50% of the substrate. This is an implicit proof of the rupture of the naphthyl ring. However, it was also found that 15–20% of 1-naphthol which was transformed remained in the growth medium, apparently without further change. This appears to indicate that at least two different pathways are involved in the degradation of 1-naphthol by the bacteria under investigation.

It could be assumed that the 1-naphthol which is completely biodegraded and results in CO_2 formation may be metabolized by the pathway proposed by Davies and Evans (1964), *i.e.*, ortho hydroxylation prior to ring cleavage. The isolation and identification of 4-hydroxy-1-tetralone suggest that at least one alternate pathway involves hydroxylation of the naphthyl ring in the 4 position, and the conversion of an aromatic ring to an aliphatic cyclic compound.

Reduction of the aromatic ring was found in the metabolism of benzoic acid by *Rhodopseudomonas palustris* (Guyer and Hegeman, 1969; Dutton and Evans, 1969), but in this case metabolic activity was only observed anaerobically in the light, and this pathway differs considerably from the known oxidative mechanisms of aromatic ring dissimilation by aerobic microorganisms.

In addition to 4-hydroxy-1-tetralone, it was possible to isolate several other metabolites which are presently under investigation for identification. When the identity of these compounds has been established, it should be possible to suggest the actual pathway and to evaluate its specific features.

ACKNOWLEDGMENT

The authors are indebted to Dr. R. O. Mumma for help in the interpretation of some of the spectral data.

LITERATURE CITED

Bollag, J.-M., Liu, S.-Y., *Soil Biol. Biochem.* **3**, 337 (1971).
Bollag, J.-M., Liu, S.-Y., *Can. J. Microbiol.* **18**, 1113 (1972).
Bowie, J. H., Cameron, D. W., Williams, D. H., *J. Amer. Chem. Soc.* **87**, 5094 (1965).
Daly, J. W., Jerina, D. M., Witkop, B., *Experientia* **28**, 1129 (1972).
Davies, J. I., Evans, W. C., *Biochem. J.* **91**, 251 (1964).
Dutton, P. L., Evans, W. C., *Biochem. J.* **113**, 525 (1969).
Fernley, H. N., Evans, W. C., *Nature (London)* **182**, 373 (1958).
Guyer, M., Hegeman, G. D., *J. Bacteriol.* **99**, 906 (1969).
Jackman, L. M., Sternhell, S., "Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry," Pergamon Press, Elmsford, N.Y., 1969.
Jerina, D. M., Daly, J. W., Witkop, B., Zaltzman-Nirenberg, P., Udenfriend, S., *Biochemistry* **9**, 147 (1969).
Jerina, D. M., Daly, J. W., Jeffrey, A. M., Gibson, D. T., *Arch. Biochem. Biophys.* **142**, 394 (1971).
Kazano, H., Kearney, P. C., Kaufman, D. D., *J. Agr. Food Chem.* **20**, 975 (1972).
Lamberton, J. G., Claeys, R. R., *J. Agr. Food Chem.* **19**, 487 (1970).
Litterst, C. L., Lichtenstein, E. P., *Arch. Environ. Health* **22**, 454 (1971).
Liu, S.-Y., Bollag, J.-M., *J. Agr. Food Chem.* **19**, 487 (1971).
Miskus, R., Gordon, H. T., George, D. A., *J. Agr. Food Chem.* **7**, 613 (1959).
Murphy, J. F., Stone, R. W., *Can. J. Microbiol.* **1**, 579 (1955).
Nakanishi, K., "Infrared Absorption Spectroscopy," Holden-Day, San Francisco, Calif., 1962.
Pasto, D. J., Johnson, C. R., "Organic Structure Determination," Prentice-Hall, Englewood Cliffs, N.J., 1969.
Stewart, N. E., Millemann, R. E., Brease, W. P., *Trans. Amer. Fish. Soc.* **96**, 25 (1967).
Tausson, W. O., *Planta* **4**, 214 (1927).
Treccani, B., Walker, N., Wiltshire, G. H., *J. Gen. Microbiol.* **11**, 341 (1954).
Walker, N., Wiltshire, G. H., *J. Gen. Microbiol.* **8**, 273 (1953).

Received for review July 1, 1974. Accepted October 25, 1974. Authorized for publication on June 12, 1974, as paper No. 4711 of the Journal Series of the Pennsylvania Agricultural Experiment Station, University Park, Pa.

Residues of Leptophos and Its Metabolites following Application to Various Crop Plants

Heinz E. Braun,*[1] Freeman L. McEwen,[2] Richard Frank,[1] and Gwen Ritcey[2]

Leptophos was applied to 13 crops in the early stage of growth and to crucifers in a weekly schedule. Samples were collected at harvest and analyzed for leptophos. Residue levels in beans, cabbage, cauliflower, celery, corn, onions, pea pods, and rutabagas were found to be less than 0.1 ppm where the insecticide had been applied in the early stage of plant growth; higher levels were found in broccoli, lettuce, and carrots; pea vines were found to contain the highest residues at harvest. Where multiple applications of leptophos were made to cabbage, cauliflower, and broccoli on a 7-day schedule, residues at 2 days after the final application ranged from 1.9 to 4.6 ppm. Residues were highest on broccoli and were still present in excess of 1 ppm after 13 days. Residue consisted predominantly of the parent compound with leptophos oxon and 4-bromo-2,5-dichlorophenol accounting for only a small proportion of the total residue recovered.

The insecticide leptophos [*O*-(4-bromo-2,5-dichlorophenyl)-*O*-methyl phosphorothioate] shows promise for control of cutworms and other lepidopterous plant feeders on a number of crop plants. Residues remaining on harvested crops have not been reported except for application to wheat where no residue was found in wheat kernels, although residues of both the O analog and the parent compound remained on straw 63 days after application (Struble and McDonald, 1973). To extend our information on residues, tests were conducted to determine the extent of residues on selected crops if this insecticide were used at the stage of plant growth when cutworms are troublesome in Ontario. In addition leptophos was applied to cabbage, cauliflower, and broccoli in a weekly schedule as required to control the cabbage looper, *Tricloplusia ni* (Hübner), and the imported cabbageworm, *Pieris rapae* (Linnaeus).

[1] Provincial Pesticide Residue Testing Laboratory, University of Guelph, Guelph, Ontario N1G 2W1, Canada.
[2] Department of Environmental Biology, University of Guelph, Guelph, Ontario N1G 2W1, Canada.

MATERIALS AND METHODS

Two formulations of leptophos, 2.7 lb/gal of emulsifia-

Table I. Seeding, Treatment, and Sampling Dates for Ten Crops Treated with Leptophos and Analyzed for Insecticide Residues[a]

Crop	Date of seeding	Date of treatment	Formulation of leptophos[b]	Sampling dates
Beans	May 25	June 17	EC	July 24
Broccoli, I	May 22	June 17	EC, WP	Aug 21
Broccoli, II	May 22	Aug 4, 11, 19, 25	EC	Aug 27, Sept 4, 7
Cabbage, I	May 22	June 17	EC, WP	Aug 21
Cabbage, II	May 22	Aug 4, 11, 19, 25	EC	Aug 27, Sept 4, 7
Carrots	May 22	June 11, 18	EC	July 19, Aug 7, 27
Cauliflower, I	May 22	June 17	EC, WP	Aug 21
Cauliflower, II	May 22	Aug 4, 11, 19, 25	EC	Aug 27, Sept 4, 7
Celery	June 1	June 11	EC, WP	July 23, Aug 13, Sept 4
Corn (field)	May 22	June 17, 25	EC	Sept 17
Corn (sweet)	May 22	June 17, 25	EC	Aug 21
Lettuce	Late May	June 11, 12	EC, WP	July 3, 11
Onions (green)	May 22	June 12, 18	EC	July 19, Aug 13
Onions (main)	May 14	June 12, 18	EC	Aug 7, 27, Sept 4
Peas	May 11	June 17, 25	EC	July 17
Rutabaga	May 22	June 17, 25	EC	July 24, Aug 9

[a] Soil type is muck throughout. [b] EC, emulsifiable concentrate; WP, wettable powder.

ble concentrate (EC) and 45% wettable powder (WP), were compared on five crops at between 0.5 and 2.0 lb/acre of active ingredient to determine residue levels by harvest time (Table I). The crops included broccoli, cabbage, cauliflower, celery, and lettuce. On seven crops leptophos as the emulsifiable concentrate was compared in one and two early applications for their effect on terminal residues by harvest time (Table I). Two systems of spray applications were employed. For the crops grown on muck land (carrots, celery, lettuce, green bunching onions, and onions), the insecticide (0.5–2.0 lb of active ingredient (ai)/acre) was applied with a knapsack sprayer at the rate of 50 gal of liquid/acre. Treatments were applied to beans, peas, cabbage, cauliflower, broccoli, sweet corn, field corn, and rutabaga with a tractor-mounted low gallonage sprayer provided with a brush boom and T-jet nozzles set 20 in. apart, when these plants were in the early stage and susceptible to attack by cutworms. The boom was adjusted to cover the entire ground area and the tractor was driven at a rate so that 34 gal of liquid/acre was applied to give 0.5, 1.0, and 2.0 lb/acre. Multiple treatments of leptophos were applied for the control of the cabbage looper and imported cabbage worm on cruciferous crops. The tractor-mounted sprayer was equipped with one overhead and two drop nozzles per row, each nozzle being of the T-jet type. The equipment was driven at a rate to deliver 100 gal/acre applying 1.0 lb/acre. Leptophos treatments were applied to plots arranged in a randomized block of four replications.

(I) Emulsifiable Concentrate *vs.* **Wettable Powder.** (*i*) *Cabbage, Cauliflower, and Broccoli* (*Experiment I*). Plots were established by direct seeding on May 22. Each plot (24 ft long) consisted of one row of cabbage (variety Penn State Ballhead), broccoli (variety Waltham 29), and cauliflower (variety Snowball Improved). The insecticide was applied on June 17 and the samples for residue analysis were collected on August 21, at which time each crop was at a marketable stage. No other pesticide was used on these plants.

Four one-quarter cabbage heads were collected from each plot. The heads were trimmed as for normal marketing with four–six wrapper leaves included before quartering.

Four cauliflower heads were collected from each plot and trimmed as for normal marketing. Wrapper leaves were left on each head with the tips trimmed above the curd. Heads were quartered and one-quarter of each of the four was included in the sample for residue analysis.

Broccoli spears were collected at random to make up a 2-lb sample. Where large spears were obtained in the sampling, these were cut and only one-half used in any one sample for residue analysis.

(*ii*) *Celery.* Plants (variety Tendercrisp) were transplanted to the field on June 1 into plots two rows 3 ft apart and 14 ft long. Insecticide treatments were applied on June 11 and representative samples were collected for early harvest on July 23 and Aug 13 and final harvest on Sept 4 for residue analysis (Table I). Stalks were trimmed as for commercial sale. Stalks were split longitudinally and four half stalks were used for analysis.

(*iii*) *Lettuce.* Plants (variety Great Lakes) were set in the field in late May in 16-in. rows. Plots were four rows 8 ft long. Insecticide treatments were applied on June 11. Samples were collected on July 3 and 11 for residue analysis—the latter date being the time at which the plants would have been ready for normal harvest (Table I). The lettuce heads were trimmed to remove coarse outside leaves and four heads were split longitudinally for each sample analysis.

(*iv*) *Beans* (variety Tender Crop) were seeded in 30-in. rows on a mineral soil on May 27. Plots consisted of two rows 50 ft long. Insecticide treatments were applied on June 17 and the samples were collected for residue analysis July 24. Samples consisted of 0.25 lb of bean pods of random size collected at the normal harvesting time.

(II) One *vs.* **Two Applications.** (*i*) *Carrots* (variety Red Core Chantenay) were seeded on May 22 in 16-in. rows. Plots were four rows 5 ft long. Leptophos was applied on June 11 and those receiving a second application were sprayed June 18. Representative samples of 1 lb of finger-sized carrots were collected for the early harvest dates (July 19 and Aug 7) and 2 lb for the final harvest date (Aug 27) when the carrots were mature (Table I).

(*ii*) *Sweet corn* (variety Seneca Chief) was planted on May 22 in rows 30 in. apart. Each plot consisted of two rows 60 ft long. Insecticide treatments were applied on June 17 and 25 and samples for residue determinations were collected on Aug 21.

Six corn ears were selected at random from each plot with the husk left on for residue analysis. Corn stalks were collected at random and chopped into 4–6 in. lengths. About 2 qt of the cut material was used for residue determinations.

(*iii*) *Field corn* was planted on May 22 and plot design was similar to that for sweet corn above except only two replications were employed. Plots were treated with insec-

Table II. Residues of Leptophos and Its Two Metabolites in Six Vegetables Treated with Two Formulations of the Insecticide for Cutworm Control

Crop	Rate, lb/acre	Days from last applic.	Emulsifiable concentrate, ppm			Wettable powder, ppm		
			Leptophos	Oxon	Phenol	Leptophos	Oxon	Phenol
Beans	1.0	37	ND	ND	ND			
	2.0	37	ND	ND	ND			
Broccoli	1.0	65	Tr	ND	ND	0.071	0.002	0.004
	2.0	65	Tr	ND	ND	0.123	0.004	0.004
Cabbage	1.0	65	ND	ND	ND	ND	ND	ND
	2.0	65	ND	Tr	Tr	ND	ND	ND
Cauliflower	1.0	65	ND	ND	Tr	0.001	Tr	ND
	2.0	65	ND	Tr	Tr	0.004	ND	ND
Celery	0.5	41–42	0.067	ND	Tr	0.098	Tr	0.001
	1 0	41–42	0.147	Tr	0.003	0.200	0.002	0.003
	2.0	41–42	0.407	0.002	0.006	0.392	0.006	0.005
	0.5	62–63	Tr	ND	ND	Tr	ND	Tr
	1.0	62–63	0.032	0.001	0.001	0.021	ND	Tr
	2.0	62–63	0.042	Tr	0.002	0.035	ND	Tr
	0.5	85	0.001	ND	Tr	ND	ND	ND
	1.0	85	0.003	ND	ND	Tr	ND	ND
	2.0	85	0.027	ND	Tr	0.008	Tr	Tr
Lettuce	0.5	21	0.204	Tr	0.001	0.097	ND	0.002
	1.0	21	0.370	Tr	0.002	0.360	0.004	0.007
	2.0	21	0.250	Tr	0.004	0.180	0.002	0.004
	0.5	30	0.011	ND	ND	0.057	ND	ND
	1.0	30	0.120	Tr	0.001	0.048	ND	0.001
	2.0	30	0.044	Tr	0.003	0.120	Tr	0.002

ticide on June 17 and 25 and sampled Sept 17 for residue analysis. Both samples of corn ears and corn stover were collected as described for sweet corn above.

(*iv*) *Green Bunching Onions.* Onions were seeded on May 22 in 16-in. rows. Plots consisted of four rows 5 ft long. Insecticide treatments were applied on June 12 and 18 and samples were collected on July 19 (early harvest) and Aug 13 (final harvest) for residue analysis. Each sample consisted of approximately 1 qt of onions (including the top) pulled at random from the test area. Soil was washed from the roots.

(*v*) *Dry Onions.* Onions (variety Autumn Spice) were seeded on May 14 in rows 16 in. apart. Plots consisted of four rows 5 ft long. Insecticide treatments were applied on June 12 and 18. Samples were collected on Aug 7, Aug 27, and Sept 4 for residue analysis. On Aug 7, onions were approximately 0.75 in. in diameter and the samples consisted of 1 qt of bulbs and tops. Samples on Aug 27 and Sept 4 consisted of dry onions (bulbs only), and 2 lb of samples was taken for each plot. Soil was rinsed from the plants.

(*vi*) *Peas* (variety Perfection) were planted on May 11 with an 8-ft drill. Plots were 8 ft wide × 29 ft long. Insecticide treatments were applied on June 17 and 25 and sampling for residue analysis was done on July 17. One quart of peas in the pod was collected from the plots for residue determination as was approximately 2 qt of vines minus pods. Each was analyzed separately.

(*vii*) *Rutabaga* (variety Laurentian) was seeded on May 22 in 30-in. rows. Plots consisted of four rows 25 ft long. Insecticide treatments were applied on June 17 and 25 and samples for residue purposes were collected on July 24 and Aug 9. In each sampling both roots and tops were collected. Root samples were obtained by pulling six roots at random from each plot. Roots were trimmed and washed to remove extraneous rootlets and soil. Roots were halved and one-half of each was included in samples on residue analysis. Samples of tops consisted of approximately 2 qt of leaves cut at random from each plot.

(III) Four Applications. (*i*) *Cabbage, Cauliflower, and Broccoli (Experiment II).* A second series of plots of cabbage, cauliflower, and broccoli was direct seeded on May 22 to provide plots for evaluating control of the imported cabbage worm and the cabbage looper with leptophos. Each plot consisted of one row of cabbage, one of cauliflower, and one of broccoli in a randomized block design of four replications and separated 20 ft between replications. Plots were buffered laterally by one row of broccoli. Leptophos treatments were applied on Aug 4, 7, 19, and 25 and samples were taken for residue analysis on Aug 27, Sept 4, and Sept 7. Samples for residue analysis were taken as described above.

(IV) Analysis Procedure. *Sample Preparation.* Intact vegetable samples were lightly rinsed with water to remove adhering soil particles and shaken dry. Samples were macerated in a food chopper and a composite sample was removed for extraction. All plant samples were extracted within 24 hr of receipt in the laboratory. Samples were macerated immediately prior to extraction.

Extraction and Partitioning. Extraction, cleanup, and determination of leptophos and its two metabolites were carried out as described by Braun (1974). A 50.0-g macerated sample was extracted by blending with 170 ml of acetonitrile and water to bring the total liquid volume to 250 ml. Blending was carried out at high speed for 5 min and a 125-ml aliquot was removed by filtration using a coarse sintered-glass funnel. The aliquot was partitioned into 100 ml of benzene by shaking vigorously for 1 min and adding 350 ml of a 1% aqueous solution of sodium carbonate and shaking again for 1 min. The benzene extract was washed by shaking for 30 sec with 50 ml of water and the washings were added to the aqueous fraction. The benzene extract and rinsings were combined and drained through absorbent cotton and concentrated by rotary vacuum for column cleanup. This fraction contained the leptophos and its oxon (fraction A).

To the aqueous extract was cautiously added 35 ml of 10% sulfuric acid until the pH was 1.0. Benzene (100 ml) was then added and the mixture was shaken vigorously for 1 min. The benzene extract was drained through absorbent cotton, rinsed, and concentrated to 5–10 ml by rotary

Table III. Residues of Leptophos and Its Two Metabolites on Several Crops Treated with Emulsifiable Concentrate Formulation to Control Cutworms

Crop	Rate, lb/acre	Days from last applic.	One application, ppm			Days from last applic.	Two applications, ppm		
			Leptophos	Oxon	Phenol		Leptophos	Oxon	Phenol
Carrots	0.5	38	0.068	Tr	0.002	31	0.162	0.002	Tr
	1.0	38	0.096	ND	Tr	31	0.315	0.002	0.002
	2.0	38	0.115	0.002	0.003	31	0.340	0.001	0.002
	0.5	57	0.018	ND	ND	50	0.056	0.004	0.002
	1.0	57	0.028	ND	ND	50	0.056	Tr	Tr
	2.0	57	0.047	ND	Tr	50	0.106	0.001	0.001
	0.5	77	0.014	ND	ND	70	0.129	Tr	Tr
	1.0	77	0.017	0.001	Tr	70	0.052	Tr	ND
	2.0	77	0.026	Tr	Tr	70	0.135	0.001	0.002
Corn cobs (sweet)	1.0	65	ND	ND	ND	57	ND	ND	ND
	2.0	65	ND	ND	ND	57	ND	ND	ND
Corn stover (sweet)	1.0	65	ND	ND	ND	57	ND	ND	ND
	2.0	65	ND	ND	ND	57	ND	ND	ND
Corn cobs (field)	1.0	92	ND	ND	ND	84	ND	ND	ND
	2.0	92	ND	ND	ND	84	ND	ND	ND
Corn stover (field)	1.0	92	ND	ND	ND	84	0.30	0.002	0.006
	2.0	92	0.024	Tr	Tr	84	0.015	Tr	0.003
Onions (green)	0.5	37	0.042	ND	0.004	31	0.075	0.002	0.002
	1.0	37	0.075	0.005	0.003	31	0.262	0.003	0.004
	2.0	37	0.262	0.004	0.005	31	0.422	0.002	0.006
	0.5	63	Tr	ND	ND	57	0.017	ND	ND
	1.0	63	0.016	ND	Tr	57	0.033	ND	Tr
	2.0	63	0.071	Tr	Tr	57	0.084	0.002	0.003
Onions (main)	0.5	56	0.005	ND	ND	50	0.010	ND	ND
	1.0	56	0.008	ND	ND	50	0.014	0.001	0.003
	2.0	56	0.042	Tr	0.002	50	0.088	0.002	0.011
	0.5	76	0.001	ND	ND	76	0.019	Tr	0.001
	1.0	76	0.018	ND	ND	76	0.042	0.001	0.003
	2.0	76	0.028	Tr	Tr	76	0.059	0.001	0.004
Pea pods	1.0	30	0.002	ND	Tr	22	0.017	Tr	0.003
	2.0	30	0.005	ND	ND	22	0.027	ND	0.007
Pea vines	1.0	30	2.05	0.017	0.103	22	10.6	0.048	0.275
	2.0	30	1.08	0.021	0.138	22	7.42	0.058	0.257
Rutabaga (tops)	0.5	37	0.020	ND	Tr	29	0.035	0.002	0.005
	1.0	37	0.034	Tr	0.001	29	0.517	0.006	0.008
	2.0	37	0.041	ND	0.001	29	0.455	0.010	0.017
	0.5	53	0.015	ND	0.002	45	0.038	0.002	0.002
	1.0	53	0.019	0.002	0.005	45	0.020	0.001	0.003
	2.0	53	0.026	Tr	Tr	45	0.066	0.001	0.004
Rutabaga	0.5	37	0.006	ND	ND	29	0.004	ND	0.001
	1.0	37	0.011	ND	Tr	29	0.040	Tr	0.001
	2.0	37	0.023	Tr	0.003	29	0.063	0.002	0.006
	0.5	53	0.001	ND	ND	45	0.013	0.002	Tr
	1.0	53	0.011	Tr	Tr	45	0.015	Tr	0.001
	2.0	53	0.008	Tr	ND	45	0.016	ND	Tr

vacuum at 50°. This fraction contains the 4-bromo-2,5-dichlorophenol (fraction B).

Column Chromatography and Silylation. Two identical columns containing 10 g of silica gel and topped with 1 cm of washed sand were prewashed with 50 ml of benzene. The concentrated benzene extracts A and B were introduced to each column.

Column A was eluted with 50 ml of benzene and the eluent contained the leptophos fraction. The O analog is hydrolyzed on the column to 4-bromo-2,5-dichlorophenol which eluted from the column with 100 ml of benzene containing 2 ml of acetic acid. Column B was eluted with 100 ml of benzene containing 2 ml of acetic acid to give the phenol metabolite. Percolation rates did not exceed 5 ml/min.

The eluates were evaporated just to dryness with rotary vacuum at 50° and the leptophos residue was redissolved in 5.00 ml of methanol for analysis by flame photometric glc. The phenol analogs were redissolved in 5.00 ml of benzene plus 0.10 ml of *N,O*-bis(trimethylsilyl)acetamide reagent (undiluted) and sealed. The mixture was allowed to stand for 30 min at room temperature, was then diluted with 4 vol of benzene, and analyzed by electron capture glc.

Gas-Liquid Chromatography. Leptophos was measured by flame photometric detection attached to a Micro Tek 220 gas chromatograph with a 5% OV-1 column operating at the following parameters: column temperature, 210° isothermal for leptophos; carrier gas, helium at 60 ml/min; injection block temperature, 225°; detector tempera-

Table IV. Residues of Leptophos and Its Two Metabolites on Cruciferous Crops when Treated with Four Applications of Leptophos for the Control of Imported Cabbage Worm and Cabbage Looper

Crop	Days from last applic.	Leptophos at 1 lb/acre of emulsifiable concentrate, ppm		
		Leptophos	Oxon	Phenol
Broccoli	2	4.60	0.042	0.038
	10	1.01	0.004	0.008
	13	1.42	0.011	0.010
Cabbage	2	2.10	0.005	0.005
	10	0.94	0.008	0.003
	13	0.88	0.007	0.007
Cauliflower	2	1.93	0.018	0.018
	10	0.72	0.005	0.007
	13	0.25	0.003	0.005

ture, 210° (with ignition); detector gas flows (ml/min), hydrogen at 150, oxygen at 20, and air at 40; injection volume, 5 μl (equivalent to a 25-mg sample).

The trimethylsilyl derivatives of the phenol were measured by electron capture detection with a 4% SE-30–6% OF-1 column operating at the following parameters: column temperature, 135° isothermal; carrier gas, nitrogen at 80 ml/min; injection block temperature, 225°; detector temperature, 275°; injection volume, 5 μl (equivalent to a 5-mg sample).

Recoveries and Extraction Efficiency. Recoveries of leptophos and its metabolites were determined at fortification levels of 0.05, 0.50, and 5.0 ppm for the parent compound and 0.01, 0.10, and 1.0 ppm for the metabolites using celery and lettuce substrates. Fortifications were made directly onto the substrate prior to the blending operation. Recoveries of the parent compound averaged 95% with a standard deviation of 6.03 from the overall average. Recoveries of leptophos oxon and 4-bromo-2,5-dichlorophenol averaged approximately 10% lower with standard deviations of 8.17 for the oxygen analog and 7.95 for the phenol.

The efficiency of extraction by blending with aqueous acetonitrile was determined by comparison with exhaustive chloroform–methanol Soxhlet extraction. No significant differences in extraction efficiency were observed for either the parent compound or the metabolites.

RESULTS

In all crops treated with leptophos the parent compound was present at much higher concentrations than either of the two metabolites—the oxon or the phenol derivative. Both metabolites were in most cases close to the limit of detection of 0.002 ppm (Tables II, III, and IV).

(I) **Emulsifiable Concentrate** *vs.* **Wettable Powder.** Five of the vegetable crops treated received equal quantities of both the emulsifiable concentrate and the wettable powder and the disappearance of leptophos appeared to be very similar irregardless of formulation.

(*i*) *Cruciferous Crops.* Leptophos and its metabolites had virtually disappeared 65 days after application of 1 and 2 lb/acre as the emulsifiable concentrate from all three cruciferous crops. Where the wettable powder was used it also disappeared from cabbage and cauliflower; however, residues of 0.07 ppm were present on broccoli where 1 lb/acre had been applied and 0.12 ppm where 2 lb was used (Table II).

(*ii*) *Celery.* The disappearance of both formulations was similar with a rapid decline as a function of time (Table II, Figure 1). Residues remaining 42 days after treatment reflected the original application rates. After 63 days residues from 1 and 2 lb/acre had dropped to between 0.02 and 0.04 ppm. Disappearance between 63 and 85 days was slow with residues ranging from nondetectable to 0.03 ppm at the 85-day interval. Between 42 and 63 days a tenfold disappearance in residue occurred and this trend continued for most treatments from 63 to 85 days.

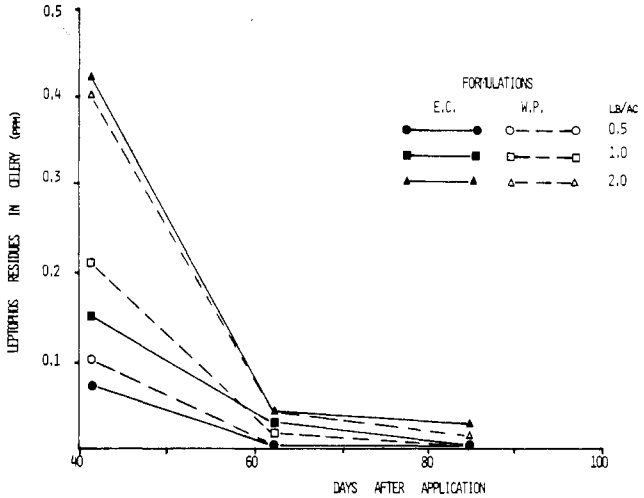

Figure 1. Leptophos residues in celery at intervals after application of two formulations at three dosage rates.

(*iii*) *Lettuce* treated with leptophos between 0.5 and 2.0 lb/acre resulted in residues that ranged from 0.10 to 0.37 ppm 21 days later. By day 30 residues had further declined by a three- to tenfold reduction and ranged from 0.01 to 0.12 ppm (Table II).

(*iv*) *Beans.* There were no residues found on green beans harvested 37 days after the application of 1 or 2 lb/acre of leptophos. At the time of spraying bean pods had not formed; therefore pods analyzed had not been exposed to spray.

(II) One *vs.* **Two Applications.** In general the rate of the original application and the days between application and harvesting had a greater effect on terminal residues than did the use of one or two applications. Carrots and onions and pea vines had terminal residues that were affected by one or two applications. Residue levels as the result of two applications of insecticides were elevated by amounts which suggested that treatments were additive.

(*i*) *Carrots.* Terminal residues 70 days after applying double treatments of 0.5–2.0 lb/acre of leptophos ranged from 0.05 to 0.14 ppm. Where only single treatments were made residues were 0.02–0.05 ppm at day 57 and 0.01–0.03 ppm at day 76. It would appear that with carrots residues from two applications were additive. It is noteworthy that after 70 days residues from some double applications were still above 0.1 ppm.

(*ii*) *Corn (Sweet and Field).* No residues were found on either cobs or stover of sweet corn treated 65 days before sampling with 1 or 2 lb/acre. With field corn residues were found on stover 84 days after treatment at residues as high as 0.3 ppm.

(*iii*) *Green Bunching Onions.* Residue levels reflect the dosage rate and number of applications. Residues present 37 days after application had been reduced by a factor of approximately 5 during the succeeding 26-day period (Table III, Figure 2).

(*iv*) *Onions (Main Crop).* The result of two applications of leptophos elevated residues at day 70 above those of a single application at day 56. Residues at day 56 ranged from 0.005 to 0.042 where treatments ranged from 0.5 to 2.0 lb/acre. Residues at day 70 ranged from 0.020 to 0.064 ppm where the same treatments were made twice at an interval of 7 days. Even so, residues were below 0.1 ppm at day 50 with both a single and a double application.

(*v*) *Peas.* Residues on pea pods reflect the rate of appli-

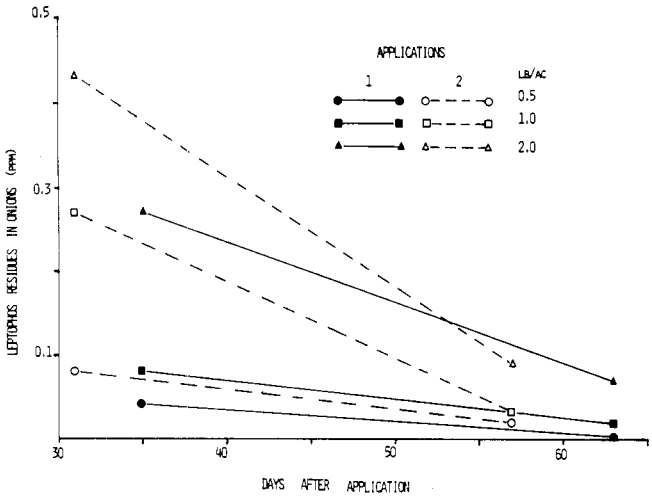

Figure 2. Leptophos residues in onions at intervals after application of two formulations at three dosage rates.

cation and number of applications; however, residues were below 0.03 ppm after 22 days from both 1- and 2-lb/acre applications applied once or twice.

Residues of leptophos in pea vines were high and significant amounts of the leptophos oxon and the phenol were present. Residues of leptophos on the pea vines did not correlate with dosage applied. However, much higher residues were found following two applications and the shorter interval than from the single application.

(*vi*) *Rutabagas.* Residues of leptophos and its metabolites were found in the tops of rutabagas at 29, 37, 45, and 53 days after application. Residues reflected the rate of application and number of applications and declined rapidly (in most instances) between the two harvest dates. Residues in the roots were not as high as those found in the tops. Residues in roots declined rapidly between the first and second harvestings and residues were below 0.02 ppm at 45 and 53 days after application.

(III) Four Applications. (*i*) *Cabbage, Cauliflower, and Broccoli.* When leptophos was applied to these crops once when the plants were very small, little residue was detectable at harvest time (Table II). Where leptophos was applied to these crops in a weekly program, initial residues following the fourth application were high. Residues declined as the interval from last application to harvest increased. In both cabbage and broccoli, levels near 1 ppm were still present 13 days after application (Table IV).

DISCUSSION

Results from the present study indicate that residues of leptophos and/or its metabolites are not present in significant amounts at harvest when this insecticide is used for cutworm control early in the season. Residues do persist, however, for a considerable time and use of leptophos near harvest will result in high residues. Intervals in excess of 13 days must be observed if residues or such cole crops as cabbage, broccoli, and cauliflower are to be less than 1 ppm.

Long persistence of trace amounts in the stover of field corn agrees with findings of Struble and McDonald (1973) with wheat straw. Whether or not such levels (0.3 ppm of corn stover; 0.07 ppm of wheat straw) pose a problem remains to be established.

ACKNOWLEDGMENT

Thanks are extended to J. Stanek and J. W. McWade for laboratory preparation, extraction, and cleanup of samples.

LITERATURE CITED

Braun, H. E., *J. Ass. Offic. Anal. Chem.* **57**, 182 (1974).
Struble, D. L., McDonald, S., *J. Econ. Entomol.* **66**, 1321 (1973).

Received for review June 26, 1974. Accepted September 30, 1974.

Bromide Residues in Apples Fumigated with Ethylene Dibromide

T. Dumas* and E. J. Bond

When three varieties of apples were fumigated with 8–24 mg/l. of ethylene dibromide for 4 hr at 13° and held at this temperature the ethylene dibromide in the edible portion (skin and pulp) desorbed to a level below 0.1 ppm in less than 13 days. In the seeds 30 ppm of ethylene dibromide was found immediately after treatment and this did not desorb during 13 days of storage at 13°. In apples stored at 4° after treatment desorption of ethylene dibromide was slower than in apples held at 13°, taking nearly 4 weeks to reach the 0.1-ppm level. The inorganic bromide residue found after the desorption of organic bromide was below 5 ppm even in apples fumigated at twice the required dosage.

Ethylene dibromide has been found effective for controlling certain insects and mites on harvested apples without causing injury to the fruit (Richardson, 1955; Sanford, 1962; Bond *et al.*, 1973). Shipments of apples requiring treatment usually have to be fumigated at cool autumn temperatures (down to 13°) with the apples subsequently being stored at about 0°. At these lower temperatures the sorption of the ethylene dibromide increases and consequently excessive residues may accumulate in the fruit. Normally unchanged ethylene dibromide and other volatile reaction products are thought to desorb to tolerable levels at higher temperatures. However, it has been found that at 13° the fumigant is retained in fruit for several days after the treatment (Dumas, 1973). Furthermore, analyses of residues using a glc method have shown that a former method of determination based on ashing and oxidation to bromate, followed by iodometric titration (Kolthoff and Yutzy, 1937), which was thought to indicate total bromide residue did not represent all of the ethylene dibromide left in the fruit. Experiments were therefore

Research Institute, Canada Department of Agriculture, University Sub Post Office, London, Ontario, N6A 5B7, Canada.