Persistence and Uptake of Cloethocarb in a Mineral Soil and Its Efficacy against the Tuber Flea Beetle, *Epitrix tuberis* Gentner

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Lance 15G (15% cloethocarb) at 2.0 and 4.0 g of ai/10 m of row was applied as an in-furrow band to potatoes (cv. Russet Burbank) at planting in a silt loam to control the tuber flea beetle, *Epitrix tuberis* Gentner. Cloethocarb persisted in the soil for more than 64 days, and its disappearance followed first-order kinetics. The calculated rate constants and half-lives were 0.032 day^{-1} and 21.7 days at the low rate and 0.023 day^{-1} and 30.1 days at the high rate. Cloethocarb was translocated readily into potato leaves. The ratio of cloethocarb concentration between the leaves and the treated soil was highest (3.35) in 37-day-old plants and lowest (0.90) in 65-day-old plants. The concentrations accumulated in the leaves were lethal to the adult beetles and effectively suppressed the populations of overwintered adults introduced to treated plants 28–30 days after planting. Almost complete adult mortality occurred within 24 h of beetle release, which resulted in a significant reduction of emergence of the next generation of adults. Control of beetles released onto treated plants 56 and 64 days after planting, however, was less effective.

INTRODUCTION

Aldicarb [2-methyl-2-(methylthio)propionaldehyde *O*-(methylcarbamoyl)oxime] has been used extensively for control of tuber flea beetle, *Epitrix tuberis* Gentner, in potatoes. In British Columbia, a furrow treatment of Temik 10G (10% aldicarb) at 2.2 kg of ai/ha is recommended for this purpose. However, repeated use of aldicarb in areas with sandy soils on Long Island, NY, and in The Netherlands has resulted in contamination of shallow wells with aldicarb including its toxic oxidative metabolites (Enfield et al., 1981; Smelt et al., 1983). Residues of aldicarb have also been detected in groundwater from wells in potato-growing areas of Prince Edward Island (Ernst et al., 1988; Lapcevic and Bobba, 1988). To reduce its use, other soil insecticides should be considered for use as alternatives.

Cloethocarb [2-(2-chloro-1-methoxyethoxy)phenyl *N*-methylcarbamate] is a systemic insecticide recently introduced by BASF AG of Germany. Its mammalian toxicity (oral LD_{50} to rat, 35.5 mg/kg) is much lower than that of aldicarb (oral LD_{50} to rat, 0.93 mg/kg). Field trials with Lance 20G (20% cloethocarb) and 15G (15% cloethocarb) have indicated that it is effective against Colorado potato beetle, *Leptinotarsa decemlineata* (Pitblado, 1985, 1986). Therefore, we conducted field trials to evaluate the efficacy of cloethocarb against the tuber flea beetle along with other soil insecticides recommended for control of the tuber flea beetle or the potato flea beetle, *Epitrix cucumeris* (Harris), and to determine the persistence and uptake of cloethocarb in a silt loam soil. The efficacy data from 1987 and 1988, and findings on the persistence and uptake of cloethocarb from 1987, are reported here. Efficacy data of aldicarb and fonofos (*O*-ethyl *S*-phenyl ethylphosphonodithioate), which are registered in Canada for control of the tuber flea beetle, are also included here for comparison.

MATERIALS AND METHODS

Field Trials. Field trials were conducted in 1987 and 1988 in a silt loam soil (Abbotsford soil series, classification Othic Humo-Ferric Podzol, pH 5.76; organic matter content 5.1%, sand 39.6%, silt 54.1%, and clay 6.3%) at the Research Sub-station, Agriculture Canada, Abbotsford, BC. The insecticides tested were Lance 15G (15% cloethocarb) at 2.0 and 4.0 g of ai/10 m, Dyfonate 10G (10% fonofos) at 1.0 and 2.0 g of ai/10 m, and Temik 15G (15% aldicarb) at 1.0 and 2.0 g of ai/10 m. All treatment plots were 3 m long rows, with potatoes (cv. Russet Burbank) planted 30 cm apart and 15 cm deep. Each row contained 10 plants and was divided in half by a 50 cm × 50 cm vertically oriented sheet of cardboard. Each insecticide was tested at two rates, paired 1 m apart, with at least 2 m between adjacent treatments. Treatments at each rate were replicated four times in a split plot design. At planting, the granular insecticides were applied alongside the seed pieces with a hand-held shaker to furrows, 20 cm wide at the top and 10 cm wide at the bottom. Since these chemicals are highly toxic, personal protective equipment including gloves and respirator must be worn by the applicator. All plots were uniformly hilled once before emergence to ensure even emergence and growth. Black plastic barricades (1.5 m high) were erected around the field perimeters to discourage tuber flea beetle immigration or emmigration.

Plots were planted on May 20, 1987, and May 24, 1988. On June 17, 1987 (28 days after planting), and on June 23, 1988 (30 days after planting), "overwintered" generation tuber flea beetles were collected from a nearby artifically infested potato planting for release into the experimental plots. In one of the two segregated halves of each treated row (i.e., five plants per half), five beetles were released on each of the middle three plants. Untreated rows of potatoes with released beetles served as controls. In 1988, additional rows of potatoes did not receive beetles to measure immigration and within-field dispersion. On July 15 in 1987 (56 days after planting) and on July 28 in 1988 (65 days after planting), a second release of beetles was made but this time into the previously uninfested half of each treatment row. Five beetles were released on each of the middle three plants in 1987, whereas only three beetles were released per plant in 1988. After all releases, beetles were allowed to feed and oviposit for a period of about 1 week, after which time parathion (*O,O*-diethyl *O*-4-nitrophenyl phosphorothioate) at 340 g of ai/ha was applied to the foliage of all plots by using a backpack sprayer.

One and six days after the first set of beetle releases, plants in each treatment plot were visually inspected for beetles by a trained observer to determine adult beetle mortality induced by feeding. On July 23, 1987, and July 29, 1988, the three consecutive plants first infested in each treatment row were cut off at ground level and removed, and the exposed areas were covered with emergence cages to quantify the emergence of the subsequent "summer" adult generation. Beetles emerging into collection

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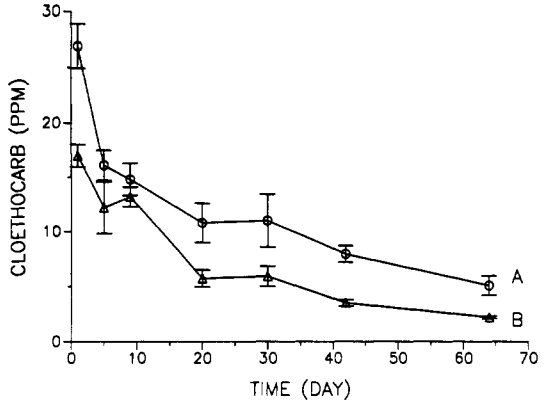

Figure 1. Concentrations of cloethocarb in silt loam soil after in-furrow band treatment at planting with Lance 15G at (A) 4.0 and (B) 2.0 g of ai/10 m of row.

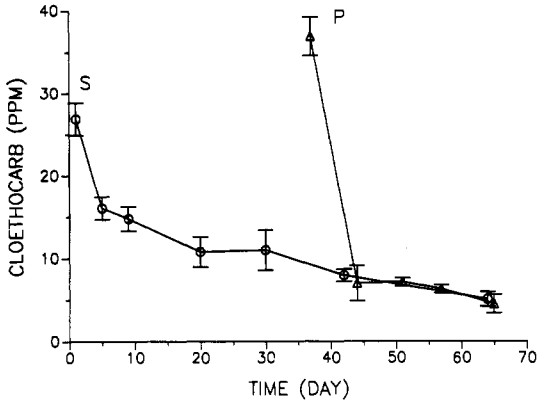

Figure 2. Concentrations of cloethocarb in leaves of potato plants (P) grown in treated silt loam soil (S) after in-furrow band treatment at planting with Lance 15G at 4.0 g of ai/10 m of row.

vials were counted daily until the number emerging in the control fell to one beetle per day. Due to the large size of the plants at the time of the second beetle release, visual samples were not taken in 1987 or 1988. Treatment efficacy of the second release of beetles was measured solely by the emergence of the subsequent adult generation into emergence cages set out on August 19, 1987, and September 2, 1988.

The persistence and uptake of cloethocarb were determined from the 1987 field trials. Concentrations of cloethocarb in soil were determined 1, 5, 9, 20, 30, 42, and 64 days after application. Twenty cores of soil (2.5 cm diameter × 20 cm deep) were taken randomly from within the cloethocarb-treated band in each replicate to form a composite sample. The four replicates of each treatment were analyzed separately for cloethocarb. Thus, there were four readings per treatment at each rate per sampling date for statistical analysis (Szeto et al., 1986), and the data given in Figure 1 are the means of those readings.

After the emergence of potato foliage, the uptake of cloethocarb in the leaves was investigated by determining the residues 37, 44, 51, 57, and 65 days after planting. Only the treatment with the high rate of Lance 15G at 4.0 g of ai/10 m of row from the 1987 field trials was studied. There were 10 plants in each replicate. One trifoliate leaf cut about 8 cm from the base of the petiole was picked from each plant near the top of the main haulm on each sampling date to form the composite sample for each replicate. The four replicates were analyzed separately, and the results given in Figure 2 are the means of those readings.

Determination of Cloethocarb. After sieving and thorough mixing, aliquots of 50 g of moist soil as collected from the field were mixed with 50 g of anhydrous Na_2SO_4 and extracted with 125 mL of ethyl acetate (pesticide grade) in 250-mL stoppered Erlenmeyer flasks by shaking for 0.5 h on a wrist-action shaker. The extracts were filtered through Whatman No. 1 filter paper into 500-mL round-bottom flasks. Extraction was repeated twice more in the same manner with 100 mL of ethyl acetate. The combined extracts were concentrated in a flash evaporator at 35 °C, and the final volumes were adjusted to 10 mL for cleanup.

Similarly, aliquots of 10 g of potato leaves were macerated with 30 g of anhydrous Na_2SO_4, and the resultant mixtures were extracted three times with 100, 75, and 75 mL of ethyl acetate by blending for 5 min in a Sorvall Omni-Mixer. The extracts were filtered through a Büchner funnel lined with a glass fiber filter paper. They were combined and concentrated in a flash evaporator at 35 °C to 10 mL for cleanup.

Aliquots of 1 mL of crude extracts equivalent to 5 g (wet weight) of soil or 1 g (fresh weight) of potato leaf were mixed thoroughly with 1 mL of cyclohexane (pesticide grade) for cleanup by gel filtration column chromatography. A Pharmacia column, Model SR 25 (45 cm × 2.5 cm i.d.), was packed with Bio-Beads S-X12. The beads were swelled in a 1:1 (v/v) mixture of dichloromethane (pesticide grade) and cyclohexane overnight before they were packed in the column. An Eldex Model B-100-S high-pressure pump was used for solvent delivery, and a Valco Model C6PX sample injection valve equipped with a 5-mL injection loop was used to inject samples onto the column. After introduction of a sample, the column was eluted with the 1:1 (v/v) mixture of dichloromethane and cyclohexane. Fraction 1, consisting of the first 58 mL, was discarded, and fraction 2, consisting of the next 150 mL, was collected. Fraction 2 contained the cloethocarb. All cleaned extracts were concentrated in a flash evaporator at 35 °C just to dryness, and the residues were dissolved in an appropriate volume of ethyl acetate for GC analysis.

GC Analysis. A Hewlett-Packard Model 5880 gas chromatograph equipped with a nitrogen/phosphorus detector was used for determination of cloethocarb. A Hewlett-Packard cool on-column inlet and a Hewlett-Packard cross-linked methyl silicone capillary column (25 m × 0.31 mm i.d.; 0.33 μm thick) were used. The operating parameters were as follows: detector temperature, 300 °C; inlet temperature, ambient. Column temperature was programed as follows: initial, 80 °C for 0.5 min; first program rate 25 °C/min to 185 °C; second program rate 5 °C/min to 225 °C and hold for 3 min. Helium was the carrier gas at 105 kPa. Detector gas consisted of hydrogen at 4 mL/min and air at 120 mL/min. Nitrogen was the makeup gas at 30 mL/min. Under the conditions described, the absolute retention time of cloethocarb was 7.51 min. Quantification was based on peak areas of the external standards injected before and after the sample.

Evaluation of Analytical Methods. Samples of soil and potato leaf were collected from the untreated plots, and they were processed and analyzed according to the described method. No gas chromatographic response that interfered with the analysis of cloethocarb was detected in any of these samples. Quadruplicate samples of 50 g of moist soil and 10 g of potato leaf were fortified by adding 0.5 mL of the stock solutions of cloethocarb in acetone to give fortification levels of 0.1, 2.0, and 10.0 ppm for soil and 0.1, 1.0, and 10.0 ppm for potato leaf. After fortification, the samples were equilibrated at room temperature in a fume hood for about 30 min before extraction. All the fortified samples were extracted, purified, and analyzed as described to determine the recovery of cloethocarb.

RESULTS AND DISCUSSION

Efficacy. In visual observations made 1 day after the initial beetle release in 1988, only one beetle was counted in the untreated check plots that did not receive beetles (dispersion check). Subsequent emergence of summer generation beetles was also significantly lower ($p < 0.05$) in the dispersion checks as compared with the checks that received beetles (infested checks). Beetles emerging from the dispersion checks began well after emergence in the infested checks. These observations suggest that (1) the potato field was relatively free of beetles prior to and shortly after the first release of beetles and (2) dispersion of beetles between plots was low. This trend did not persist for the second beetle release in 1988, however, with emergence from the dispersion checks being no different from that in the infested checks.

In 1987, significantly fewer ($p < 0.05$) beetles were visually counted in plots treated with the two rates of

Table I. *E. tuberis* Counted on the Foliage and Subsequent Emergence of Next Generation Beetles from Potatoes Grown in a Silt Loam and Treated at Planting with Granular Insecticides at Abbotsford, BC, 1987 and 1988

		E. tuberis release 1 (June 17, 1987)[a]						*E. tuberis* release 2 (July 15, 1987)[a]	
		visual counts[b]				emergence[c]		emergence	
treatment	rate, g of ai/10 m	day 1	SE	day 6	SE	mean	SE	mean	SE
cloethocarb 15G	2.0	0.0 a	0.0	0.0 a	0.0	2.3 a	1.7	10.3 NS	7.3
	4.0	0.0 a	0.0	0.0 a	0.0	5.8 ab	2.6	14.3	5.3
aldicarb 15G	1.0	3.0 b	1.4	0.8 a	0.5	12.0 b	2.4	15.0	1.3
	2.0	0.3 a	0.3	0.0 a	0.0	4.3 ab	2.6	12.3	6.0
fonofos 10G	1.0	15.5 c	1.2	10.5 b	0.5	52.0 c	6.3	20.8	6.6
	2.0	13.5 c	1.6	9.5 b	2.1	49.5 c	10.8	11.8	3.4
infested check		15.8 c	0.9	11.3 b	1.4	89.3 d	1.9	15.0	4.1
		E. tuberis release 1 (June 23, 1988)[a]						*E. tuberis* release 2 (July 28, 1988)[a]	
		visual counts[b]				emergence[c]		emergence	
treatment	rate, g of ai/10 m	day 1	SE	day 6	SE	mean	SE	mean	SE
cloethocarb 15G	2.0	0.8 a	0.3	0.3a	0.3	4.0 ab	1.7	5.8 abcd	2.6
	4.0	0.0 a	0.0	0.0a	0.0	0.3 a	0.3	0.3 a	0.3
aldicarb 15G	1.0	6.0 bc	1.6	0.3a	0.3	9.8 ab	5.4	10.3 bcd	2.7
	2.0	3.5 b	0.6	0.3a	0.3	5.8 ab	1.3	4.8 abcd	1.4
fonofos 10G	1.0	9.3 c	1.8	4.0b	1.1	23.5 bc	12.4	2.0 ab	0.4
	2.0	7.5 bc	1.7	5.8b	2.4	45.8 cd	16.6	3.0 abc	1.1
infested check		9.5 c	1.8	6.8b	1.4	71.3 d	15.6	17.5 d	7.3
dispersion check		0.3 a	0.3	0.0a	0.0	12.8 b	3.8	11.8 cd	2.6

[a] Numbers followed by the same letter in each column are not significantly different. NS, not significantly different (Duncan's multiple range test, $p < 0.05$). Data were transformed $[(X + 0.5)^{1/2}]$ for analysis of variance. [b] Mean numbers (±SE) observed on the specified days after beetle release. [c] *E. tuberis* release 1, 1987: emergence was over a 23-day period beginning 38 days after release. Release 2, 1987: emergence was over a 20-day period beginning 35 days after release. Release 1, 1988: emergence was over a 23-day period beginning 40 days after release. Release 2, 1988: emergence was over a 40-day period beginning 36 days after release.

either cloethocarb or aldicarb than in the infested checks as early as 1 day after beetle release (Table I). Dead beetles were often found alongside feeding holes on potato leaves in these systemic treatments. One day after beetle release in 1988, significantly fewer ($p < 0.05$) beetles were counted in the cloethocarb treatments than in all other treatments including aldicarb. By the sixth day, beetles counted in all cloethocarb and aldicarb plots in both years were not significantly different from each other ($p < 0.05$) but were significantly lower ($p < 0.05$) than the fonofos or infested checks. Predictably, there was no significant difference between the numbers of beetles counted in plots treated with fonofos, a nonsystemic, and the infested checks (Table I). These results show that in-furrow band treatment of cloethocarb at planting in silt loam was at least as effective as aldicarb in controlling overwintered adults feeding on plants between 28 and 30 days after planting (Table I).

The excellent control of the initial release of overwintered adults by cloethocarb and aldicarb was reflected in the subsequent emergence of the summer generation. In 1987, emergence from the systemic treatments was significantly lower ($p < 0.05$) than from the fonofos or infested check plots (Table I). This trend was also apparent in the 1988 field trial, except that emergence from the fonofos lower rate treatment was not significantly different ($p > 0.05$) from all systemic treatments except cloethocarb at the high rate (Table I). Emergence from the fonofos-treated plots was intermediate between the systemic and infested check plots and was generally significantly lower ($p < 0.05$) than the infested checks (Table I). These data suggest that fonofos is efficacious against tuber flea beetle larvae feeding below ground, since levels of the initial release of parental overwintered adults were not significantly reduced as compared with the infested checks (Table I). The possibility that the systemic treatments also controlled tuber flea beetle larvae could not be determined from this study.

The total efficacy of the granular treatments in controlling a second release of tuber flea beetles (56 and 65 days after planting in 1987 and 1988, respectively) and their below-ground progeny was measured by emergence of the subsequent adult generation (Table I). In 1987, there was no significant difference ($p > 0.05$) in emergence among any of the treatments. In 1988, however, emergence from plots treated with cloethocarb at the high rate was significantly lower ($p < 0.05$) than from the low-rate aldicarb treatment and from the infested checks (Table I). Emergence from aldicarb plots was not significantly different ($p > 0.05$) from the infested checks in either years (Table I). Emergence in the fonofos plots was significantly lower ($p < 0.05$) than in the infested checks in 1988 (Table I).

These data indicate that the systemic insecticides cloethocarb and aldicarb will provide excellent control of tuber flea beetles attacking potato plants between 28 and 30 days after planting. At an undisclosed time thereafter, the efficacies of these systemics as well as the nonsystemic fonofos became more variable. These data also suggest that Lance 15G (15% cloethocarb) at 2.0 and 4.0 g of ai/10 m of row is a highly promising soil insecticide applicable in integrated pest management programs for potato.

Persistence and Uptake. The percentage recoveries of cloethocarb from soil and potato leaves at three fortification levels are given in Table II, and they ranged from 92.3 to 114%. The data from the 1987 and 1988 field trials are reported here without correction in accordance with recoveries.

The mean concentrations and standard deviations of cloethocarb in soil from the four replicates at each rate are given in Figure 1. On the basis of the bulk density and moisture content of the soil, samples taken from the treated band (10 cm wide at the bottom) to a depth of 20 cm would have initial concentrations of about 12.5 and 25.0 ppm

Table II. Percent Recovery of Cloethocarb from Fortified Soil and Potato Leaves

	% recovery ± SD (n = 4)	
fortification, ppm	soil	leaves
10	94.6 ± 2.8	99.5 ± 2.1
2.0	92.3 ± 6.2	
1.0		109 ± 5.0
0.1	99.8 ± 9.1	114 ± 4.1

(dry weight), respectively, at the application rates of 2.0 and 4.0 g of ai/10 m of row. The actual mean concentrations 1 day after treatment were 17.0 ppm at the low rate and 26.9 ppm at the high rate. The latter was in close agreement with the theoretical value, but the former was somewhat higher (Figure 1).

Cloethocarb persisted in the silt loam. The mean concentrations in the treated band 64 days after application were 2.18 ppm at the low rate and 5.07 ppm at the high rate, which were 12.8 and 18.8% of the corresponding concentrations 1 day after application (Figure 1). To better evaluate the persistence of cloethocarb here, residue concentrations were transformed to natural logarithms for regression analysis. Linearity of the relationship between ln (concentration, y) and time (x) was tested by adding x^2 to the regression. There were no statistically significant deviations from linearity ($p > 0.05$) in either treatment. Therefore, a first-order process was assumed. The calculated rate constants and half-lives were 0.032 day^{-1} and 21.7 days for the low rate and 0.023 day^{-1} and 30.1 days for the high rate. The persistence of cloethocarb was compared with that of phorate, a systemic insecticide currently registered in Canada against potato flea beetle and Colorado potato beetle. In a concurrent study in the same field in 1987, the persistence and uptake of phorate were investigated. At 3.2 g of ai/10 m of row the rate constants for the disappearance and half-lives of phorate were 0.011 day^{-1} and 65.0 days in silt loam planted with potatoes and 0.014 day^{-1} and 49.2 days in unplanted silt loam (Szeto et al., 1990). It is apparent that cloethocarb is significantly less persistent ($p < 0.05$) than phorate.

Cloethocarb translocated readily from the treated silt loam into potato leaves (Figure 2). The ratio of cloethocarb concentration between the leaves and the treated silt loam was highest (3.35) in 37-day-old plants and lowest (0.90) in 65-day-old plants. The availability of cloethocarb did not appear to be a limiting factor in the uptake, as 5.07 ppm of cloethocarb remained in the soil 64 days after treatment (Figure 2). Growth dilution was probably the major factor contributing to the decrease in the ratio of uptake by the maturing plants. Cloethocarb appeared to be more readily translocated from the treated soil than phorate. In a concurrent study with phorate, the ratio of total phorate between the leaves and treated silt loam was highest (0.85) in 37-day-old plants and lowest (0.23) in 65-day-old plants (Szeto et al., 1990). The systemic property of cloethocarb is a major contributing factor for its efficacy against overwintered tuber flea beetles discussed earlier under Efficacy.

In view of the lower mammalian toxicity of cloethocarb, results from our field trials of 1987 and 1988 in a silt loam indicate that cloethocarb is a highly promising alternative to aldicarb for tuber flea beetle control. However, research on the movement of cloethocarb in soil is necessary to ascertain if it would or would not leach and contaminate groundwater before this chemical could be recommended as an alternative to aldicarb. Furthermore, more research is needed to evaluate further its performance in other mineral and organic soils and to investigate the extent of microbial degradation, which cloethocarb has been shown to enhance (Gauger et al., 1986; Reed et al., 1987; Racke and Coats, 1988).

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