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Survival of entomopathogenic nematodes within host cadavers in dry soil

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Abstract

Our objectives were to determine whether entomopathogenic nematode emergence from host cadavers is influenced by soil moisture, whether the nematodes can survive adverse desiccating conditions in the soil by remaining within the host cadaver, and whether differences in such an adaptation occur among species. In the first experiment, wax moth larvae killed by *Steinernema glaseri*, *Steinernema carpocapsae*, *Steinernema riobravis*, or *Heterorhabditis bacteriophora* were placed in soil water potentials ranging from −500 MPa (very dry) to −0.006 MPa (moist). No infective juveniles (IJs) emerged from cadavers at −500 MPa, and only few *S. glaseri* and *S. carpocapsae* emerged at −40 MPa. Large numbers of IJs emerged at ≥ −5 MPa from cadavers containing *S. carpocapsae*, *S. glaseri*, or *H. bacteriophora*. *S. riobravis* emerged only at ≥ −0.3 MPa. In the second experiment, cadavers were left in dry soil (−40 MPa) for various periods of time before being rehydrated. The number of IJs emerging per cadaver and the infectivity of the emerged IJs were determined. IJ emergence declined with the time that the cadavers were left in dry soil. Regression analysis predicted that IJ emergence from cadavers with *S. glaseri*, *S. carpocapsae*, *H. bacteriophora*, or *S. riobravis* would stop after 27, 62, 80, and 111 days, respectively, in dry soil. We hypothesize that *S. carpocapsae*, a sit-and-wait forager, survives longer than *S. glaseri* because it is adapted to infect insects near the soil surface, whereas *S. glaseri*, an actively searching forager, is adapted to infect insects deeper in the soil profile. Cadavers colonized by *S. carpocapsae*, therefore, are more likely to be exposed to dehydrating conditions. *H. bacteriophora*, an actively searching forager, may survive longer within cadavers because the gummous consistency of its host cadavers retains moisture very efficiently. *S. riobravis* may survive for considerable lengths of time within cadavers in adaptation to the subtropical, semiarid climate of its geographic area of origin. © 1997 Elsevier Science B.V.

Keywords: *Steinernema*; *Heterorhabditis*; Soil ecology; Soil water potential; Biological control

1. Introduction

Terrestrial nematode activity and survival are affected by many abiotic soil factors (Wallace, 1966) among which soil moisture is considered central.

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Low soil moisture can adversely affect nematode activity and survival, but a considerable number of terrestrial nematode species survive some degree of dehydration if the drying process is gradual (Womersley, 1987). Such drying conditions are common in soil because the relative humidity (RH) in the interstitial spaces stays close to 100% and drops drastically only in extremely dry conditions.

A limited degree of dehydration resistance has been observed with entomopathogenic nematodes in the genera *Steinernema* and *Heterorhabditis*. The third-stage infective juvenile (IJ), the only free-living stage of entomopathogenic nematodes, does not survive rapid desiccation in laboratory experiments under low RH regimes (Simons and Poinar, 1973; Kung et al., 1991; Womersley, 1990), but it can persist considerable lengths of time in dry soil (Kung et al., 1991). The gradual desiccation in soil may provide IJs enough time to adapt physiologically into a partially desiccated and immobilized quiescence state (Womersley, 1990).

Most studies of the effects of drying on nematodes have been on free-living species. Entomopathogenic nematode IJs, however, are found both free in the soil or within a cadaver. IJs that find a suitable insect host, enter it through natural openings and release symbiotic bacteria that propagate in the host's hemocoel, killing it within 2 days. The nematodes feed on the bacteria and host tissue, go through two or three parasitic generations, and finally emerge from the depleted host cadaver as IJs (Poinar, 1990). During the development within the cadaver which usually takes between one and three weeks, the external environmental conditions may change drastically. Rapid decline in soil moisture can occur in the upper soil layers, especially in sandy soils with little water retention capacity; entomopathogenic nematodes are very frequently isolated from such soils (Hara et al., 1991; Stock, 1995; Strong et al., 1996). In addition, the infected host may move to areas with low soil moisture or RH before succumbing to the infection. In such situations, the host cadaver may retain moisture better than the surrounding soil and protect the IJs from dehydration until the moisture conditions become conducive for IJ dispersal and host finding. Accordingly, the objectives of the present study were to determine the influence of soil moisture in different entomopathogenic nematode species on emergence from host cadavers and nematode survival within host cadavers under adverse desiccating conditions.

2. Material and methods

Experiments were conducted in incubators at 20 ± 1°C unless indicated otherwise. A loamy sand (87% sand, 7% silt, 6% clay; 0.3% organic matter; pH 6.9) that had been autoclaved and then stored at least 2 weeks was used. The moisture release curve of this soil was established using the filter paper method for determination of soil matric potential (Hamblin, 1981). *Steinernema carpocapsae* All strain, *Steinernema glaseri* NC strain, *Sternernema riobravis* Texas strain, and *Heterorhabditis bacteriophora* NC1 strain were cultured in last instar larvae of the greater wax moth, *Galleria mellonella*, and the IJs were harvested from White traps and stored in sterilized distilled water at 10°C for 5–21 days before use (Woodring and Kaya, 1988).

2.1. Experiment 1

The effect of soil moisture on IJ emergence was determined in this experiment. Soil was prepared at different moisture levels, thoroughly mixed, and was kept in double plastic bags to allow for moisture equilibration for 4 days before use. The soil water potentials (w/w soil moistures) used in this experiment were: −500 MPa (1%), −40 MPa (2%), −5 MPa (3%), −0.3 MPa (4%), −0.1 MPa (5%), −0.03 MPa (7%) and −0.006 MPa (13%).

To obtain nematode-infected cadavers, last instar wax moth larvae weighing between 190 and 210 mg were placed individually (on two layers of filter paper) in an inverted petri dish (60 × 15 mm). To each dish, 20 IJs of *S. carpocapsae*, *S. glaseri* or *S. riobravis*, or 40 IJs of *H. bacteriophora* were added. The larvae were incubated for 3 days to allow for the development of the typical signs of nematode infections (i.e., death and characteristic coloration of the cadaver). For each species, eight cadavers were dissected to determine the number of nematodes that had established per wax moth larva.

The experimental arena was a 220-ml plastic cup filled with 150 cm^3 of soil to a height of 3 cm.

Nematode-killed cadavers were placed individually in cups at 1 cm soil depth. There were 14 replicates per soil moisture level and nematode species. The cups were placed on trays which were sealed in two plastic bags containing moist paper tissue to reduce moisture loss. Two days before the earliest expected emergence of IJs (based on preliminary studies), daily observations of the cadavers started. Each cadaver was removed from the soil and placed under a dissecting microscope to examine its body surface for IJ emergence. To minimize moisture loss, the cups were recapped during the cadaver observation period. Before replacing the cadavers, the soil in the cup surrounding the site where the cadavers had been placed was also examined for IJs. When no more IJs were observed to emerge from a cadaver for three consecutive days, it was dissected to determine the condition of any enclosed nematodes. Cadavers from which no IJ emergence was observed were dissected 14 days after termination of emergence from all other cadavers within a treatment.

2.2. Experiment 2

In this experiment, IJ survival within cadavers in dry soil was determined by their ability to emerge and infect after rehydration of the cadavers. Based on Experiment 1, a soil moisture was selected at which no significant emergence was expected. Cadavers of nematode-infected wax moth larvae were obtained and soil was prepared as described in Experiment 1. The cadavers were placed in the dry soil (−40 MPa) for various periods of time (Table 1) depending on preliminary observations made for each nematode species. The experiment was conducted twice; the time intervals the cadavers were exposed to dry soil were changed in the second trial if the observation in the first trial suggested changes.

At each sampling time for each nematode species, eight cadavers were placed individually on White traps. Control cadavers were placed in moist soil (−0.006 MPa) and transferred to White traps one day before the earliest expected IJ emergence. The cadavers were checked daily for IJ emergence until emergence stopped. The emerging IJs were transferred every second day to tissue flasks and kept at 10°C. The total number of IJs that emerged per cadaver was determined by counting four subsamples from each cadaver. Cadavers without IJ emergence were dissected to confirm nematode infection and determine the condition of the enclosed nematodes. As a measure of infectivity, we determined the penetration efficiency of the emerged IJs. For steinernematids, we added 100 IJs to each of three petri dishes (60 × 15 mm) per replicate cadaver and added five wax moth larvae per dish. After 4 days, the dead insects were rinsed with tap water, dissected in a 0.5% Pepsin solution, and incubated at 37°C for 2 h to digest the insects' tissues (Mauleon et al., 1993). Then, the number of nematodes penetrated into each larva was counted. Because *H. bacteriophora* has a very low penetration efficiency on filter paper (Koppenhöfer, personal observation), it was evaluated by adding 200 IJs to each of three plastic cups filled with 100 ml of moist soil (−0.006 MPa) per replicate. After 6 days, the insects were processed as above.

2.3. Experiment 3

For the warm temperature adapted *S. riobravis* (Grewal et al., 1994), the same procedures as described for Experiment 2 were used at 30°C in two trials. To ensure a more uniform infection, wax moth larvae were exposed to 50 IJs. We also shortened the exposure time to 2 days because we expected the higher temperature to accelerate nematode development (Grewal et al., 1994).

2.4. Statistical analysis

In Experiment 1, the effect of soil moisture on the number of cadavers with IJ emergence was analyzed by nematode species using a *G*-test (Sokal and Rohlf, 1981). The date of first emergence in different soil moistures was analyzed by nematode species using analysis of variance (ANOVA) (PROC GLM) and post hoc means separation tests (Tukey's test) (SAS Institute, 1988). Replicates and treatments in which no nematodes emerged were not included in the analysis. Duration of emergence was analyzed similarly. In Experiments 2 and 3, replicates in which no IJs emerged were included in the means for number of emerged IJs but not in the means for percentage infectivity, date of first IJ emergence, or duration of emergence from cadavers. The relationship between number of IJs emerging or percentage infectivity of the emerged IJs and the time the cadavers were kept

Table 1
Experiment 2 (Trial 2). Days to first emergence and duration of emergence of IJs from cadavers of wax moth larvae infected with *S. glaseri*, *S. carpocapsae*, *H. bacteriophora*, or *S. riobravis*, kept in dry soil at −40 MPa water potential for various periods of time at 20°C before placement on White traps [a]

	Days in dry soil	Cadavers with emergence [b]	Days to 1st emergence	Duration of emergence [c]
S. glaseri	0 [d]	7	8.1 ± 0.8	–
	7	6	1.0 ± 0.0a [e]	–
	14	6	1.0 ± 0.0a	–
	20	3	2.0 ± 0.0a	–
	25	4	1.5 ± 0.2a	–
	29	6	1.7 ± 0.4a	–
S. carpocapsae	0	8	15.1 ± 0.4	38.1 ± 0.6
	14	8	3.5 ± 0.5a	29.9 ± 2.1a
	28	8	3.3 ± 0.9a	29.6 ± 1.7a
	42	7	3.4 ± 0.6a	17.6 ± 2.4b
	56	8	4.6 ± 1.2a	8.0 ± 0.7c
	72	3	1.0 ± 0.0b	4.7 ± 0.2c
H. bacteriophora	0	8	18.0 ± 0.2	6.5 ± 0.2
	21	8	1.0 ± 0.0b	3.0 ± 0.0b
	35	8	1.0 ± 0.0b	3.1 ± 0.1b
	49	8	1.5 ± 0.2b	4.5 ± 0.3a
	63	2	3.0 ± 0.3a	3.5 ± 0.3b
	70	0	– [f]	–
S. riobravis	0	8	13.4 ± 0.3	5.5 ± 0.3
	21	7	1.0 ± 0.0a	5.4 ± 0.2a
	42	6	1.3 ± 0.2a	4.7 ± 0.2a
	56	8	1.8 ± 0.4a	4.1 ± 0.6a
	70	6	1.8 ± 0.4a	4.3 ± 0.4a
	91	6	1.5 ± 0.3a	6.5 ± 0.4a

[a] Wax moth larvae were exposed to IJs in petri dishes for 3 days at 20°C before placement in soil.
[b] Each nematode species/exposure period combination had 8 replicates.
[c] Observations on duration of emergence were not taken for *S. glaseri*.
[d] Control cadavers (0) were transferred from infection dishes onto White traps. Controls were not included in analysis.
[e] Means ± SE of same species within columns followed by the same letter are not significantly different ($P < 0.5$).
[f] No IJs emerged from any cadaver in treatment.

in dry soil was determined with regression analysis (Sigmaplot, Jandel Scientific). The dates of first IJ emergence from cadavers after different times of exposure to dry soil and the duration of emergence from the cadavers were analyzed using ANOVA (PROC GLM) and Tukey's test. Data are presented as means ± SE. Differences between means are considered significant at $P < 0.05$.

3. Results

3.1. Experiment 1

IJ emergence from host cadavers was significantly affected by soil moisture ($G > 29$; df = 6; $P < 0.001$). For all four species, no IJs emerged from the cadavers in soil prepared at −500 MPa water potential, and only few IJs of *S. carpocapsae* and *S. glaseri* emerged from some cadavers at −40 MPa (Table 2). At −5 MPa, large numbers of *S. carpocapsae*, *S. glaseri*, and *H. bacteriophora* IJs emerged from all cadavers but no *S. riobravis* emerged. At ≥ −0.3 MPa, IJs of all nematode species emerged in large numbers from the cadavers. The first day of IJ emergence from cadavers was delayed by low soil moisture in *S. carpocapsae* ($F = 4.2$; df = 5,59; $P < 0.01$), *H. bacteriophora* ($F = 7.88$; df = 4,59; $P < 0.001$), and *S. riobravis* ($F = 4.3$; df = 3,40; $P < 0.05$) but not in *S. glaseri* ($F = 0.66$; df = 5,49; $P = 0.6$) (Table 2). Duration

Table 2
Experiment 1. Days to first emergence and duration of emergence of IJs from cadavers of wax moth larvae infected with *S. glaseri*, *S. carpocapsae*, *H. bacteriophora*, or *S. riobravis*, and kept at different soil water potential at 20°C [a]

Nematode species	Water potential (MPa)	Cadavers with emergence [b]	Days to 1st emergence	Duration of emergence
S. glaseri	−500	0	– [c]	–
	−40	3	10.3 ± 1.1a [d]	1.0 ± 0.3a
	−5	13	10.2 ± 0.9a	3.8 ± 0.5b
	−0.3	11	8.9 ± 0.5a	3.9 ± 0.3b
	−0.1	12	9.0 ± 0.4a	3.3 ± 0.4b
	−0.03	11	9.2 ± 0.5a	2.5 ± 0.2b
	−0.006	13	9.7 ± 0.7a	3.0 ± 0.3b
S. carpocapsae	−500	0	–	–
	−40	4	14.4 ± 0.5a	3.8 ± 0.5a
	−5	14	15.1 ± 0.6a	5.9 ± 0.5b
	−0.3	13	12.7 ± 0.5b	8.3 ± 0.6c
	−0.1	13	12.5 ± 0.5b	7.2 ± 0.4bc
	−0.03	13	12.7 ± 0.6b	6.9 ± 0.3bc
	−0.006	13	12.9 ± 0.6b	6.5 ± 0.4bc
H. bacteriophora	−500	0	–	–
	−40	0	–	–
	−5	14	18.0 ± 0.4ab	6.7 ± 0.6a
	−0.3	14	18.8 ± 0.4a	5.4 ± 0.6a
	−0.1	14	18.6 ± 0.2	7.1 ± 0.6a
	−0.03	14	16.9 ± 0.2b	6.4 ± 0.5a
	−0.006	13	16.9 ± 0.3b	5.3 ± 0.6a
S. riobravis	−500	0	–	–
	−40	0	–	–
	−5	0	–	–
	−0.3	13	17.6 ± 0.2a	14.6 ± 0.2a
	−0.1	13	17.8 ± 0.3a	13.7 ± 0.2a
	−0.03	12	17.0 ± 0.3ab	13.0 ± 0.2a
	−0.006	13	16.6 ± 0.3b	4.6 ± 0.3b

[a] Wax moth larvae were exposed to IJs in petri dishes for 3 days at 20°C before placement in soil.
[b] Each nematode species/soil moisture combination had 14 replicates.
[c] No emergence was observed in treatment.
[d] Means ± SE of same species within columns followed by the same letter are not significantly different ($P < 0.5$).

of IJ emergence was affected in different ways among species. *S. glaseri* ($F = 3.7$; df = 5,50; $P < 0.01$) and *S. carpocapsae* ($F = 6.41$; df = 5,59; $P < 0.001$) emerged for a significantly shorter period at the lowest soil moistures, whereas *H. bacteriophora* emergence was not affected and *S. riobravis* emergence was significantly shorter at the highest soil moisture ($F = 490.0$; df = 3,40; $P < 0.001$) (Table 2).

3.2. Experiment 2

The emergence and infectivity of IJs surviving within cadavers in dry soil (Table 3) as well as timing and duration of IJ emergence were affected in similar ways in both trials. In the first trial, however, *S. carpocapsae* and *S. riobravis* showed longer survival in the cadavers than we had predicted, and consequently, we did not have a treatment with no or minimal IJ emergence. Accordingly, we extended the observation periods in the second trial. Because of differences in sampling dates and IJ numbers and infectivity, the data from the two trials were not pooled for analysis. We will present the data of the second trial, and will indicate differences between the trials where necessary.

For *S. glaseri*, the mean number of nematodes that established in the wax moth larvae was 8.0 ± 0.7.

Table 3
Experiment 2. Linear regression analysis of number of IJs emerging from cadavers infected with *S. glaseri*, *S. carpocapsae*, *H. bacteriophora*, or *S. riobravis*, and percentage of emerged IJs that infected wax moth larvae vs. time in days (t) the cadavers were placed in soil at −40 MPa water potential

Nematode species	Temperature (°C)	Trial [a]	IJ emergence (×1000)		Infectivity (%)	
			Equation	r^2	Equation	r^2
S. glaseri	20	1	$19.1 - 0.7t$	0.93	$24.2 + 0.8t$	0.71
		2	$15.5 - 0.6t$	0.90	$19.4 - 0.6t$	0.52
S. carpocapsae	20	1	$237.9 - 4.9t$	0.81	$54.3 - 0.9t$	0.87
		2	$117.9 - 1.9t$	0.89	$25.6 - 0.3t$	0.85
H. bacteriophora	20	1	$172.1 - 2.2t$	0.96	$20.3 - 0.1t$	0.46
		2	$200.1 - 2.5t$	0.86	$35.2 - 0.1t$	0.10
S. riobravis	20	1	$55.4 - 0.5t$	0.73	$38.8 - 0.2t$	0.35
		2	$89.5 - 0.8t$	0.84	$46.0 - 0.1t$	0.06
S. riobravis	30	1	$145.9 - 3.6t$	0.60	$41.7 + 0.3t$	0.25
		2	$112.2 - 4.0t$	0.57	$34.6 + 0.7t$	0.48

[a] Sampling dates differed between Trials 1 and 2.

As had been observed in Experiment 1 at −40 MPa soil water potential, low numbers of IJs emerged from some cadavers while they were still in the soil. The number of emerging IJs declined rapidly to almost zero after 20 days exposure but did not completely stop even after 29 days (Fig. 1). Dissection of the cadavers from which IJ emergence had stopped or never occurred revealed exclusively dead IJs. Infectivity of emerged IJs increased with exposure time in trial 1 but decreased in trial 2 (Table 3). The number of days until the *S. glaseri* IJs started emerging from cadavers after transfer from dry soil to White traps was not affected by exposure time (Table 1). No observation on duration of emergence was taken in trial 2. In trial 1, exposure to −40 MPa for 5–20 days did not affect the duration of IJ emergence (3.0–4.4 days), but no IJs emerged after 25 days of exposure, and after 29 days emergence was only observed from two cadavers (duration 1.0 ± 0.0 days).

For *S. carpocapsae*, the mean number of nematodes that established in the wax moth larvae was 8.6 ± 1.1. Although the number of IJs emerging from cadavers and their infectivity were approximately twice as high in trial 1 than in trial 2, the trends were similar (Table 3). Both number of IJs emerging and IJ infectivity declined with longer exposure to dry soil (Fig. 1) and IJ emergence was close to zero after 72 days. The number of days until the first IJs emerged from cadavers was significantly shorter after 72 days of exposure than after 14–56 days of exposure (Table 1) ($F = 4.67$; df = 4,29; $P < 0.01$). Dissection of the cadavers from which IJ emergence had stopped or never occurred revealed exclusively dead IJs. The duration of emergence was considerably shorter in trial 1 (10 days for 0 day of exposure and 8–10 days for up to 30 days of exposure to dry soil) than in trial 2. In trial 2, duration of emergence significantly decreased at the longer exposure periods ($F = 35.56$; df = 4,29; $P < 0.001$) (Table 1).

For *H. bacteriophora*, the mean number of nematodes that established in the wax moth larvae was 3.8 ± 0.2. IJ numbers emerging from the cadavers declined gradually with exposure to −40 MPa (Fig. 1). No more emergence was observed on sampling day 70. Dissection of the cadavers from which IJ emergence had stopped or never occurred revealed exclusively dead IJs. Infectivity of the emerged IJs did not change (Table 3, Fig. 1). The number of days until the *H. bacteriophora* IJs started emerging was significantly longer on sampling day 63 than on the earlier sampling days (Table 1) ($F = 12.30$; df = 3,22; $P < 0.001$). The data for duration of emergence showed no trend.

For *S. riobravis*, the mean number of nematodes

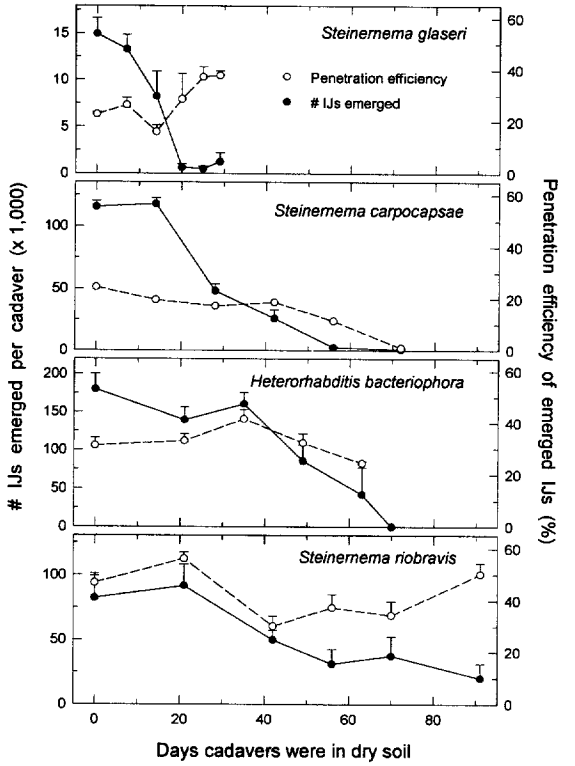

Fig. 1. Number of IJs of four species of entomopathogenic nematode emerging (●) and percentage infectivity of IJs emerged (○) from cadavers placed in soil at −40 MPa water potential for various periods of time at 20°C.

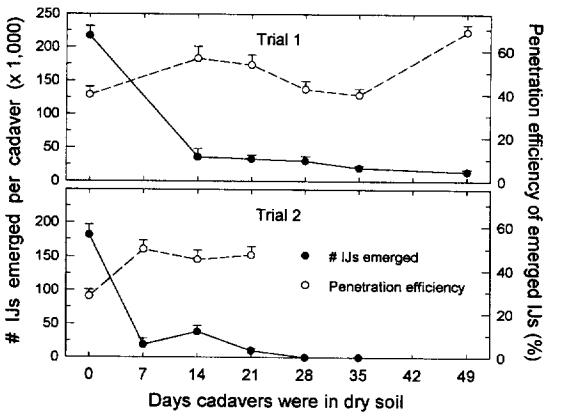

Fig. 2. Number of IJs of *S. riobravis* emerging (●) and percentage infectivity of IJs emerged (○) from cadavers placed in soil at −40 MPa water potential for various periods of time at 30°C in two trials.

that established in the wax moth larvae was 7.2 ± 1.5. IJ emergence declined gradually with exposure to −40 MPa (Fig. 1) but had not stopped on the last sampling day; regression analysis predicted that emergence would stop after 111 days. Infectivity of the emerged IJs did not change (Table 3, Fig. 1). The number of days until *S. riobravis* IJs started emerg-

Table 4
Experiment 2. Days to first emergence and duration of emergence of *S. riobravis* IJs from cadavers of wax moth larvae kept in soil at −40 MPa water potential for various periods of time at 30°C before placement on White traps [a]

Days in 2% soil moisture	Trial 1		Trial 2	
	Days to 1st emergence	Duration of emergence	Days to 1st emergence	Duration of emergence
0 [b]	6.5 ± 0.2	7.5 ± 0.2	5.8 ± 0.2	11.0 ± 0.7
7	– [c]	–	1.0 ± 0.0	5.4 ± 0.6a
14	1.0 ± 0.0	6.0 ± 0.2a [d]	1.0 ± 0.0	4.9 ± 0.7a
21	1.0 ± 0.0	3.6 ± 0.2c	1.0 ± 0.0	4.1 ± 0.4a
28	1.0 ± 0.0	3.6 ± 0.3c	– [e]	–
35	1.0 ± 0.0	3.0 ± 0.3c	–	–
49	1.0 ± 0.0	4.9 ± 0.1b	– [c]	–

[a] Wax moth larvae were exposed to IJs in petri dishes for 2 days at 30°C before placement in soil. Each treatment had 8 replicates and all cadavers showed IJ emergence.
[b] Control cadavers were immediately transferred from infection dishes onto White traps. Control was not included in analysis.
[c] No observations were taken on this day in the respective trial.
[d] Means ± SE within columns followed by the same letter are not significantly different ($P < 0.5$).
[e] No IJs emerged from any cadaver in treatment.

ing and duration of emergence were not significantly affected by exposure time (Table 1).

3.3. Experiment 3

At 30°C, decline in *S. riobravis* IJ emergence with exposure to −40 MPa was accelerated compared to the trials at 20°C. The mean number of nematodes that established in the wax moth larvae was 21.2 ± 1.2 and 22.4 ± 1.4 in trials 1 and 2, respectively. In both trials, the number of IJs emerging had already dropped dramatically compared to the control when the first sample was taken after 14 and 7 days of exposure to dry soil, respectively (Fig. 2). Thereafter, the IJ numbers declined gradually. In trial 1, IJ emergence had not stopped on the last sampling day; the regression analysis predicted that emergence would stop after 39 days (Table 3). In trial 2, emergence stopped after 28 days (Fig. 2). In both trials, the infectivity of the emerging IJs increased (Table 3, Fig. 2). With the exception of the controls, IJs started emerging 1.0 ± 0.0 days after placement on the White traps (Table 4). Differences between sampling days in duration of emergence were significant in trial 1 ($F = 32.41$; df = 4,35; $P < 0.001$) but not in trial 2; there was no trend in the data (Table 4).

4. Discussion

We showed that emergence of entomopathogenic nematode IJs from host cadavers is influenced by soil moisture, that IJs can survive for considerable lengths of time within desiccating host cadavers in dry soil, and that different nematode species are affected in different ways by the low soil moisture. We observed no or only very limited IJs emergence from cadavers in dry soil (−500– − 40 MPa), and IJ emergence at the lowest soil water potential at which emergence occurred (−5 MPa) was delayed by 1–2 days in all nematode species tested. *S. carpocapsae*, *S. glaseri*, and *H. bacteriophora*, however, emerged in high numbers at −5 MPa which is still unconducive for nematode infection (Koppenhöfer et al., 1995). Only *S. riobravis* completely evaded unfavorable moisture conditions by remaining within the cadavers below −0.3 MPa.

It is not clear whether IJ persistence within the cadaver is an adaptation to low soil moisture conditions or whether the nematodes are simply trapped in the cadaver. There appears to be some correlation between persistence within cadavers and the ecology of the different nematode species. The species that persisted the shortest time, *S. glaseri*, is an actively searching foraging strategist or cruiser (Campbell and Gaugler, 1993), and appears to have a close association with scarabaeid grubs as hosts (Wang et al., 1994, Wang et al., 1995). These hosts occur below the soil surface where soil moisture conditions are more stable than closer to the surface. In contrast, *S. carpocapsae* is a sit-and-wait strategist forager or ambusher (Campbell and Gaugler, 1993) and adapted to infect hosts on or close to the soil surface. Therefore, it is more likely to be exposed to dehydration within a host cadaver. In our study, *S. carpocapsae* persisted approximately twice as long as *S. glaseri*.

H. bacteriophora persisted for long periods within the cadavers despite it being an actively foraging species. *H. bacteriophora*-infected wax moth larvae have a gummous consistency and this may contribute to the better moisture retention of these cadavers compared to *Steinernema*-infected cadavers. For the most persistent nematode species in our study, *S. riobravis*, on the other hand, high desiccation tolerance may have a very high survival value. This species is endemic to the subtropical, semiarid lower Rio Grande Valley (Raulston et al., 1992) where it naturally parasitizes corn earworm and fall armyworm prepupae and pupae in corn fields. The distribution of *S. riobravis* in noncultivated areas, however, has not been studied.

S. riobravis survival in the cadavers was drastically reduced at 30°C compared to 20°C. At 30°C, the cadavers dried out much faster. After the initial rapid decline in IJ survival, the decline slowed down to a rate similar to that observed at the lower temperature. The shape of the IJ survival curves at 20°C of *S. glaseri* and *S. carpocapsae* in both trials and of *S. riobravis* in trial 2 was similar but stretched over a longer period. We hypothesize that the outer layers of the cadaver desiccate faster than its interior. The desiccated layers, starting with the cuticle and probably also including dead IJs, may function as a buffer reducing further desiccation of the cadaver. This

mechanism is also supported by the observation that in most cases the infectivity of the surviving IJs remained high despite the decline in IJ emergence.

IJs can also persist for long periods as individuals in the soil (Kung et al., 1991) and IJs of the ambusher *S. carpocapsae* are better adapted to the more variable soil moisture conditions close to the soil surface than the cruiser *S. glaseri*. *S. carpocapsae* infects (Koppenhöfer et al., 1995) and persists (Kung et al., 1991) better in drier soil than *S. glaseri*. However, if the IJs complete their development inside a host cadaver located in unfavorably dry soil, they probably have no choice but to endure within the cadaver for two reasons. First, if they manage to exit the cadaver, they are instantly exposed to the low moisture without time to adapt physiologically into a quiescence state. Under such rapid desiccation regimes, IJs survive only for a few days or even hours (Schmiege, 1963; Kamionek et al., 1974; Kung et al., 1991). Second, the cuticle of the host dries out and hardens at a rate and to a degree that probably restricts the escape of the IJs from the cadaver until it is rehydrated.

By retaining moisture and functioning as a buffer, the host cadaver may serve as a means for nematode populations to persist through dehydration conditions. We do not know how commonly this occurs under field conditions where moisture and temperature conditions are more variable and different insect species serve as hosts. For short periods with insufficient moisture, remaining inside the host cadaver could be an efficient mechanism for entomopathogenic nematode survival.

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