

# Survival and Horizontal Movement of Infective Stage *Neoaplectana carpocapsae* in the Field

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**Abstract:** Infective stage *Neoaplectana carpocapsae* were applied to a furrow of loam soil in the field. At regular intervals over a 7-week period, soil samples were taken at distances of 0, 10, 20, 30, and 40 cm on either side of the furrow. Live, infective nematodes were recovered from the soil at all sampling dates over the 7-week period, and nematodes slowly spread out from the original line of application, averaging a distance of 4.35 cm/day.

**Key words:** entomogenous nematode, dispersal in soil.

Steinernematid nematodes show great promise as biological control agents of insect pests and are now being commercially produced for this purpose (8). One of the important aspects regarding their use is their survival and distribution after application in the field. Various workers have studied the migration of infective stage juveniles of *Neoaplectana* under laboratory conditions (1-3,5,6). This study was conducted to determine the survival and distribution of *N. carpocapsae* infective juveniles when applied to field soil. We prefer to maintain the name *Neoaplectana carpocapsae* for this nematode. Reasons for this decision have already been presented (7).

## MATERIALS AND METHODS

The experiment was conducted on the Oxford plots of the University of California at Berkeley. The soil is a Dublin loam (40% sand, 33% silt, 26% clay). The soil

has a CEC (cation exchange) of 20.1 meq/100 g, a pH of 5.2-5.6, and an organic matter content of 0.7-1.24%.

A plot of land 3 m wide by 15 m long was selected for the experiment. A furrow 5 cm deep was made lengthwise through the middle of the plot. The experiment was initiated on 8 October 1984 with dry, barren soil. The plot was irrigated by a sprinkler on 7 October 1984.

Nematodes used were the 42 strain of *Neoaplectana carpocapsae* Weiser (supplied by Biosis, Palo Alto, Calif.). The nematodes had been reared in *Galleria mellonella* larvae and stored at 8 C for 3 months. Infective juveniles suspended in 8 liters of water were poured into the furrow at a rate of 1 million nematodes per linear meter. Care was taken to agitate the suspension during application and to apply the suspension uniformly in the furrow.

Before the furrow was covered, one corn seed was placed every 6 cm in the furrow. The plants were used as indicators of soil moisture content. Only when the plants wilted was the plot irrigated.

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TABLE 1. *Neoaplectana carpocapsae* infective juveniles recovered from soil samples collected at various distances from the line of nematode application.

Date (1984)	Distance (cm)									
	0	10 N	10 S	20 N	20 S	30 N	30 S	46 N	46 S	Total
8 October (immediately after application)	11,966	0	0	0	0	0	0	0	0	11,966
15 October	1,080	520	1,240	165	546	178	0	0	0	3,729
22 October	538	921	288	262	114	80	96	58	90	2,447
29 October	416	390	300	110	92	104	100	76	96	1,684
7 November	392	288	270	116	130	44	54	33	83	1,410
13 November	308	245	144	285	196	135	90	36	97	1,536
28 November	284	345	220	167	115	260	92	93	45	1,621

Each figure represents the number of nematodes recovered from a combination of five samples taken on the line of application (0), north of the line (N) or south of the line (S) of application.

Immediately after application of the nematodes and on days 7, 14, 21, 30, 36, and 51 following, soil samples were taken to monitor nematode movement.

Soil samples were taken with a 5-cm-d soil auger to a depth of 15.3 cm. At each sampling date, a transect line was made (north-south) to the original furrow (east-west) and samples were taken at 0, 10, 20, 30, and 46 cm on either side of the line of application. At each sampling date, five replicate soil samples were collected at each distance from both sides of the original furrow. All five samples collected on each side of the furrow at each distance were combined for extracting and counting nematodes.

Nematodes were extracted from the now combined 1,500 cm<sup>3</sup> soil samples by the Cobb sieving and gravity method. Because of the high clay content of the soil, the final sieving was through a 38- $\mu$ m-pore sieve, and the residue was placed in a Baermann funnel to allow the nematodes to migrate through the debris.

The five samples of nematodes from the soil collected at each distance were combined and agitated, and three 1-ml samples were counted and averaged. The average was adjusted to the total volume of suspension to estimate the total number of nematodes recovered.

After counting, approximately 100 nematodes were placed in petri dishes lined with filter paper together with 10 wax moth larvae (*Galleria mellonella*). After 7 days, the moth larvae were examined for nematode infection.

Soil temperature at 15 cm deep was 9–19 C, and air temperature was 13–21 C, over the sampling period. Irrigation of 2

cm water was provided each time on 8 and 12 October. Autumn rains started on 19 October and provided adequate moisture for the remainder of the experiment. The corn grew to 15 cm tall by the conclusion of the experiment.

Before the application of nematodes, plot soil was sampled for natural populations of *Neoaplectana*. Soil samples were separated into two portions; one portion was extracted as described earlier and the nematodes were examined. Only a few rhabditid and juvenile plant parasitic nematodes were recovered. The second portion was placed in containers with larvae of *Galleria mellonella* to "bait" any *Neoaplectana* present. No insects died from nematode infection.

Only the infective stages of *Neoaplectana*, which on microscopic examination can be distinguished from the juveniles of microbotrophic rhabditids, were counted.

## RESULTS

Viable juveniles of *N. carpocapsae* were recovered from soil during the entire 7 weeks of the experiment (Table 1). Seven days after application, nematodes were recovered 30.5 cm from the point of application indicating that some juveniles moved as much as 4.35 cm a day. Two weeks after application, infective juveniles were recovered from soil 46 cm from the point of application.

Assuming no nematodes dispersed deeper than 15.3 cm, the initial sampling should have collected all the nematodes in 25 linear cm of sampling area. In subsequent sampling, some loss was expected due to dispersion of the nematodes outside the sampling area. In order to correct for this factor, Oliver's formula (unpubl.) for col-

TABLE 2. Total infective juveniles of *Neoaplectana carpocapsae* from Table 1 corrected for loss of nematodes in samples taken after 8 October.

Date (1984)	Totals (corrected)	Recovery (%)	Loss (%)
8 October	11,966	100	0
15 October	9,561	80	20
22 October	6,274	52	48
29 October	4,317	36	64
7 November	3,615	30	70
13 November	3,938	33	67
28 November	4,156	35	65

lection efficiency was used, where  $d$  = diameter of sample size (5 cm in this case) and  $s$  = distance between samples (10 cm in this case). This formula for collection efficiency

$$\frac{\pi d^2}{4ds} = \frac{\pi d}{4s} = \frac{\pi}{8} = 0.39$$

then provides a correction factor which gives corrected sampling values presented in Table 2. Losses obtained after 8 October are then attributed to death by predation or desiccation, since reduction of numbers due to dispersion has been taken into account with the correction factor for collection efficiency.

Throughout the study period, nematodes extracted from field soil were infective to *Galleria* larvae in the laboratory.

#### DISCUSSION

This study shows that infective juveniles of *N. carpocapsae* can remain viable and infective for at least 7 weeks after application to field soil under soil, temperature, and humidity conditions reported here and that they disperse from the point of application. The soil was barren and dry at the beginning of the experiment, and since insect hosts were scarce or absent (none were recovered during nematode extraction of soil), the nematodes would be expected to migrate in search of hosts as has been noted in laboratory experiments (2,3).

Because of the nature of the soil extraction process, these experiments monitored only living nematodes. Also, the extraction efficiency of the sieving-Baermann funnel method for soil nematodes is low (4). On 8 October 11,966 nematodes were extracted immediately after application of 1 million nematodes/linear meter. This represents five samples (each sample = 5 linear cm), for a total of 25 linear cm of furrow.

Since approximately 1 million nematodes were applied per linear meter, or 10,000 per linear cm, recovery of 479 nematodes/cm would yield an extraction efficiency of 4.8%. When applied, some of the nematodes probably washed outside the range of the soil auger, and others probably passed through the 38- $\mu$ m-pore sieve, resulting in a recovery rate lower than the extraction efficiency of 10–25% in previous reports (4).

Horizontal movement of neoaplectanids in soil was previously measured only under laboratory conditions (1,5). Georgis and Hague (1) noted that in 5 days infective juveniles of *N. carpocapsae* moved 7 cm in sterilized forest soil, providing a dispersal rate of 1.4 cm/day, whereas Moyle and Kaya (5) observed that they moved up to 14 cm in 2 days in sand, giving a dispersal rate of 7 cm/day. Our observed rate of 4.35 cm/day in field clay loam soil falls between these two values.

The two known methods of active movement by infective juvenile neoaplectanids are crawling and leaping (9). It appears that *N. carpocapsae* is fairly efficient at dispersal horizontally under field conditions.

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