# Behavioral Effects of Carbofuran and Phenamiphos on *Pratylenchus vulnus*. III. Penetration and Development

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*Abstract:* Penetration of bean roots by *Pratylenchus vulnus* was inhibited by continuous exposure of the nematode to carbofuran and phenamiphos and by drenches of higher concentrations of these chemicals. The inhibition was explicable by inhibition of motility, dispersion, and attraction. If incubated in aerated distilled water, nematodes treated with carbofuran and phenamiphos recovered and reproduced as well as untreated nematodes. Foliar treatments were ineffective. Apparently, no basipetal transport of carbofuran and phenamiphos occurs in beans. Both nematicides arrested nematode development by interfering with egg production and transitions between life stages. *Key Words:* beans, root-lesion nematodes, control mechanism.

Nonfumigant nematicides are being explored intensively for replacement and supplementation of fumigant nematicides. Among the compounds being tried currently for this purpose are carbofuran (2,3-dihydro-2,2-dimethyl-7-benzofuranyl methylcarbamate; Niagara Chemical Division, F.M.C. Corp., Middleport, New York) and phenamiphos (ethyl 4-(methylthio)-m-tolyl isopropylphosphoroamidate; Mobay Chem. Corp., Kansas City, Missouri). Little is known about their mode of action against nematodes, particularly at sublethal concentrations. Previous reports have suggested that direct contact of nematodes with carbamate or organophosphate nematicides or their metabolites can influence behavioral responses and development (5,6,11,12,16, 18). Investigation has been made of the effects of carbofuran on penetration by *Tylenchorhynchus claytoni* (7), of oxamyl or phenamiphos on penetration by *Pratylenchus penetrans* (6), of carbofuran and phenamiphos on the development of *Aphelenchoides rutgersi* (16), and of carbofuran on the development of *Acrobeloides nanus* (25). We (14,15) have established the threshold concentrations at which carbofuran and phenamiphos inhibit *P. vulnus* motility, dispersion, and attraction to bean roots, and we have found conditions under which *P. vulnus* recovers normal activity after the toxicant is removed. This study investigates the influence of carbofuran and phenamiphos on the penetration and development of *P. vulnus* in bean roots.

## GENERAL MATERIALS AND METHODS

Technical-grade carbofuran and phenamiphos, and *Phaseolus vulgaris* cv. Kentucky Wonder (Northrup King Seed Co., Minneapolis, Minn. 55413) were used with *Pratylenchus vulnus* obtained from carrot cultures (14). Acetone was used to help dissolve the chemicals, and Tween® 80 (Atlas Chemical, Wilmington, Del.) promoted leaf-wetting in foliage treatments.

Seedlings germinated to 3 to 5 cm high in vermiculite in a greenhouse were transplanted singly to 30-ml disposable plastic cups containing steam-sterilized quartz sand (150-250 $\mu$m). The cups were placed in a growth chamber regulated to maintain the sand at 23 C with a photoperiod of 10 h light and 14 h darkness. Plants were watered with half-strength Hoagland's nutrient solution and inoculated with *P. vulnus* when they were 8 to 10 cm tall by pouring onto the sand a 2-ml aqueous suspension containing the appropriate number of treated or untreated nematodes.

To evaluate the population structure of *P. vulnus*, seedlings were removed from the plastic cups after appropriate incubation periods. Then the roots were washed free of sand, fixed with 5% formalin, and stained in 0.5% acid fuchshin-lactophenol. Each stained root system was chopped into small pieces and macerated with a Waring blender for 30 seconds in 25 ml of water. Large root fragments were removed with a screen with 0.246-mm aperture, and life stages were counted under a stereoscope at 125× magnification.

A randomized block design was used in each experiment, and data were subjected to analysis of variance and Duncan's

multiple-range tests except as specified otherwise.

## UNTREATED NEMATODES CHARACTERIZED

The life cycle of *P. vulnus* is poorly understood, as are other factors affecting penetration and development in the host. It was obligatory to establish the characteristics of several parameters, e.g. inoculation density, duration of incubation, differential penetration of life stages etc., necessary to the selection of a set of conditions under which the influence of the nematicides could be assessed.

*Plant exposure time:* To assess penetration as a function of exposure period, seedlings were inoculated with 500 nematodes (39% $L_2$, 24% $L_3$-$L_4$, 15% male and 21% female) for six different exposure periods (Fig. 1). At the end of each incubation period, root systems (eight replicates) were washed free of sand, fixed, and stained and the nematodes were counted.

Results: Penetration increased logarithmically with exposure time until it reached a maximum at 48 h (Fig. 1).

*Inoculum density:* To assess penetration as a function of inoculum density (Fig. 2), beans were inoculated with different numbers of nematodes (population structure as above) and placed in the growth chamber. After 48 h, roots were processed, and the nematode population structure was determined.
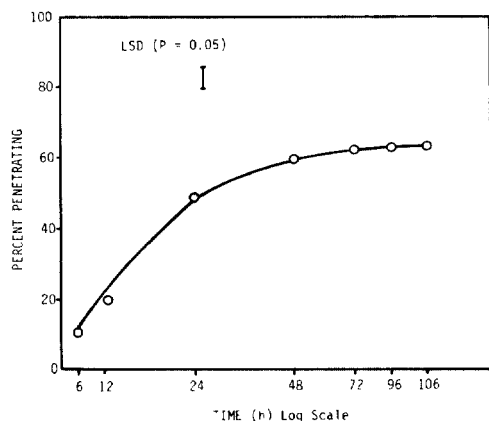
Results: Percentage penetration was not a function of inoculum density ($P = 0.01$) for the 48-h incubation period except for the lowest inoculum level tested (Fig. 2), at which the method was unreliable.

*Life stages:* To evaluate penetration of bean roots as a function of nematode life stage, bean seedlings were inoculated with known numbers of each stage alone, or in combination with other stages, and allowed 48 h for penetration. Thirty individuals of each stage, hand-picked under a dissecting microscope, were used to inoculate single seedlings. There were five replicates. For comparative purposes, a mixed-stage suspension (as above) of *P. vulnus* was also used.

Results: The percentage penetration for each stage was characteristic and consistent whether the inoculum was composed of single stages, or mixtures of stages (Table 1). Although all stages of *P. vulnus* penetrated bean roots, the capability varied: females > males > $L_3$-$L_4$ > $L_2$. Percentage penetration increased with length of incubation period up to 48 h and remained constant thereafter. Poor penetration by $L_2$ can be explained, in part, as a consequence of a lower capacity to disperse (14) and to respond to bean root emanations (15). These results are consistent with observations with *Pratylenchus penetrans* on alfalfa (22,24). The absence of correlation between the penetration of *P. vulnus* and the inoculum density, however, was not consistent with direct correlations reported on wheat roots with *P. thornei* (3), or red clover with *P. penetrans* (8), and on maize with *P. brachyurus* and *P. zea* (19).



Fig. 1. Percentage penetration of bean roots by *P. vulnus* (ca 500, mixed stages) over different exposure periods at 23 ± 0.5 C.
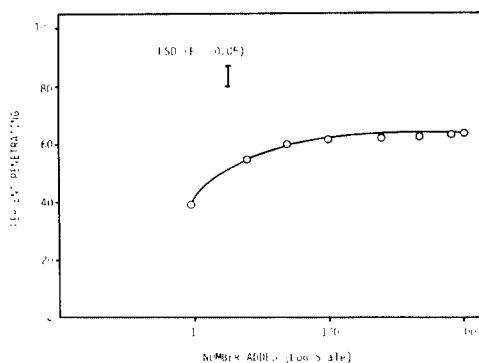


Fig. 2. Percentage penetration of bean roots with different inoculum numbers of *P. vulnus* (all stages) after 48 h at 23 ± 0.5 C.

Table 1. The percentage penetration of bean roots by each of four stages of *P. vulnus*, alone or in combination during a 48-h exposure period at 23 ± 0.5 C.

| Life stages | No. | Percentage penetrating | | | |
|---|---|---|---|---|---|
| | | $L_2$ | $L_3$-$L_4$ | Males | Females |
| $L_2$, | 30 | 18 | — | — | — |
| $L_3$, $L_4$, | 30 | — | 70 | — | — |
| Males | 30 | — | — | 73 | — |
| Females | 30 | — | — | — | 81 |
| Equal numbers of all stages | | 21 | 72 | 73 | 80 |
| Normal inoculum[a] | 500 | 24 | 65 | 77 | 86 |
| | 5000 | 22 | 66 | 75 | 83 |
| LSD ($P = 0.05$) | | NS | NS | NS | NS |

[a]Population structure: $L_2$ (39%), $L_3$-$L_4$ (24%), males (15%), females (21%).

## CHEMICAL TREATMENTS OF NEMATODES

*Effects on penetration:* To assess the ability of *P. vulnus* ($L_3$, $L_4$, adults) to penetrate bean roots while continuously exposed to nematicide, seedlings growing singly in 30-ml cups were drenched with about 90 ml of nematicide solution, placed in the growth chamber for 24 h, and then inoculated with 500 nematodes pretreated 12 h at the same concentration. There were eight replicates for each concentration. After 48 h, nematodes were extracted from the roots and counted. Solute concentrations in these experiments were 0.12M acetone; 0.05, 0.025, 0.005, 0.001, and 0.0003 mM carbofuran; and 0.003, 0.001, 0.0005, 0.0003, and 0.0001 mM phenamiphos.

Results: Under continuous exposure to nematicide, nematode penetration of bean roots was inhibited by exposures to much lower concentrations of phenamiphos than of carbofuran under identical conditions (Fig. 3). Sixteen times as much carbofuran as of phenamiphos was needed for full inhibition of nematode penetration. Inhibition increased with the concentration of each nematicide.

Discussion: Penetration of bean roots was inhibited when the system was continuously exposed to carbofuran and
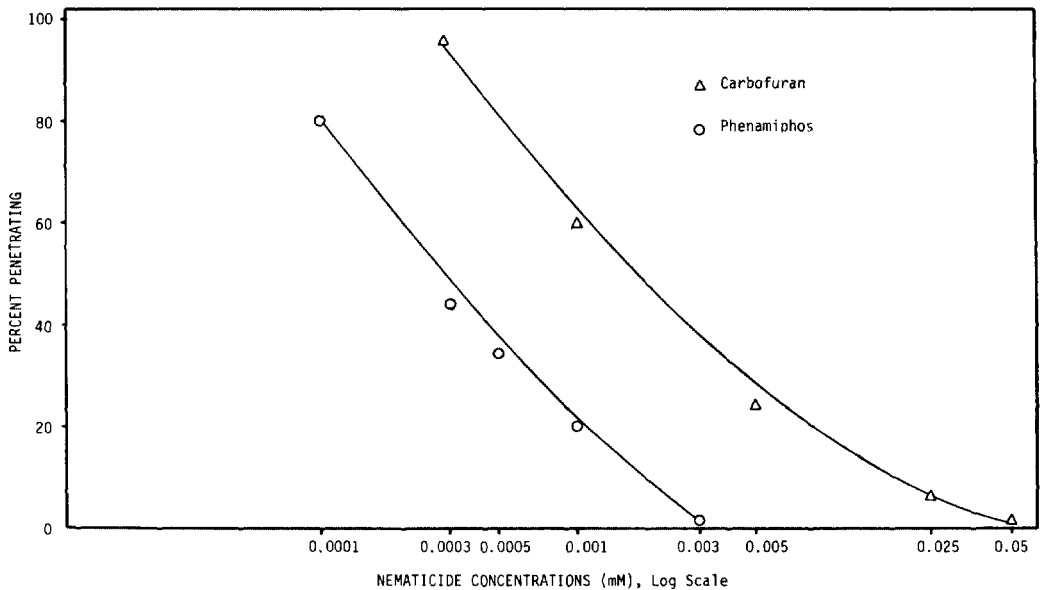


Fig. 3. Percent penetration (relative to controls) of bean roots by *P. vulnus* continuously exposed to carbofuran or phenamiphos solutions. Bean roots were pretreated with nematicides, then inoculated with a pretreated 2-ml suspension (ca 500, $L_3$, $L_4$, adults) per seedling. Nematodes recovered from bean roots 48 h after inoculation.

phenamiphos. The degree of inhibition depended on nematicide concentration. However, no particular behavioral characteristics intrinsic to the immediate act of penetration were observed to be affected by carbofuran or phenamiphos. Nematode responses inhibited could be motility, dispersion, or attraction. At higher concentrations where dispersion was inhibited, penetration was also inhibited. At a lower concentration (0.0001 mM phenamiphos, with full inhibition of attraction and no inhibition of dispersion), penetration was suppressed. At still lower concentrations, where there was no inhibition of motility, dispersion, or attraction, penetration was equivalent to that by untreated controls.

*Effects of development and reproduction:* In trials involving continuous exposure to chemicals, seedlings grown in 30-ml cups in the growth chamber were inoculated with a 2-ml suspension containing about 1,000 nematodes (all stages) and incubated for 24 h. After incubation, six replicates were used to determine the number penetrating and the population structure. The remaining seedlings were removed, and each was placed in a 150-ml glass funnel containing 0.12 M acetone; 0.01 and 0.001 mM carbofuran; and 0.001 and 0.0001 mM phenamiphos solutions. There were 12 replicates for each concentration. The root system of each seedling was supported in the funnel just below the solution surface by a stopper in the plastic lid covering the funnel. Every 5 days, nematodes accumulating at the funnel tip were collected and counted, and the solution in the funnel was replaced with fresh preparation. At 10 and 20 days of incubation, the roots of six replicates were harvested to determine the *P. vulnus* population increase and structure.

An additional experiment was devised to determine the *P. vulnus* population increase and structure as affected by a one-time drench (discontinuous treatment). Infested seedlings were transplanted into 60-ml cups of sterilized sand and drenched with 180 ml of treatment solution. Treatments replicated 12 times, included 0.6 M acetone; 1.0, 0.1, and 0.01 mM carbofuran; and 0.1 and 0.01, and 0.001 mM phenamiphos, and seedlings were grown in a growth chamber. During the growth period, dis-

tilled water was added as needed to maintain adequate moisture. Twenty days after inoculation, roots of six replicates were harvested, fixed, stained, and macerated for determinations of nematode population and structure. Roots of the remaining six replicates were chopped and placed in a mist chamber for 7 days to collect nematodes for analysis of population structure.

Results: The development and reproduction of *P. vulnus* exposed continuously to carbofuran or phenamiphos varied with the nematicide and its concentration (Fig. 4). Within 10 days of exposure, development and reproduction were arrested by 0.001 mM phenamiphos and slightly suppressed by 0.01 mM carbofuran, as can be seen by the reduction of egg numbers. The 20-day exposure amplified the differences among treatments. Carbofuran at 0.001 mM was equivalent to untreated controls with respect to population structure and reproduction. At 0.0001 mM phenamiphos, population structure was unchanged from those untreated, but actual numbers of mixed stages and eggs were significantly ($P = 0.05$) reduced (Fig. 4). Phenamiphos at 0.0001 mM completely inhibited development and reproduction; the numbers within each stage and the total number of mixed stages were equivalent to the initial population in the roots. No nematodes abandoned the root system at this concentration throughout the 20 days. At other concentrations the number of nematodes leaving the roots was inversely proportional to the increasing concentration of nematicides (Table 2). The one-time drench, in a higher concentration range, effected responses similar to those in the continuous-exposure experiments (Fig. 5). Again, phenamiphos was found to be active at lower concentrations than carbofuran. With both compounds, development and reproduction were arrested or inhibited, depending on concentration. Population structure in the one-time drench at the end of the 20-day period indicated that 10 times as much phenamiphos (0.01 vs 0.1 mM) and 100 times as much carbofuran (0.01 vs 1.0 mM) was needed to cause an effect similar to that obtained by the continuous exposure. The greater nematode counts obtained under mist extraction probably resulted from continued or resumed development and repro-
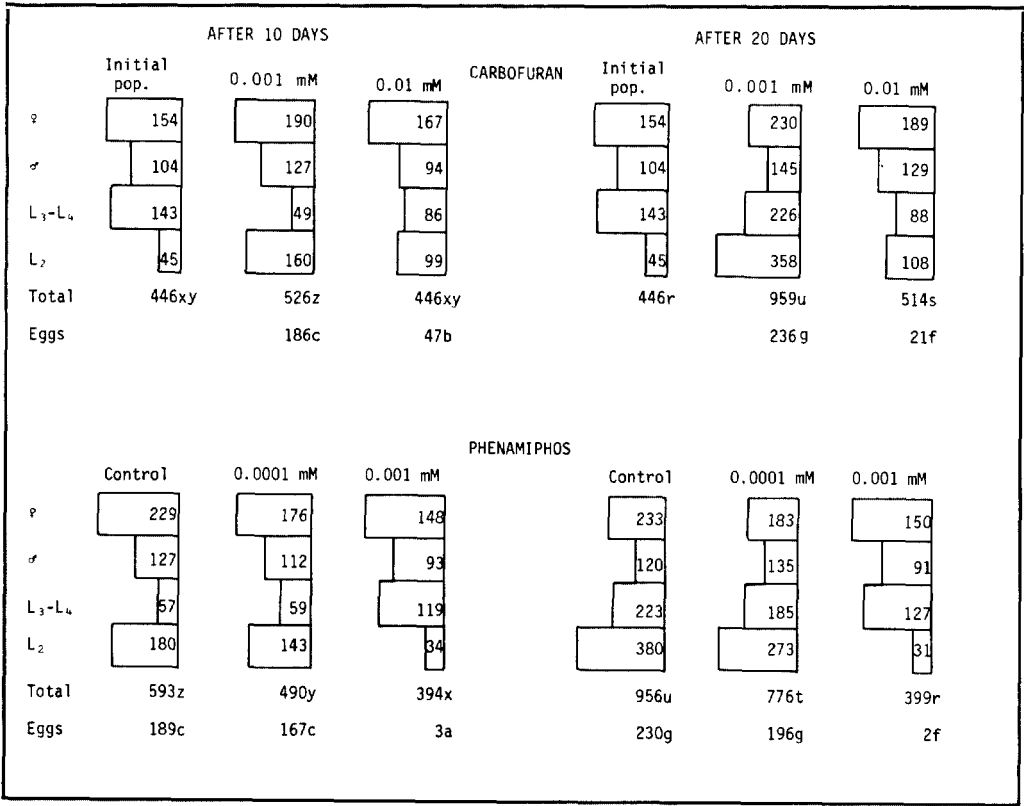
Fig. 4. Proportion of nemas in each life stage (number = counts) from infested bean roots, after 10 and 20 days of continuous exposure to carbofuran or phenamiphos solutions, as indicated. For each exposure period, unlike letters are significantly different (*P* = 0.05) according to Duncan's multiple-range test.

duction occurring after recovery, and during the additional 7 days of extraction.

Discussion: Carbamate and organophosphate nematicides may affect different behavioral aspects directly associated with nematode development: feeding (13), attraction of males by females (11), or hatching (2,4,11,12,20,23). The experiments show that a population of *P. vulnus* within bean roots is affected by continuous or discon-

Table 2. The numbers of *P. vulnus* (all stages) leaving bean roots/time when continuously exposed to nematicide solutions.

| Treatment (mM) | Days after incubation | | | | Total |
|---|---|---|---|---|---|
| | 5 | 10 | 15 | 20 | |
| Acetone 0.6 M | 15 | 9 | 12 | 12 | 48 |
| Carbofuran 0.001 | 12 | 13 | 10 | 12 | 47 |
| Carbofuran 0.01 | 3 | 4 | 2 | 2 | 11 |
| Phenamiphos 0.0001 | 4 | 4 | 3 | 2 | 13 |
| Phenamiphos 0.001 | 0 | 0 | 1 | 0 | 1 |

tinuous immersion in nematicide solutions and that development and reproduction are inhibited under suitable conditions. The effect of both chemicals appears to be similar but varies with concentration; at equal concentrations, phenamiphos is consistently more active.

It is apparent that population structures of untreated *P. vulnus*, given adequate food and proper conditions, attain a remarkably constant structure in population numbers from about 1,000 up to 250,000 (Fig. 6). The populations are about 60% larvae (35% L$_2$, 28% L$_3$-L$_4$) and 40% adults (15% males and 25% females).

The ratio of males to females (2:3) both in treated and untreated populations remained remarkably constant even though actual numbers changed (Figs. 4, 5). This suggests that there is no differential susceptibility between sexes. It is evident that these chemicals interfere with reproduction and larval development.
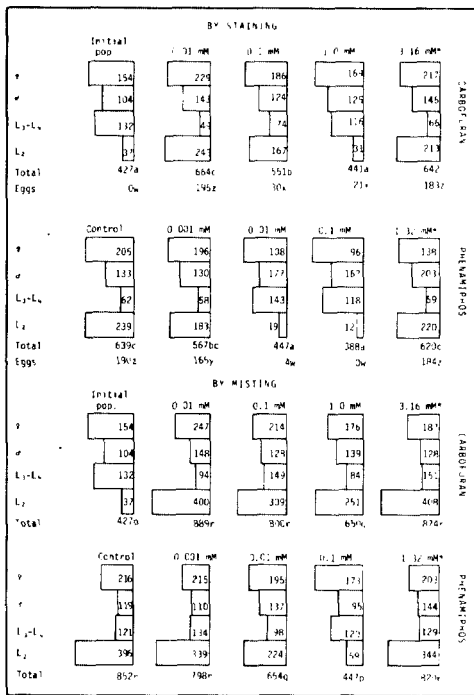
Fig. 5. Proportion of nemas of each life stage (numbers = counts) after one drench in carbofuran or phenamiphos solutions. Graphs indicate population structures 20 days after *P. vulnus* inoculation of bean roots (killed and stained); lower grouping illustrates population structures of nematodes obtained from roots mist-extracted 7 days after 20 days of growth. * indicates 20-day structures after bean foliage dips 12 h prior to inoculation and 4 times (every 5 days) subsequently. In each group, unlike letters are significantly different ($P = 0.05$), according to Duncan's multiple-range test.

Alteration of *P. vulnus* reproduction was indicated by the reduction in eggs released into root tissues at high concentrations of carbofuran (0.01, 0.1 mM) and phenamiphos (0.001, 0.01 mM) in the continuous exposure or the one-drench experiments (Fig. 5). The reduced production of eggs and egg-laying could be explained in part by the inhibition of dispersion at the same concentrations. Reduced dispersion would likely result in reduced contact between males and females but would not account for the changes in population structure. For example, though egg production was greatly reduced with carbofuran, $L_2$ production proceeded well with continuous and discontinuous exposure, thus hatching was not inhibited. Nonetheless, in these partially arrested populations, the $L_2$ did not make the transition to $L_3$-$L_4$. The $L_3$-$L_4$ group, however, continued to develop to adults, so this group was reduced in number from the initial population level. Therefore, the effect of carbofuran seems to have been to arrest the development of $L_2$ to $L_3$. Furthermore, it could be interpreted that $L_4$ was unaffected and able to make the transition to adulthood; however, the adult was inhibited in egg production, possibly because the reduced dispersion interfered with normal mating.

Phenamiphos at high concentration, with both continuous and discontinuous exposure test, not only influenced egg production but obviously inhibited transitions between life stages. This resulted in a population unchanged in number and structure for the initial inoculation. Interference of phenamiphos with dispersion was verified by the lack of any nemas, regardless of stage, abandoning the roots during the continuous-exposure period.

Once the nemas were released from the environment containing nematicide, normal egg production and larval development resumed. The one exception observed was with nemas exposed to a high concentration (0.1 mM) of phenamiphos as a one-time drench. The 7 days under mist apparently leached nematicides from the nematodes. The nematicide concentration inside the body depends on the concentration in the surrounding medium (11,17). Dilution by misting would reduce the internal concentration, releasing the nematode from the nematicide, and thus enhancing recovery. This observation was consistent with other experiments on recovery after nematicidal exposure. These results were significant since leaching of nematicides from plants, soil, and nemas could result in loss of nematicidal activity under field conditions, especially after heavy rains or normal irrigation.

*Nematode recovery:* To assess the capacity of *P. vulnus* to recover and penetrate bean roots after nematicide treatment, 500 nematodes ($L_3$, $L_4$, and adults) were placed in individual wells with 1 ml of nematicide solution. Each treatment was replicated six times. Treatment concentrations consisted of 0.6 M acetone; 6.25, 2.75, 1.25, 0.7, and 0.27 mM carbofuran; and 0.05, 0.009, 0.005, and 0.0005 mM phenamiphos. The treatment was either 1 or 7 days. During treat-
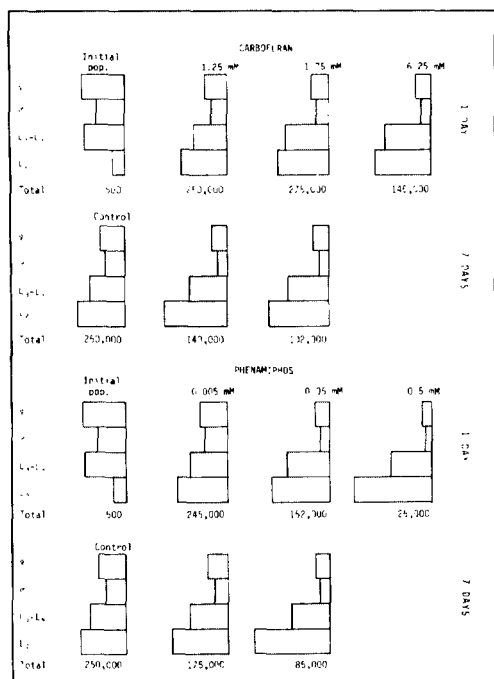
Fig. 6. Numbers of *P. vulnus* (ca 500 inoculum) and population structure after 90 days of development in carrot cultures at 23 ± 0.5 C. Nematodes were treated in carbofuran or phenamiphos solution for 1 or 7 days and subsequently washed in aerated distilled water before incubation.

ment the wells were kept in a high-humidity box placed in a growth chamber.

Recovery immediately after treatment was determined by washing each aliquant three times with aerated distilled water (ADW), inoculating to bean seedlings, and allowing 48 h for penetration. Then the seedlings were harvested, and the roots were stained by procedures already described. For longer recovery periods, treatment concentrations (7 days' exposure) were 0.6 M acetone; 6.25, 3.25, 1.25, and 0.25 mM carbofuran; and 0.5, 0.05, 0.005, and 0.0005 mM phenamiphos. After washing three times, the nematodes were returned to ADW (replaced daily at 15C). At 2-day intervals aliquants (6 replicates) were tested for penetration.

To assess whether short-term recovery estimates reflected recovery of nematodes to full potential, population increase and structure after 90 days of culture was used. Treatment solutions consisting of 0.6 M acetone; 6.25, 2.75, 1.25, 0.7, and 0.25 mM carbofuran; and 1.0, 0.5, 0.25, 0.05, 0.009, and 0.005 mM phenamiphos were applied

to 500 nematodes in individual wells for a period of 1 or 7 days. After treatment, each aliquant of nematodes was washed three times with ADW, treated with Aretan, (Plant Protection Limited, Yalding, Kent, England) 133 ppm, for 8 hours, washed aseptically in sterile ADW, and treated for 12 h with 4,000 ppm streptomycin sulfate before placing the nematodes onto sterile carrot discs for culture. After 90 days of incubation at 25C, nematodes were extracted and counted, and the population structure was determined.

Results: Penetration increased with a logarithmic decrease in concentration of nematicide solution (Fig. 7). The response curves for 1- and 7-day treatments were linear over the times and concentrations plotted. Phenamiphos was more active at lower concentrations than was carbofuran, but, with both nematicides, the longer treatment period was more effective in inhibiting penetration than the shorter treatment at the same concentration. In these tests the opportunity to recover was limited to the three washings in ADW after removal from the nematicidal environment and the 48 h in which the bean roots were exposed for penetration. Additional nematodes recovered from the 1.25-mM, 7-day carbofuran (Fig. 8) treatment when additional time up to 6 days was allowed in ADW before inoculation of beans. Additional time in ADW did not increase recovery from treatment at the next-higher or -lower concentration of carbofuran. With phenamiphos (Fig. 9) there was substantial additional recovery, up to 6 days, when incubated in ADW, but more than 6 days in ADW gave no added benefit. At 0.5 mM phenamiphos or 6.25 mM carbofuran no recovery could be demonstrated.

When recovery occured, it was real and permanent, since reproduction and population increases after treatments were normal (Fig. 6). The population increases relative to controls (Fig. 10) followed a pattern similar to penetration evaluations and depended upon treatment concentrations for both 1-day and 7-day treatments. The nearly parallel regression lines of percentage penetration (Fig. 7) after exposure to phenamiphos (1, 7 days) suggested that inhibition was not a simple function of concentration and length of exposure but was more likely
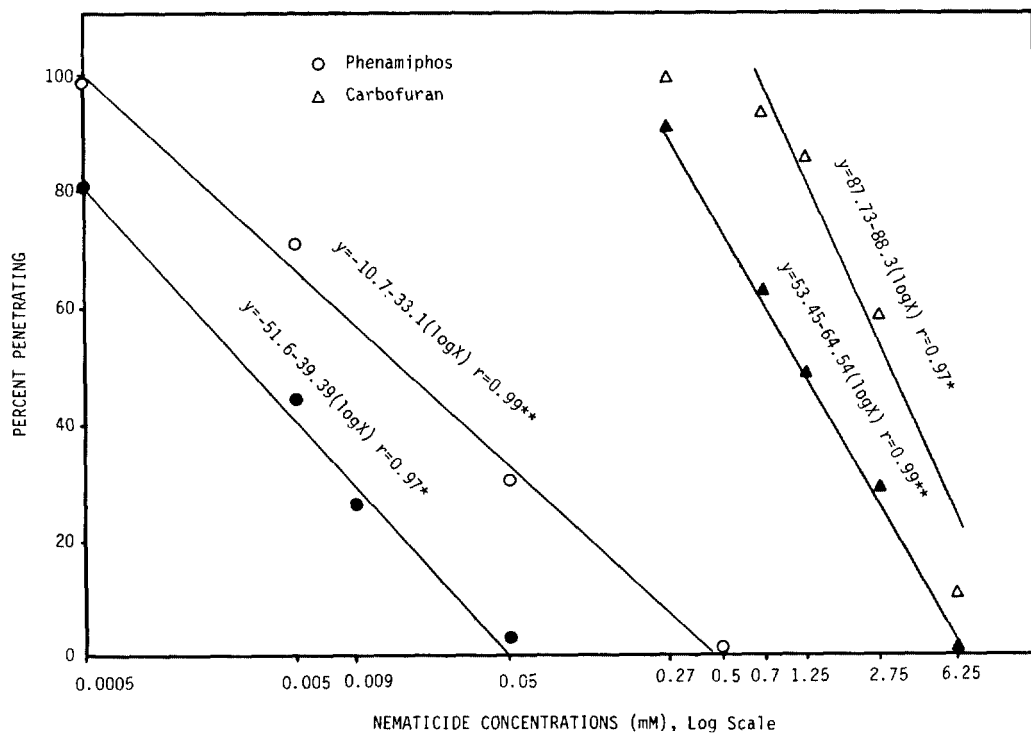
Fig. 7. Percentage penetration (relative to control) in bean roots by *P. vulnus* ($L_3$, $L_4$, adults) treated with various concentrations of nematicides for one (open symbols) or seven days (filled symbols), followed by three washings in aerated distilled water. Penetration was determined after 48 h of exposure.

the product of concentration and time of exposure over most of the active range. The corresponding curves for carbofuran had a greater slope and the product relation between the two curves did not hold as it did for phenamiphos. For carbofuran the toxicological activity as represented by relative percent penetration was more a function of concentration than of time. Furthermore the concentration range reflecting changing activity was much narrower than for phenamiphos. The sensitivity of *P. vulnus* was about two orders of magnitude greater for phenamiphos than for carbofuran. The extended recovery experiments (Figs. 8, 9)
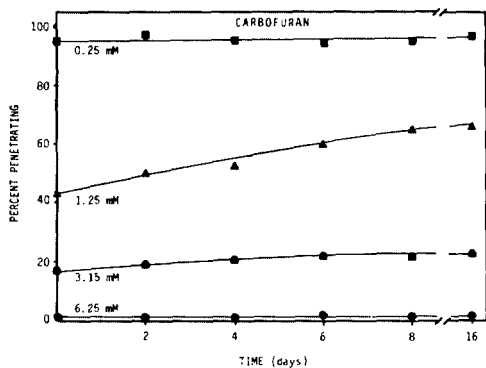


Fig. 8. Percent penetration of bean roots by *P. vulnus* ($L_3$, $L_4$, adults) relative to controls, after seven days of exposure to carbofuran and subsequent washings with aerated distilled water. Penetration was determined for six recovery periods in aerated distilled water.
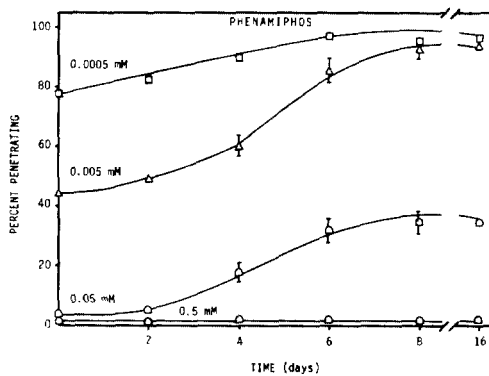


Fig. 9. Percent penetration of bean roots by *P. vulnus* ($L_3$, $L_4$, adults) relative to controls, after seven days of exposure to phenamiphos and subsequent washings with aerated distilled water. Penetration was determined for six recovery periods in aerated distilled water. Bars indicate 95% confidence limits for the means.
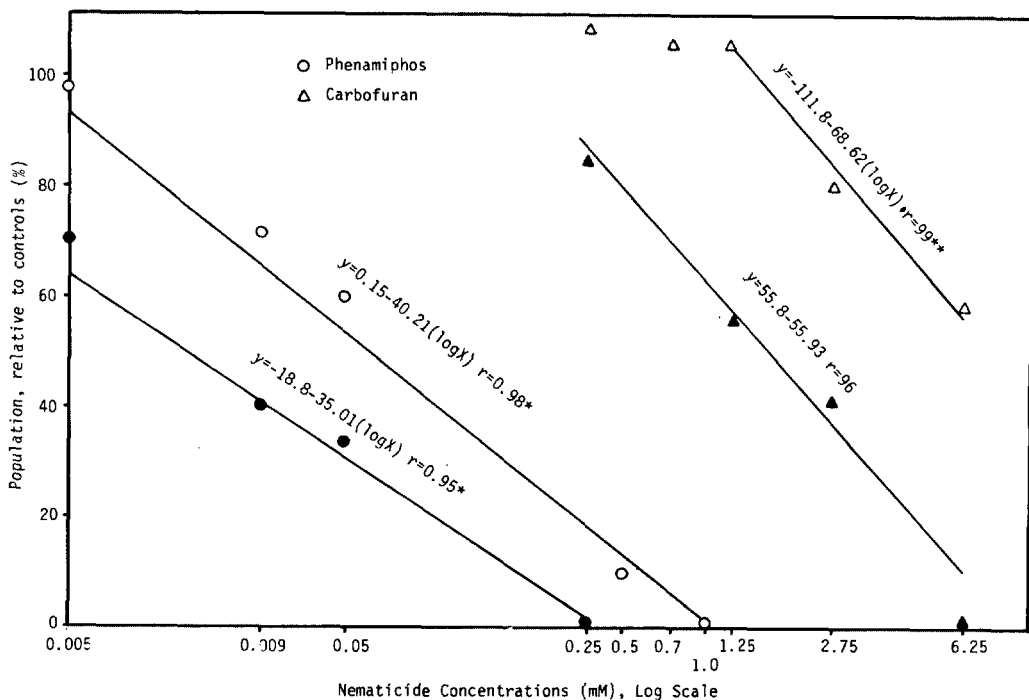
Fig. 10. Population of *P. vulnus*, as percent of control, developed on carrot cultures in 90 days at 23 ± 0.5 C from inocula treated with different concentrations of nematicides as indicated, for one (open symbols) or seven days (filled symbols). Nemas washed in aerated distilled water before incubation on carrot culture. * $P \leq 0.05$; ** $P \leq 0.01$.

indicated that with both nematicides there existed a rapid recovery (within 48 h of exposure for penetration). With phenamiphos there was an additional slow recovery, complete within 6 days of incubation in ADW. With carbofuran there appeared to be no slow recovery except at 1.25 mM concentration. Since recovery increased to a determinate value, less than 100%, and depended upon the concentration of the nematicide treatment, the *P. vulnus* population consisted of two groups of nematodes: those with a capacity to recover and those without. Of those able to recover, one subgroup recovered quickly, while another group recovered more slowly, depending on the nematicide and the concentration. These observations suggested that all individuals of a population of *P. vulnus* were not physiologically equivalent in their reaction to these nematicides, and that there were perhaps three factors (quick recovery, slow recovery, no recovery) that influenced their immobilization and penetration.

The 90-day population-increase experiments on carrots confirmed that recovered nematodes possessed the full potential of untreated nematodes (Fig. 10). The sensitivity of *P. vulnus* to phenamiphos, demonstrated in penetration experiments, was reflected in population increase studies and was about two orders of magnitude greater than for carbofuran.

## NEMATICIDE TREATMENT OF FOLIAGE

The bean seedlings used were 10 cm. tall. For assessing the effects of foliage nematicide treatments on nematode penetration, all foliage 3 centimeters above the sand level was immersed for 60 seconds in the nematicidal solutions. Plants were dipped one to six times at 12-h intervals for each concentration. Five hundred *P. vulnus* (L$_3$, L$_4$, adults) were inoculated onto the sand of each pot 72 h after each treatment and allowed to penetrate for 48 h. There were six replicates of each treatment. Stock solutions were prepared as described for other experiments with the addition of 0.1% wetting agent, and were diluted to: 4.06, 1.35, 0.45, 0.15, and 0.05 mM. To prevent chemical contamination of the soil, the seedlings were

Table 3. Summary of results of bean (var. Kentucky Wonder) foliage dip treatments in carbofuran and phenamiphos solutions.

| Conc. (mM) | Nematicide | DIPPINGS (60 seconds) | | | | |
|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 5 | 6ᵃ |
| 4.06 | Carbofuran | — | — | — | — | — |
| | | | | | very phytotoxic | |
| | Phenamiphos | — | — | — | — | — |
| | | phytotoxic | | very phytotoxic | | |
| 1.35 | Carbofuran | — | — | — | — | — |
| | | | | | phytotoxic | |
| | Phenamiphos | — | — | — | — | — |
| | | | | phytotoxic | | very phytotoxic |
| 0.45 | Carbofuran | — | — | — | — | — |
| | Phenamiphos | — | — | — | — | — |
| | | | | phytotoxic | | |
| 0.15 | Carbofuran | — | — | — | — | — |
| | Phenamiphos | — | — | — | — | — |
| 0.05 | Carbofuran | — | — | — | — | — |
| | Phenamiphos | — | — | — | — | — |

ᵃ = 12-h interval between dip treatment.
— = no inhibition of nematode penetration.

held inverted until the foliage dried. Stems were wrapped with filter paper to avoid drainage of water of guttation or condensation from the foliage or petioles to the sand.

For determining the influence of nematicides on development and reproduction, pot cultures were maintained for 20 days. The foliage was dipped into nematicide solutions (3.16 mM carbofuran or 1.32 mM phenamiphos) once 12 h before inoculation, and subsequently four more times at 5-day intervals. Seedlings were grown in a growth chamber under conditions described previously.

Results: All foliar treatments were ineffective. No inhibition of penetration (Table 3) or of development of reproduction (Fig. 5) was observed, even at phytotoxic concentrations of the chemicals. The results from attraction, penetration, and development experiments with foliar applications of carbofuran or phenamiphos indicated that basipetal transport did not occur in bean seedlings under the conditions of these experiments.

Discussion: These results are consistent with results for phenamiphos on tomato (*L. esculentum*) and broad bean (*Vicia faba*) plants (6,21) but inconsistent with other reports on pineapple (1,10) and sugar beets (9) and with observations of carbofuran on

tomatoes (21). Basipetal movement of a "systemic" nematicide applied to foliage would be a useful control method for nematode pest problems in foliage, stem, or roots. With current granular nonfumigant nematicides, acropetal movement is much more common than the desirable basipetal one. Because of the importance of the problem, the apparent inconsistencies of various reports merit further study.

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