conditions at both locations were similar. At Melfort 32.2 cm of rain was recorded for the duration of the experiment and 12.3 cm was noted at Regina.

At both sites the amounts of residues recovered from each of the four replicated plots were reproducible (Table III), which indicates that removal of entire soil levels from small treated plots is a satisfactory method for the study of herbicide persistence under field conditions.

In all cases less than 2% of the applied trifluralin was detected in the 5- to 10-cm soil levels, thus confirming that trifluralin is not readily leached under field conditions (Parka and Tepe, 1969).

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Movement and Fate of Dyfonate in Soils under Leaching and Nonleaching Conditions

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The degradation and movement of ^{14}C-Dyfonate in different soil types was tested in the laboratory under leaching and nonleaching conditions, at different temperatures and various incubation times. Different apparatuses are described which had been especially designed for testing the movement of pesticidal chemicals in soils with and without water. Although Dyfonate did not move to an appreciable extent through an agricultural loam soil, its transport with water through soils was, to a large extent, a function of the soil type. Water, after its percolation through a silt loam, did not contain measurable amounts of the insecticide, while water after its percolation through a sandy loam soil was toxic to mosquito larvae and contained 4% of the dosage originally applied to the top soil layer. After degradation of ^{14}C-Dyfonate in soils, radiocarbon was lost from the soil by volatilization of the ethoxy moiety and partially by transport of the ring moiety with water. When no water was percolated the insecticide also degraded and moved into untreated soil, but to a lesser degree.

Studies on the persistence and fate of insecticides after application to crops and soils have, to a large extent, been conducted with chemicals of the chlorinated hydrocarbon type. In an attempt to reduce the application of persistent pesticides, more degradable compounds are now being used for insect control. Concurrent with this use, more information is needed as to the fate of these insecticides after application. In addition to persistence and metabolism studies with organophosphorus and carbamate insecticides, problems related to the migration and potential transport of these chemicals with water through soils should be investigated. Concern has particularly been expressed regarding the potential movement of insecticidal residues with water from upper soil strata into uncontaminated soil and ground water below. Lake and river water can undoubtedly be contaminated with runoff water from adjacent agricultural fields. In this case soil particles to which insecticidal residues are adsorbed are being washed off the soil. However, one of the most important questions is whether water in deeper soil strata can be contaminated by insecticidal residues.

To study the movement of pesticide chemicals in soils, various experiments were conducted in the past, primarily with chlorinated hydrocarbon insecticides (Beran and Guth, 1965; Bowman *et al.*, 1965; Harris, 1969; Lichtenstein and Schulz, 1958). Lichtenstein *et al.* demonstrated in 1967 that water, after its percolation through aldrin- or parathion-treated loam soils, did not contain measurable amounts of aldrin but did contain small amounts of parathion, whose concentration in the water was below the water solubility of the insecticide. The amount of parathion in percolated water, though, was a function of the concentration of the insecticide in the soil.

In the present study, laboratory experiments were conducted with the organophosphorus insecticide Dyfonate (*O*-ethyl *S*-phenyl ethylphosphonodithioate) to study its persistence, metabolism, and potential movement in different soil types under leaching and nonleaching conditions. Dyfonate was selected because it is not as persistent as many of the chlorinated hydrocarbon insecticides (Schulz and Lichtenstein, 1971) but is relatively volatile in comparison to other organophosphorus compounds (Lichtenstein and Schulz, 1970). It has a water solubility of 13 ppm, which is greater than the solubility of most chlorinated hydrocarbon insecticides and smaller than that of most of the organophosphorus compounds. Although these experiments were conducted with one model insecticide, procedures developed and described here could serve as a model for testing the persistence and

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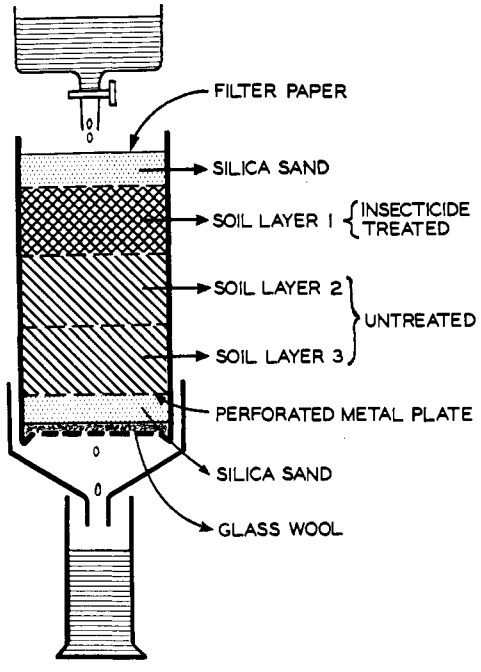

Figure 1. Diagram depicting apparatus used for the percolation of water through insecticide-treated and lower untreated soil layers

Table I. Characteristics of Soil Types Used in Experiments

	Percent			
	Organic matter	Sand	Silt	Clay
Plano silt loam	4.7	5	71	24
Fox fine sandy loam	1.0	84	12	4
Plainfield sand	0.8	91	3	6
Quartz sand	0.0	100	0	0

movement of other pesticidal chemicals in soil under leaching and nonleaching conditions.

MATERIALS AND METHODS

Chemicals. Dyfonate, ^{14}C(ethoxy)-Dyfonate (specific activity of 5.75 mCi/mmol), ^{14}C(ring)-Dyfonate (specific activity 4.7 mCi/mmol), dyfoxon (*O*-ethyl *S*-phenyl ethylphosphonothiolate), *O*-ethylethanephosphonothioic acid, *O*-ethylethanephosphonic acid, thiophenol, methyl phenyl sulfoxide, methyl phenyl sulfone, 4-hydroxyphenyl methyl sulfone, and 2-(hydroxyphenyl)methyl sulfone were obtained through the courtesy of the Stauffer Chemical Co. Solvents used were methyl alcohol, diethyl ether, and redistilled benzene, hexane, and acetone.

Soils. Experiments were conducted with three soil types and a quartz sand. Their major characteristics are shown in Table I.

Extraction Methods. Soils from experiments described below under Ia, IIa, and IIc were extracted twice in a Lourdes homogenizer with a 1:1 mixture of methyl alcohol and acetone, followed by a third extraction with a 1:1:1 mixture of benzene, methanol, and acetone. The solvent–soil ratio was 1.5 ml per gram per extraction. The extracts and the soil were filtered under suction, resulting in a "dry soil" and a filtrate which was concentrated in a flash evaporator at 35°C until most of the solvents had been evaporated. The remaining extract was then adjusted to 100 ml with water and shaken three times with 50-ml portions of benzene, thus finally resulting in a benzene and a water extraction phase. To extract organic solvent soluble compounds further from the water extraction phase, it was acidified to pH 2 with 1 *N* HCl and shaken three times with 50-ml portions of diethyl ether. This then yielded an ether fraction and a water fraction of the water-extraction phase.

Soils from experiments Ib and IIb were extracted with a 1:1 mixture of hexane–acetone (2 ml/g of soil). After removal of the acetone with water, the hexane was dried over anhydrous sodium sulfate and adjusted to volume for analyses.

Water (250 ml) collected after its percolation through soil columns in experiment Ia, was extracted three times with 75 ml portions of benzene and in experiment Ib with three 100 ml portions of hexane. Both the organic and the water phase were then analyzed as described below.

Analytical Methods. Liquid Scintillation Counting (lsc). To determine the radiocarbon content in the organic solvent fractions obtained from soil or water extracts, 2-ml aliquots of extract were added to 14 ml of scintillation mixture (6 g of PPO, 0.25 g of dimethyl POPOP in 1000 ml of toluene). The radiocarbon in unextracted or extracted water was determined by adding 2 ml of this water to 14 ml of a second scintillation mixture (4 g of PPO, 0.05 g of dimethyl POPOP, 120 g of naphthalene and 880 ml of 1,4-dioxane). These mixtures were then counted for 10 to 100 min in a Model 3320 Packard Tri-Carb scintillation spectrometer. Data were corrected for background, counter efficiency, and dilution. The radiocarbon remaining in soils after their extraction was determined by combusting 200-mg aliquots of the dry soil (Expt. Ia, IIa, and IIc) in an oxygen-filled Schöniger flask according to a method described by Kelly *et al.* (1961). The $^{14}CO_2$ produced was absorbed in 3 ml of a 1:2 mixture of phenylethylamine:methanol which was then dissolved in 14 ml of the toluene-based scintillation mixture for analyses by lsc.

Gas–Liquid Chromatography. Glc was performed with the organic solvent phases utilizing a Packard Model 7834 gas chromatograph as previously described (Lichtenstein and Schulz, 1970).

Thin-Layer Chromatography. Tlc was performed with the benzene phases obtained after extraction of both the soils and percolated water in Exp. Ia and the hexane extracts of soils in Expt. IIb. Aliquots of the extracts were concentrated and applied to silica gel precoated plates (Brinkmann Instruments, Inc., N.Y.), followed by development in chloroform:ethyl acetate (1:1) (McBain *et al.*, 1970). Compounds on the developed plates were visualized by immersing them in iodine vapors for 10 min, then spraying with ammonium molybdate–ammonium chloride reagent (Block *et al.*, 1958), and heating at 100°C for 10 min. In this way a blue color was observed for the following reference grade compounds: Dyfonate, dyfoxon, *O*-ethylethanephosphonothioic acid, thiophenol, 2-hydroxyphenyl methyl sulfone, and 4-hydroxyphenyl methyl sulfone. Additional spraying of the plates with 0.5% DCQ reagent (Menn *et al.*, 1957) resulted in an orange color where *S*-phenyl-containing compounds were present. To visualize radioactive compounds, sprayed plates were covered with Saran wrap and exposed for 30 days to Kodak No-Screen X-ray films.

The potential toxicity to insects of soils or water percolated through soils was tested with vinegar flies (*Drosophila melanogaster* Meig.) or with mosquito larvae (*Aedes aegypti* L.). In experiment Ib vinegar flies were exposed to the dry residue of the hexane fraction of soil extracts. Mosquito larvae were exposed to 15-ml aliquots of water which had been percolated through various soil columns (Expt. Ia and b).

Table II. Movement of ^{14}C(Ethoxy)-Dyfonate with Water Through a Plano Silt Loam Soil at 27 ± 1°C

Soil layers	Day	Recovered in percent of ^{14}C or μg of Dyfonate found in the treated soil 1 day after the insecticide application at 20 ppm: Soil extract, Benzene, lsc[b]	Benzene, glc[b]	Water, Ether, lsc	Water, Ether, glc	Water, lsc	Bound,[a] lsc
I. Treated	29	45.0 ± 1.0	45.0 ± 1.0	0.8 ± 0.5	0.4 ± 0.3	0.1 ± 0	8.2 ± 0.9
	105	19.0 ± 0.9	19.4 ± 0.9	1.7 ± 0.2	1.4 ± 0.2	0.3 ± 0	11.1 ± 0.4
II. Untreated	29	14.0 ± 0.5	14.0 ± 0.3	0.2 ± 0.0	0.1 ± 0.0	0.1 ± 0	2.7 ± 0.6
	105	6.5 ± 1.1	6.2 ± 1.6	0.1 ± 0.0	TR[c]	0.1 ± 0	5.0 ± 0.2
III. Untreated	29	0.4 ± 0.2	0.5 ± 0.2	TR	ND[c]	TR	0.5 ± 0.2
	105	0.3 ± 0.1	0.3 ± 0.1	TR	TR	TR	0.45 ± 0.05

	Percolated water[d]: Benzene, lsc	Benzene, glc	Water, lsc	BA–A[e]
	1	0.1 ± 0.0	ND	0.45 ± 0.05	0/72
	15	0.1 ± 0.0	TR	0.45 ± 0.05	0/72
	29	0.2 ± 0.1	0.1 ± 0.0	0.25 ± 0.05	0/72
	44	0.3 ± 0.0	0.2 ± 0.0	0.15 ± 0.05	35/72
	105	0.1 ± 0.0	TR	TR	0/72

Total ^{14}C in soil = 72% (after 29 days) and 44.5% (after 105 days) of ^{14}C in treated soil on day 1.
Total ^{14}C in water = 1.55% (after 29 days) and 2.1% (after 105 days) of ^{14}C in treated soil on day 1.
Total Dyfonate by glc = 60.1% (after 29 days) and 27.6% (after 105 days) of insecticide in treated soil on day 1.

[a] Determined by combustion of previously extracted soil. [b] Lsc = ^{14}C determined by liquid scintillation counting. Glc = Dyfonate determined by gas–liquid chromatography. Results are averages of duplicate experiments. [c] TR = Trace. ND = nondetectable. [d] Water (330 ml) collected after its percolation through soils 1, 15, 29, 44, or 105 days after treatment of upper soil layer with ^{14}C-Dyfonate. [e] Percent mortality observed with larvae *Aedes aegypti* exposed for 72 hr to 15-ml aliquots of percolated water.

Table III. Movement of ^{14}(Ring)-Dyfonate with Water Through a Plano Silt Loam Soil at 27 ± 1°C

Soil layers	Day	Recovered in percent of ^{14}C or μg of Dyfonate found in the treated soil 1 day after the insecticide application at 20 ppm: Soil extract, Benzene, lsc[b]	Benzene, glc[b]	Water, Ether, lsc	Water, Ether, glc	Water, lsc	Bound,[a] lsc
I. Treated	29	50.0 ± 1	49.0 ± 8	4 ± 3	4 ± 3	0.25 ± 0.05	13 ± 1
II. Untreated	29	12.0 ± 1	13.0 ± 1	1 ± 0	1 ± 0	0.1 ± 0.00	3 ± 0
III. Untreated	29	0.8 ± 0.2	0.1 ± 0.08	TR[c]	TR	TR	0.1 ± 0

	Percolated water[d]: Benzene, lsc	Benzene, glc	Water, lsc	BA–A[e]
	1	TR	ND[c]	3.65 ± 0.05	0/72
	15	TR	0.1 ± 0	1.5 ± 0.00	0/72
	29	0.65 ± 0.15	0.1 ± 0	0.1 ± 0.00	0/72

Total ^{14}C in soil = 84.2% of ^{14}C in treated soil on day 1
Total ^{14}C in water = 6.0% of ^{14}C in treated soil on day 1
} 90.2% (including 16.1% bound ^{14}C).
Total Dyfonate by glc = 67.3% of insecticide in treated soil on day 1.

[a] Determined after combustion of previously extracted soil. [b] Lsc = ^{14}C determined by liquid scintillation counting. Glc = Dyfonate determined by gas–liquid chromatography. Results are averages of duplicate experiments. [c] TR = Trace. ND = nondetectable. [d] Water (330 ml) collected after its percolation through soils 1, 15, and 29 days after soil treatment. [e] Percent mortality observed with larvae of *Aedes aegypti* exposed for 72 hr to 15-ml aliquots of percolated water.

EXPERIMENTAL PROCEDURES

I. Movement of Dyfonate in Soils with Water. (a) MOVEMENT AND PERSISTENCE OF ^{14}C-DYFONATE IN A LOAM SOIL. Plano silt loam was treated with an acetone solution of ^{14}C-(ethoxy)-Dyfonate, previously diluted with nonradioactive insecticide. To obtain an "aged" residue, the treated soil was kept in a sealed container in an incubator at 27°C for 1 day, when aliquots were extracted and analyzed by lsc and glc for their radiocarbon and Dyfonate content. At that time 13.5 μg (0.011 μCi) of ^{14}C(ethoxy)-Dyfonate per g of dry soil was determined. This relatively high concentration of Dyfonate was utilized to facilitate measurement of the insecticide or its potential metabolites in untreated soil layers and percolated water. In this experiment (Table II), four 1-qt ice cream cartons (7 in. × 3.5 in. diam, 17 cm × 8.5 cm diam) (Figure 1) were prepared by perforating the bottoms and placing a layer of glass wool and 200 g of hexane-washed quartz sand inside. Two 200-g portions of insecticide-free loam soil were placed on top of the sand. The initial thickness of the 200-g soil layer was 1.5 in. (3.7 cm). A 200-g portion of ^{14}C(ethoxy)-Dyfonate-treated soil was then placed on top of the two untreated layers in each of the four cartons. To facilitate equal distribution of water dripped from above, 200 g of hexane-washed quartz sand was finally placed on top of the treated

Table IV. Movement of Dyfonate with Water Through Three Soil Types at 22 ± 2°C

Soil layers	Day	Recovered in percent of Dyfonate applied[a] to upper soil layer: P. silt loam glc[b]	P. silt loam BA-D[c]	F. sandy loam glc	F. sandy loam BA-D	Quartz sand glc	Quartz sand BA-D
I. Treated	17	37.3 ± 0.3	100/0.3	15.8 ± 1.2	100/48	0.05 ± 0	66/48
II. Untreated	17	8.9 ± 0.2	60/48	19.4 ± 0.1	85/48	<0.05 ± 0	5/48
III. Untreated	17	0.05 ± 0	39/48	14.5 ± 0.5	68/48	<0.05 ± 0	0/48
Total (Soil)		46.25		49.7		<0.15	
		Percolated water[d]: glc	BA-A[e]	glc	BA-A	glc	BA-A
	0	ND[f]	0/48	TR[f]	0/48	37.2 ± 3.2	100/.5
	7	ND	0/48	1.1 ± 0.1	35/48	10.0 ± 0.5	95/48
	17	ND	0/48	2.7 ± 0.2	100/18	5.0 ± 0.0	95/48
Total (water)		ND		3.8		52.2	
Total (soil and water)		46.25		53.5		52.35	

[a] Dyfonate applied at 20 ppm (4000 μg/200 g of soil). [b] Dyfonate determined by gas–liquid chromatography. Results are averages of duplicate experiments. [c] Percent mortalities/hours observed with *Drosophila melanogaster* after exposure to the dry residue of the hexane fraction of soil extracts, equivalent to 0.5 g of soil. [d] Water (330 ml) collected after its percolation through soils initially (0), 7, and 17 days after soil treatment with Dyfonate. [e] Percent mortalities/hours observed with larvae of *Aedes aegypti* after exposure to 15-ml aliquots of percolated water. [f] ND = Nondetectable. TR = trace.

soil and covered with two layers of filter paper. All sand and soil layers were separated by perforated stainless steel plates, which made it possible to separate each soil layer at the end of the experiment.

Water was then added from a glass container onto each of the four soil columns at a rate of 1 drop/5 sec. Approximately 235 ml of water were needed to saturate each soil column, until water started to drip out at the lower end. Percolation was continued until 330 ml of water were collected from each column. A total of 565 ml of water were added onto each column until 330 ml of percolated water had been collected. This amount represented a rainfall of 3.91 in. or 9.95 cm. Eighty milliliters of the percolated water from each soil column were used for bioassay tests with mosquito larvae. The remaining 250 ml of water were extracted and analyzed as described.

After the initial percolation (day 1) the soil columns were left undisturbed for 14 days in an incubator at 27 ± 1°C. After that (day 15) the percolation was repeated. Approximately 450 ml (3.12 in. or 7.93 cm of rain) were added onto the soil columns until 330 ml was collected from each column. After 14 additional days in the incubator (29 days after the insecticide application) the percolation procedure was repeated. Two of the four cartons were then frozen, the carton was removed, and the three soil layers in each carton were carefully separated. Because of the presence of the perforated metal plates it was possible to minimize the contamination of one soil layer by another. Each of the soil layers and the percolated water samples from days 1, 15, and 29 were extracted for analysis as described. The two remaining cartons were kept in the incubator for a total of 105 days and water percolation was repeated after 44 and 105 days as described. Finally the soil layers from these two columns and the percolated water were also extracted and analyzed. Results were ultimately expressed as radiocarbon content or μg of Dyfonate in soils or water in percent of the content recovered from the Dyfonate-treated soil 1 day after application of the insecticide.

The identical experiment was repeated with two soil columns over a 29-day incubation period (Table III), except that the upper soil layer had been treated with ^{14}C(ring)-Dyfonate, which contained, after 1 day of aging, 15 μg (0.008 μCi) of ^{14}C-Dyfonate per g of dry soil.

(b) Movement and Persistence of Dyfonate in Different Soil Types. Two soils (Plano silt loam and Fox fine sandy loam) and a quartz sand were treated at 20 ppm with an acetone solution of Dyfonate. After mixing of the soil or sand, aliquots were taken for initial analyses. Duplicate columns with silt loam, sandy loam, and quartz sand were then constructed as described. Water was percolated through these columns initially (day 0) and 7 and 17 days later (Table IV). Between percolations the columns were covered and kept at room temperature (22 ± 2°C). After 17 days the soil or sand layers were removed and prepared for extraction. Soil, sand, and water extracts were analyzed by glc and expressed as μg of Dyfonate found in percent of the dosage applied to the upper soil or sand layer. In addition, soils, sand, and water were tested for insect toxicity by utilizing *Drosophila* or *Aedes* bioassay procedures.

II. Movement of Dyfonate in Soils under Nonleaching Conditions. (a) Vertical Movement and Persistence of ^{14}C-Dyfonate in a Silt Loam. To study the vertical movement of Dyfonate in a silt loam soil in the absence of percolating water, four soil columns were set up as described in Ia above, two with ^{14}C(ethoxy)-Dyfonate and two with ^{14}C-(ring)-Dyfonate-treated soil (Table V). No water was percolated through these columns, but the initial weight of each column was maintained during the 29 days of incubation by sprinkling water onto the top soil layer as necessary. After that the treated and the two untreated soil layers in each carton were separated as described and prepared for extraction and analysis. Results were finally expressed as the radiocarbon content or μg of Dyfonate found in each soil layer in percent of the content determined in the treated soil 1 day after the insecticide application.

(b) Vertical and Horizontal Movement of ^{14}C(Ethoxy)-Dyfonate in Two Soil Types under Nonleaching Conditions. To study both the vertical and the horizontal movement of Dyfonate with two different soils, duplicate five-layer soil columns were constructed with Plainfield sand and Plano silt loam as described. The two upper and the two lower soil layers were free of insecticide and had a weight of 200 g each, while the 100-g center layer was treated with ^{14}C(ethoxy)-Dyfonate at 20 ppm (0.049 μCi per g of dry sandy soil and 0.053 μCi per g of dry silt loam). The top and bottom covers of each carton were perforated. To prevent excessive loss of

Table V. Vertical Movement of ^{14}C-Dyfonate Through a Loam Soil in the Absence of Percolating Water (29 Days Incubation at 27 ± 1°C)

Soil layers	Recovered in percent of ^{14}C or μg of Dyfonate found in soil 1 day after its treatment with insecticide at 20 ppm			
	Soil extract			
	Benzene		Water, lsc	Bound,[a] lsc
	lsc[b]	glc[b]		
	Ethoxy-label			
I. Treated	58.2 ± 6.2	62.7 ± 6.6	0.4 ± 0.0	7.0 ± 0.2
II. Untreated	6.2 ± 0.4	6.4 ± 0.0	0.45 ± 0.05	1.0 ± 0.0
III. Untreated	0.2 ± 0.1	0.35 ± 0.25	TR[c]	0.1 ± 0.0
Total ^{14}C in soil = 73.6% of ^{14}C in treated soil on day 1 (including 8.1% bound ^{14}C). Total Dyfonate by glc = 69.54% of insecticide in treated soil on day 1.				
	Ring-label			
I. Treated	62.9 ± 3.8	60.4 ± 2.8	3.0 ± 0.2	13.9 ± 2.5
II. Untreated	6.8 ± 0.1	7.1 ± 0.0	1.3 ± 0.5	2.7 ± 0.9
III. Untreated	0.1 ± 0.1	0.1 ± 0.1	TR	TR
Total ^{14}C in soil = 90.7% of ^{14}C in treated soil on day 1 (including 16.6% bound ^{14}C). Total Dyfonate by glc = 67.6% of insecticide in treated soil on day 1.				

[a] Determined by combustion of previously extracted soil. [b] Lsc = ^{14}C determined by liquid scintillation counting. Glc = Dyfonate determined by gas–liquid chromatography. Results are averages of duplicate experiments. [c] TR = trace.

moisture, the four soil columns were placed in polyethylene bags and kept for 14 days in the dark at room temperature (22 ± 2°C). During that time one carton of sand and one of loam soil were kept in a vertical position while the other two columns were kept in a horizontal one. At the end of the incubation period each soil column was dismantled and the individual soil layers were extracted and analyzed. Results were finally expressed as radiocarbon content or μg of Dyfonate recovered in percent of the dosage initially applied to the center soil layer.

(c) Horizontal Movement of Dyfonate in Rectangular and Circular Containers. To study further the horizontal movement of Dyfonate, two types of apparatus were used. One of them (Figure 2) consisted of a rectangular stainless steel container, $12^1/_4$-in. × $7^1/_8$-in. and $2^3/_{16}$-in. high (31.2-cm × 18.0-cm and 5.5-cm high) into which three compartments, made of 20-mesh stainless steel screen, were inserted. This arrangement resulted in a 1-in. wide center compartment and three 1-in.-wide compartments to either side.

The second container (Figure 3) consisted of a round Pyrex glass dish, $7^1/_2$-in. diameter and 4-in. high (18.5-cm diameter and 9.7-cm high) into which three 20-mesh stainless steel screen containers were inserted, resulting in four 1-in. wide circular compartments.

Dyfonate treated (50 ppm) field moist (15 water) silt loam was placed into the center compartments of two rectangular and two circular containers, while the remaining compartments were filled with untreated field moist soil. The amount of soil in each of the compartments of the rectangular containers was 360 g on a dry weight basis. The amounts of treated soil in the center layer of the round container was 27 g on a dry weight basis and 232, 600, and 1200 g in the inner, middle, and outer rings, respectively. A preliminary experiment was then conducted with silt loam soil treated with Dyfonate at 50 ppm in two rectangular and in two round containers. One of each shape was incubated for 42 days at 15 ± 1°C and the two remaining ones at 30 ± 1°C. During incubation, moisture levels were maintained by sprinkling water onto the soils as necessary. After incubation, each soil layer was extracted and analyzed by glc, tlc, and bioassay procedures.

Finally, tests were conducted in duplicate with a Plano silt loam to which ^{14}C(ethoxy)-Dyfonate had been applied at 50 ppm (Table VI). To determine the actual radiocarbon and Dyfonate content of this soil, aliquots were removed for extraction and analysis immediately after soil treatment. The center compartments of two rectangular and two round containers were then filled with the insecticide-treated soil, while the remaining compartments were filled with insecticide-free soil. To prevent loss of water and excessive volatilization,

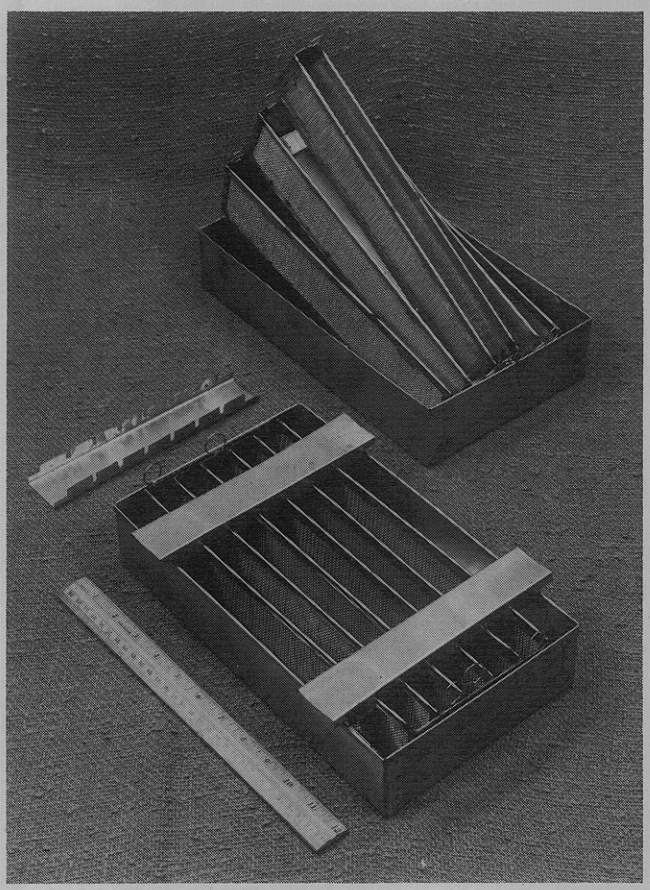

Figure 2. Rectangular stainless steel containers used for measuring the horizontal movement of an insecticide from a treated soil layer in the center compartment into untreated soil within the adjacent compartments

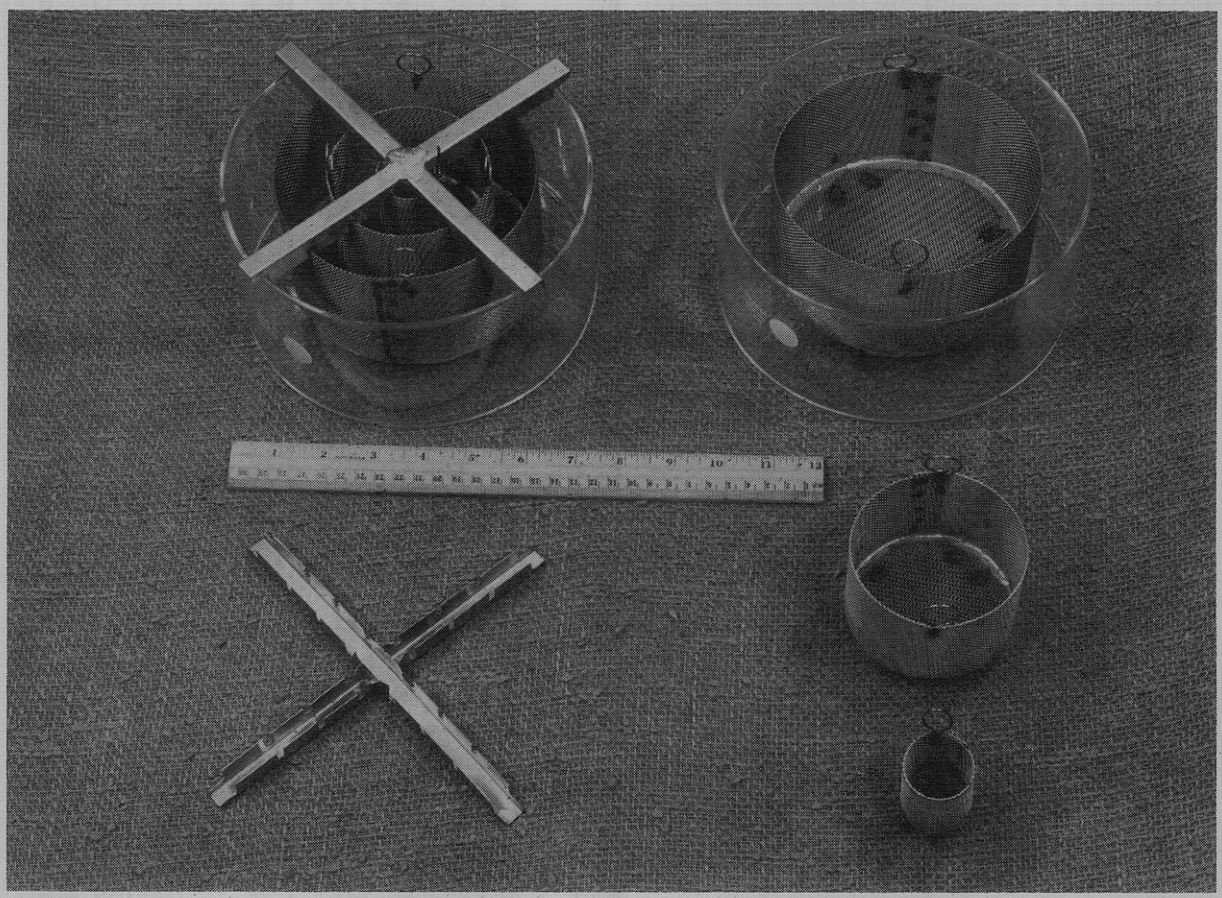

Figure 3. Round containers used to measure the movement of an insecticide from treated soil within the center stainless steel mesh compartment into untreated soil in the surrounding circular soil layers

Table VI. Horizontal Movement of ^{14}C(Ethoxy)-Dyfonate from an Insecticide-Treated (50 ppm) Silt Loam into Surrounding Untreated Soil Layers during a 21-Day Incubation Period at 22 ± 2°C. Results Are Averages of Duplicated Tests

Extraction phase	Recovered from soil as ^{14}C or μg of Dyfonate in percent of the amounts initially present in the center soil layer				
	Rectangular soil layers (Figure 2)				
	Center	Inner	Middle	Outer	All layers
Benzene ^{14}C[a]	59.8 ± 1.1	17.5 ± 0.1	0.9 ± 0.1	0.5 ± 0.1	78.6 ± 1.1
D-%[b]	60.8 ± 0.5	18.6 ± 0.2	1.0 ± 0.1	0.5 ± 0.0	80.9 ± 0.4
D-ppm[c]	31.6 ± 0.2	5.2 ± 0.01	0.3 ± 0.01	0.1 ± 0.01	
Water ^{14}C	0.2 ± 0.0	Trace	Trace	Trace	0.2 ± 0.0
Bound[d] ^{14}C	4.4 ± 0.2	0.8 ± 0.6	0.1 ± 0.0	Trace	5.3 ± 0.4
Total ^{14}C	64.4	18.3	1.0	0.5	84.1
% Distribution[e]	76	21.6	1.3	0.2	100
	Round soil layers (Figure 3)				
Benzene ^{14}C	35.5 ± 3.2	27.8 ± 0.2	1.4 ± 0.1	0.9 ± 0.0	65.6 ± 3.0
D-%	38.4 ± 5.2	28.7 ± 0.9	1.4 ± 0.0	0.9 ± 0.0	69.4 ± 6.6
D-ppm	20.0 ± 2.7	1.7 ± 0.05	0.03 ± 0.0	0.01 ± 0.0	
Water ^{14}C	0.2 ± 0.05	Trace	Trace	Trace	0.2 ± 0.05
Bound ^{14}C	4.2 ± 0.1	3.2 ± 0.1	0.4 ± 0.1	0.2 ± 0.2	7.9 ± 0.2
Total ^{14}C	39.9	31.0	1.8	1.1	73.8
% Distribution	54.5	42.0	2.4	1.1	100

[a] ^{14}C determined by liquid scintillation counting. [b] D-% = Dyfonate, determined by gas–liquid chromatography, in percent of initial. [c] D-ppm = Concentration of Dyfonate as determined by gas–liquid chromatography in various soil layers. [d] Determined by combustion of soil after extraction. [e] % Distribution = Distribution of ^{14}C in various soil layers in percent of the total recovered from all layers.

each container was covered with aluminum foil and then held in the dark for 21 days at room temperature (22 ± 2°C). At the end of the incubation period each soil layer was extracted and analyzed by lsc and glc. Results were expressed as radiocarbon content or Dyfonate in each soil layer in percent of the amounts that had been found in the soil after the insecticidal application.

RESULTS AND DISCUSSION

I. Movement of Dyfonate in Soils with Water. (a) MOVEMENT AND PERSISTENCE OF ^{14}C-DYFONATE IN A PLANO SILT LOAM SOIL. After 29 days of soil incubation, 60% of the ^{14}C(ethoxy)-Dyfonate found in the treated soil 1 day after its application was recovered as Dyfonate from the total soil columns and the percolated water as determined by glc

analyses (Table II). Only 28% could be recovered after 76 additional days (105 days of incubation), thus indicating a toal loss of 72% of the insecticide during the 105-day period. The radiocarbon determination of soil and water, however, revealed that 73 and 47% of the originally determined dosage was present after 29 and 105 days, respectively. These figures include unextractable radioactivity of 11.4 and 16.5% (29 and 105 days), as determined by soil combustion procedures.

Although the water solubility of Dyfonate is 13 ppm, the insecticide did not move appreciably through the soil columns with percolating water (Table II). Most of the radiocarbon and Dyfonate (as determined by glc) was found in the benzene phase of the soil extract. Of these amounts, three-fourths were still in the upper insecticide-treated layer, close to one-fourth in the second untreated layer and only small amounts in the lower soil layer.

The amounts of ^{14}C(ethoxy)-Dyfonate recovered from the percolated water were extremely small and were far below its water solubility. The total radiocarbon which passed through the soil columns and was found in the water amounted to 1.55% (after three percolations) and 2.1% (after five percolations) of that recovered from the treated soil 1 day after application. This water was nontoxic to mosquito larvae, except in one case (Table II, 44-day percolate), where 35% of the exposed mosquito larvae died over a 72-hr exposure period to this water. In view of the other data, this latter result cannot be explained. Three-fourths of the radiocarbon recovered from the percolated water remained in the water phase after extraction with benzene. Since the radiocarbon content in the benzene extraction phase of the percolated water was larger than the amounts of Dyfonate, as determined by glc, some metabolism of the insecticide apparently occurred. Further analysis of this benzene phase by tlc and autoradiography indicated the presence of dyfoxon in addition to Dyfonate.

Analyses of the columns whose top layer had been treated with ring-labeled ^{14}C-Dyfonate indicated (Table III) that 90.2% of the originally present radiocarbon could be recovered from the soils and percolated water after 29 days of soil incubation. Since with the ethoxy label this figure amounted to only 73.5% (Table II) and since data obtained by glc were similar in both cases, it appears that the loss of radiocarbon occurred primarily with the ethoxy part of the molecule after Dyfonate metabolism.

One of the major differences in results obtained with ethoxy and ring-labeled Dyfonate was the larger radiocarbon content in water which had percolated through ^{14}C(ring)-Dyfonate treated soil (Table III). This showed that the insecticide was to some extent metabolized into a water-soluble compound which contained the ring moiety of the molecule, thus accounting for the higher radioactivity. Based on analyses by tlc, benzene extracts of water (percolated on day 29 through ^{14}C(ring)-labeled Dyfonate-treated soil) contained Dyfonate (R_f 0.70), dyfoxon (R_f 0.42) and two unknowns (R_f 0.22 and 0.53).

(b) Movement and Persistence of Dyfonate in Different Soil Types. These experiments were conducted with nonradioactive Dyfonate in a manner similar to the previously described tests, except that in addition to a silt loam, a sandy loam and a quartz sand were used and the incubation of the soil columns was conducted at room temperature over a 17-day period. Results (Table IV) indicated that the amount of Dyfonate transported with water through the soils was a function of the soil type. With the Plano silt loam, no Dyfonate could be detected in the percolated water and no toxicity to mosquito larvae was indicated. Forty-six percent of the dose applied to the top soil layer was still present in the total column after 17 days, and four-fifths of the recovered amount was located in the upper layer, close to one-fifth in the middle untreated layer, while only small amounts could be detected in the lower soil layer.

With a sandy loam soil the total amount of Dyfonate recovered was similar to the amount found in the silt loam soil. However, the insecticide had moved to some extent through the column since both lower untreated soil layers contained amounts of Dyfonate similar to that which had remained in the top treated layers. Water which had percolated through these sandy loam columns contained initially only traces of the insecticide and also was nontoxic to mosquito larvae. However, Dyfonate could be measured in increasing amounts in water which had percolated through this soil 7 or 14 days later and it became increasingly toxic to mosquito larvae. During the first percolation of water through the soil column some of the insecticide was apparently transported into the lower untreated soil layers, yet not enough of the compound appeared in the water to be toxic. With increasing water movement, Dyfonate was finally leached through the soil and appeared in the percolated water.

When these experiments were conducted with quartz sand of low sorptive capacity, the total amount of Dyfonate recovered from the sand columns and the water was 52.35% of the dosage originally applied to the upper sand layer. However, nearly all of this (52.2% of applied) was recovered from the percolated water. Bioassays and analyses by glc indicated that the major portion of the insecticide (over two-thirds of the Dyfonate recovered during the 17-day period) was leached through the quartz sand columns with the first water percolation. These results were not surprising in view of the fact that the quartz sand is not a soil, its organic matter and silt content are zero, and its sorptive capacity is negligible. It indicates that the movement of a pesticide through soil with water, in addition to being dependent on its water solubility, is to a large extent a function of the soil type. The persistence of Dyfonate was smallest in the silt loam which probably possessed the highest activity of microorganisms.

II. Movement of Dyfonate in Soils under Nonleaching Conditions. (a) Vertical Movement and Persistence of ^{14}C-Dyfonate in a Silt Loam. Results from experiments conducted in conjunction with experiment Ia with ^{14}C-ethoxy or ^{14}C(ring)-labeled Dyfonate-treated loam soil, but in the absence of percolating water, are summarized in Table V. Under these nonleaching conditions, the insecticide also moved into lower soil strata, possibly in vapor form due to its volatility properties (Lichtenstein and Schulz, 1970). This movement, however, was not as evident as when water was percolated through the soil columns (Tables II and III). Sixty to 63% of the applied insecticide was still in the upper treated soil layer under nonleaching conditions as opposed to 45 and 49% under leaching conditions. The total radiocarbon content was smallest in those columns whose upper soil layer had been treated with ^{14}C(ethoxy)-Dyfonate. It appears, therefore, that once the insecticide had been broken down in the soil, loss of ^{14}C from the ethoxy moiety of the molecule contributed to the lower recoveries of radiocarbon from these soil columns. This was further indicated by the smaller amounts of unextractable radioactivity and the small amounts of water-soluble radiocarbon which was recovered from the water extraction phase of extracts from ^{14}C(ethoxy)-Dyfonate-treated soils (Table V).

(b) Vertical and Horizontal Movement of ^{14}C(Ethoxy)-

DYFONATE IN TWO SOIL TYPES. After untreated soils (Plainfield sand or Plano silt loam) had been placed both above and below a ^{14}C(ethoxy)-Dyfonate treated soil as described, columns were held for 14 days in both vertical and horizontal positions. Results indicated that the insecticide moved to some extent into the adjacent untreated soil layers, but this movement was similar in both vertical and horizontal directions. In Plainfield sand columns, 32 to 40% of the applied insecticide was still located in the treated layers, 12.3 to 14.5% was located in the adjacent untreated layer, and 0.1 to 1.9% was in the outer untreated layers. Once the insecticide reached the outer soil layer it could volatilize at the soil–air interphase. Since under these experimental conditions results obtained by glc and lsc were similar, the major reason for the loss of the insecticide from these soil columns was probably through volatilization, thus accounting for a total recovery of only 62 and 66% of the applied dosage.

Due to the larger sorptive capacity of the loam soil, less insecticidal movement was noticeable. Sixty-nine to 72% of the applied dosage was still located in the insecticide-treated layer, 4.1 to 8.5% was located in the adjacent untreated layers, and no Dyfonate could be detected in the outer silt loam layers, thus preventing insecticide loss through volatilization. All this apparently accounts for the total recovery of 83 to 84% of the applied dosage.

(c) HORIZONTAL MOVEMENT OF DYFONATE IN RECTANGULAR AND CIRCULAR CONTAINERS. Preliminary tests conducted with Dyfonate-treated silt loam soil, incubated in rectangular or round containers (Figures 2 and 3) at 15 and 30°C indicated two major points: the total loss of the insecticide was largest at 30°C, where 73.2 and 67.6% of the applied insecticide was lost from the rectangular and round containers, respectively, while at 15°C these figures were 23.7 and 46.9%, respectively. In addition, a sideward movement of the insecticide into untreated soils was noticeable and was most pronounced at a temperature of 30°C. After 42 days of soil incubation, 22 and 57% of the totally recovered insecticide was located in the three untreated soil layers of the rectangular and round containers, respectively, while these figures amounted to only 7 and 29% at a temperature of 15°C.

Results of duplicated tests conducted with ^{14}C(ethoxy)-Dyfonate at room temperature are summarized in Table VI. Although experimental conditions were quite different with soils in rectangular and round containers, conclusions pertaining to results obtained from both experimental setups can be drawn. Since data obtained by glc and lsc were nearly identical, metabolism of Dyfonate under these experimental conditions was negligible. This was also indicated by the fact that nearly all the recovered radioactivity was located in the benzene phase of the soil extract. Of the radiocarbon content which was recovered from all soil layers, 24% were located in the three untreated soil layers of the rectangular containers and 55.5% was in the three untreated soil layers of the round containers. The major portion of these amounts, though, was located in the inner untreated soil layer which was adjacent to the insecticide-treated soil. When data were calculated in ppm and expressed as concentration of Dyfonate in each soil layer (D-ppm, Table VI), it also became evident that the insecticide concentrations in the untreated soil layers were relatively small.

Although under nonleaching conditions Dyfonate did not move appreciably in soils within all three of the described containers, these different experimental designs could be useful with other pesticidal chemicals.

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