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Persistence, Degradation, and Distribution of Deltamethrin in an Organic Soil under Laboratory Conditions

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Deltamethrin [(*S*)-α-cyano-3-phenoxybenzyl *cis*-(1*R*,3*R*)-2,2-dimethyl-3-(2,2-dibromovinyl)cyclopropanecarboxylate] labeled with ^{14}C at the methyl or benzylic position was applied to an organic soil at 10 mg/kg in a laboratory incubation study. A steady decrease of extractable ^{14}C residues was accompanied by a corresponding increase of bound ^{14}C residues over a 180-day incubation period. The degradation of deltamethrin was slower under the anaerobic than aerobic condition. The insecticide was degraded in the organic soil by hydrolysis at the ester linkage followed by the formation of oxidative products. A small proportion of bound ^{14}C residues in the incubated soil was identified as 3-(2,2-dibromovinyl)-2,2-dimethylcyclopropanecarboxylic acid, whereas the remainder constituted unidentified products. The soil containing bound ^{14}C residues (19% of initial ^{14}C applied) was fractioned into humic substances. The humin fraction contained an appreciable amount of bound ^{14}C residues.

Deltamethrin [(*S*)-α-cyano-3-phenoxybenzyl *cis*-(1*R*,3*R*)-2,2-dimethyl-3-(2,2-dibromovinyl)-2,2-dimethylcyclopropanecarboxylate] is a pyrethroid that is also known as decamethrim and NRDC 161. The insecticide is active against a wide range of insects that attack crops, animals, and man. While there is ample information available in the literature about its photochemistry (Ruzo et al., 1977) and metabolism in plants (Ruzo and Casida, 1979) and animals (Ruzo et al., 1978, 1979), little is known about its persistance and degradation in soil. The limited published work reveals that deltamethrin is immobile in mineral soils (Kaufman et al., 1981; Hill, 1983) and a half-life in the range of 1–8 weeks has been reported (Chapman et al., 1981; Miyamoto and Mikami, 1983; Hill, 1983).

Deltamethrin has been recommended for foliar applications on various vegetable and field crops (FAO, 1981). It is possible that this insecticide will be found useful to control numerous species of insects in vegetable crops grown in organic soils that occur widely in eastern Canada and are of great economic value for vegetable production.

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Pesticides are applied to organic soils in doses usually higher than those of mineral soils. The high carbonaceous and water contents of organic soils cause adsorption and dilution of pesticides that may result in decreased bioactivity and increased persistance of the additive. Since in most foliar applications a large portion of the chemical may come in contact with the soil, it is necessary to determine its fate in the soil environment. The purpose of this investigation was to examine in laboratory incubation experiments the persistence, degradation, and bound residue formation of deltamethrin in an organic soil.

MATERIALS AND METHODS

Chemicals. Deltamethrin (^{14}C labeled and unlabeled) was a gift from Roussel-Uclaf-Procida through its subsidiary Hoescht of Canada Ltd. The radiochemical purity (determined by thin-layer chromatography) and specific activity of methyl-labeled and benzylic-labeled deltamethrim was 98 and 96% and 56 and 58 mCi/mmol, respectively. A portion of methyl-labeled and benzylic-labeled material was individually mixed with purified deltamethrin and dissolved in acetone to give a concentrations of 484 μg/mL (4.64×10^5 dpm) and 483 μg/mL (5.43×10^5 dpm), respectively. Base-catalyzed hydrolysis of deltamethrin followed by thin-layer chromatography purification gave pure 3-(2,2-dibromovinyl)-2,2-dimethylcyclopropanecarboxylic acid (Br_2CA). 3-Phenoxybenzaldehyde (PBald), 3-phenoxybenzoic acid (PBacid), and

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3-phenoxybenzyl alcohol (PBalc) were purchased from Aldrich Chemical Co., Milwaukee, WI. 3-(4-Hydroxyphenoxy)benzoic acid (HO-PBacid) and 3-(4-hydroxyphenoxy)benzyl alcohol (HO-PBalc) were prepared following the procedure of Unai and Casida (1977).

Soil. Soil was collected from the 0–15-cm layer of a field at the St. Clotide experimental farm (Quebec). The soil was air-dried, mixed, and passed through 2-mm screen before use. The soil contained 16.8% mineral matter, 40.6% carbon, and 2.6% nitrogen. The field capacity of the soil was 300% and the pH was 5.7.

Aerobic Incubation Experiment. Soil samples (30 g on an oven-dry weight basis) were placed in Erlenmeyer flasks (500 mL) to which 500 μL of hexane containing labeled (0.5 μCi) and unlabeled deltamethrin was added to give an insecticide concentration of 10 mg/kg. The solvent was allowed to evaporate and the soil was thoroughly mixed. The moisture content of the soil was adjusted to 70% of field capacity. A scintillation vial containing 10 mL of 0.1 M KOH was placed in each soil flask, and an Ascarite vial was attached to the flask as described by Loss et al. (1980). The flasks were incubated at 21 ± 1 °C in the dark until required. Distilled water was added as necessary to maintain the initial moisture content of the soil samples. The flasks containing methyl-labeled treated soil were removed at intervals during a period ranging from 0 to 6 months whereas the benzylic-labeled treated samples were removed only at the end of a 90- and 180-days incubation period. The evolved trapped $^{14}CO_2$ was determined and soil samples were then analyzed. In addition, microbial population was determined by the plate count method. Soil extract actidione agar was used for counting bacteria and actinomycetes, and Rose Bengal–streptomycin agar used for counting fungi (Pramer and Schmidt, 1964).

Anaerobic Incubation Experiment. An Erlenmeyer flask containing treated soil as described above was placed in Gas Pack anaerobic jars, and the lid was secured (Willis, 1969). The jar was purged with a stream of nitrogen, and then a steady flow of hydrogen was maintained for 10–15 min, the top was then closed, and the jar was incubated at 21 ± 1 °C. At the end of the incubation period the flask was removed and processed as before.

Determination of Extractable Residues. Preliminary experiments indicated that nearly quantitative recoveries of deltamethrin were obtained from the fortified samples when the soil had a moisture content at about 200% prior to extraction with solvent. Therefore, no attempt was made to air-dry the soil before extraction. Soil removed at intervals was extracted with 125 mL of a hexane–acetone mixture (1:1) in a mechanical shaker for 3 h, and the extract was filtered under suction. The extraction was repeated 1 more time and filtered under suction into the same flask. The soil residue was then washed with the extracting solution (3 × 50 mL) and combined with the filtrate. The soil was then transferred in an Erlenmeyer flask, extracted with 125 mL of acidic methanol (25% HCl), and filtered under suction into a separate flask. This was followed by washing the soil with acidic methanol (3 × 40 mL), and the washes were combined with the methanol extract. The hexane–acetone or acidic methanol extract obtained above was concentrated to a small volume and ^{14}C determined. Further extraction of soil with the solvent system did not remove any measurable ^{14}C residues. Extraction of the incubated soil with acetonitrile, acetone, methanol, or hexane did not increase the extraction efficiency. Thus, the procedure employed in this study was considered effective in removing all the extractable ^{14}C residues from the soil. The extracts were partitioned between hexane and aqueous acetone. Aliquots of the hexane phase were used for the analysis of deltamethrin by gas chromatography (GC) and for ^{14}C by liquid scintillation counting (LSC). The remaining portion of hexane phase was taken to just dryness, and the residue was dissolved in a small volume of acetone and analyzed by TLC. The aqueous acetone phase containing 0.5% of the initially applied ^{14}C was discarded. Residual solvent remaining in the extracted soil was allowed to evaporate, and the soil was stored for determining the nonextractable (bound) ^{14}C residues as described later.

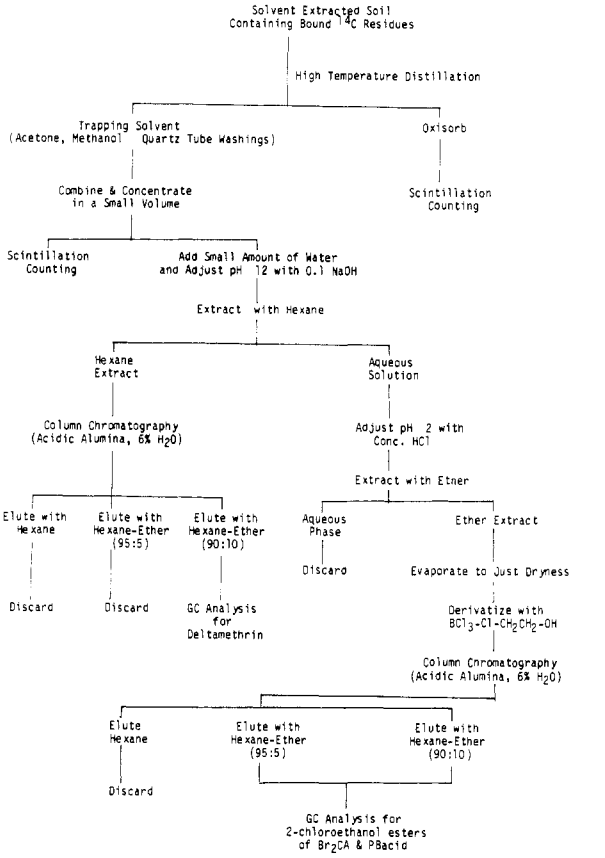

Figure 1. Schematic diagram for the analysis of nonextractable (bound) residue from soil treated with [^{14}C]deltamethrin.

Determination of Nonextractable (Bound) Residues. A portion of the extracted soil was combusted to $^{14}CO_2$ for determining the total bound ^{14}C residues. The other portion was subjected to the high-temperature distillation (HTD) technique as described earlier (Khan and Hamilton, 1980). Acetone and methanol were used as the trapping solutions for the materials purged with helium during HTD. The quartz tube was first washed with acetone and then with methanol, and the washings were combined with the trapping solutions. The material in different traps was then processed as depicted in Figure 1.

Fractionation of Extracted Soil. The extracted soil was fractionated into humic materials by the method described in a previous publication (Khan, 1982).

Determination of Radioactivity. Combustion of dried soil or humic materials was done in a Packard sample oxidizer, Model 306, to produce $^{14}CO_2$. Aliquots of various extracts or solution described above were analyzed by LSC as described earlier (Khan and Hamilton, 1980).

Chromatography and Analysis. Analytical and preparative TLC was performed with 20 × 20 cm precoated silica gel GF chromatoplates with 0.25- and 1.0-mm

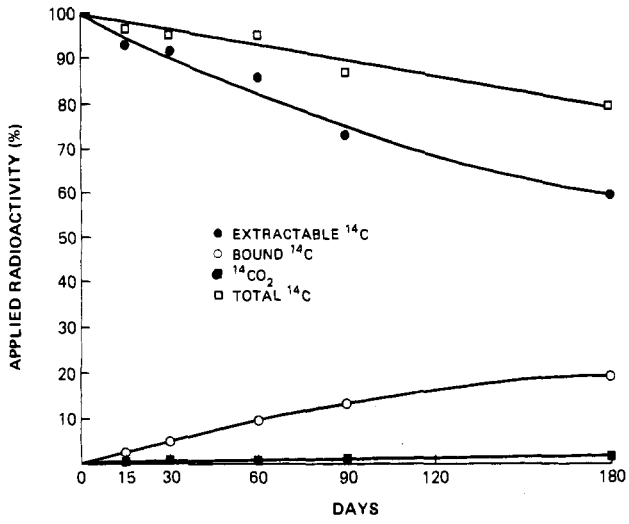

Figure 2. Extractable and bound ^{14}C residues in an organic soil aerobically incubated with methyl-labeled [^{14}C]deltamethrin for 180 days.

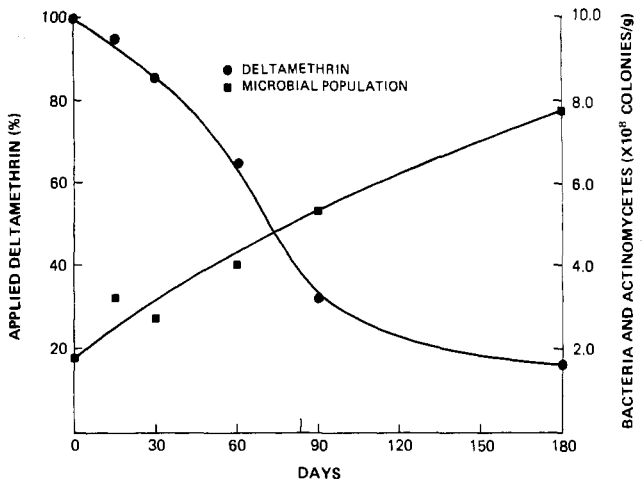

Figure 3. Persistence of deltamethrin and microbial population in an organic soil aerobically incubated for 180 days.

gel thickness (Analtech, Inc., Newark, DE), respectively. The solvent systems used were (A) ethyl acetate–formic acid–water (35:2:2), (B) toluene (saturated with formic acid)–diethyl ether (10:3), (C) toluene–hexane–acetic acid (15:3:2), (D) benzene–carbon tetrachloride (1:1), and (E) A × B, indicating development in the first direction with solvent system A and in the second direction with solvent system B. Unlabeled reference chemicals were detected by ultraviolet fluorescence quenching. Radioactive spots were detected by radioautography and scraped free from the glass support for LSC. The products were recovered by extraction of the gel with suitable solvents and analyzed by GC and TLC and finally by cochromatography with unlabeled standards. The fractions containing Br_2CA and PBacid were derivatized to their 2-chloroethanol esters prior to GC.

Column chromatography was carried out with a glass column (20 cm × 0.8 cm) containing 4 g of deactivated acidic alumina (6% H_2O) packed in hexane. A 0.5-mL sample was applied to the column and eluted with 4 mL of hexane, 10 mL of hexane–ethyl ether (19:1), and 10 mL of hexane–ethyl ether (9:1). Each collected eluate was concentrated by evaporation and analyzed by TLC and GC.

The gas chromatograph was a Varian Model 3700 equipped with a ^{63}Ni detector. Two columns consisting of a 2.5 m × 0.25 cm i.d. glass tube packed with 3% SE-30 on Chromosorb WHP and 3% OV-210 on Gas-Chrom Q were used. For the analysis of deltamethrin and the isomer (αR)-deltamethrin, the SE-30 and OV-210 columns were operated at 240 and 230 °C with nitrogen carrier gas at a flow rate of 40 and 30 mL/min, respectively. The analysis of 2-chloroethanol esters of Br_2CA and PBacid were carried out by operating the OV-210 column isothermally at 130 and 170 °C, respectively, or programming the SE-30 column from 130 to 210 °C at the rate of 8 °C/min. The nitrogen carrier flow rate, injector port temperature, and detector temperature were 30 mL/min, 210 °C, and 340 °C, respectively.

RESULTS AND DISCUSSION

The amounts of extractable ^{14}C residues recovered from the methyl-labeled deltamethrim treated soil decreased over an aerobic incubation period of 180 days (Figure 2). This, in turn, corresponded to an increase in the formation of soil-bound ^{14}C residues and a very small increase in the release of $^{14}CO_2$. Thus, by the end of the incubation period, extractable ^{14}C residues decreased to 59.0% while the bound ^{14}C residues increased to 19.2% of the initially added ^{14}C. The amounts of $^{14}CO_2$ released from the methyl- or benzyl-labeled deltamethrin during the incubation period were small (0.23–1.5%), thereby indicating very little mineralization occurred in soil. The total radiocarbon initially added to the organic soil could not be accounted for only by extractable ^{14}C, bound ^{14}C, and $^{14}CO_2$. It is possible that this loss in radiocarbon occurred due to the formation of some volatile degradation products during the incubation.

The degradation of methyl-labeled deltamethrin in the organic soil was compared under aerobic and anaerobic conditions after the 180-day incubation period. The degradation of deltamethrin appeared to be slower under the anaerobic condition as indicated by the presence of about 87% extractable ^{14}C of that initially added. Furthermore, the amount of radioactivity present in bound form and $^{14}CO_2$ was 3.3 and 0.21%, respectively.

Figure 3 shows the persistence of deltamethrin (determined by GC) in the aerobically incubated organic soil. The insecticide degraded less rapidly after 90 days of incubation. Thus, the time required for reducing the concentration of deltamethrin to half of the original value was found to be 72 days. For indoor-incubated mineral soils half-lives of deltamethrin in the range of 23–34 days have been reported (Miyamoto and Mikami, 1983; Hill, 1983). Thus, deltamethrin appears to be less susceptible in organic soil than in mineral soil. Similar observations have been reported by Chapman et al. (1981). The data also demonstrate that the disappeance of deltamethrin was more rapid in the aerobic than anaerobic incubated samples. Thus, by the end of the 180-day incubation period, the recoveries of deltamethrin, as determined by GC, under the aerobic and anaerobic conditions were 16 and 82%, respectively, further indicating that microbial action was involved in the deltamethrin disappearance.

The data show (Figure 3) that the bacteria and actinomycetes number increased steadily in the treated soil, and at the end of the incubation period the increase over the initial number was by a factor of approximately 4. No appreciable changes in numbers of bacteria and actinomycetes occurred in the incubated untreated soil. Furthermore, the fungal population remained relatively unaffected during the incubation period (6.1×10^4–6.6×10^4 numbers/g).

The relative amounts of the extractable degradation products obtained from the aerobic incubation of deltamethrin in organic soil are shown in Table I. The products

Table I. Extractable Degradation Products of [^{14}C]Deltamethrin Aerobically Incubated with an Organic Soil

	% of the applied radioactivity			
	90-day incubation		180-day incubation	
compd	methyl label	benzylic label	methyl label	benzylic label
deltamethrin	32.9	24.7	21.4	15.4
Br_2CA	29.6		17.4	
PBacid		8.3		1.8
PBald		1.9		1.4
PBalc		0.2		0.3
HO-PBacid		0.2		0.2
HO-PBalc		<0.1		<0.1

were identified and quantitated by a combination of preparative and analytical TLC, cochromatography, radioautography, and GC of the derivatized products. The main metabolites were Br_2CA and PBacid. These two metabolites along with PBald decreased with time and appear to have no tendency to accumulate in the organic soil. Some other unknown radiolabeled products that appeared on TLC plates could not be identified because of their low levels and unavailability of the reference standards. The data show that the degradation pathway of deltamethrin in the organic soil follows hydrolysis at the ester linkage. The compounds thus produced undergo further degradation, resulting in the formation of oxidative products, such as HO-PBacid, HO-PBalc, and PBald. The latter were present only in trace amounts (Table I). Contrary to the observations reported for phenothrin (Nambu et al., 1980) and permethrin (Kaneko et al., 1978), very little oxidation of the benzene ring was noted in this study for deltamethrin. Roberts and Standen (1977) also reported that phenyl ring oxidation was the minor degradation route for the metabolism of a structurally similar insecticide, cypermethrin.

In a recent report Hill (1983) observed that (αS)-deltamethrin was converted to the αR epimer in a mineral soil to a very small extent (2–5% of the applied material). However, no such conversion of αS to αR occurred in the incubated organic soil as evidenced by the GC analysis of the extracts. Miyamoto and Mikami (1983) also observed no $\alpha S/\alpha R$ interconversion in other mineral soils.

The identity of the unextracted or bound ^{14}C residues in the organic soil was determined by the HTD technique (Khan and Hamilton, 1980). In preliminary experiments it was observed that HTD of the reference standard of [^{14}C]deltamethrin resulted in a recovery of about 87–91% radioactivity in the distillates. Furthremore, HTD of air-dried extracted control soil to which [^{14}C]deltamethrin (10 ppm) was added resulted in 80–84% recovery of the radioactivity. Very little thermal decomposition of [^{14}C]deltamethrin occurred during the HTD analysis (0.3–5.7% $^{14}CO_2$). The HTD distillates were processed as depicted in Figure 1. Analysis of the distillates as described in Figure 1 indicated the presence of deltamethrin only in the form of αR and αS epimers. The total radioactivity recovered by HTD from the extracted soil sample taken after the 180-day incubation was about 35% of the total bound ^{14}C. HTD of the soil containing bound ^{14}C residues under different conditions, such as temperature, helium flow rates, treatment of soil with HCl and H_2O, or different solvents in traps, did not increase the recovery of radioactivity. The remaining radioactivity was found to be present in the coal-like polymerized burned solid material left in the porcelain boat after HTD. The distillates subjected to various cleanup and extraction procedures (Figure 1) were analyzed by TLC and GC to determine the identity of the bound residues. The presence of small amounts (<0.1 ppm) of bound Br_2CA was confirmed by the methods employed. However, due to the analytical difficulties, such as loss of the material due to surface adsorption of pyrethroids (Helmuth et al., 1983; Akhtar, 1982), and interferences from HTD codistillates, the low concentrations of other compounds, which may be present as bound residues, could not be positively identified.

Table II. Distribution of Bound ^{14}C Residues in Humic Materials Fractionated from an Organic Soil Treated with [^{14}C]Deltamethrin

	% of the bound radioactivity	
humic material	methyl label	benzylic label
humic acid	21.7	24.8
fulvic acid	16.8	7.1
humin	58.5	65.6

The organic soil contained 19.2% bound ^{14}C of the total applied radioactivity following an incubation period of 180 days. The bound residues became distributed among the various soil organic matter fractions. The proportions of total bound radioactivity in HA, FA, and humin fractions are shown in Table II. Similar observations have been reported by Roberts and Standen (1977) for bound [^{14}C]cypermethrin, a structurally similar insecticide, distribution in humic materials. They reported that the FA fraction contained small amounts of PBacid, HO-PBacid, and Cl_2CA depending on the radiolable used. These observations are of considerable interest as FA is considered to be the dominant soluble organic fraction present in soil solution under field conditions. It is known that low molecular weight humic materials, such as FA, can be taken up by roots and translocated in plants (Schnitzer and Khan, 1978). Thus, FA-bound residues would be expected to become easily bioavailable to plants. The bound ^{14}C residues in HA appeared to be very stable as only less than 10% ^{14}C was released by HTD.

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Registry No. Deltamethrin, 52918-63-5; (1*R*,3*R*)-3-(2,2-dibromovinyl)-2,2-dimethylcyclopropanecarboxylic acid, 53179-78-5.

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Distribution and Metabolism of *cis*- and *trans*-Resmethrin in Lactating Jersey Cows

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Resmethrin labeled with radiocarbon in either the acid or alcohol moiety and administered orally to lactating Jersey cows at 10 mg/kg was rapidly absorbed, metabolized, and excreted. The cis isomer was eliminated primarily in feces, but the trans isomer was eliminated primarily in urine. Tissue residues at 48 h posttreatment were low (<1 ppm) except in liver and kidney and were generally higher with the alcohol-labeled compounds. Only very low levels of radiocarbon were secreted into milk. Unmetabolized resmethrin appeared in trace amounts in tissue and as the major residue in milk and feces. The major metabolites from both isomers arise from ester hydrolysis and subsequent oxidation of the hydrolytic products and include chrysanthemic acid (free and conjugated with glucuronic acid), chrysanthemumdicarboxylic acid, 5-benzyl-3-furoic acid (free and conjugated with glucuronic acid or glycine), and 5-(α-hydroxybenzyl)-3-furoic acid.

The synthetic pyrethroid insecticide resmethrin is a mixture of (1*RS*)-cis and (1*RS*)-trans isomers of (5-benzyl-3-furyl)methyl 2,2-dimethyl-3-(2-methyl-1-propenyl)cyclopropanecarboxylate. Resmethrin combines very low mammalian toxicity (Verschoyle and Barnes, 1972; Gray and Conners, 1980) with excellent insecticidal activity (Elliott, 1971; Okuno et al., 1969). Previous studies have examined the metabolism of related pyrethroids in ruminants (Gaughan et al., 1977; Ivie and Hunt, 1980) and the metabolism of resmethrin has been studied in vitro (Ueda et al., 1975a) and in the laboratory rat (Miyamoto et al., 1971; Ueda et al., 1975b).

Among the projected uses for resmethrin is its utilization as a space spray for fly control in and around dairy facilities. The current studies were designed to obtain data on the distribution of resmethrin isomers and their metabolites in the body tissues, excreta, and milk of orally dosed lactating cattle. Remethrin metabolites have been resolved, quantitated, and identified where possible.

MATERIALS AND METHODS

Chemicals. Radiocarbon-labeled preparations of resmethrin (RES) were supplied by the S. B. Penick Corp. (Lyndhurst, NJ) as follows: acid [^{14}C]-(1*RS*)-*cis*-resmethrin (CAC) and acid [^{14}C]-(1*RS*)-*trans*-resmethrin (TAC), both labeled in the carbonyl group of the acid moiety, and alcohol [^{14}C]-(1*RS*)-*cis*-resmethrin (CAL) and alcohol [^{14}C]-(1*RS*)-*trans*-resmethrin (TAL), both labeled in the carbon 2 position of the furan ring. Analysis of each preparation by gas–liquid chromatography/mass spectroscopy (GLC/MS) showed each of the chemicals to be of >98% isomeric purity. Thin-layer chromatography (TLC) in several systems (B–G) (vide infra) showed each preparation to be of >96% radiochemical purity. The chemical identitites of the isomers were verified by mass spectroscopy. Unlabeled samples of (*RS*)-*cis*- and (*RS*)-*trans*-resmethrin and certain analogues for use as possible metabolite standards were also supplied by the Penick Corp. and included 5-benzyl-3-furoic acid (BFCA), (5-benzyl-3-furyl)methanol (BFA), *cis*-chrysanthemic acid (*c*-CA), and *trans*-chrysanthemic acid (*t*-CA).

Chromatography. Initial resolution of resmethrin metabolities from all samples was accomplished by TLC, using precoated silica gel plates (0.25 mm gel thickness, 20 × 20 cm, with fluorescent indicator, Brinkmann) and various combinations of solvent systems as follows: (A) benzene (saturated with formic acid)–tetrahydrofuran (10:1), developed 3 times; (B) benzene (saturated with formic acid)–ether (10:3), developed 2 times; (C) hexane–ether (5:1); (D) carbon tetrachloride–hexane–ether (8:1:1), developed 2 times; (E) benzene–ethyl acetate–methanol (6:1:1); (F) carbon tetrachloride–ether (3:1); (G) benzene–hexane (1:1); (H) 1-propanol–acetic acid–water (6:1:1); (I) methanol–water–acetic acid (8:3:1).

High-performance liquid chromatography (HPLC) was used in some cases to further resolve and chromatographically characterize the metabolites of the resmethrin isomers. A Waters Associates Model 440 instrument with UV detector at 254 nm and Waters chromatography pump, Model M-6000, were used with a 5-μm Supelco C-18 column (15-cm length, 4.6 mm i.d., Supelco, Inc.) preceded by a Whatman C-18 guard column (Pierce Chemical Co.). The mobile phase was varied for optimum resolution of the various metabolites and included (HPLC-A) acetonitrile–water–formic acid (30:70:0.5), (HPLC-B) acetonitrile–water–formic acid (35:65:1), and (HPLC-C) acetonitrile–water (70:30). Radiocarbon quantiation of the eluents was accomplished by collection of the appropriate

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