

Effects of Selected Nematicides on Hatching of *Heterodera schachtii*

ARNOLD E. STEELE¹

Abstract: Aldicarb, carbofuran, fensulfothion, and phenamiphos were tested in concentrations of 1–100 $\mu\text{g/ml}$ for their effects on hatching of *Heterodera schachtii*. Exposure of cysts to 1 μg aldicarb or carbofuran/ml stimulated hatch whereas phenamiphos and, to a lesser degree, fensulfothion inhibited hatch. Addition of aldicarb to sugarbeet root diffusate or 4 mM zinc chloride suppressed activities of these hatching agents. Transfer of cysts previously treated with aldicarb or carbofuran to zinc chloride or water rapidly initiated hatch which finally exceeded the hatch from cysts not treated with the nematicides. **Key words:** sugarbeet diffusate, zinc chloride, aldicarb, carbofuran, fensulfothion, phenamiphos, hatch, *Beta vulgaris*, sugarbeet nematode.

Journal of Nematology 15(3):467-473. 1983.

The action and fate of carbamate and organophosphate insecticide-nematicides in soil and in plants are of considerable interest to agriculturalists concerned with minimizing crop losses through pest management. A few of the relatively new pesticides, e.g., aldicarb, are systemically active in plants and effective against pests in soil,

and minimum effective doses may vary with the developmental stage of the target organism (4). At low concentrations some of these materials are not nematicidal but do inhibit hatching of juveniles (10,11) and disrupt nematode movement (4). Steele (10,11) suggested that low concentrations of aldicarb, aldicarb sulfoxide, and aldicarb sulfone may stimulate hatching of *Heterodera schachtii* Schm. Hatch stimulation by sublethal doses of nematicides is an action which may be contrary to the intended action. Since concentration of pesticides tends to decrease with increased distance from the point of application, row treatments may provide conditions favorable to hatch-

Received for publication 24 May 1982.

¹Zoologist, U. S. Department of Agriculture, Agricultural Research Service, Western Region, P. O. Box 5098, Salinas, CA 93915.

The author gratefully acknowledges the contribution of chemicals for this study by Union Carbide Corporation, Jacksonville, Florida, and Mobay Chemical Corporation, Agricultural Chemicals Division, Kansas City, Missouri.

Mention of a proprietary product does not constitute approval, guarantee, or warranty by the U. S. Department of Agriculture.

ing at sites distant from points of application. Consequently, a study was undertaken to obtain additional information on the effects of selected concentrations of carbamate and organophosphate materials on nematode hatch.

MATERIALS AND METHODS

Technical grade aldicarb, carbofuran, fensulfthion, and phenamiphos were tested to determine if they stimulate or inhibit hatch of *Heterodera schachtii* Schm., 1871 during chemical treatment or after cysts were removed from aqueous solutions of the chemicals. Hatchability during and after treatments with nematicides was evaluated with sugarbeet root diffusate or zinc chloride hatching agents (3).

Populations of *H. schachtii* were maintained on sugarbeets (*Beta vulgaris* L.) in a greenhouse. Cysts were recovered from 60–90-day-old plants and those suitable for experimentation (8) were stored at 8 C until needed. An experimental replicate consisted of 33–35 cysts supported on a sieve immersed in 5 ml of treatment solution within a capped portion cup (9). Experiments were conducted in high-humidity cabinets maintained at constant 24 C. Sieves were transferred to fresh solutions at weekly intervals. Changes of treatment solution

were performed after three rinses of the sieves and their cyst contents in distilled water. Excess water was removed by blotting with filter paper. The influence of treatment solutions on hatch was measured at weekly intervals by counting the number of juveniles that had emerged from cysts and migrated through the sieves.

Effects of aldicarb on hatching: This test compared the emergence of *H. schachtii* juveniles from cysts exposed continuously for 9 weeks to tap water or 4 mM zinc chloride with that obtained from cysts exposed for 4 weeks to one of several concentrations of aldicarb followed by 5 weeks exposure to water or 4 mM zinc chloride (Table 1). All treatments were replicated seven times and each replication included 33 cysts.

Effects of aldicarb on hatching agents: These tests were designed to determine if aldicarb (5 µg/ml), alone or in combination with 4 mM zinc chloride or sugarbeet root diffusate, altered the stimulatory effects of these two hatching agents. The effects were measured after a 1-week exposure and again after a 6-week exposure to the various treatments (Table 2). Three treatments were applied at 8 C to suppress hatching during the initial 1-week treatment period, and all other treatments were applied to

Table 1. Effects of various treatments on hatch of *Heterodera schachtii*.

Treatment (1st 4 wk/ last 5 wk)	Data group*	Conc. of aldicarb (µg/ml)	Hatch†		
			First 4 weeks	Last 5 weeks	Total 9 weeks
Water/water	(1)	...	1,181 ab	140a	1,321 a
ZnCl ₂ /water	(1)	...	2,277 bc	324 ab	2,601 ab
Aldicarb/water	(1)	1	3,516 c	636 b	4,152 c
Aldicarb/water	(1)	2	2,429 bc	623 b	3,052 bc
Aldicarb/water	(1)	3	1,759 ab	1,405 c	3,164 bc
Aldicarb/water	(1)	4	832 a	1,920 d	2,752 abc
Aldicarb/water	(1)	5	558 a	1,918 d	2,539ab
Water/ZnCl ₂	(2)	...	1,147 a	3,278 c	4,425 bc
ZnCl ₂ /AnCl ₂	(2)	...	2,908 b	1,412 ab	4,320 bc
Aldicarb/ZnCl ₂	(2)	1	3,910 c	1,120 a	5,030 c
Aldicarb/ZnCl ₂	(2)	2	2,307 b	1,119 a	3,426 ab
Aldicarb/ZnCl ₂	(2)	3	2,715 b	1,248 a	3,963 abc
Aldicarb/ZnCl ₂	(2)	4	994 a	2,149 ab	3,143 ab
Aldicarb/ZnCl ₂	(2)	5	609 a	2,178 b	2,787 a

*For statistical analysis, data within each column were separated into groups (1) or (2). Values within a group not followed by the same letter differ significantly according to Duncan's multiple-range tests ($P = 0.05$).

†Mean of seven replicates of 33 cysts each.

Table 2. Effects of aldicarb and zinc chloride alone and in combination on hatching of *Heterodera schachtii*.

First treatment* (1 week)	Mean hatch†	Second treatment* (6 weeks)	Mean hatch†	Total hatch
Water	519 b‡	Water	394 a‡	913 bcd‡
Water	588 b	ZnCl ₂	4,197 c	4,785 f
ZnCl ₂	814 c	Water	186 a	1,000 cd
ZnCl ₂ §	1 a	Water	144 a	145 ab
ZnCl ₂	878 c	ZnCl ₂	4,557 c	5,434 fg
ZnCl ₂ §	1 a	ZnCl ₂	5,677 d	5,678 g
Aldicarb	4 a	Water	336 a	340 abc
Aldicarb	2 a	ZnCl ₂	5,886 d	5,888 g
Aldicarb	3 a	Aldicarb	13 a	16 a
Aldicarb + ZnCl ₂	115 a	Water	2,014 b	2,129 c
Aldicarb + ZnCl ₂	67 a	ZnCl ₂	5,316 d	5,383 gf
Aldicarb + ZnCl ₂	67 a	Aldicarb + ZnCl ₂	1,486 b	1,553 ed

*Cysts treated 1 week with water, 4 mM ZnCl₂, 5 µg aldicarb/ml or 5 µg aldicarb/ml + 4 mM ZnCl₂ and then 6 weeks with second treatment solutions. Concentrations of solutes for the two treatment periods were the same.

†Mean of seven replicates of 35 cysts each.

‡Values in a column not followed by the same letter differ significantly according to Duncan's multiple-range tests ($P = 0.05$). See Fig. 3.

§Cysts with solutions stored at 8 C during first treatment period only.

cysts at 24 C. All treatments were replicated seven times and each replication included 35 cysts.

Effects of carbofuran, fensulfothion, and phenamiphos: This test compared the emergence of *H. schachtii* juveniles from cysts exposed continuously for 7 weeks to tap water or 4 mM zinc chloride with that obtained from cysts exposed for 1 week with one of several concentrations of carbofuran, fensulthion, or phenamiphos followed by 6 weeks exposure to water or 4 mM zinc chloride (Tables 3–6). All tests were replicated seven times and included 35 cysts per replication.

RESULTS

Effects of aldicarb on hatching: The number of hatched juveniles that emerged from cysts treated with 1 µg aldicarb/ml for 4 weeks was significantly greater than the number from cysts exposed to tapwater (Table 1). Also, the cumulative emergence through the test period was highest from cysts initially treated with 1 µg aldicarb/ml.

Although initial treatments of cysts with 4 mM zinc chloride stimulated hatch as

measured by emerged juveniles, subsequent treatment of these cysts with water resulted in immediate decline in hatch rate. The decline indicates that hatch was stimulated only as long as the cysts were exposed to the hatching agent.

Effect of addition of aldicarb to hatching agents: Although aldicarb suppressed the activity of zinc chloride, replacement of the combined treatment with water resulted in a total hatch that was significantly greater than the total hatches initiated by water following treatments of either aldicarb or zinc chloride alone (Table 2). Significantly higher numbers of juveniles emerged from cysts first exposed to aldicarb for 1 week and followed by exposure to zinc chloride for 6 weeks than from cysts exposed to water for 1 week followed by zinc chloride for 6 weeks. Also, emergence from cysts exposed for 1 week to aldicarb alone, or in combination with root diffusate or to root diffusate at 8 C, was significantly lower than emergence from cysts exposed to water or root diffusate at room temperature (Table 3). Treatments ending with diffusate resulted in total hatches significantly

Table 3. Effects of aldicarb and sugarbeet root diffusate alone and in combination on hatch of *Heterodera schachtii*.

Initial treatment* (1 week)	Mean hatch†	Second treatment* (6 weeks)	Mean hatch†	Total hatch
Water	243 b‡	Water	793 bc‡	1,036 b‡
Diffusate§	12 a	Water	1,598 d	1,610 bc
Diffusate	2,817 c	Diffusate	974 c	3,791 d
Aldicarb	10 a	Water	306 ab	316 a
Aldicarb	15 a	Diffusate	4,818 e	4,833 e
Aldicarb	5 a	Aldicarb	5 a	10 a
Aldicarb + diffusate	2 a	Water	1,944 d	1,956 c
Aldicarb + diffusate	10 a	Diffusate	4,843 e	4,853 e
Aldicarb + diffusate	15 a	Aldicarb + diffusate	29 a	44 a

*Concentration of aldicarb was 5 µg/ml.

†Mean of seven replicates of 35 cysts each.

‡Values in a column not followed by the same letter differ significantly according to Duncan's multiple-range tests ($P = 0.05$).

§Cyst with solutions stored at 8 C during first treatment period only.

greater than those from continuous treatment with diffusate; the initial inhibition was followed first by an exceptionally high rate of hatch (during the second and third weeks) and then by a rapidly decreasing rate of hatch.

Effects of carbofuran, fensulfothion, and phenamiphos: Emergence of juveniles from cysts exposed to 1 µg carbofuran/ml for 1 week was slightly better than emergence from cysts exposed to water (Table 4). Exposure to proportionally greater concentrations of carbofuran had a proportionally

greater inhibitory effect ($r = .99$). However, if cysts were exposed to carbofuran for 1 week and were then treated with zinc chloride for up to 7 weeks, juvenile emergence was increased in proportion to the concentration of carbofuran up to 20 µg/ml.

Hatch was inhibited to some degree by all concentrations of fensulfothion or phenamiphos during the initial 1 week of treatment period (Tables 5 and 6). For fensulfothion, inhibition tended to increase with increasing concentration of the chemical within the range of 1–5 µg/ml, but the

Table 4. Effect on hatch of treating *Heterodera schachtii* cysts initially with carbofuran and then with 4 mM zinc chloride.

Treatment (1st wk/last 6 wk)	Conc. of carbofuran (µg/ml)	Cumulative hatch*		
		1 week	3 weeks	7 weeks
Water/ZnCl ₂	—	291 c	2,409 a	3,791 a
ZnCl ₂ /ZnCl ₂	—	495 d	3,387 bc	4,515 ab
Carbofuran/ZnCl ₂	1.0	434 d	2,557 a	3,891 a
Carbofuran/ZnCl ₂	2.5	217 bc	2,673 ab	4,040 a
Carbofuran/ZnCl ₂	5.0	149 b	3,649 cde	4,970 bc
Carbofuran/ZnCl ₂	10.0	19 a	4,103 cde	5,208 bc
Carbofuran/ZnCl ₂	20.0	18 a	4,420 dc	5,495 c
Carbofuran/ZnCl ₂	40.0	3 a	3,853 cde	5,033 bc
Carbofuran/ZnCl ₂	80.0	4 a	4,555 e	5,358 c
Carbofuran/ZnCl ₂	100.0	3 a	4,402 de	5,446 c

*Mean of seven replicates of 35 cysts each. Values in a column not followed by the same letter differ significantly according to Duncan's multiple-range tests ($P = 0.05$).

Table 5. Effect on hatch of treating *Heterodera schachtii* cysts initially with fensulfothion and then with 4 mM zinc chloride.

Treatment (1st wk/last 6 wk)	Conc. of fensulfothion ($\mu\text{g/ml}$)	Cumulative hatch*		
		1 week	2 weeks	7 weeks†
Water/ ZnCl_2	—	271 c	3,759 cd	5,763 a
$\text{ZnCl}_2/\text{ZnCl}_2$	—	2,487 d	3,714 cd	6,201 a
Fensulfothion/ ZnCl_2	1.0	173 b	3,548 c	5,639 a
Fensulfothion/ ZnCl_2	2.5	138 b	4,397 de	5,750 a
Fensulfothion/ ZnCl_2	5.0	19 a	4,053 cd	5,378 a
Fensulfothion/ ZnCl_2	10.0	10 a	4,886 c	6,322 a
Fensulfothion/ ZnCl_2	20.0	8 a	4,425 de	6,242 a
Fensulfothion/ ZnCl_2	40.0	6 a	3,266 c	5,480 a
Fensulfothion/ ZnCl_2	80.0	7 a	2,286 b	5,866 a
Fensulfothion/ ZnCl_2	100.0	8 a	1,158 a	5,777 a

*Mean of seven replicates of 35 cysts each. Values in a column not followed by the same letter differ significantly according to Duncan's multiple-range tests ($P = 0.05$).

†Means in this column not significantly different.

effect was not proportional ($r = -0.57$). However, when 1 week of exposure to fensulfothion was followed by exposure to zinc chloride for 6 weeks, hatch increased; at the end of 7 weeks, the cumulative hatches did not reflect the initial nematicide treatment. Similarly, 6 weeks of treatment with zinc chloride markedly increased juvenile emergence from cysts exposed to 1–20 μg phenamiphos/ml (Table 6). But emergence from cysts previously exposed to 40 and 80 μg phenamiphos/ml for 1 week followed by 6 weeks in zinc chloride was significantly ($P = 0.05$) lower than in all other treatments. While some, but not all, juveniles eventually recovered from the

treatment with 80 μg phenamiphos/ml, the data suggested that the nematicidal action of phenamiphos may increase with increased concentrations above 20 $\mu\text{g/ml}$.

DISCUSSION AND CONCLUSIONS

The increased hatches stimulated by 1 μg aldicarb or carbofuran/ml are likely responses to low rates of these chemicals. Similar findings have been reported. Batterby (1) observed hyperactivity in second-stage juveniles of *H. schachtii* exposed to 2–40 μg aldicarb sulfoxide/ml. He also noted that juveniles exposed to 2, 5, and 10 μg aldicarb/ml exhibited earlier coordi-

Table 6. Effect of hatching of treating *Heterodera schachtii* cysts initially with phenamiphos and then with 4 mM zinc chloride.

Treatment (1st wk/last 6 wk)	Conc. of phenamiphos ($\mu\text{g/ml}$)	Cumulative hatch*		
		1 week	2 weeks	7 weeks
Water/ ZnCl_2	—	313 b	2,990 b	5,127 c
$\text{ZnCl}_2/\text{ZnCl}_2$	—	2,783 c	4,216 c	5,875 cd
Phenamiphos/ ZnCl_2	1.0	41 a	3,530 bcd	5,897 cd
Phenamiphos/ ZnCl_2	2.5	8 a	4,149 de	6,139 d
Phenamiphos/ ZnCl_2	5.0	3 a	4,397 e	6,112 d
Phenamiphos/ ZnCl_2	10.0	1 a	3,801 cde	6,042 d
Phenamiphos/ ZnCl_2	20.0	1 a	3,188 bc	5,464 cd
Phenamiphos/ ZnCl_2	40.0	1 a	251 a	3,833 b
Phenamiphos/ ZnCl_2	80.0	1 a	2 a	1,872 a

*Mean of seven replicates of 35 cysts each. Values in a column not followed by the same letter differ significantly according to Duncan's multiple-range tests ($P = 0.05$).

nated movement than those exposed to aldicarb sulfoxide or aldicarb sulfone. Hough and Thomason (4) found that 0.48 μg aldicarb/ml increased hatching of *Meloidogyne javanica*, and Kämpfe (5) reported that when *H. schachtii* cysts were treated with 8 μg aldicarb/ml for 10 days and subsequently placed in rapeseed root diffusate, hatching was stimulated fourfold.

Concentrations of 5 μg /ml or greater of aldicarb, carbofuran, or fensulfotion inhibited hatch of *H. schachtii*, but phenamiphos effectively inhibited hatch at 1 μg /ml, the lowest concentration tested. However, the effectiveness of phenamiphos in controlling *H. schachtii* in soil would be influenced by the rate of its degradation to nontoxic materials and the degree to which its adsorption on soil particles restricts its movement. Although the hatch of *H. schachtii* was low during treatment of the cyst with 4 to 5 μg aldicarb/ml, subsequent treatment with water resulted in a final hatch that was not significantly different from that from water alone. In soil aldicarb is depleted in 1 week and the half-life of its oxidative metabolite, aldicarb sulfoxide, is 2 weeks (2). The apparent loss of activity in this study may have been due to the oxidation of aldicarb or aldicarb sulfoxide to aldicarb sulfone, a compound shown to be less effective than aldicarb against *H. schachtii* (1). Also, addition of aldicarb to sugarbeet root diffusate or zinc chloride suppressed the activity of these hatching agents. Exposure of cysts to zinc chloride or water after treatment with aldicarb carbofuran, or a mixture of aldicarb and a hatching agent, accelerated the hatching rate of *H. schachtii* juveniles. The residual activities of carbamates were distinctly different from those of the organophosphates fensulfotion and phenamiphos. Treatments with zinc chloride followed by fensulfotion did not give a significant increase in hatch over that initiated by water followed by zinc chloride. A similar treatment of phenamiphos followed by zinc chloride resulted in decline of hatch proportional to the concentration of the nematicide in the initial treatments.

These observations suggest that as aldicarb or carbofuran degrade to levels below those that are hatch inhibiting, the hatch

rate increases markedly, although temporarily. Whether such an increase occurs under field conditions is not known. Even if it does, the juveniles which emerge from cysts after degradation of carbamates may not be able to invade host plant roots as effectively as juveniles not exposed to the pesticides. Hough and Thomason (4) reported that movement of second-stage juveniles of *M. javanica* and *H. schachtii* was inhibited by 1 μg aldicarb/ml and that males treated with 0.01 μg aldicarb/ml failed to migrate toward females. The attraction of *Pratylenchus vulnus* Allen and Jensen, 1951 to bean roots is inhibited by aqueous solutions of carbofuran or phenamiphos at concentrations below those necessary to inhibit motility and dispersion (6,7). Steudel et al. (12) and Thielemann and Steudel (13) investigated the effects of aldicarb on the population dynamics of *H. schachtii* on sugarbeet during a 9-year period. They found that aldicarb retarded development of the nematode during the first months after application but the treatment had no effect on the final population density, which stabilized at 3,000 eggs and larvae per 100 cm^3 of soil. After plowing nematode population levels were similar in treated and nontreated plots.

The present study suggests that reported failures of aldicarb to consistently control *H. schachtii*, especially in areas relying on natural rainfall to release the chemical from its carrier, may be partly due to an increased hatching of nematode eggs exposed to low concentrations of the material.

LITERATURE CITED

1. Batterby S. 1979. Toxic effects of aldicarb and its metabolites on second stage larvae of *Heterodera schachtii*. *Nematologica* 25:377-384.
2. Bull, D. L. 1968. Metabolism of UC-21149 [2-methyl-2-(methylthio) propionaldehyde O-(methylcarbamoyl)oxime] in cotton plants and soil in the field. *J. Econ. Entomol.* 61:1598-1602.
3. Clark, A. J., and A. M. Shepherd. 1965. Zinc and other metallic ions as hatching agents for the beet cyst nematode, *Heterodera schachtii*, Schm. *Nature (London)* 208:502.
4. Hough, A., and I. J. Thomason. 1975. Effects of aldicarb on the behavior of *Heterodera schachtii* and *Meloidogyne javanica*. *J. Nematol.* 7:221-229.
5. Kämpfe, L. 1975. Untersuchungen zur Wirkung von Aldicarb auf Nematoden. In Vortragstagung (1) zu Aktuellen Problemen der Phytonematologie Am 29.5 1975 Gesellschaft der DDR,

Sektion Phytopathologie und Universität Rostock, Rostock, DDR.

6. Marban-Mendoza, N., and D. R. Viglierchio. 1980. Behavioral effects of carbofuran and phenamiphos on *Pratylenchus vulnus* I. Motility and dispersion. *J. Nematol.* 12:102-114.

7. Marban-Mendoza, N., and D. R. Viglierchio. 1980. Behavioral effects of carbofuran and phenamiphos on *Pratylenchus vulnus* II. Attraction to bean roots. *J. Nematol.* 12:114-118.

8. Steele, A. E. 1972. Evaluation of cyst selection as a means of reducing variation in sugarbeet nematode inocula. *J. Am. Soc. Sugar Beet Technol.* 17:22-29.

9. Steele, A. E. 1976. Improved methods of hatching *Heterodera schachtii* larvae for screening chemicals. *J. Nematol.* 8:23-25.

10. Steele, A. E. 1977. Effects of selected carba-

mates and organophosphate nematicides on hatching and emergence of *Heterodera schachtii*. *J. Nematol.* 9:149-154.

11. Steele, A. E., and L. R. Hodges. 1975. In-vitro and in-vivo effects of aldicarb on survival and development of *Heterodera schachtii*. *J. Nematol.* 7:305-312.

12. Steudel, W., R. Thielemann, and W. Haufe. 1978. Der einfluss von aldicarb auf die vermehrung des Rubenzystenälchens (*Heterodera schachtii* Schmidt) und den ertag von Zukerrüben in der Köln-Aachener Bucht. *Nematologica* 24:361-374.

13. Thielemann, R. and W. Steudel. 1970. Neunjährige Erfahrungen mit Monokulturen von Zukerrüben auf mit *Heterodera schachtii* (Schmidt) verseuchtem Boden. *Nachrichtenbl. Dtsch. Pflanzenschutzdienstes Braunsch.* 25:145-149.