02	0.02 0.03	97 95-100 86 -	2.7% -	3 1	<0.02 (3)	not verified	R007982 (trial) 1969-1973 USA Anal 1969-74

GC-FPD, method 1 (1976-1984)

GC-FPD, method 1 is published (Hunt *et al.*, 1981), and was used in US trials on cucumbers (C032715), tomatoes (C032715) and potatoes (C033867). The method is based on extraction with hexane, clean-up on a silicic acid column using 10% acetone in n-hexane as final eluent and determination by GC-FPD (glass column, 15% Carbowax 20M, temperature 190°C). The method was validated for snap beans, tomato, cucumber, lettuce, onion, turnip root, turnip leaf and radish. The results for root and fruiting vegetables are shown in Table 32.

Table 32. Validation results for the determination of ethoprophos using GC-FPD method 1.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
tomato	0.005	0.01 0.1	89 88-90 82 -	- -	2 1	-	not verified	C032715 (trial/ method validation) 1976 USA
cucumber	0.005	0.01 0.1	89 78-100 100 -	- -	2 1	-	not verified	C032715 (trial/ method validation) 1976 USA
turnip root	0.01	0.01 0.1	82 82-82 96 86-105	- ns	2 3	-	not verified	C032715 (method validation)
radish	0.005	0.01 0.1	108 - 105 -	- -	1 1	-	not verified	C032715 (method validation)
potatoes	0.01	0.5 2.5	92 - 100 -	- -	1 1	<0.3LOQ (2)	not verified	C033867 (trial) 1983 USA

GC-FPD, method 2 (1978)

In the analytical method used in Dutch trials on cucumbers (C034084) and tomato (C034084) crops were blended with sodium sulfate and hexane. The homogenate was filtered, evaporated to dryness and dissolved in hexane. Ethoprophos was determined by GC-FPD (glass, 10% DC200 + 15% QF1 (1+1) on chrom Q 80/100, temperature 190°C, phosphorus mode). Validation results from supervised residue trials are summarized in Table 33.

Table 33. Validation results for the determination of ethoprophos using GC-FPD method 2.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	no	Control (mg/kg)	Linearity	Ref.
Cucumber	0.01	0.01	85 80-90	2	<0.3LOQ (7)	not verified	C034084 (trial/ method validation) Anal. Oct/Nov 78
		0.02	95 95-95	2			
		0.04	97 -	1			
		0.06	97 -	1			
		0.1	95 -	1			
		0.4	96 -	1			
Tomato	0.01	0.01	68 55-82	2	<0.3LOQ (5)	not verified	C034084 (trial/ method validation) Anal. Oct/Nov 78
		0.03	90 84-97	2			
		0.1	104 97-112	2			

Mobil GC method (1980-1981)

The Mobil GC method was used in supervised residue trials carried out in 1980 in Canada on cucumber (C032713). No description was available. Concurrent validation results are shown in Table 34.

Table 34. Validation results for the determination of ethoprophos using Mobil GC method.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	no	Control (mg/kg)	Linearity	Ref.
Cucumber	0.01	0.1	99 94-104	2	<0.3LOQ (2)	not verified	C032713 (trial) 1980 Canada Anal. Aug 80
Cucumber	0.01	0.1	98 91-104	2	<0.3LOQ (1)	not verified	C032713 (trial) 1980 Canada Anal. Oct 81

GC-FPD, method MP-RE-08-83 (1983)

Method MP-RE-08-83, based on Mobil method MP 12-38, was used in Dutch trials on potatoes (R007979) and is based on extraction with hexane, clean-up (filtration) and determination by GC-FPD in the phosphorus mode (OV 17 column, temperature 200°C) (Rhône-Poulenc, 1983). Calibration was by 4 single external standards in the range 0.1-2.0 mg/l (mg/kg sample equivalent not stated). The method was validated for cucumber, maize (grains, whole plants), potatoes, and green tobacco. Validation results for potatoes are shown in Table 35.

Table 35. Validation results for the determination of ethoprophos using GC-FPD method MP-RE-08-83.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
Potato	0.02	0.02 0.2	85 74-91 94 86-100	11% 7.6%	3 3	<0.02	4 single points, range 0.1-1.0 mg/l; linear by graph	R007979 (method validation)
Potato	0.02	0.02 0.2	94 - 95 -	- -	1 1	<0.02 (2)	not verified	R007979 (trial)

GC-FPD method 3 (1984-1987)

GC-FPD method 3 was used in Brazilian trials on tomato (C033859, banana (C033861/C033862) and sugar cane (C033863/C033864). Only a summary description was available. Samples were extracted with n-hexane and passed through sodium sulfate. Ethoprophos was determined by GC-FPD (glass column, 5% SE-30 on Chromosorb WAW-DMCS 80-100 mesh, 150°C or 160°C, phosphorus mode). The reported LOQ was 0.05 mg/kg. Recovery at 0.05 mg/kg was 98% for tomato, 97% for banana pulp, 111% for whole banana and 86% for sugar cane stalks. No matrix interferences were found (<0.05 mg/kg, n=1 for each sample). Further details were not available.

Method 175 (1985-1991)

Method 175-1985 and modifications thereof were used in trials on sweet potatoes (C032642) and potatoes (R008006 and R008010) and in a storage stability study on broccoli, cabbage, dry peanut hay, and green and cured tobacco (R009168). Six different procedures (8.1-8.6) were developed depending on the sample. The method was based on extraction with hexane, clean-up (by silica gel or Florisil column, or partitioning into ACN) and determination by GC-FPD (phosphorus mode, 5% EGSS-X on 100/120 GCQ, 175°C for cole crops, 5% EGSS-X on 100/120 GCQ, 160°C or 10% DC-200 on 80/100 chrom WHP, 175°C for oily crops, 15% Carbowax 20M on 60/80 mesh GCP at 190°C or 10% DC-200 on 80/100 chrom WHP at 168°C for potatoes, 3% OV-17 on 80/100 GCQ, 180°C for tobacco, green and dry) (Perez *et al.*, 1985).

Calibration was by external standardization in the range 0.1-2.0 mg/l in hexane (corresponding to 0.01-0.2 mg/kg).

Method 175-1990 was used in UK trials on potatoes (R008006). Modification of the 1985 method using clean-up on Florisil Sep-Pak with 15% ethyl acetate in toluene as final eluent. Ethoprophos was determined by GC-FPD (5% OV 101 column, temperature programme 140-190°C, phosphorus mode).

Method 175-1991 was used in other UK trials on potatoes (R008010). The chromatographic column was modified (glass OV-17 WCOT microbore capillary, temperature 150 - 210°C, phosphorus mode).

Table 36. Validation results for the determination of ethoprophos using GC-FPD method 175.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
cabbage	0.01	0.01	104 -	-	1	-	not verified	C032642/R009168 (method validation) 175-1985, procedure 8.1
broccoli	0.01	0.01	91 -	-	1	-	not verified	idem
peanut hay	0.01	0.01 0.02 0.05 0.1	110 - 84 - 87 - 92 -	- - - -	1 1 1 1	-	not verified	C032642/R009168 (method validation) 175-1985, procedure 8.2
potato	0.01	0.01 0.05 0.1	87 - 90 - 77 -	- - -	1 1 1	-	not verified	C032642/R009168 (method validation) 175-1985, procedure 8.3
tobacco, green	0.01	0.02 0.05	99 - 94 -	- -	1 1	-	not verified	C032642/R009168 (method validation) 175-1985, procedure 8.4
tobacco, dry	0.01	0.02 0.05	96 - 96 -	- -	1 1	-	not verified	idem
sweet potato	0.01	0.01 0.1	90 80-100 104 98-109	- -	2 2	<LOQ (2)	not verified	C032642 (trial) Anal. 29 July 85 175-1985, procedure 8.3
peanut hay	0.01	0.05	96 86-101	7.4%	4	<0.3LOQ (4)	not verified	R009168 (storage stability) 175-1985, procedure 8.2
cabbage	0.01	0.05	102 92-109	8.6%	3	<0.3LOQ (3)	not verified	R009168 (storage stability) 175-1985, procedure 8.1
broccoli	0.01	0.05	98 92-105	6.0%	4	<0.3LOQ (4)	not verified	R009168 (storage stability) 175-1985, procedure 8.1
tobacco (green)	0.01	0.05	90 73-106	16%	5	<0.3LOQ (5)	not verified	R009168 (storage stability) 175-1985, procedure 8.4
tobacco (cured)	0.01	0.05	86 76-98	9.3%	5	<0.3LOQ (5)	not verified	R009168 (storage stability) 175-1985, procedure 8.4
potato	0.01	0.01 - 20 ¹	92 77-101	ns	3	<LOQ (1)	single point 0.05 mg/l	R008006 (trial) Anal. 29 Jun 1990 175-1990
potato	0.01	0.01 0.1	79 - 96 -	- -	1 1	<0.3LOQ (1)	single point 0.05 mg/l	R008010 (trial) Anal. 15 May 1991 175-1991

¹ Individual levels not shown in the study report

Method AR 52-87 (1987-2004)

Method AR 52-87 and modifications thereof were used in trials on potatoes (R007984, R007988, R008000, R007970, C013482, C015231, C019660, C019661), tomatoes (R008853, R008899, R016050, C016512, C023919), sweet peppers (R016050, R009798, C023543, C024789, C036690, C036691), strawberries (R008903, R008027), cucumbers (R004197, C025160, C036689), melons (R004456, C025152, C036692; C036693), a storage stability study on potatoes and tomatoes (R011312 and 02-120) and a processing study on potatoes (R016070). The method was based on extraction with MeOH, clean-up and determination by GC-FPD, GC-TSD (=GC-NPD), GC-PFPD, GC-MS or GC-MS-MS (Dupont and Soun, 1987, 1988, Maestracci, 1998e, Barbier, 2000, Meilland and Kieken, 2003, Bourgade and Rosati, 2003). The results are shown in Table 37.

Table 37. Validation results for the determination of ethoprophos using method AR 52-87.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
potato, whole	0.01	0.01 0.02 0.04	100 84-110 115 - 82 -	12% - -	4 1 1	<0.01 (12)	3 single points; range 0.05-0.2 mg/l; in hexane; linear, by graph	R007984 (trial) 1986 Germany; analysis Mar 87; 1987: GC-FPD
potato, whole	0.01	0.01 0.04	96 - 100 -	- -	1 1	<0.01 (2)	not verified	R007988 (trial) 1987 Germany analysis Jan 88 1988a: GC-FPD
potato, whole	0.01	0.01 0.02 0.06	96 - 87 - 87 -	- - -	1 1 1	<0.01 (20)	not verified	R008000 (trial) 1989 Germany anal. Mar 90 1990: GC-FPD
potato, whole	0.01	0.01 0.05 0.1 0.2	104 85-111 102 93-112 87 - 83 -	12% - - -	4 2 1 1	<0.3LOQ - 0.0045 (8)	not verified	R007970 (trial/ method validation) 1995 UK anal. Mar 96 1996: GC-FPD
tomato	0.01	0.01 0.2	84 73-98 81 -	12% -	5 1	<0.01 (6)	5 single points; range 4-20 ug/L; in hexane; linear, $r > 0.999$	R008853 (trial) 1996 ES Anal. Sept 96 1996b: GC-TSD
tomato	0.01	0.01 0.02 0.05	101 93-112 119 - 94 89-98	8.7% - -	4 1 2	<0.01 (8)	not verified	R008899 (trial) 1997 ES Anal Sept 97 1997: GC-FPD
tomato	0.01	0.01 0.02	79 - 78 -	- -	1 1	<0.01 (2)	not verified	R008904 (trial) 1997 IT Anal. 1997: GC-FPD
plum	0.01	0.01 0.02 0.05	100 99-101 88 - 87 85-89	- - -	2 1 2	<0.01 (5)	not verified	C013119 (method validation) Anal. Sept 97 1997: GC-FPD
strawberry	0.01	0.02	88 -	-	1	<0.01 (2)	not verified	R008903 (trial) 1997 IT Anal. Oct 97 1997: GC-FPD
sweet pepper	0.01	0.01	88 -	-	1	<0.01 (2)	not verified	R016050 (trial) 1997 IT Anal Oct 97 1997: GC-FPD
potato	0.01	0.01 0.1	84 84-85 87 74-95	- 11%	2 5	<0.01 (13)	5 double or triple points; range 0.025-0.50 mg/l; in hexane $r^2 > 0.99$ (n=2)	R011312 (storage) Anal Mar-Dec 99 1997: GC-FPD

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
cucumber	0.01	0.01 0.02 0.05	96 90-103 102 - 86 73-100	- - -	2 1 2	<0.01 (13)	6 single points; range 0.025-1.0 mg/l; in hexane; linear, by graph	R004197 (trial) 1998 ES Anal Sept 98 1998a: GC-FPD
melon, whole	0.01	0.01 0.025 0.10	107 - 98 - 92 -	- - -	1 1 1	<0.01 (3)	6 single points range 0.025-1.0 mg/l; in hexane; linear, by graph	R004456 (trial) 1998 ES Anal Dec 98 - Jan 99 1998a: GC-FPD
melon, peel	0.01	0.01 0.25	83 - 97 -	- -	1 1	<0.01 (2)	idem	R004456 (trial) 1998 ES Anal Dec 98 - Jan 99 1998a: GC-FPD
melon, pulp	0.01	0.01 0.025	89 - 94 -	- -	1 1	<0.01 (2)	idem	R004456 (trial) 1998 ES Anal Dec 98 - Jan 99 1998a: GC-FPD
strawberry	0.01	0.01 0.025 0.050	101 - 117 - 70 -	- - -	1 1 1	<0.01 (3)	5 single points; range 0.025-0.5 mg/l; in hexane; linear, by graph	R008027 (trial) 1998 IT Anal Dec 98- Jan 99 1998a: GC-FPD
green pepper	0.01	0.01 0.02 0.05 0.1	109 96-122 108 - 118 - 93 -	- - - -	2 1 1 1	<0.01 (5)	6 single points; range 0.025-1.0 mg/l; in hexane; linear, $r^2 > 0.999$	R009798 (trial) 1998 ES Anal Sept 98 - Feb 99 1998b: GC-FPD
potato, whole	0.005	0.005 0.1	84 72-105 84 75-100	9.6% 8.7%	15 14	<0.005 (13)	6 single points; range 0.003-0.1 mg/l; in hexane; linear, by graph	R016070 (trial/processing/ method validation) 1999 UK anal Dec 99-Mar00 1999: GC-FPD
potato, peel	0.005	0.005 0.1	78 - 68 64-72	- -	1 2	<0.005 (2)	idem	idem
potato, peeled	0.005	0.005 0.1	83 - 61 -	- -	1 1	<0.005 (1)	idem	idem
potato, baked	0.005	0.005 0.1	75 - 82 -	- -	1 1	<0.005 (1)	idem	idem
potato	0.01	0.01 0.1	101 97-104 95 88-101	3% 6%	5 5	<0.3LOQ (2)	6 single points; range 0.025-0.50 mg/l; in hexane; linear, $R^2 > 0.999$	C01520; Method validation 2000: GC-PFPD
grape	0.01	0.01 0.1	102 93-109 94 91-100	6% 4%	5 5	<0.3LOQ (2)	idem	idem
potato	0.01	0.01	106 100-110	5.0%	3	<0.01 (8)	6 single points; range 0.025-0.50 mg/l; in hexane; linearity not verified	C013482 (trial) 2000 France Anal Jan 2001 2000: GC-PFPD
potato	0.01	0.01	101 95-107	-	2	<0.01 (6)	6 single points; range 0.025-0.50 mg/l; in hexane; linear, $R^2 > 0.999$	C015231 (trial) 2000 Spain/Greece Anal Apr 2001 2000: GC-PFPD
tomato	0.01	0.01 0.02 0.04 0.1	77 70-88 102 - 70 - 98 95-101	11% - - -	4 1 1 2	<0.01 (21)	6 single points range 0.025-1.0 mg/l; in hexane linear $r^2 > 0.999$ (n=2)	C016512 (trial) 2000 ES, IT Anal May-June 2001 2001a: GC-FPD
tomato	0.005	0.005 0.01 0.02 0.025 0.05	105 102-108 87 - 74 - 78 - 92 88-97	2.5% - - - -	5 1 1 1 2	<0.005 (30)	6 duplicate points range 0.01-0.5 mg/l; in hexane linear, $r^2 > 0.999$	C023919 (trial) 2001 ES, P Anal March 2002 2001a: GC-FPD
sweet pepper	0.005	0.005 0.01	76 72-80 71 -	4.4% -	4 1	<0.005 (18)	5 single point range 0.0125-0.2 mg/l;	C023543 (trial/method

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
		0.015 0.05	71 - 90 87-94	- -	1 2		in hexane; linear, $r^2 > 0.99$	validation) 2001 FR, ES, I, GR Anal March 2002 2001a: GC-FPD
sweet pepper	0.005	0.005 0.01 0.02 0.025 0.05	78 75-85 71 - 72 - 73 - 94 -	4.6% - - - -	7 1 1 1 1	<0.005 (36)	5 duplicate points range 0.0125-0.2 mg/l; in hexane; linear, $r^2 > 0.99$	C024789 (trial/method validation) 2001 ES, I Anal Mar 02 2001a: GC-FPD
potato	0.01	0.01	88 82-95	5.5%	6	<0.01 (12)	6 single points; range 0.020-0.50 mg/l; in hexane; linear: $R^2 > 0.999$	C019660 (trial/ method validation) 2001 FR, GR Anal Nov 2001 2001b: GC-PFPD
potato	0.005	0.005 0.005	84 65-94 91 75-111	19% 13%	3 15	<0.005 (41)	6 single points; range 0.020-0.50 mg/l; in hexane; linear: $R^2 > 0.9999$ (PFPD) $R^2 > 0.99$ (MS-MS)	C019661 (trial/ method validation) 2001 UK, DE, FR Anal Aug-Oct 2001 2001c: GC- PFPD/MS
cucumber	0.005	0.005 0.005 0.05	85 85-86 87 70-111 94 -	0.7% 14% -	3 - 1	<0.005 (52)	8 single points range 0.005-0.50 mg/l; in hexane; linear: $R^2 > 0.9999$ (TSD) $R^2 > 0.999$ (PFPD)	C025160 (trial/ method validation) 2001 FR, I, ES, P, GR Anal June 2002 2001d: GC- PFPD/TSD
melon, peel	0.005	0.005 0.05	79 70-95 86 -	10% -	8 1	<0.005 (9)	8 single points range 0.005-0.5 mg/l; in hexane; linear $r^2 > 0.999$ (n=2)	C025125 (trial/method validation) 2001 FR, I, ES, GR Anal July 2002 2001d: GC-TSD
melon, pulp	0.005	0.005 0.05	90 73-103 98 -	11% -	8 1	<0.005 (9)	idem	C025125 (trial/method validation) 2001 FR, I, ES, GR Anal July 2002 2001d: GC-TSD
potato	0.005	0.005 0.05	82 71-96 90 84-101	12% 8%	5 5	<0.3LOQ (2)	4 single points; range 0.02-0.2 ug/L; in hexane; linear, $R^2 > 0.99$	C028919; method validation; 2003a: GC-MS
tomato	0.005	0.005 0.05	84 75-90 88 79-101	8% 11%	5 5	<0.3LOQ (2)	idem	idem
lettuce	0.005	0.005 0.05	80 68-93 99 88-108	12% 9%	5 5	<0.3LOQ (2)	idem	idem
tomato	0.005	0.1	93 87-102	6%	5	<0.01 (4)	not verified	02-120 (storage stability) 2003a: GC-MS
potato	0.005	0.1	92 78-111	14%	6	<0.01 (4)	idem	idem
sweet pepper	0.005	0.005 0.05	102 101-104 74 71-78	- -	2 2	<0.3LOQ (1)	4 duplicate points range 0.02-0.2 ug/L in hexane; linear, $R^2 > 0.99$	C033190 (method validation) Anal. Apr/May 2003 2003b: GC-MS
melon	0.005	0.005 0.05	94 85-102 78 76-79	- -	2 2	<0.3LOQ (1)	idem	idem
cucumber	0.005	0.005 0.05	105 101-109 84 81-86	- -	2 2	<0.3LOQ (1)	idem	idem
lettuce	0.005	0.005 0.05	96 95-96 84 82-86	- -	2 2	<0.3LOQ (1)	idem	idem

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
pepper	0.005	0.005 0.05	100 93-105 75 71-78	4.3% 5.0%	8 3	<0.005 (27)	5 duplicate points; range 0.02-0.25 ug/L; in hexane; R ² >0.99	C036690 (trial/method validation) 2002 FR, ES, I, GR Anal. Jul/Nov 2003 2003b: GC-MS
pepper	0.005	0.005 0.15 0.05	99 91-111 63 - 80 71-98	13% - 15%	4 1 4	<0.005 (18)	5 duplicate points; range 0.02-0.25 ug/L; in hexane; R ² >0.99	C036691 (trial/method validation) 2002 FR, ES, I Anal. June 2003 2003b: GC-MS
melon, whole	0.005	0.005 0.010 0.015 0.040 0.050	101 85-112 114 - 99 - 86 85-88 78 76-79	11% - - - -	4 1 1 2 2	<0.005 (25)	5 duplicate points; range 0.02-0.25 ug/L; in hexane; R ² >0.999	C036693 (trial) 2002 FR, ES, I Anal Aug-Sept 2003 2003b: GC-MS
melon, pulp	0.005	0.005 0.015 0.040	99 - 104 99-109 71 -	- - -	1 2 1	<0.005 (10)	idem	C036693 (trial) 2002 FR, ES, I Anal Aug-Sept 2003 2003b: GC-MS
cucumber	0.005	0.005 0.01 0.04 0.05	104 76-117 94 90-100 89 - 81 76-86	13% 5.9% - 6.2%	12 3 1 3	<0.005 (49)	5 single points; range 0.02-0.25 ug/L; in hexane; linearity not verified	C036689 (trial/method validation) 2002 FR, ES, I Anal. Sept-Oct 2003 2003b: GC-MS
melon, whole	0.005	0.005 0.05	89 79-102 76 -	13% -	3 1	<0.005 (4)	5 duplicate points; range 0.02-0.25 ug/L; in hexane; r ² >0.99	C036692 (trial/method validation) 2002 FR, ES, I Anal Oct 2003 2003b: GC-MS
melon, peel	0.005	0.005	92 90-94	2.1%	4	<0.005 (18)	idem	C036692 (trial) 2002 FR, ES, I Anal Oct 2003 2003b: GC-MS
melon, pulp	0.005	0.005	84 72-104	19%	4	<0.005 (18)	idem	C036692 (trial) 2002 FR, ES, I Anal Oct 2003 2003b: GC-MS

JFRL GC-FPD method (1988)

The analytical method used in Phillipine trials on bananas (peel and pulp, R011296) was based on extraction with acetone, clean-up and determination by GC-FPD (3% OV 17 on chromosorb WHP 80-100 mesh, 195°C, phosphorus mode). Individual recoveries were not reported. The average recovery was 100% (0.01 mg/kg). Linearity was not reported. Proposed LOQ was 0.005 mg/kg and control samples were <LOQ (n=3).

GC-TSD method 1 (1988)

The analytical method used in a sugar cane processing study (C036561) was based on extraction with hexane, clean-up and determination by GC-TSD (5% OV-101 on chromosorb G, 160°C) (Brockelsby *et al.*, 1988; Parthasarathy, 1989). Individual recoveries were not reported. Average recoveries were 89%-93% (0.02-0.05 mg/kg). Linearity and results for control samples were not reported. The reported LOQ was 0.01 mg/kg for sugar cane stalks, leaves and juice.

GC-FPD, method 4 (1991-)

The analytical method used in processing studies on sugar cane (R016038) and potatoes (R007960), storage stability studies on sugar cane commodities (R008872) and various other commodities (R008020), trials on cucumbers (R009784) and a field rotational crop study (R008901) was modified several times. The method is based on extraction of the parent and metabolite mA with MeOH, clean-up, derivatisation of metabolite A with diazomethane and determination by GC-FPD (Eng, 1992; Rhone-Poulenc, 1994a-e; Thiem, 1994). Only the validation results for the parent are shown in Table 38. The proposed LOQ was 0.005-0.01 mg/kg, but because brassica commodities and some batches of diazomethane contained compounds that interfered with quantification of the parent, the valid LOQ should be increased to at least 0.05 mg/kg.

Independent validation: In separate experiments, ethoprophos and metabolite mA were quantitatively recovered from resin/nuchar/attaclay (100%), GPC columns (98%-122%) and silica gel columns (84%-98%). Physical characteristics of GPC columns changed during several weeks of non-use and it is recommended to eliminate this step. In the ILV a DB-5 column (temperature 60-260°C) was used. The linear range was 7-25 pg injected instead of 5-200 pg as in the original method. An interference was found at the retention time of ethoprophos owing to impurities in the diazomethane used. Changes in GC conditions and GC columns did not resolve the problem, so ethoprophos was determined separately without the methylation step. This resulted in average matrix interferences of 0.076 mg/kg apparent ethoprophos, which is unacceptably high.

Method performance for ethoprophos and metabolite mA is generally better for crops of high to moderate moisture. The method will generally fail for mA if dry and oily crops are not hydrated before extraction. Chromatograms were characterized by numerous peaks which made interpretation difficult, especially for cruciferous plants (e.g. brassicas). Certain batches of diazomethane contained compounds that interfered with quantification of parent ethoprophos. Derivatization efficiency and precision for metabolite mA were not verified. Because of matrix interferences, the LOQ for parent ethoprophos should be increased to at least 0.05 mg/kg.

Table 38. Validation results for the determination of ethoprophos (parent) using GC-FPD method 4.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
cabbage	0.005	0.005 0.01 0.05 0.5	87 74-95 96 80-113 99 85-107 89 78-109	8.0% 13% 8.1% 13%	7 7 5 5	<0.3LOQ- 0.0028 (8)	6 single points; range 0.005-0.1 mg/l; in hexane linear; R2>0.98	R009735 (method validation) Anal Oct/Nov 91 Original version
sugar cane, stalks	0.01	0.01 0.05 0.5	83 72-94 85 81-92 88 84-91	11% 4.4% 3.0%	6 6 6	0.0032 (1)	4 single points; range 5-50 ug/L; in iso-octane linear by graph	R016038 (method validation, processing study) Anal Feb 93 Version 3.0
sugar cane, bagasse	0.01	0.01 0.05 0.5	93 82-102 82 74-86 87 85-90	9.2% 5.3% 8.1%	6 6 6	<0.3LOQ (1)	idem	idem
sugar cane, sugar	0.01	0.01 0.05 0.5	104 96-114 103 97-108 99 93-101	7.5% 3.7% 3.3%	6 6 6	<0.3LOQ (1)	idem	idem
sugar cane, molasses	0.01	0.01 0.05 0.5	86 70-95 89 79-104 96 90-101	13% 11% 5.0%	6 6 6	<0.3LOQ (1)	idem	idem
sugar cane, syrup	0.01	0.01 0.05 0.5	99 84-120 96 85-105 100 78-111	15% 7.8% 11%	9 9 9	<0.3LOQ (1)	idem	idem
sugar cane, clarified juice	0.01	0.01 0.05 0.5	91 79-105 86 87-92 95 86-99	11% 6.0% 5.8%	6 6 6	<0.3LOQ (1)	idem	idem
sugar cane,	0.01	0.01	83 72-92	10%	5	0.0043 (1)	idem	idem

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
mixed juice		0.05 0.5	91 80-104 98 91-102	8.8% 3.8%	6 6			
sugar cane, clarifier mud	0.01	0.01 0.05 0.5	100 89-107 87 79-95 93 83-100	8.3% 8.0% 7.5%	6 6 6	<0.3LOQ (1)	idem	idem
potato tubers	0.01	0.01 0.05 0.5	97 94-100 99 95-106 94 90-97	2.4% 4.2% 3.9%	6 6 6	<0.3LOQ (1)	4 single points; range 5-50 ug/L; in iso-octane linear by graph	R007960 (method validation, processing study) Anal Jul/Aug 93 Version 7.0
potato, dry peel	0.01	0.01 0.05	97 93-103 97 90-105	4.7% 5.4%	4 4	<0.3LOQ (1)	idem	idem
potato, chips	0.01	0.01 0.05	108 99-116 106 103-109	7.1% 2.7%	4 4	<0.3LOQ (1)	idem	idem
potato, wash water	0.01	0.01 0.05	94 90-98 99 96-102	3.7% 2.4%	4 4	<0.3LOQ (1)	idem	idem
lima bean pods	0.005	0.02 0.1	93 93-94 80 74-91	- 9.5%	2 4	0.065-0.088 (4)	5 single points range 7-25 pg injected; in iso-octane; r>0.98	C037530 (ILV) Anal 1994 Version 2.0, without methylation step
cabbage	0.02	0.2	100 95-104	4.5%	3	<0.3LOQ- 0.016 (9)	5 single points; range not stated; in iso-octane; r ² >0.99	R008020 (method validation, storage stability) Anal. 1993-94 Version 3.0
potato tuber	0.02	0.2	99 95-104	4.8%	3	<0.3LOQ- 0.018 (12)	idem	idem
pineapple bran	0.02	0.2	103 98-107	4.6%	3	<0.3LOQ- 0.015 (12)	idem	idem
pineapple fruit	0.02	0.2	102 98-104	3.2%	3	<0.3LOQ- 0.014 (11)	idem	idem
pineapple feed pulp	0.02	0.2	84 80-86	4.1%	3	<0.3LOQ- 0.017 (10)	idem	idem
pineapple juice	0.02	0.2	102 92-110	9.0%	3	<0.3LOQ- 0.016 (7)	idem	idem
peanut kernels	0.02	0.2	97 90-101	6.3%	3	<0.3LOQ- 0.012 (12)	idem	idem
peanut meal	0.02	0.2	86 80-96	10%	3	<0.3LOQ- 0.031 (9)	idem	idem
peanut vines	0.02	0.2	108 100-117	7.9%	3	<0.3LOQ- 0.007 (9)	idem	idem
peanut hay	0.02	0.2	94 93-96	1.8%	3	<0.3LOQ- 0.012 (10)	idem	idem
peanut hulls	0.02	0.2	114 110-117	3.1%	3	<0.3LOQ (9)	idem	idem
peanut refined oil	0.02	0.2	86 72-104	19%	3	<0.3LOQ- 0.022 (9)	idem	idem
peanut crude oil	0.02	0.2	96 83-111	15%	3	<0.3LOQ- 0.005 (8)	idem	idem
peanut soap stock	0.02	0.2	64 60-66	3.4%	9	<0.3LOQ- 0.013 (13)	idem	idem
corn starch	0.02	0.2	100 94-106	6.0%	3	<0.3LOQ- 0.011 (11)	idem	idem
corn fodder	0.02	0.2	112 109-116	3.1%	3	<0.3LOQ (11)	idem	idem
corn meal	0.02	0.2	99 98-100	1.0%	3	<0.3LOQ- 0.014 (9)	idem	idem
corn grain	0.02	0.2	108 105-113	4.3%	3	<0.3LOQ- 0.014 (13)	idem	idem
corn grain dust	0.02	0.2	85 81-91	6.5%	3	<0.3LOQ- 0.015 (9)	idem	idem
corn forage	0.02	0.2	100 92-105	7.2%	3	<0.3LOQ (10)	idem	idem

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
corn refined oil	0.02	0.2	78 72-81	6.7%	3	<0.3LOQ-0.014 (9)	idem	idem
corn crude oil	0.02	0.2	80 77-81	2.9%	3	<0.3LOQ-0.012 (9)	idem	idem
sugar cane stalks	0.005	0.2	86 73-96	13%	4	<0.3LOQ-0.041 (41)	5 single points 0.01-0.25 mg/l in iso-octane $r^2 > 0.99$	R008872 (method validation, storage stability) Anal 1994. Version 3.0
sugar cane molasses	0.005	0.2	87 78-96	9.2%	4	<0.3LOQ-0.051 (41)	idem	R008872 (method validation, storage stability) Anal 1994 Version 3.0
sugar cane refined sugar	0.005	0.2	97 93-100	3.1%	4	<0.3LOQ-0.12 (41)	idem	R008872 (method validation, storage stability) Anal 1994 Version 3.0
cucumber	0.01	0.01	89 86-92	2.4%	5	-	4 single points 15-150 pg injected in iso-octane; loglinear by graph	R009784 (method validation) Version 6.0
		0.05	89 87-92	2.2%	5			
		0.5	90 84-95	4.9%	5			
cucumber	0.01	0.01	95 87-108	5.8%	13	<0.01 (12)	4 single points 15-150 pg injected in iso-octane; $r^2 > 0.99$ (log-log)	R009784 (method validation/trial/storage stab) Anal Oct 93/Feb 94 Version 6.0
		0.05	95 82-106	8.0%	11			
		0.5	99 97-101	-	2			
field corn grain	0.01	0.01	96 90-98	3.1%	6	<0.3LOQ (1)	4 duplicate points 5-50 ug/L in iso-octane; linear graph (log-log)	R008901 (method validation) Anal May 94 Version 13.0
		0.05	94 91-97	2.1%	6			
		0.5	90 88-96	3.2%	6			
field corn forage	0.01	0.01	98 88-105	6.3%	6	<0.3LOQ (1)	idem	idem
		0.05	98 96-101	2.0%	6			
		0.5	95 91-99	3.5%	6			
field corn fodder	0.01	0.01	107 105-111	2.5%	6	<0.3LOQ (1)	idem	idem
		0.05	95 92-98	2.3%	6			
		0.5	88 84-93	3.7%	6			
collards	0.01	0.01	92 -	-	1	<0.01 (1)	4 duplicate points 5-50 ug/L in iso-octane $r^2 > 0.98$ (log-log)	R008901 (rotational crop study) Anal Aug-Nov 96 Version 13.0
		0.5	83 -	-	1			
pea forage	0.01	0.01	100 87-112	10%	4	<0.01 (4)	idem	idem
		0.5	96 87-109	10%	4			
pea seeds	0.01	0.01	80 71-85	9.7%	3	<0.01 (3)	idem	idem
		0.5	76 75-78	-	2			
pea straw	0.01	0.01	82 77-90	8.8%	3	<0.01 (3)	idem	idem
		0.5	76 74-79	3.0%	3			
mustard forage	0.01	0.01	94 90-101	6.1%	3	<0.01 (3)	idem	idem
		0.5	88 84-92	4.5%	3			
radish roots	0.01	0.01	87 78-97	8.6%	7	<0.01-0.013 (7)	idem	idem
		0.5	83 63-95	13%	7			
radish tops	0.01	0.01	96 86-104	6.8%	7	<0.01 (7)	idem	idem
		0.5	91 80-99	6.2%	7			
red leaf lettuce	0.01	0.01	78 70-90	13%	6	<0.01 (6)	idem	idem
		0.5	81 73-92	13%	6			
wheat forage	0.01	0.01	96 92-102	4.7%	4	<0.01 (4)	idem	idem
		0.5	90 83-98	6.8%	4			
wheat grain	0.01	0.01	86 83-93	6.4%	3	<0.01 (3)	idem	idem
		0.5	76 74-78	2.6%	3			
wheat straw	0.01	0.01	86 82-93	6.5%	3	<0.01 (3)	idem	idem
		0.5	78 73-84	7.0%	3			

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
sorghum forage	0.01	0.01 0.5	95 87-101 87 81-95	7.8% 8.0%	3 3	<0.01 (3)	idem	idem
sorghum grain	0.01	0.01 0.5	87 77-96 81 77-86	- -	2 2	<0.01 (2)	idem	idem
sorghum straw	0.01	0.01 0.5	88 77-96 81 77-86	- -	2 2	<0.01 (2)	idem	idem
soya bean forage	0.01	0.01 0.5	100 94-106 87 86-88	- -	2 2	<0.01 (2)	idem	idem
soya bean grain	0.01	0.01 0.5	89 - 82 -	- -	1 1	<0.01 (1)	idem	idem
soya bean straw	0.01	0.01 0.5	80 - 74 -	- -	1 1	<0.01 (1)	idem	idem

GC-NPD method 1 (1996)

GC-NPD method 1 (Capri *et al.*, 1998) was used in Italian trials on tomato (R008029). Crops were blended with diatomaceous earth and ethyl acetate. After filtration, the extract was concentrated and ethoprophos was determined by GC-NPD (DB-5 column, 60-280°C). The reported LOQ was 0.01 mg/kg in tomato (fruit and plant), and the reported recoveries $\pm$ RSD 90% $\pm$ 13 for 1 ng and 90% $\pm$ 10% for 2 ng (equivalent mg/kg sample and individual values were not reported).

AOAC method 970.52

AOAC method 970.52 was used in a duplicate diet study carried out in the USA (Fenske *et al.*, 2002), where residues <LOQ were found. Homogenised samples (20 g fruits or vegetables, 300 ml beverages, 100 ml dairy products) were extracted with ACN. The extract was washed with 30% aqueous sodium chloride and hexane. Water was added and residues were partitioned into DCM. Samples were concentrated and brought to 2 ml in hexane/acetone (1:1). Ethoprophos was determined using GC-PFPD (DB-1 column). Calibration was by external standardization at 3-5 concentration levels. Performance characteristics (n=3-6) were as follows.

- fresh fruits and vegetables: LOQ 0.005 mg/kg, mean recovery 77% at 0.025 mg/kg. This food group was represented by a mixture of equal portions of cored apples and peeled bananas.
- fruit juices and beverages: LOQ 0.033 mg/kg, mean recovery 91% at 0.067 mg/g, represented by a mixture of apple juice, grape juice and orange juice (with pulp).
- dairy products: LOQ 0.005 mg/kg, mean recovery 100% at 0.020 mg/kgs food, represented by whole milk.
- processed foods: LOQ 0.005 mg/kg, mean recovery 67% at 0.025 mg/kg, represented by a mixture of cookies, sugar pops, corn tortillas, bologna, corn chips, oatnut bread, macaroni and cheese.

Further details were not available.

Multi-residue GC method

The multi-residue GC method was used in a residue monitoring study carried out in Belgium, where no residues were found. The method is used for determination of organochlorine, organophosphorus, and nitrogen-containing pesticides in vegetables and fruits. Samples are extracted with petroleum etheracetone followed by liquid-liquid partitioning with water. Apolar pesticides are found in the petroleum ether-phase, polar pesticides are extracted from the aqueous layer with DCM. Analysis is by GC-ECD, GC-FPD or GC-TSD. Further details were not available.

Analytical methods for environmental studies

The Meeting received information on analytical methods for the determination of ethoprophos and mA in soil used in soil degradation and rotational crop studies. Various other methods for analysis of soil, air and water are not included, because the corresponding studies were not evaluated.

GC-FPD method 5 (1984)

GC-FPD method 5 was used in soil degradation study 1. Soil was extracted with MeOH. The MeOH extract was mixed with 10% (w/v) NaCl and partitioned into hexane. The hexane extract was filtered through anhydrous sodium sulfate and concentrated. Ethoprophos was determined by GC-FPD (10% OV101 on diatomite CQ (80/100 mesh), temperature 190°C, phosphorus mode). Calibration was by external standardization. Validation results are shown in Table 40.

Table 40. Validation results for the determination of ethoprophos using GC-NPD method 5.

Sample	LOQ reported mg/kg	Spike mg/kg	Recovery, % mean range	RSD	no	Control (mg/kg)	Linearity	Ref.
sandy clay loam	-	0.53	84 78-89	-	2	-	10 single points;	R009121 (soil degradation) method validation
sandy loam	-	1.1	93 90-96	-	2	-	range 0-5 mg/l	
	-	5.3	84 79-90	-	2	-	in hexane;	
	-	14	90 86-95	-	2	-	linear: r>0.999	

Rhone Poulenc method 172 (1984-1991)

Rhone Poulenc method 172 (Perette *et al.*, 1984) is a modification of Mobil Chemical method 96-78 (25 Sept 1978). Dry soil is extracted with MeOH. The filtered MeOH extract is mixed with 10% aqueous NaCl and partitioned into hexane. The hexane extract is filtered through anhydrous sodium sulfate and concentrated. Ethoprophos is determined by GC-FPD (3% Silar 5CP on 80/100 GCQ or 3% OV-17, temperature 220°C, phosphorus mode). Calibration is by external standardization.

Validation results are shown in Table 41. In addition, samples were fortified with radiolabelled ethoprophos and analysed both by GC-FPD and LSC. Results were similar (ratio 0.88-1.1).

Method 172, version 1991, was used in soil degradation study 3 (R009121). The detection system was changed to GC-NPD (capillary OV-1, temperature programme 150-245°C).

Table 41. Validation results for the determination of ethoprophos using method 172.

Sample	LOQ reported mg/kg	Spike mg/kg	Ratio FPD/LSC mean range	Recovery, % mean range GC-FPD	RSD	no	Control (mg/kg)	Linearity	Ref.
loam	0.01	0.01	1.1 0.95-1.1	107 97-115	5.8%	7	-	6 single points;	R009187 (method validation) original version
sandy loam		0.1	1.0 0.88-1.1	100 91-107	5.2%	8	-	range 0.1-2.0 mg/l;	
		1	1.0 0.94-1.1	100 91-107	6.8%	5	-	in hexane;	
		10	1.0 0.91-1.1	91 82-98	7.2%	5	-	not verified	
humic sand	0.01	0.01	-	88 -	-	1	<0.01	-	R009121 (soil degradation) version 1991
sandy loam		0.05	-	90 88-91	-	2	(3)	-	
loamy silt		0.1	-	91 -	-	1	-	-	
		0.25	-	91 -	-	1	-	-	
		0.50	-	94 -	-	1	-	-	
		1.0	-	92 -	-	1	-	-	
		5.0	-	80 -	-	1	-	-	

Stability of pesticide residues in stored analytical samples

The Meeting received data on the stability of residues in crops with high water content (pineapple, broccoli, cabbage, potatoes, sweet potatoes, tomato), dry crops with starch and protein (maize), dry crops with fat or oil, starch and protein (peanut), miscellaneous crops (sugar cane, tobacco (green and cured)), processed commodities (pineapple juice, peanut and maize oil, maize starch, refined cane sugar) and feed items (pineapple bran and feed pulp, peanut hulls, meal, vines and dry hay, maize meal, forage, fodder, and grain dust, sugar cane molasses) stored frozen. Crops with high water and high acid content (citrus fruits) were not investigated.

Study 1. Broccoli, cabbage, dry peanut hay, and green and cured tobacco were spiked with ethoprophos at 0.05 mg/kg and stored at -20°C over a 9-18 months period (R009168, Perez, 1986, non-GLP). Sweet potatoes from a field trial (Ville Pratte, LA, USA) were treated with 10 kg ai/ha at lay-by (3 Aug 1984) and harvested after 101 days. They contained 0.06 mg/kg ethoprophos as incurred residues at initial analysis (259 days after harvest). Duplicate samples were analysed by method 175-1985 (procedures 8.1-8.4). Results were not corrected for concurrent recoveries (73%-109%), nor for interferences (<0.3 LOQ). They are shown in Table 42.

Table 42. Frozen storage stability of 0.05 mg/kg ethoprophos on various commodities (n=2) stored at -20°C .

Sample	Storage (months)	Ethoprophos remaining, %		Ethoprophos, concurrent recovery %
		mean	range	
Broccoli	0	108	105-111	105
	6	76	69-82	99
	9	74	68-80	94
	12	56	54-58	92
Cabbage	0	107	104-108	104
	10	94	86-101	109
	12	84	84-84	92
Sweet potato ¹	0	100	0.058 mg/kg	-
	9	94	0.048-0.061 mg/kg	103
Peanut hay	0	91	90-92	96
	6	44	43-45	101
	9	42	40-44	101
	12	41	38-44	86
Tobacco green	0	90	84-96	84
	6	75	70-80	106
	9	113	106-120	73
	12	96	94-97	103
	18	92	88-96	84
Tobacco cured	0	82	82-82	84
	6	57	56-58	98
	9	92	90-94	76
	12	95	94-96	84
	18	61	54-68	86

¹ contained 0.058 mg/kg ethoprophos as incurred residues at initial analysis (259 days after harvest)

Study 2. Pineapple, cabbage, potato, peanut, maize, their processed products and their feed byproducts were spiked with either ethoprophos or mA at a concentration of 0.20 mg/kg (R008020, Ibrahim, 1995). Samples were stored for a period of 12 months. One set was stored at -5°C and another set at -20°C . Samples stored at -5°C were analysed first and if the residues were stable at this temperature, the set stored at -20°C was not analysed. Samples were analysed using GC-FPD, method 4, version

3.0. Results were not corrected for concurrent recoveries (37%-118%), but were corrected for matrix interferences (up to 0.031 mg/kg for ethoprophos and up to 0.007 mg/kg for mA). Results from the –20°C set are shown in Table 43.

Table 43. Frozen storage stability of 0.2 mg/kg ethoprophos or 0.2 mg/kg mA on various commodities (n=3) stored at –20°C or -5°C (pineapple juice).

Sample	Storage (months)	Ethoprophos remaining %			Ethoprophos, concurrent recovery %	mA remaining %			mA, concurrent recovery %
		mean	range	RSD		mean	range	RSD	
Pineapple fruit	0	85	70-97	16	95	75	72-81	7	80
	3	83	80-86	4	97	80	78-82	3	89
	6	83	81-85	3	91	76	75-77	2	85
	9	81	76-86	6	89	70	69-71	1	79
	12	62	57-69	10	75	67	64-70	5	79
Pineapple juice (at -5°C)	0	97	94-102	5	93	90	86-93	4	89
	3	93	76-103	16	97	92	90-95	6	96
	6	75	71-77	4	81	79	79-80	1	89
	9	99	90-106	8	113	83	80-86	4	96
	12	86	83-89	4	79	74	73-75	2	79
Pineapple bran	0	99	90-115	14	83	83	68-96	17	71
	3	70	62-78	11	91	57	55-60	5	86
	6	78	78-79	1	99	61	59-64	4	87
	9	81	76-87	7	104	55	48-60	12	84
	12	58	52-64	10	90	48	41-55	15	78
Pineapple feed pulp	0	118	110-127	7	116	84	80-86	4	88
	3	67	66-69	3	91	59	58-62	4	82
	6	73	67-78	8	91	61	58-64	5	84
	9	78	72-82	7	87	61	57-64	6	83
	12	55	52-59	7	87	50	46-53	7	69
Cabbage	0	85	71-93	14	82	87	86-91	2	86
	9	99	94-104	5	108	72	71-75	3	85
Potato	0	122	77-188	48	89	86	79-90	7	84
	2 ¹	94	94-94	-	108	81	80-82	-	95
	3	78	65-94	19	90	78	75-80	3	91
	6	77	67-85	12	86	56	49-61	11	82
	9	95	92-100	4	101	55	53-56	3	79
	12	85	83-87	3	94	54	53-54	1	82
Peanut kernels	0	77	70-82	8	82	83	79-85	4	84
	1.5	78	76-81	3	90	91	90-92	1	89
	3	53	52-56	4	84	70	68-72	3	89
	6	48	47-51	5	71	68	67-68	1	72
	9	68	67-70	2	102	81	79-84	3	95
	12	69	66-73	5	84	75	75-75	0	83
Peanut crude oil	0	77	75-79	3	70	86	85-88	2	73
	6	78	73-83	6	78	63	60-65	4	68
Peanut refined oil	0	82	77-85	5	73	95	92-98	3	91
	3	81	80-83	2	89	81	80-83	2	89
	6	89	86-90	3	88	65	64-66	2	77
	9	80	74-84	7	85	59	52-64	11	80
	12	84	82-86	3	79	73	73-74	1	76
Peanut hull	0	94	90-100	6	92	86	86-87	1	90
	3	76	74-79	4	94	55	54-56	2	86
	6	72	69-73	3	95	42	40-45	7	84
	9	56	54-60	6	93	35	32-37	8	83
	12	61	57-64	6	89	34	30-39	14	77
Peanut meal ²	0	95	93-100	5	93	81	80-84	3	81
	3	58	52-61	9	75	53	51-57	6	80
	6	48	40-54	15	83	39	37-41	5	71
	9	58	49-63	13	94	50	44-54	11	83
	12	50	46-53	7	96	43	42-43	1	79

Sample	Storage (months)	Ethoprophos remaining % mean range RSD	Ethoprophos, concurrent recovery %	mA remaining % mean range RSD	mA, concurrent recovery %
Peanut soapstock	0	61 61-62 1	63	72 72-73 1	74
	3	58 53-64 9	61	48 33-71 43	37
	6	64 59-69 8	72	76 60-99 26	111
Peanut vines	0	96 87-105 9	96	87 85-91 4	88
	3	91 87-98 7	100	29 28-30 3	91
	6	89 87-91 2	90	19 18-19 3	78
	9	89 85-92 4	91	13 12-15 11	78
	12	80 78-84 4	92	9 7-11 24	82
Peanut hay	0	94 91-99 5	93	90 87-93 3	91
	3	86 80-90 6	100	29 27-30 5	83
	6	76 73-80 5	93	17 16-18 6	78
	9	67 59-74 11	88	18 17-19 6	76
	12	68 68-68 0	83	17 16-18 6	78
Corn grain	0	111 109-114 2	114	91 89-93 2	97
	1.5	94 90-99 5	108	54 48-60 11	88
	3	79 72-85 8	90	39 35-44 16	91
	6	85 81-88 4	97	35 35-36 2	81
	9	86 83-90 4	107	36 36-37 2	89
Maize starch	0	102 98-108 5	101	79 76-82 4	91
	3	115 106-125 8	106	28 26-31 9	77
	6	97 93-105 7	105	25 21-29 16	77
	9	104 99-108 4	118	21 19-24 13	69
	12	83 81-84 2	91	18 17-19 6	66
Maize crude oil	0	71 69-73 3	72	81 80-83 2	80
	3	71 69-75 5	68	80 78-82 3	84
	6	70 66-73 5	74	68 67-69 2	75
	9	88 86-92 4	90	72 70-73 2	88
	12	82 80-87 5	83	73 71-75 3	82
Maize meal	0	104 103-105 1	91	89 88-89 1	80
	3	90 83-99 9	83	58 52-62 9	67
	6	104 100-112 6	105	68 63-74 8	88
	9	106 99-111 6	105	64 60-66 5	74
	12	90 89-91 1	95	54 53-54 1	67
Maize forage	0	95 89-102 7	92	85 83-88 3	84
	3	81 77-84 4	93	66 64-69 4	91
	6	82 81-84 2	86	55 53-56 3	75
	9	78 71-83 8	109	52 48-56 8	87
	12	74 67-81 10	93	51 45-55 10	75
Maize fodder	0	98 96-100 2	90	92 90-94 2	86
	1.5	81 76-86 6	91	67 65-70 4	87
	3	75 72-78 4	91	59 58-60 2	87
	6	86 83-90 4	98	56 54-58 4	83
	9	85 81-88 4	105	47 45-49 5	86
Maize grain dust	0	74 67-81 10	93	51 45-55 10	75
	3	71 70-72 1	88	42 39-45 7	71
	6	86 75-97 13	85	79 68-90 14	77
	9	79 72-83 8	90	66 63-68 4	85
	12	87 85-89 2	97	68 66-70 3	82
	0	85 75-92 11	113	61 54-68 12	88
	3	85 75-92 11	113	61 54-68 12	88
	12	91 89-93 2	98	70 69-70 1	79

¹ Sample analysed in duplicate, not in triplicate² Peanut meal control samples contained matrix interferences at 0.039 mg/kg ethoprophos eq.

Study 3. Samples of sugar cane and its processed commodities were spiked with either ethoprophos or mA at a concentration of 0.20 mg/kg (R008872, Eng, 1996) and stored at -20°C for 15 months. Triplicate samples were analysed using GC-FPD, method 4, version 3.0. Results were not corrected for concurrent recoveries (68%-105%), but were corrected for matrix interferences (<0.3 LOQ-0.12 mg/kg for ethoprophos, <0.3 LOQ for mA). Results are shown in Table 44.

Table 44. Frozen storage stability of 0.2 mg/kg ethoprophos or 0.2 mg/kg mA on sugar cane and its processed commodities (n=3) stored at -20°C .

Sample	Storage (days)	Ethoprophos remaining %			Ethoprophos, concurrent recovery %	mA remaining %			mA, concurrent recovery %
		mean	range	RSD		mean	range	RSD	
sugar cane	0	91	90-91	1	85	84	83-84	1	72
	44	73	70-76	4	95	77	73-79	4	87
	92	65	55-71	13	69	63	60-67	6	73
	167	80	75-87	8	93	74	73-76	2	79
	276	82	76-87	7	92	76	72-79	5	89
	358	73	62-91	22	81	67	62-74	9	91
	453	73	60-88	19	89	83	69-93	15	79
molasses	0	83	70-89	13	86	79	73-84	7	78
	44	86	75-94	12	88	88	80-93	8	87
	92	75	68-83	10	84	62	59-65	5	72
	166	86	74-94	12	81	74	63-83	14	91
	275	86	75-92	11	77	85	73-94	13	74
	357	109	104-113	4	91	96	73-108	21	103
	453	102	98-105	4	105	60	57-65	7	68
refined sugar	0	88	82-93	6	90	76	75-77	1	76
	44	89	86-92	3	95	71	67-73	5	74
	92	67	65-71	4	89	63	58-65	6	68
	163 ¹	79	68-93	16	100	68	61-72	9	72
	276	92	88-96	4	97	87	86-87	1	83
	361	68	59-78	14	84	80	78-83	4	87
	451	81	72-88	10	84	82	75-89	9	93

¹ Apparent ethoprophos in control sample of refined sugar at 163 days of storage is 0.58 LOQ.

Study 4. Untreated potato samples were spiked with ethoprophos at 0.10 mg/kg and stored at -18°C for 9 months (Quintelas, 2000). Duplicate spiked samples were analysed using method AR 52-87, 1997, GC-FPD. Results were not corrected for concurrent recoveries (74%-95%), nor for interferences (<0.01 , n=4). Results are shown in Table 45.

Table 45. Frozen storage stability of 0.1 mg/kg ethoprophos on potatoes (n=2) stored at -18°C .

Storage (months)	Remaining %		concurrent recovery % at 0.1 mg/kg
	mean	range	
0	95	95-95	95
3	54	47-60	74
6	93	92-94	95
9	76	71-80	84

Study 5. Cucumbers in trial 93-0089 were treated with a single 13 kg ai/ha pre-planting soil EC application and three replicate field samples were harvested 69 days after treatment (Kowite, 1994a). The samples were stored for 105 days at -18°C and then analysed for the first time using GC-FPD method 3, 1994, version 6.0. Results were not corrected for interferences (<0.01 mg/kg) nor for concurrent recoveries (86%-98%). Samples were stored at -18°C for another 125 days (230 days since harvest). Results are shown in Table 46.

Although metabolite mA remained stable during the test period, no information is available for the first 105 days of storage. Therefore no conclusions can be drawn on overall storage stability.

Table 46. Frozen storage stability of incurred ethoprophos and metabolite mA in cucumbers stored at -18°C .

Storage (days)	mg/kg parent	mg/kg mA	% mA remaining	concurrent recovery % parent; mA
105	<0.01 (3); mean <0.01	0.047; 0.060; 0.078; mean 0.062	100 ¹	89; 86
111	<0.01 (3); mean <0.01	0.058; 0.071; 0.080; mean 0.070	113	96; 89
230	<0.01 (3); mean <0.01	0.055; 0.061; 0.079; mean 0.065	105	98; 90

¹ % remaining set at 100% at 105 days of storage; storage stability for the first 105 days not known.

Study 6. Potato and tomato samples were fortified with ethoprophos at 0.10 mg/kg (Uceda, 2004). Duplicate spiked samples were stored at -18°C and analysed for ethoprophos at day 0, and 2, 12 and 19 months using method AR 52-87, 2003a, GC-MS. Results, uncorrected for concurrent recoveries (82%-111%) or interferences (<0.01, n=4) are shown in Table 46A.

Table 46A. Frozen storage stability of 0.1 mg/kg ethoprophos on potato and tomato stored at -18°C .

Sample	Storage period	Remaining % mean range	Concurrent recovery % at 0.1 mg/kg
Potato	0 day	98 95-100	-
	2 months	88 78-84	82
	12 months	70 59-84	85, 78
	19 months	83 80-86	111
Tomato	0 day	88 87-89	-
	2 months	98 96-100	94
	12 months	96 91-101	94
	19 months	88 84-93	102

USE PATTERN

Ethoprophos is an organophosphorus insecticide and nematicide registered in over 58 countries for use on a wide variety of crops and field-grown ornamentals. The crops selected for the 2004 JMPR review encompass the ones, which represent the highest market segments worldwide (strawberry, banana, pepper, tomato, melon, cucumber, potato, sweet potato, and sugar cane). Other uses of ethoprophos, not evaluated here, are on fruit trees (e.g. citrus, orange, lemon, grapefruit, apple, pear, peach, mulberry), fruit and vine crops, pistachios, olives, passion fruit, pineapple, tree tomato, radish, garlic, red onion, onion, aubergine, courgette, okra, baby marrows, butternuts, patti pans, head cabbage, cauliflower, endive, lettuce, spinach, artichokes, asparagus, leeks, peas, chickpeas, beans, green beans, French beans, snap beans, Lima beans, field beans, soya beans, peanuts, cotton, African oil palm, cereals (wheat, barley, rye, triticale, oats, spelt), sweet corn, maize (corn), field corn, rice, sorghum, coffee plants, grass, beets, sugar beets, tobacco, pine trees, horticulture (flowers, ornamentals, rose, chrysanthemum, carnation) and nurseries (fruit trees, olives, citrus).

Ethoprophos is a non-systemic, contact product to be used exclusively as a soil treatment. The use of any of the ethoprophos formulated products (GR, MG, EC, gel) must be followed by thorough soil incorporation (soil depth down to about 20 cm in some cases). Incorporation can be achieved either by watering-in (for banana for example) or by mechanical incorporation using suitable tillage equipment. The incorporation step is mandatory to enhance the effectiveness of the product as it is used to eradicate mainly soil-dwelling insects and nematodes.

Granular formulated products are applied either at pre-planting, or pre emergence or planting stages. For the crops for which a transplanting step is required (such as fruiting vegetables), ethoprophos is applied before or at the transplanting stage. For perennial crops such as banana and sugar cane, ethoprophos can be applied during the vegetative stages of the crops. Application of granular formulations is usually restricted to one application per growing cycle except banana to which the product can be applied twice a year. The granular product is applied either as a band or as an overall application.

Emulsifiable concentrates can be applied either at the pre-plant/planting stages (usually once) or post-planting/transplanting stages. In the latter case the liquid formulation is applied through the drip irrigation water, when the number of application can be up to 4 –5 per growing cycle depending on the crop.

Emulsifiable concentrate gel products are registered only in Mexico. The product is packaged as a water-soluble bag to be diluted in the proper amount of water before spraying using conventional ground equipment. Gel application is usually performed either at planting (potato) or post-planting (cucumber).

Tables 47 to 56 outline the current world-wide use patterns for ethoprophos. The manufacturer provided the GAP data in summarized form, as original labels and as English translations. No labels were available for Tunisia or Algeria, but they are the same as for Morocco. No labels were available for Ireland, but they are the same as for the UK.

For some crops ethoprophos is applied out either overall (broadcast over the entire surface) or as a band (also referred as row application). In the USA (except for banana/plantain) the maximum number of applications is restricted to 1 per year for the investigated crops. For band application, the dose rate per hectare can be expressed either on the entire surface area or the treated area. The calculation of the dose rate based upon the treated area is performed according to the label's instructions (bandwidth, row spacing) whenever possible. If overall or band is not specified on the label, the method of application is referred to as "soil treatment".

Application rates for bananas are given only as g ai/tree. For fruiting vegetables with edible peel (cucumber, sweet pepper, tomato) the PHI specified on the labels is either 30 days or 60 days. A PHI of 30 days does not seem relevant for a fruiting crop if the application is either pre-planting or at planting.

Table 47. Registered uses of ethoprophos on strawberries.

Country	Site	Formulation (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No	PHI (days)
Austria	F/I	GR 100	Soil treatment	6.0	na	ns	ns
Spain	F/I ¹	EC 200	Soil treatment (15 days before transplanting)	6.0	ns	1	60

F: open field; I: indoor, may be greenhouse or plastic cover;

na: not applicable, direct treatment with granular formulation

ns: not stated on label

¹ almost all commercial farms in Spain cultivate strawberries under plastic tunnels. On the label no distinction is made between indoor and outdoor crops (Barriere, 2004a)

Table 48. Registered uses of ethoprophos on bananas and plantains in the field.

Country	Formulation (g ai/kg; g ai/l)	Application method	Dose rate g ai/tree	Spray conc. kg ai/hl	No; (interval, days)	PHI (days)
Brazil	GR 100	Half moon around the stem	3.0	na	2	3

Country	Formulation (g ai/kg; g ai/l)	Application method	Dose rate g ai/tree	Spray conc. kg ai/hl	No; (interval, days)	PHI (days)
		(at beginning and end of rainy season)				
Cameroon	MG 100	Around the stem (0.50 m radius)	4.5 (57 kg ai/treated ha)	na	2-3; reapply every 4-6 months	ns
Central America ¹	GR 100; GR 150; EC 720	Soil treatment (at beginning of rainy season)	2.9-3.0	na/ns	1	30
Columbia	GR 50	Around the stem (0.75 m radius)	4.0 (23 kg ai/treated ha)	na	2; reapply every 6 months	ns
Columbia	GR 100	Around the stem (0.75 m radius)	3.0 (17 kg ai/treated ha)	na	2; reapply every 6 months	ns
Columbia	GR 110 Biodac	On the shoot about to sprout	3.3	na	3; reapply every 4 months	ns
Columbia	GR 150 Biodac	Around the stem (0.40 m radius)	3.0 – 3.8 (60-75 kg ai/treated ha)	na	3; reapply every 4 months	ns
Ecuador	GR 150 Biodac	Soil treatment	3.0	na	2 - 3	0
France	MG 100	Around the stem	4.0-4.5	na	ns	ns
Côte d'Ivoire	GR 200	Around the stem (0.30-0.40 m radius)	4.0-8.0 (80-283 kg ai/treated ha)	na	2-3	ns
Morocco*	GR 100	Overall application	5.0	na	ns	ns
Morocco	EC 200	Drip irrigation (during growing season: 1 st at planting)	2.0 kg ai/ha (max 10 kg ai/ha)	ns	5 (7 days interval)	30
Peru	GR 150	Soil treatment	3.0 – 4.5	na	1	7
Philippines @	GR 100	Around the stem (0.75 m radius)	4.0 – 5.0 (23-28 kg ai/treated ha)	na	2 (6 months)	ns
Portugal (Madeira)	GR 100	Around the stem (0.30 – 0.80 m radius)	2.0 – 4.5 (22-71 kg ai/treated ha)	na	2 (reapply every 6 months)	56
Spain ²	EC 200	Drip irrigation	3.0 – 3.6	ns	1	60
Spain ²	EC 200	Flood irrigation	5.0 – 6.0	ns	1	60
USA	GR 150	Around the stem (0.75 m radius)	6.0 (34 kg ai/treated ha)	na	2 (180)	ns
USA	EC 200	Around the stem (0.75 m radius)	5.8 (33 kg ai/treated ha)	ns	2 (180)	ns
Venezuela	GR 100	Around the stem (0.40 m radius)	4.0	na	2 - 3	30
Venezuela	GR 150	At sowing (in the plant hole) or around the stem for established crops	2.6-3.0	na	2-3	30

na: not applicable, direct treatment with granular formulation

ns: not stated on the label

*: no printed label or registration certificates available (confirmed by manufacturer)

@: There is no available commercial label for the Philippines, because the product is imported from the USA. Values listed in the Table are from the blueprint.

¹ Central America includes Guatemala, El Salvador, Honduras, Nicaragua, Costa Rica, Panama, Dominican Republic and Belize.

² Bananas are cultivated on a commercial basis in the Canary Islands. About 20% of the cultivated surface is under protected covers (plastic or mesh) while the remaining 80% is open field (Barriere, 2004a).

Table 49. Registered uses of ethrophosphos on vegetables (without further specification).

Country	Site	Formulation (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No	PHI (days)
Austria	F/I	GR 100	Soil treatment	6.0	na	ns	ns
Algeria ^{1*}	I	GR 100	Soil treatment	5.0	na	2-3	ns

Country	Site	Formulation (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No	PHI (days)
			(application period ns)				
Cameroon	ns	MG 100	Overall application (pre planting/pre-transplanting)	4.5	na	1	ns
Cameroon	ns	MG 100	Band application ¹ (pre planting / pre-transplanting)	- (4.5 kg ai/treated ha)	na	1	ns
Chile	F/I	EC 720	Overall application (pre planting)	3.6 – 8.6	ns	1	ns
France	F	EC 200	Overall application (10 days before planting upto planting)	4.0-10	0.50 – 1.7	1	ns
Greece	F	GR 100	Overall application (a few days before - at sowing/ 1 week before-at transplanting)	6.0 – 8.0	na	1	60
Greece	F	EC 720	Drip irrigation	7.2	ns	1	60
Morocco ¹ *	ns	GR 100	Soil treatment (localised) (application period ns)	10	na	ns	ns
Tunisia ¹ *	ns	GR 100	Soil treatment (application period ns)	5.0 - 10	na	ns	ns
Venezuela	F	GR 100	Band application (0.40-0.50 m wide) 1-3 weeks pre-planting	2.0-2.5	na	1	30

F: open field

I: indoor, may be greenhouse or plastic cover

na: not applicable, direct treatment with granular formulation

ns: not stated on label

* no printed label or registration certificates available (confirmed by manufacturer)

¹ Examples of vegetables, mentioned on label: peppers, cucurbits, cabbages.

Table 50. Registered uses of ethoprophos on cucumber.

Country	Site	Form. (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No (inter- val, days)	PHI (days)
Central America ¹	F	GR 100; GR 150; EC 720	Soil treatment (at planting)	2.0 – 4.2	na/ns	1	30
Italy	F/I	GR 100	Overall application (one week pre planting)	3.0 – 10	na	1	30
Italy	F/I	GR 100	Band application (one week pre planting)	- (3.0 – 10 kg ai/treated ha)	na	1	30
Italy	F	EC 172.9	Overall application (pre-sowing / pre-planting)	6.9 – 8.6	0.069- 0.43	1	30
Italy	F	EC 172.9	Band application (pre-sowing / pre-planting)	- (6.9 – 8.6 kg ai/treated ha)	0.069- 0.43	1	30
Côte d'Ivoire	ns	GR 200	Overall application (at planting / at transplanting)	6.0 – 12	na	1	ns
Côte d'Ivoire	ns	GR 200	Band application (width 0.35-0.65 m) (at planting / at transplanting)	- (6.0 – 12 kg ai/treated ha)	na	1	ns
Mexico	F	GR 150	Band application ² (1 week before transplanting up to transplanting)	2.0 – 3.0	na	1	ns
Mexico	F	gel 720	Soil treatment (at planting)	2.2	ns	1	ns

Country	Site	Form. (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No (interval, days)	PHI (days)
Mexico	F	gel 720	Soil treatment (1 st after seedtime until crop has 2 true leaves)	2.2	ns	1-2 (15-20)	ns
Portugal	F/I	GR 100	Overall application (pre planting)	8.0	na	1	56
Spain	F/I	GR 100	Overall application (pre-sowing or pre- transplanting)	6.0 – 8.0	na	1	60
Spain	F/I	EC 200	Soil treatment (15 days before planting/transplanting)	6.0	ns	1	60
Spain	F/I	EC 200	Drip irrigation (post transplanting)	0.6 (max total 6.0)	ns	1-10 (7-10)	60
USA	ns	GR 150	Band application ³ (pre planting/ at planting)	2.2 (12-15 kg ai/treated ha)	na	1	ns
USA	ns	EC 720	Band application ³ (pre planting/at planting)	1.7 (9.7 – 12 kg ai/treated ha)	ns	1	ns

F: open field

I: indoor, may be greenhouse or plastic cover

na: not applicable, direct treatment with granular formulation

ns: not stated on the label

¹ Central America includes Guatemala, El Salvador, Honduras, Nicaragua, Costa Rica, Panama, Dominican Republic and Belize.

² Band 0.30-0.40 m wide, 1.5-2.0 m row spacing.

³ Band 0.30-0.38 m wide, 2.1 m row spacing

Table 51. Registered uses of ethrophosphos on sweet and chili peppers.

Country	Site	Form. (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No; (interval, days)	PHI (days)
Central America ¹	F	GR 100; GR 150; EC 720	Soil treatment (at planting)	2.0 – 4.2	na/ns	1	30
Italy	F/I	GR 100	Overall application (one week pre planting)	3.0 – 10	na	1	30
Italy	F/I	GR 100	Band application (one week pre planting)	- (3.0 – 10 kg ai/treated ha)	na	1	30
Italy	F	EC 172.9	Overall application (pre-sowing / pre-planting)	6.9 – 8.6	0.069- 0.43	1	30
Italy	F	EC 172.9	Band application (pre-sowing / pre-planting)	- (6.9– 8.6 kg ai/treated ha)	0.069- 0.43	1	30
Italy (green pepper)	F	EC 172.9	Drip irrigation (post transplanting)	1.7 – 3.5 (max total 8.6)	ns	3-4 (20-30)	30
Côte d'Ivoire*	F	GR 200	Overall/band application (at planting)	9.0	na	1	ns
Korea (red pepper)	F/I ²	GR 50	Overall application (1 week before transplanting)	4.0-5.0	na	1	ns
Peru (chili pepper)	F	GR 150	Overall application (pre transplanting)	3.8 – 4.5	na	1	7
Portugal	F	GR 100	Overall application (pre planting)	8.0	na	1	56
Spain	F/I	GR 100	Overall application (pre-sowing/ pre- transplanting)	6.0 – 8.0	na	1	60
Spain	F/I	EC 200	Soil treatment (15 days before	6.0	ns	1	60

Country	Site	Form. (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No; (interval, days)	PHI (days)
			sowing/transplanting)				
Spain	F/I	EC 200	Drip irrigation (post transplanting)	0.6 (max. total 6.0)	ns	1-10 (7-10)	60
Thailand (pepper)	F	GR 100	Around the stem, followed by watering	40	na	1	ns

F: open field; I: indoor: may be greenhouse or plastic cover

na: not applicable, direct treatment with granular formulation

ns: not stated on the label

*: no printed label or registration certificates available (confirmed by manufacturer)

¹ Central America includes Guatemala, El Salvador, Honduras, Nicaragua, Costa Rica, Panama, Dominican Republic and Belize.

² In Korea, red peppers are cultivated both in open field and in vinyl houses.

Table 52. Registered uses of ethoprophos on tomatoes.

Country	Site	Formulation (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No; (interval, days)	PHI (days)
Central America ¹	F	GR 100; GR 150; EC 720	Soil treatment (at planting)	2.0 – 4.2	na/ns	1	30
Chile	F/I	EC 720	Overall application (pre planting)	3.6 – 8.6	ns	1	ns
Columbia	F/I	GR 50; GR 100	Band application (width 0.45-0.60 m) (pre planting until at transplanting)	6.8-9.0 (12.5–19 kg ai/treated ha)	na	1	ns
Columbia	F/I	GR 50; GR 100	Overall application (pre planting until at transplanting)	6.8 - 9.0	na	1	ns
Ecuador	F	EC 69.6	Soil treatment (1 week before transplanting)	0.56-0.70	ns	1	90
France	F	EC 200	Overall application (10 days before planting upto planting)	4.0-10	0.50 – 1.7	1	ns
Italy	F/I	GR 100	Overall application (one week pre planting)	3.0 – 10	na	1	30
Italy	F/I	GR 100	Band application (one week pre planting)	- (3.0 – 10 kg ai/treated ha)	na	1	30
Italy	F	EC 172.9	Overall application (pre-sowing / pre-planting)	6.9 – 8.6	0.069-0.43	1	30
Italy	F	EC 172.9	Band application (pre-sowing / pre-planting)	- (6.9 – 8.6 kg ai/treated ha)	0.069-0.43	1	30
Italy	F	EC 172.9	Drip irrigation (post transplanting)	1.7 – 3.5 (total max 8.6)	ns	3-4 (20-30)	30
Côte d'Ivoire	ns	GR 200	Overall application (at planting / at transplanting)	6.0 – 12	na	1	ns
Côte d'Ivoire	ns	GR 200	Band application (width 0.35-0.65 m) (at planting / at transplanting)	- (6.0 – 12 kg ai/treated ha)	na	1	ns
Morocco	F/I	EC 200	Drip irrigation (during growing season: 1 st treatment 10 days before planting)	5.0 (1 st treatment) 1.0 (later treatments) (total max 10)	ns	2-6 (5)	30
Peru	F	GR 150	Overall application (pre transplanting)	3.8 – 4.5	na	1	7
Portugal	F	GR 100	Overall application	8.0	na	1	56

Country	Site	Formulation (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No; (interval, days)	PHI (days)
			(pre planting)				
Spain	F/I	GR 100	Overall application (pre planting/pre- transplanting)	6.0 – 8.0	na	1	60
Spain	F/I	EC 200	Drip irrigation (during growing season: 1 st treatment 15 days after transplanting)	0.8 – 2.0 (total max 6.0)	ns	several (15-20)	60
Venezuela	F	GR 100	Band application (0.40-0.50 m width) (1-3 weeks pre-planting)	2.0 – 2.5	na	1	30

F: open field

I: indoor, may be greenhouse or plastic cover

na: not applicable, direct treatment with granular formulation

ns: not stated on the label

¹ Central America includes Guatemala, El Salvador, Honduras, Nicaragua, Costa Rica, Panama, Dominican Republic and Belize.

Table 53. Registered uses of ethoprophos on melons.

Country	Site	Formulation (g ai/kg; g ai/l)	Application method	Dose rate kg ai/ha	Spray conc. kg ai/hl	No	PHI (days)
Central America ¹ (watermelon)	F	GR 100; GR 150; EC 720	Soil treatment (at planting)	2.0 – 4.2	na/ns	1	30
Ecuador (cantaloupe)	F	GR 150 Biodac	Soil treatment (at planting/transplanting)	0.23 – 0.30 g ai/plant	na	1	ns
Italy (melon, watermelon)	F/I	GR 100	Overall application (one week pre planting)	3.0 – 10	na	1	30
Italy (melon, watermelon)	F/I	GR 100	Band application (one week pre planting)	- (3.0 – 10 kg ai/treated ha)	na	1	30
Italy (melon, watermelon)	F	EC 172.9	Overall application (pre-sowing / pre-planting)	6.9 – 8.6	0.069-0.43	1	30
Italy (melon, watermelon)	F	EC 172.9	Band application (pre-sowing / pre-planting)	- (6.9 – 8.6 kg ai/treated ha)	0.069-0.43	1	30
Côte d'Ivoire	ns	GR 200	Overall application (at planting / at transplanting)	6.0 – 12	na	1	ns
Côte d'Ivoire	ns	GR 200	Band application (width 0.35-0.65 m) (at planting/at transplanting)	- (6.0 – 12 kg ai/treated ha)	na	1	ns
Portugal	F	GR 100	Soil treatment (pre planting)	8.0	na	1	56
Peru (melon, watermelon)	F	GR 150	Overall application (pre transplanting)	3.8 – 4.5	na	1	7

F: open field

I indoor, may be greenhouse or plastic cover

na: not applicable, direct treatment with granular formulation

ns: not stated on the label

¹ Central America includes Guatemala, El Salvador, Honduras, Nicaragua, Costa Rica, Panama, Dominican Republic and Belize.