Metabolism of Fenamiphos in 16 Soils Originating from Different Geographic Areas

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The metabolism of the plant protectant Nemacur (1; common name fenamiphos) was investigated in 16 soils from 11 countries with different climates. The metabolism study was conducted under aerobic conditions at two different temperatures. The active ingredient used was phenyl ring-1-^{14}C-labeled. In all soils the degradation products fenamiphos sulfoxide (2), fenamiphos sulfone (3), 3-methyl-4-(methylsulfinyl)phenol (5), 3-methyl-4-(methylsulfonyl)phenol (6), and 3-methyl-4-(methylsulfonyl)-anisole (7) as well as $^{14}CO_2$ could be detected in various amounts. Although the soils had been selected for their diverse heterogeneity, the degradation pathway was the same in all soils. After incubation for 90 days at a temperature of 22 °C, between 2.3% and 66.8% of the total toxic residues (TTR; 1 + 2 + 3) were still detectable in the soil. During this period, 1.1–39.0% of the applied radioactivity was mineralized and between 18.5% and 61.8% bound to the soils. After application and incubation of ^{14}C-labeled metabolites 3-methyl-4-(methylsulfinyl)phenol (5), 3-methyl-4-(methylsulfonyl)phenol (6), and 3-methyl-4-(methylthio)phenol (4), in a Parabraunerde from Germany, a large proportion of the radioactivity was no longer extractable.

INTRODUCTION

For the official registration of a plant protectant in the Federal Republic of Germany the rate of its degradation and metabolism in soil must be investigated as one part of its environmental fate. For these studies a standard soil proposed by the federal registration office (BBA) and one of our own choice is used for these studies. The most important metabolites, including the volatile organic portions and the CO_2 formed, must be identified to determine the degradation pathway (Schinkel et al., 1986). However, plant protectants are often used worldwide, and the question arises as to what extent the degradation pathway which has been established for one soil can be extrapolated to a multitude of soil types originating from different geographic areas. Metabolism studies of plant protectants in different soils, originating from the same region, did not show any quantitative differences in metabolism (Guth, 1980).

The organophosphorus compound fenamiphos is a plant protectant with nematicidal activity being used worldwide on a great number of agricultural crops (Homeyer, 1971).

In the present study the metabolism of the active ingredient [*phenyl*-1-^{14}C]fenamiphos was investigated in 16 soils from 11 countries. The objective was to clarify whether the quantitative degradation of this compound would be the same in all of the soils investigated. Two representative temperatures were chosen for each soil to reflect the climatic zones of their origin. In addition, the behavior of the predicted phenolic metabolites 4–6 was investigated in a Parabraunerde of Germany since they had been identified previously.

The structures of metabolites 2, 3, 5, and 6 have been described previously (Waggoner and Khasawinah, 1974; Lee et al., 1986; Ou and Rao, 1986).

MATERIALS AND METHODS

Soils. Sixteen soils from 11 countries representing a broad spectrum of different soil types from important cultivation areas of the world were used. Soil samples were collected to a depth of 0–15 cm in a moist condition and transported by air mail to Bayer AG, Leverkusen, Germany. After the soils were seeded with Welsch grass (*Lolium multiflorum* Lam.), they were kept in a greenhouse at 18 °C in winter and summer. Shortly before the study was started, soils were sieved through a 2-mm sieve. Soil properties, determined according to VDLUFA (1990), are given in Table I. None of the soils investigated had been treated with fenamiphos in the previous 5 years.

Chemicals. [*phenyl*-1-^{14}C]Fenamiphos (1; *O*-ethyl-*O*-[3-methyl-4-(methylthio)phenyl] isopropylamidophosphate; specific activity 2.32 MBq/mg), [*ring*-1-^{14}C]-3-methyl-4-(methylthio)phenol (4; specific activity 1.88 MBq/mg), [*ring*-1-^{14}C]-3-methyl-4-(methylsulfinyl)phenol (5; specific activity 1.71 MBq/mg), and [*ring*-1-^{14}C]-3-methyl-4-(methylsulfonyl)phenol (6; specific activity 1.56 MBq/mg) were obtained from Bayer AG, Leverkusen, Germany, as well as analytical grade fenamiphos (>99% pure) and the six metabolite standards fenamiphos sulfoxide (2), fenamiphos sulfone (3), 3-methyl-4-(methylthio)phenol (4), 3-methyl-4-(methylsulfinyl)phenol (5), 3-methyl-4-(methylsulfonyl)phenol (6), and 3-methyl-4-(methylsulfonyl)anisole (7). The radiochemical purity of all compounds was greater than 99% as shown by thin-layer chromatography, except for the radiolabeled compound 5 (>97%).

Soil Treatment. To avoid any effects of the solvents upon the biological activity of the soils, they were treated as follows: The total amount of soil needed for one temperature treatment was weighed into a polyethylene bag. An aliquot of the soil (2%) was dried, ground in a porcelain dish, and treated with radiolabeled and nonradiolabeled fenamiphos dissolved in acetone (0.5 mL/100 g of soil). The amount of active ingredient per 100 g of soil was 0.77 mg, corresponding to a maximum use rate of 10 kg/ha with the ratio of radiolabeled/nonradiolabeled fenamiphos being 1:10 by weight. The resulting total amount of radioactivity per flask was approximately 180 kBq. The water content was adjusted to 75% of the maximum retention capacity at a vacuum of 33 kPa and checked gravimetrically every 2 weeks. After homogenization, replicates of the soils (100 g, based on dry substance) were incubated (in triplicate) in 250-mL Erlenmeyer flasks in a closed system as described by Anderson (1975). The evolved $^{14}CO_2$ was trapped in a soda lime cartridge (10 g). Flasks containing soils from temperate climate zones were incubated in the dark at both a standard temperature of 22 °C and a second temperature of 16 °C, which reflected the mean annual temperature of their geographic origin. Flasks containing soils from tropical and subtropical zones were incubated at 22 and 28 °C. In sterile controls the active ingredient was applied to the soil surface in the flasks after autoclaving (121 °C) for 1 h. After evaporation of the acetone, the soil was thoroughly homogenized using a spatula. The deionized water applied was treated with

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Table I. Soil Properties

no. soil	country/region	texture % clay	texture % silt	texture % sand	% H_2O (33 kPa)	pH (H_2O)	% C_{org}	% N	CEC,[a] mequiv/100 g
I	Canada/Alberta	22.7	47.6	29.7	43.5	7.27	6.52	0.65	47.7
II	Sweden/Halland	8.5	9.0	82.5	11.7	6.33	1.23	0.13	7.4
III	Germany/Bavaria	14.1	74.9	11.0	25.7	6.98	1.21	0.11	13.6
IV	Germany/Rheinl Pfalz	4.5	14.1	81.4	18.3	6.45	3.22	0.24	12.0
V	Netherlands/Zeeland	19.3	58.0	22.7	23.3	6.60	1.60	0.16	15.2
VI	France/Champagne	28.1	39.9	32.0	22.8	7.96	1.58	0.15	19.1
VII	U.S.A./Indiana	12.0	25.6	62.4	12.1	6.42	0.95	0.07	10.8
VIII	U.S.A./Nebraska	27.0	69.8	3.2	30.8	6.68	1.51	0.23	22.5
IX	Japan/Toyoda	9.8	48.1	42.1	58.3	5.97	3.53	0.31	31.0
X	U.S.A./Florida	1.3	3.3	95.4	5.2	6.58	0.77	0.06	4.4
XI	Costa Rica/Azul d. Turrialba	29.6	41.6	28.8	51.8	6.03	4.76	0.53	39.5
XII	Brazil/Passo Fundo	44.4	24.4	31.2	23.1	5.79	1.63	0.16	13.6
XIII	Brazil/Parana	53.5	30.9	15.6	31.3	6.52	2.28	0.33	23.2
XIV	Thailand/Chachoengsao	55.3	43.0	1.7	37.0	5.72	1.63	0.15	23.8
XV	Philippines/Mindanao	15.2	42.3	42.5	24.9	5.83	0.73	0.07	16.9
XVI	Japan/Saitama	10.6	47.2	42.2	59.8	7.10	3.58	0.39	38.3

[a] CEC, cation-exchange capacity.

Table II. R_f Values of Fenamiphos and Related Compounds and Mass Spectral Data of Metabolites Isolated from Soil XII

compound	TLC R_f A[a]	TLC R_f B[b]	TLC R_f C[c]	EI-MS data[d] M′	m/z	m/z	m/z	m/z
fenamiphos (1)	0.49	0.56	0.75					
fenamiphos sulfoxide (2)	0.04	0.03	0.18	319 (27)	304 (100)	196 (42)	122 (85)	80 (57)
fenamiphos sulfone (3)	0.30	0.31	0.61	335 (6)	320 (100)	292 (70)	249 (8)	80 (18)
3-methyl-4-(methylthio)phenol (4)	0.91	0.91	0.89					
3-methyl-4-(methylsulfinyl)phenol (5)	0.16	0.17	0.29	170 (65)	155 (100)	94 (90)	65 (61)	45 (96)
3-methyl-4-(methylsulfonyl)phenol (6)	0.75	0.82	0.74	186 (90)	171 (100)	123 (58)	107 (98)	77 (77)
3-methyl-4-(methylsulfonyl)anisole (7)	0.81	0.84	0.82	200 (90)	185 (95)	121 (100)	91 (52)	77 (47)

[a] Diethyl ether/acetone/diisopropyl ether 2:1:1 (v/v/v). [b] Diethyl ether/acetone 9:1 (v/v). [c] Chloroform/acetone (19:3/10:1 (v/v). [d] Key ions and relative intensities (in parentheses).

50 ppm thimerosal (sodium ethylmercurithiosalicylate) to keep the soils sterile during the course of the study.

Recovery and Extraction of Radioactivity. The initial amount of radioactivity applied was determined by combustion of four aliquots of the soil immediately after treatment. This result served as a reference value for the total mass balance. At each sampling time (days 15, 50, and 90) three flasks from each treatment were removed for analysis. The $^{14}CO_2$ adsorbed by the soda lime was liberated with 18% aqueous HCl in a closed system and quantitatively transferred into an absorption cocktail (400 mL of toluene, 330 mL of phenylethylamine, 220 mL of methanol, 7 g of butyl-PBD, 50 mL of water). Two 0.5-mL aliquots were transferred into a Beckman scintillation cocktail (10 mL) and counted by a Beckman counter (LS 8000). Liquid scintillation counting (LSC) was performed using an external ^{137}Cs standard for correcting the counting efficiency. Nonextracted residues were determined by combusting two or three aliquots (0.5 g) from each replicate in a Harvey Biological Materials (OX 300) oxidizer.

The soil in each flask was extracted three times for 15 min in an ultrasonic bath in centrifuge beakers, twice with 150 mL of acetone/methanol (1:1) and once with 150 mL of chloroform/methanol (1:1). After each extraction step, the soil was centrifuged and the clear supernatant decanted and filtered. The filtrate was concentrated to 20 mL on a rotary evaporator and analyzed for radioactivity content by LSC and for metabolites by thin-layer chromatography (TLC).

Metabolite Identification. The degradation of fenamiphos and its metabolites was monitored using thin-layer chromatography (TLC). Aliquots of the organic extracts were analyzed by TLC using Merck 60 F_{254} silica gel plates and developed in the solvent systems given in Table II.

The plates were air-dried and the radioactive compounds detected using a TLC linear analyzer (Berthold Model 2832, Wildbad, Germany). The positions of the radioactive peaks detected were compared to those of standard unlabeled compounds chromatographed in the same manner and visualized with UV light. Results obtained using solvent system A were used for quantification as this system gave the best separation of the metabolites. In some cases, when poor separation was obtained using solvent system A due to matrix effects, solvent system C was used. Autoradiographs (AGFA Curix RP1) of the TLC plates were also prepared.

For further confirmation of the structure of fenamiphos metabolites the organic soil extracts of two soils (IV and XII) were purified by TLC and subjected to GC/MS analysis using a Finnigan MAT mass spectrometer operating in the electron impact (EI) mode (Table II).

Degradation Studies with Phenols. For studying the metabolism of the three phenolic metabolites of fenamiphos, samples of Parabraunerde (soil III, 1.3 ppm) were individually treated with ring-1-^{14}C-labeled 4–6 and incubated at 22 °C and analyzed on days 30, 60, and 90. Incubation and analysis procedures were the same as described above. The experiments were carried out in duplicate.

RESULTS AND DISCUSSION

The results of the metabolism of fenamiphos in the nine soils originating from zones of moderate climate are summarized in Table III and those for the seven soils from the subtropic and tropic regions in Table IV. Recovery of the applied radioactivity was 96.8 ± 6%.

Identification of the Metabolites. The degradation products **2**, **3**, **5**, and **6**, already reported in the literature, were detected in all soils at both temperatures. The structures of the metabolites isolated from the soils were confirmed by mass spectrometry (Table II).

After 90 days, a new degradation product (**7**) could be detected in 14 of the soils. The mass spectrum of metabolite **7** is shown in Figure 1. In two soils (IX and XIV), this metabolite was only detected in traces after 160 or 176 days, respectively. Degradation products generated by a similar methylation step to the formation of **7** have also been reported for fenitrothion (Spillner et al., 1979), 2,4,5-T (McCall et al., 1981), and 2,4-D (Smith, 1985).

Table III. Aerobic Soil Metabolism of Fenamiphos in Nine Soils from Cool to Temperate Geographic[a] Regions: Recovery and Distribution of Applied Radioactivity at 16 and 22 °C

temp, °C	day	extr[b]	1	2	3	5	6	7	TTR[c]	nonextr	$^{14}CO_2$	recovery
						Canada (I)						
22	15	74.5	14.6	45.0	6.2	3.9	4.8	0.0	65.8	18.5	1.3	94.3
	50	53.5	2.9	24.3	6.3	3.7	14.9	1.4	33.5	36.2	8.3	97.7
	90	45.3	2.2	27.7	4.3	2.7	16.7	2.7	34.2	34.8	17.0	97.1
16	15	79.9	33.5	41.2	2.1	2.1	1.0	0.0	76.8	14.8	0.6	95.3
	50	70.1	10.0	37.8	7.2	4.8	10.3	0.0	55.0	21.5	3.4	95.0
	90	54.3	5.3	25.4	4.5	3.4	14.4	1.3	35.2	31.6	10.4	96.3
						Sweden (II)						
22	15	85.7	7.8	65.3	9.0	1.3	2.3	0.0	82.1	6.3	1.7	93.7
	50	70.8	1.5	35.6	19.0	1.1	11.9	1.7	56.1	12.2	8.1	91.1
	90	55.7	0.8	21.0	14.1	0.9	14.8	3.9	35.9	18.5	16.2	90.4
16	15	87.8	18.0	65.7	3.6	0.5	0.0	0.0	87.3	3.7	0.6	92.1
	50	81.5	8.6	53.9	11.7	2.1	5.2	0.0	74.2	8.1	3.1	92.7
	90	77.7	4.9	49.8	14.3	2.0	6.0	0.7	69.0	9.1	4.8	91.6
						Germany/Bavaria (III)						
22	15	77.4	2.6	56.6	5.3	2.6	10.3	0.0	64.5	19.8	4.0	101.2
	50	41.8	0.1	24.1	2.1	0.8	12.3	2.4	26.3	40.3	21.2	103.2
	90	19.2	0.4	7.1	0.5	0.4	7.0	3.3	8.0	48.2	32.9	100.3
16	15	79.8	3.9	66.8	3.0	2.4	3.7	0.0	73.7	12.7	1.2	93.7
	50	52.6	0.2	29.6	3.1	1.1	17.3	1.3	32.9	23.7	11.3	87.6
	90	33.9	0.5	12.6	1.2	0.4	17.0	1.7	14.3	35.1	21.0	90.0
						Germany/Rheinl Pfalz (IV)						
22	15	84.5	15.7	52.3	4.3	7.6	4.1	0.0	72.3	10.1	2.0	96.6
	50	68.4	2.3	34.9	5.9	11.1	12.3	1.2	43.1	19.8	8.7	96.9
	90	57.7	0.8	23.4	5.2	10.1	16.2	2.0	29.4	25.4	13.2	96.3
16	15	86.5	24.0	54.2	2.5	4.2	0.9	0.0	80.7	8.5	0.1	95.1
	50	74.9	4.4	44.0	7.4	9.1	8.9	0.5	55.8	13.6	4.1	92.6
	90	66.6	1.7	28.7	7.3	8.1	18.4	1.7	37.7	20.6	9.0	96.2
						Netherlands (V)						
22	15	89.3	2.6	68.1	9.6	1.1	7.9	0.0	80.3	12.7	3.4	105.4
	50	49.4	0.3	24.3	5.7	0.7	14.9	3.1	30.3	29.5	21.8	100.7
	90	22.0	0.5	10.8	2.2	0.4	5.2	2.9	13.5	33.8	39.0	94.8
16	15	90.1	5.1	79.1	3.9	0.0	2.0	0.0	88.1	9.0	1.1	100.2
	50	72.5	1.7	37.8	11.6	1.2	19.0	1.2	51.1	18.3	7.8	98.6
	90	42.7	0.0	18.7	4.7	0.0	15.8	3.5	23.4	28.6	23.4	94.7
						France (VI)						
22	15	65.3	5.5	36.8	1.8	5.4	14.2	0.8	44.1	29.8	2.5	97.6
	50	23.1	1.1	4.8	0.3	0.8	12.2	3.3	6.2	69.1	16.8	109.0
	90	10.5	0.6	1.5	0.2	0.2	2.9	3.7	2.3	61.8	32.2	95.5
16	15	79.4	6.8	52.9	0.9	4.4	14.4	0.0	60.6	23.3	1.6	104.3
	50	52.2	3.8	27.7	0.8	2.8	16.0	1.1	32.3	42.5	7.2	101.9
	90	47.3	3.9	22.4	0.6	2.8	15.2	1.5	26.9	46.4	9.8	104.0
						U.S.A./Indiana (VII)						
22	15	79.2	7.3	47.3	6.4	3.7	14.5	0.0	61.0	18.4	1.5	99.1
	50	56.6	1.3	25.5	6.6	2.1	19.5	1.6	33.4	29.1	7.7	93.4
	90	45.9	1.1	12.5	2.5	1.8	24.7	3.3	16.1	33.8	14.0	93.7
16	15	90.8	24.7	58.6	2.6	2.3	2.6	0.0	85.9	11.7	0.8	103.3
	50	77.8	13.1	48.7	4.8	3.2	8.0	0.0	66.6	19.6	2.8	100.2
	90	72.6	4.4	45.5	5.4	4.3	12.3	0.7	55.3	25.9	4.1	102.6
						U.S.A./Nebraska (VIII)						
22	15	70.7	6.1	58.1	5.5	1.0	0.0	0.0	69.7	13.1	1.0	84.8
	50	67.9	2.1	33.8	22.6	1.6	7.1	0.7	58.5	23.5	6.6	98.0
	90	58.0	0.8	23.4	19.2	1.4	11.5	1.7	43.4	26.3	12.8	97.1
16	15	79.4	12.3	65.5	1.6	0.0	0.0	0.0	79.4	13.0	0.4	92.8
	50	70.9	3.8	54.3	11.4	0.8	0.6	0.0	69.5	14.9	1.7	87.5
	90	71.0	2.2	37.8	22.8	1.8	6.4	0.0	62.8	18.9	4.8	94.7
						Japan/Toyoda (IX)						
22	15	82.9	48.5	34.4	0.0	0.0	0.0	0.0	82.9	17.5	0.1	100.5
	50	76.7	17.6	48.9	7.1	2.0	1.1	0.0	73.6	25.6	0.3	102.6
	90	73.4	7.3	45.6	13.9	2.8	4.1	0.0	66.8	35.5	1.1	110.0
16	15	86.9	69.1	17.8	0.0	0.0	0.0	0.0	86.9	10.4	0.0	97.3
	50	85.2	34.7	48.8	0.6	1.1	0.0	0.0	84.1	19.0	0.1	104.3
	90	80.3	24.8	49.3	3.1	2.5	0.6	0.0	77.2	23.9	0.4	104.6

[a] Average of triplicate samples. [b] TLC polar products (1%) included. [c] TTR, total toxic residues (compounds 1 + 2 + 3).

Table IV. Aerobic Soil Metabolism of Fenamiphos in Seven Soils from Subtropical and Tropical Geographic[a] Regions: Recovery and Distribution of Applied Radioactivity at 22 and 28 °C

temp, °C	day	extr[b]	1	2	3	5	6	7	TTR[c]	nonextr	$^{14}CO_2$	recovery
						U.S.A./Florida (X)						
22	15	74.5	30.7	34.4	8.1	1.3	0.0	0.0	73.2	10.0	0.6	85.1
	50	60.2	7.6	30.7	15.5	2.0	4.4	0.0	53.8	22.7	2.7	85.6
	90	52.7	6.7	20.2	16.3	1.8	6.9	0.5	43.2	24.8	4.8	82.3
28	15	69.6	16.5	38.1	11.4	1.7	1.9	0.0	66.0	14.3	1.2	85.1
	50	51.3	5.3	21.0	16.2	1.8	6.2	0.5	42.5	24.3	5.2	80.8
	90	43.7	2.9	16.4	13.4	2.0	7.6	1.0	32.7	36.3	8.6	88.6
						Costa Rica (XI)						
22	15	73.0	15.5	45.4	5.9	3.7	2.5	0.0	66.8	21.6	2.0	96.6
	50	55.8	5.9	28.9	7.0	5.2	8.2	0.6	41.8	37.1	7.5	100.4
	90	47.5	3.5	21.3	5.5	5.4	10.5	1.3	30.3	39.6	12.0	99.1
28	15	66.9	14.0	37.1	5.2	6.5	4.1	0.0	56.3	28.9	4.4	100.2
	50	43.2	4.6	20.9	3.6	6.3	6.8	1.0	29.1	47.3	12.4	102.9
	90	36.8	2.6	17.6	3.9	5.0	6.2	1.5	24.1	39.3	16.1	92.2
						Brazil/P. Fundo (XII)						
22	15	81.1	11.2	53.5	6.7	5.4	4.3	0.0	71.4	18.1	2.7	101.9
	50	58.8	2.0	26.2	9.7	5.5	13.5	1.9	37.9	33.4	13.3	105.5
	90	44.3	1.2	13.0	3.5	4.2	18.9	3.5	17.7	32.1	20.8	97.2
28	15	74.5	4.0	49.6	9.2	5.9	5.8	0.0	62.8	21.4	5.0	100.9
	50	42.6	1.7	15.2	3.7	3.9	14.6	3.5	20.6	33.3	22.4	98.3
	90	23.3	0.7	5.8	1.0	2.4	8.4	5.0	7.5	46.4	36.5	106.2
						Brazil/Parana (XIII)						
22	15	71.6	16.9	40.6	5.2	4.5	4.4	0.0	62.7	28.2	1.1	99.9
	50	49.2	5.1	17.6	5.2	3.8	16.6	0.9	27.9	39.4	9.4	98.0
	90	32.4	2.2	7.8	2.0	2.7	15.6	2.1	12.0	48.5	18.1	99.0
28	15	63.7	11.0	32.3	6.9	5.1	8.4	0.0	50.2	35.7	3.8	103.2
	50	33.7	2.7	10.6	2.1	2.9	13.3	2.1	15.4	50.5	19.0	103.2
	90	18.2	1.1	4.3	0.9	1.3	8.0	2.6	6.3	52.3	33.7	104.2
						Thailand (XIV)						
22	15	86.9	24.6	60.9	0.3	1.1	0.0	0.0	85.8	13.8	0.4	101.1
	50	74.3	5.0	52.5	7.7	5.2	3.9	0.0	65.2	20.6	3.0	97.9
	90	61.2	2.9	39.6	8.5	6.2	4.0	0.0	51.0	24.6	4.3	90.1
28	15	94.2	25.0	64.2	2.5	2.5	0.0	0.0	91.7	16.1	0.6	110.9
	50	70.3	3.7	45.6	11.5	4.9	4.6	0.0	60.8	27.1	4.4	101.8
	90	50.1	1.9	28.0	9.7	4.2	6.3	0.0	39.6	30.7	9.9	90.7
						Philippines (XV)						
22	15	75.4	6.4	61.7	4.4	1.6	1.3	0.0	72.5	11.5	1.8	88.6
	50	56.8	1.2	31.7	10.6	1.7	9.6	1.4	43.5	27.0	11.2	95.0
	90	37.7	0.3	18.0	6.7	0.9	9.3	2.1	25.0	35.7	24.2	97.6
28	15	73.4	3.5	57.2	7.5	2.2	3.0	0.0	68.2	18.3	4.6	96.3
	50	38.8	0.4	22.5	6.4	0.9	5.6	2.5	29.3	30.8	23.3	92.9
	90	19.9	0.2	11.1	2.8	0.6	1.8	3.0	14.1	39.7	40.0	99.6
						Japan/Tsurug (XVI)						
22	15	73.4	14.8	56.6	1.1	0.9	0.0	0.0	72.5	20.0	0.3	93.7
	50	66.2	4.8	45.1	9.3	3.1	3.9	0.0	59.2	25.4	1.2	92.8
	90	58.7	1.9	33.7	11.3	3.2	8.0	0.6	46.9	33.2	3.5	95.4
28	15	64.0	5.6	51.0	4.0	2.3	1.1	0.0	60.6	31.4	0.5	95.9
	50	51.3	2.3	29.3	9.5	2.7	6.7	0.8	41.1	32.8	4.4	88.5
	90	43.7	1.2	22.0	6.6	3.3	8.5	2.1	29.8	44.8	9.9	98.4

[a] Average of triplicate replicates. [b] TLC polar products (1%) included. [c] TTR, total toxic residues (compounds 1 + 2 + 3).

Degradation Pathway. Fenamiphos is oxidized quickly to metabolite 2 and to a lesser extent to metabolite 3. After hydrolysis, the phenols formed are further converted to metabolite 7 and bound to the soil matrix or mineralized to CO_2. On the basis of the products found, the degradation pathway for fenamiphos in soil under aerobic conditions shown in Figure 2 can be deduced.

After 90 days, a maximum of 81.6% of the parent compound had been oxidized to metabolite 2 in the sterile treatments. In some of the sterile soils small amounts (<2.5%) of metabolite 5 could be detected (data not listed). This implies that while 1 is predominantly degraded microbially, significant nonmicrobial oxidation occurred.

As the same degradation pathway was found under aerobic conditions in all soils and at all temperatures, it can be concluded that similar microorganisms and/or enzyme systems were present in each soil. Thus, the basic degradation mechanisms of fenamiphos such as oxidation, ester hydrolysis, transmethylation, and ring cleavage were observed for all soils. This study shows that the main soil metabolites of a plant protectant can be determined using only a few soils and from these results the degradation

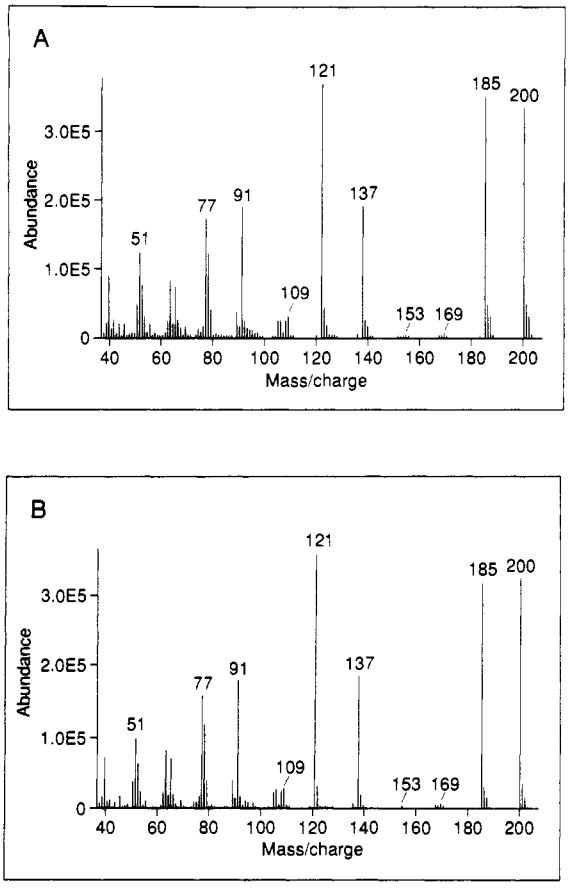

Figure 1. Mass spectrum of 3-methyl-4-(methylsulfonyl)anisole (7) isolated from soil XII.

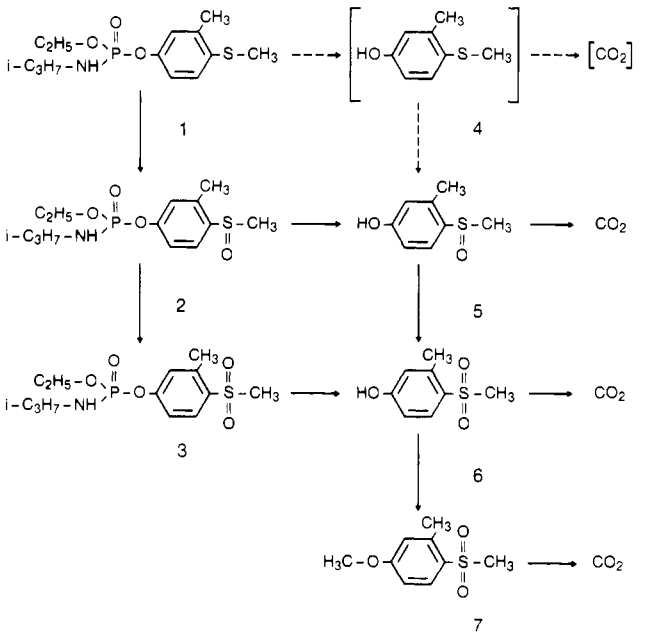

Figure 2. Proposed degradation pathway of fenamiphos in soil under aerobic conditions.

pathway can be deduced. Even at the second temperatures investigated, there were no indications that other degradation products were formed.

Quantitative Aspects. Quantitative differences between the individual soils were large. The available data do not allow the calculation of any half-lives for fenamiphos as they were below 15 days at 22 °C in all soils. If the residues of 1 and the two metabolites 2 and 3 which were all nematicidally active are combined as TTR (total toxic

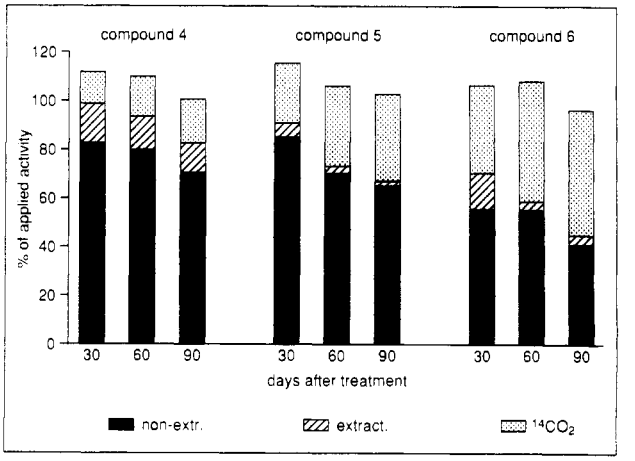

Figure 3. Distribution of applied radioactivity in a para brown earth incubated with [*ring*-1-^{14}C]-3-methyl-4-(methylthio)phenol (4), [*ring*-1-^{14}C]-3-methyl-4-(methylsulfinyl)phenol (5), and [*ring*-1-^{14}C]-3-methyl-4-(methylsulfonyl)phenol (6).

residues) according to the method of Ou and Rao (1986), these compounds were still detectable after 90 days at 22 °C at levels between 2.3% and 77.2% of the applied radioactivity depending upon the soil type used [see Table III, soil France (VI) and soil Japan/Toyoda (IX)]. The main metabolite was compound 2, and at later extraction dates compound 6 became significant. In most of the soils an increasing trend in the formation of 6 was noticeable during the experiment. In those soils in which fenamiphos was quickly mineralized and/or only small portions of bound residues were formed, lower concentrations of metabolite 6 were detected (soils III, V, VI, and XII; see Tables III and IV). The maximum amounts of metabolites 5 and 7 observed were 11.1% [Table III, soil Germany/Rheinl Pfalz (IV)] and 5.0% (Table IV, soil Brazil/P. Fundo (XII)], respectively.

The mineralization rate greatly depended on the soil used. Between 1.1% (soil IV) and 39.0% (Table III, soil V) of the radioactivity applied was detected as $^{14}CO_2$ after 90 days at 22 °C. The highest proportion of unextractable residues (48.2–61.8%) was formed in soils III, VI (Table III), and XII (Table IV). There was no correlation between the formation of nonextractable residues and the organic content of the soils.

Metabolism of the Phenols. To determine the significance of the production of the phenolic metabolites 5 and 6, which were detected in all soils, their formation of unextractable residues and mineralization were investigated after application to a Parabraunerde from Puch, FRG (Table I, soil III). Metabolite 4 was included in this experiment to determine why this compound was not detected in any of the soils.

In these experiments, metabolite 6 showed the highest degree of mineralization (Figure 3). All three compounds showed a significant trend in the formation of unextractable residues. In the case of compound 4 only 12.2% of the applied radioactivity could be extracted after 90 days, of which 3% was metabolite 4, 6.0% polar components, and 3.2% unidentified. In the case of compound 5 only 2.1% was still extractable, of which 0.2% was metabolite 5, 0.4% metabolite 6, 0.6% metabolite 7, 0.6% polar compounds, and 0.3% unidentified. In the case of compound 6, the extractable portion after 90 days amounted to only 3.7% (6, 2.1%; 7, 1.2%; polar, 0.2%; unidentified, 0.2%). Thus, in these experiments after 90 days, compound 4 was detectable even at the highest level, although it could not be found in the extracts of any of the soils in the metabolism studies. The lack of formation

of this theoretical metabolite may be explained by application of the Hammett relationship to the hydrolysis of phenyl phosphorodithionate esters (Johnson, 1980). The Hammett relationship predicts that the hydrolysis of fenamiphos should occur about an order of magnitude slower than either of the more electron deficient metabolites fenamiphos sulfoxide or fenamiphos sulfone. This slow rate of hydrolysis coupled with the rapid oxidative metabolism of fenamiphos would preclude the formation of fenamiphos phenol (4).

Conclusions. The results of the metabolism of fenamiphos showed that reactions such as oxidation, ester hydrolysis, ring cleavage, and transmethylation which frequently occur during the degradation of plant protectants in soil also take place in soils from different regions of the globe with a very different soil genesis. This indicates that the main degradation pathway of an active ingredient in aerobic soils can be deduced with sufficient accuracy in metabolism studies involving very few soils.

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