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Interactions of two idiobiont parasitoids (Hymenoptera: Ichneumonidae) of codling moth (Lepidoptera: Tortricidae) with the entomopathogenic nematode *Steinernema carpocapsae* (Rhabditida: Steinernematidae)

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Abstract

Simultaneous use of parasitoids and entomopathogenic nematodes for codling moth (CM) control could produce an antagonistic interaction between the two groups resulting in death of the parasitoid larvae. Two ectoparasitic ichneumonid species, *Mastrus ridibundus* and *Liotryphon caudatus*, imported for classical biological control of cocooned CM larvae were studied regarding their interactions with *Steinernema carpocapsae*. Exposure of *M. ridibundus* and *L. caudatus* developing larvae to infective juveniles (IJs) of *S. carpocapsae* (10 IJs/cm^2; $\approx LC_{80–90}$ for CM larvae) within CM cocoons resulted in 70.7 and 85.2% mortality, respectively. However, diapausing full grown parasitoid larvae were almost completely protected from nematode penetration within their own tightly woven cocoons. *M. ridibundus* and *L. caudatus* females were able to detect and avoid ovipositing on nematode-infected cocooned CM moth larvae as early as 12 h after treatment of the host with IJs. When given the choice between cardboard substrates containing untreated cocooned CM larvae and those treated with an approximate LC_{95} of *S. carpocapsae* IJs (25 IJs/cm^2) 12, 24, or 48 h earlier, ovipositing parasitoids demonstrated a significant preference for untreated larvae. The ability of these parasitoids to avoid nematode-treated larvae and to seek out and kill cocooned CM larvae that survive nematode treatments enhances the complementarity of entomopathogenic nematodes and *M. ridibundus* and *L. caudatus*.

Keywords: *Steinernema carpocapsae*; Biological control; Ichneumonidae; *Cydia pomonella*; Codling moth; Interaction; *Mastrus ridibundus*; *Liotryphon caudatus*; Intraguild predation

1. Introduction

Codling moth (CM), *Cydia pomonella* is the principal insect pest of apple and a significant pest of other fruit in several countries worldwide (Barnes, 1991). Most conventional orchardists tolerate less than 1% damage due to this pest. To achieve this goal, broad spectrum insecticides are regularly used to eliminate or drastically reduce CM populations. One of the several negative environmental impacts of broad spectrum insecticides is the severe reduction of nontarget organisms including natural enemies of tree fruit pest insects. Alternative interventions that minimize the use of pesticides are encouraged by legislative action, and environmental and food safety concerns. A truly integrated approach for using the various components of IPM, including biological control agents, will be required to obtain the maximum effect from a given intervention or practice without interfering with the effectiveness of others. A wide variety of natural enemies including predators, parasites and pathogens have been reported from CM (Cross et al., 1999a,b; Falcon and Huber, 1991; Lacey et al., 2000; Zimmermann and Weiser, 1991). Hymenopterous parasitoids imported from the putative center of origin of CM show potential as classical biological control agents of cocooned CM larvae in the Western United States (Bezemer and Mills, 2001; Unruh, 1997). The efficacy of entomopathogenic nematodes

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as inundatively applied biological control agents has also been demonstrated against cocooned CM larvae in orchards (Kaya et al., 1984; Nachtigall and Dickler, 1992; Unruh and Lacey, 2001) and in infested fruit bins (Cossentine et al., 2002; Lacey and Chauvin, 1999).

Entomopathogenic nematodes (Steinernematidae and Heterorhabditidae) have a good safety record especially regarding their effects on predators and pollinators (Akhurst, 1990; Akhurst and Smith, 2002; Bathon, 1996). However, the host range of certain species is sufficiently broad to warrant some concern for nontarget insects in cryptic habitats under conditions that favor good survival of the infective stage of the nematodes (Barbercheck and Millar, 1999). Upon exiting fruit, full grown CM larvae seek out cryptic habitats in which to spin their cocoons. *Mastrus ridibundus* and *Liotryphon caudatus* are external idiobiont parasitoids of cocooned CM larvae that actively search for CM hosts in cryptic habitats. These adult parasitoids paralyze their hosts upon which the larval parasitoids feed. After feeding externally on the CM host larva and killing it, the fully developed parasite larvae spin cocoons inside the host cocoon and overwinter as diapausing larvae. Concomitant use of parasitoids and nematodes in cryptic habitats could produce antagonistic interaction between the two groups resulting in intraguild predation and death of parasitoid larvae. The ability of parasitoids to detect and avoid nematode-infected hosts would help to reduce a negative interaction. The objective of our study was to determine: (a) the direct effects of exposure of developing and diapausing parasitoid larvae to infective juveniles (IJs) of *Steinernema carpocapsae* and (b) if prior infection of CM larvae by IJs affected host selection by *M. ridibundus* and *L. caudatus*.

2. Materials and methods

2.1. Source and production of organisms

2.1.1. Production of cocooned codling moth larvae

The CM colony at the Yakima Agricultural Research Laboratory was started from locally collected CM and has been maintained over the past three decades. The CM larvae used in our experiments were produced on artificial diet (Toba and Howell, 1991) under diapausing conditions (20 °C; 8L:16D) and provided perforated 15.2 cm^2 cardboard substrates (8 cm × 1.9 cm; double faced, B flute, Weyerhauser, Tacoma, WA) in which to spin their cocoons as described by Lacey and Unruh (1998). Each strip contained ≈20 cocooned larvae, and were stored at 2 °C until used.

2.1.2. Production of nematodes

The Sal strain of *S. carpocapsae* that was used in all of our experiments was originally obtained from James Cate (Integrated BioControl Systems, Aurora, IN, USA) from a culture originally isolated in an apple orchard in Indiana, USA. IJs of the nematode were produced in full grown *Galleria mellonella* larvae and harvested on White traps using procedures described by Kaya and Stock (1997). The IJs were stored at 10 °C until used. IJs less than 2 weeks old were used for all experiments.

2.1.3. Production of parasitoids

Liotryphon caudatus was collected in Southern Russia in the fall of 2000 and passed through quarantine to release in the spring of 2001. *M. ridibundus* was collected in Kazakstan, near Almaty in the fall of 1994 and passed through quarantine to release in the spring of 1995 (Unruh, 1997). Both species were reared at 22 °C and 16L:8D on cocooned diapausing CM larvae in cardboard rolls. Exposure of the CM to the wasp parasitoids occurred in screen cages and the wasps were provided with water, wicked from a vial, and honey, streaked on the side of the screen cage. Both species diapause as mature larvae inside a paper-like cocoon. Developing larvae used for direct exposure experiments were produced by exposing groups of 10 or more perforated 15.2 cm^2 cardboard substrates containing cocooned CM larvae to ovipositing parasitoids for 24 h in colony cages of either *M. ridibundus* or *L. caudatus*. The exposures resulted in 48.8 and 27.8% parasitism of CM by *M. ridibundus* and *L. caudatus*, respectively. The number of *M. ridibundus* larvae per parasitized CM larva ranged from 1 to 8 (mean 3.0). The number of *L. caudatus* larvae per parasitized CM larva was always one, due to cannibalism when more than one egg was deposited per host larva. The cardboard substrates were then incubated at 25 °C for 3 days, after which time the mid-sized larvae were used in experiments. Diapausing larvae were obtained after up to 3 weeks of exposure of the substrates in colony cages under cooler (20 °C) and shorter light (8L:16D) conditions. The larvae were subsequently incubated at 10 °C for 1–2 weeks until they had spun cocoons. The cardboard substrates containing cocooned diapausing parasitoid larvae were stored at 10 °C until they were used in experiments.

Mated females, 3–5 days old, of both parasitoid species were used for all choice experiments. After emerging, the parasitoids were allowed to mate and have access to honey and cocooned CM larvae in cardboard substrates. Twenty-four hours prior to each choice experiment, 2, 4, or 8 *M. ridibundus* females, depending on the type of experiment, were placed in large petri dishes (15 cm diameter) with a source of honey, but without host larvae. *L. caudatus* females were individually placed in 0.5 L plastic containers 24 h before experiments due to their size and the aggressive nature of the females toward one another in a more confined space.

2.2. Direct effect of exposing developing and diapausing, cocooned parasitoid larvae to infective juveniles of S. carpocapsae

For each replicated experiment for each parasitoid species, five substrates containing developing parasitoids were each treated with 152 IJs in 1 mL water (10 IJs/cm^2; $\approx LC_{80-90}$ for cocooned CM larvae) spread evenly over the surface of the strip. One milliliter of water was applied to each of five control substrates. All substrates were then individually sealed in 9 cm petri dishes with Parafilm and incubated for 6 days at 25 °C. The Parafilm was removed after the first 24 h. Mortality of parasitoid larvae was assessed after 5 days by moistening and gently separating the layers of the cardboard substrates and removing CM and parasitoid larvae from the CM cocoons. The experiment was repeated on two separate dates for each parasitoid species. The effect of *S. carpocapsae* IJs on diapausing, cocooned parasitoid larvae of each species was tested in an identical manner to that described above and the experiments were repeated twice.

2.3. Effect of nematode infection on host selection

Diapausing cocooned CM larvae in 25 cardboard substrates were treated with an approximate LC_{95} of *S. carpocapsae* IJs (380 IJs/strip = 25 IJs/cm^2), and incubated in sealed petri dishes (9 cm diameter) for 12, 24, or 48 h. Four mated female *M. ridibundus* were added to each of five large petri dishes (15 cm diameter) containing one treated and one untreated cardboard strip for each nematode incubation period. Four mated female *M. ridibundus* were also added to each of five petri dishes each containing two untreated substrates of cocooned CM larvae. Parasitoids were left in contact with treated and untreated cardboard substrates for 24 h at 25 °C (16L:8D) after which time they were removed and the substrates incubated for an additional 5 days. Five treated and five untreated substrates were incubated for 5 days at 25 °C as controls to monitor CM mortality in the absence of parasitoids. After incubation, the cardboard substrates were opened and the number of moribund, dead, and wasp-parasitized CM larvae were separately recorded for nematode-treated and untreated substrates. CM larvae were counted as dead if there was no response to probing with a dissecting needle. Moribund larvae were barely responsive to probing and were included with the dead for statistical analysis. Tests with *L. caudatus* females were conducted in a similar manner except that the substrates were exposed to the parasitoids in 0.5 L plastic containers. The experiments were repeated three times for each parasitoid species.

Another test was conducted with *M. ridibundus* to determine the effect of the number of parasitoid females per large petri dish on oviposition pressure on nematode-treated (25 IJs/cm^2) and untreated cardboard substrates containing cocooned CM larvae. The substrates were incubated for 24 h prior to introduction of parasitoids. The number of female *M. ridibundus* was 2, 4, or 8 females per each of five large petri dishes. Controls consisted of five nematode-treated and five untreated cardboard substrates containing cocooned CM larvae, as described above, and groups of five large petri dishes for each parasitoid density with each dish containing two untreated substrates. The experiments were repeated on four separate dates.

The progression of mortality in cocooned CM larvae treated with 25 IJs/cm^2 after 12, 24, and 48 incubation at 25 °C was determined using the bioassay procedure of Lacey and Unruh (1998). Fifteen cardboard substrates containing $\approx$20 larvae each were each treated with 380 *S. carpocapsae* IJs in 1 mL of water, sealed in 9 cm petri dishes and incubated at 25 °C. Five substrates were removed from incubation after 12, 24, and 48 h and mortality was assessed. Control substrates were treated with 1 mL of water and incubated in the same manner.

2.4. Statistical analysis

The mean mortality of developing and diapausing parasitoid larvae treated with nematodes was compared between species using a two-way factorial ANOVA. Mortality and parasitism (%) comparisons in choice tests (nematode-treated and untreated hosts) at 12, 24, and 48 h were based on one way ANOVA of means of the replicate petri dishes from each date of the bioassay for treated, untreated, and in the case of 12 h, control treatments or three to four replicates per condition (PROC GLM; SAS Institute, 1998). Each time interval after nematode treatment was analyzed separately because of significant time by treatment interactions ($p < 0.05$). Tukey's honestly significant difference test was used for mean comparisons. For studies of the effect of crowding on host utilization and acceptance by *M. ridibundus* (2, 4, or 8 wasps per dish), a factorial ANOVA was employed with nematode treatment and wasp density representing crossed fixed effects; again we used means from each test date, pooling the petri dishes on a given date. Tukey's HSD was used for mean comparisons. The angular transformation (arc-sine of the square root) was employed in all analyses, although the qualitative pattern of significance was identical when raw data were analyzed (not shown). Raw, not back-transformed, means are presented in the results but significance levels and mean comparisons are based on transformed data.

3. Results

3.1. Effect of direct exposure of parasitoid larvae to infective juveniles of S. carpocapsae

Developing larvae of *M. ridibundus* and *L. caudatus* were highly susceptible to direct infection by *S. carpocapsae* when exposed to 10 IJs/cm^2, a dosage that has been reported to kill 80–90% of cocooned codling moth larvae (Lacey and Unruh, 1998). The susceptibility of *M. ridibundus* larvae was not significantly different from that of *L. caudatus* (Table 1) ($F_{1,4} = 2.47,\ P = 0.19$). Diapausing larvae of both species were protected from infection within their cocoons ($F_{1,4} = 37.04,\ P = 0.004$; Table 1). The interaction between species and developmental status for susceptibility was not significant ($F_{1,4} = 0.7,\ P = 0.80$). Mortality of treated, diapausing larvae of *M. ridibundus* was not significantly different from that of controls (t_{18} stat -0.719, $P = 0.239$). However, slight but significant mortality was observed in treated diapausing *L. caudatus* larvae (t_{18} stat -2.496, $P = 0.009$).

3.2. Effect of nematode infection on host selection

When given the choice between cardboard substrates containing cocooned CM larvae that had been left untreated and those treated with an LC_{95} of *S. carpocapsae* IJs 12, 24, or 48 h earlier, ovipositing *M. ridibundus* demonstrated a significant preference for untreated larvae. Parasitism at 12 h was similar in the untreated test substrates (from the choice test) and the untreated control substrates (no choice) and these were greater than that in nematode-treated substrates (12 h: $F_{2,6} = 9.19,\ P = 0.015$) (Fig. 1a). Differences in parasitism between treated and untreated test substrates at 24 and 48 h were also significant (24 h: $F_{1,4} = 27.36,\ P = 0.006$; 48 h: $F_{1,4} = 85.09,\ P = 0.0008$). The degree of parasitism in the treated substrates markedly declined with increasing time after nematode treatment. By 48 h after application of nematodes, *M. ridibundus* females demonstrated a 21-fold preference for untreated larvae over nematode-treated larvae. A small sample ($\approx$10 larvae) of individuals that were parasitized in the nematode-treated substrates at 48 h were dissected and found not to be infected with nematodes, indicating selection of healthy CM larvae within substrates where 92–96% of the larvae were infected by nematodes. Mortality associated with *M. ridibundus* was between 8 and 18% greater than that due to parasitism alone in the untreated or control substrates (Figs. 1a and b). Mortality in treatment effects mirrored that for parasitism, but explained much more of the variation in the data and were significant (12 h: $F_{2,6} = 90.8,\ P = 0.0001$; 24 h: $F_{1,4} = 172.7,\ P = 0.0002$; and 48 h: $F_{1,4} = 148.8,\ P = 0.0003$). Death in unparasitized CM larvae that were not treated with nematodes was ostensibly due to both predation and probing of the host without subsequent oviposition. Most of the dead CM larvae in the untreated substrates that were not parasitized by *M. ridibundus* showed signs of being stung by the parasite; melanized black spots were very abundant on some.

Both mortality and parasitism were significantly influenced by both wasp density (mortality: $F_{2,27} = 4.38,\ P = 0.023$; parasitism: $F_{2,27} = 7.44,\ P = 0.003$) and nematode-treatment (mortality: $F_{2,27} = 147.9,\ P < 0.0001$; parasitism: $F_{2,27} = 7.20,\ P = 0.003$), (Figs. 2a and b) and interactions between these effects were not significant (mortality: $F_{4,27} = 1.94,\ P = 0.13$; parasitism: $F_{4,27} = 0.82,\ P = 0.53$). Parasitism increased with wasp density in control, treated, and untreated substrates (Fig. 2a). Mortality increased with wasp density in untreated and control substrates but not in treated substrates (Fig. 2b). When parasitism was subtracted from mortality and the remainder analyzed, wasp density became insignificant in the model ($F_{2,27} = 2.49,\ P = 0.102$) while nematode-treatment remained highly significant.

Parasitism by *L. caudatus* in untreated CM larvae versus those that had not been treated with nematodes was generally similar to that observed with *M. ridibundus*. Unlike that seen with *M. ridibundus*, parasitism by *L. caudatus* at 12 h was statistically similar in all three treatments although the general pattern followed *M. ridibundus* in that parasitism was numerically higher in

Table 1
Effect of direct exposure of developing larvae and cocooned diapausing larvae of *M. ridibundus* and *L. caudatus* to infective juveniles of *S. carpocapsae*[a]

Insect species[c]	% Mortality ± SE[b]			
	Developing larvae		Cocooned diapausing larvae[d]	
	Nematode-treated	(Controls)	Nematode-treated	(Controls)
Mastrus ridibundus	70.7 ± 7.0	(0)	1.8 ± 0.1*	(0 a)
Liotryphon caudatus	85.2 ± 6.4	(1.7 ± 0.1)	8.8 ± 0.3	(0 a)

[a] Ten infective juveniles/cm^2, 25 °C.
[b] Larval development states were significantly different (Tukey's HSD, $\alpha = 0.05$).
[c] Insect species were not significantly different in overall response to nematode treatments (Tukey's HSD, $\alpha = 0.05$).
[d] Means followed by an asterisk are not significantly different from their respective control (Student's t test, $P > 0.05$).

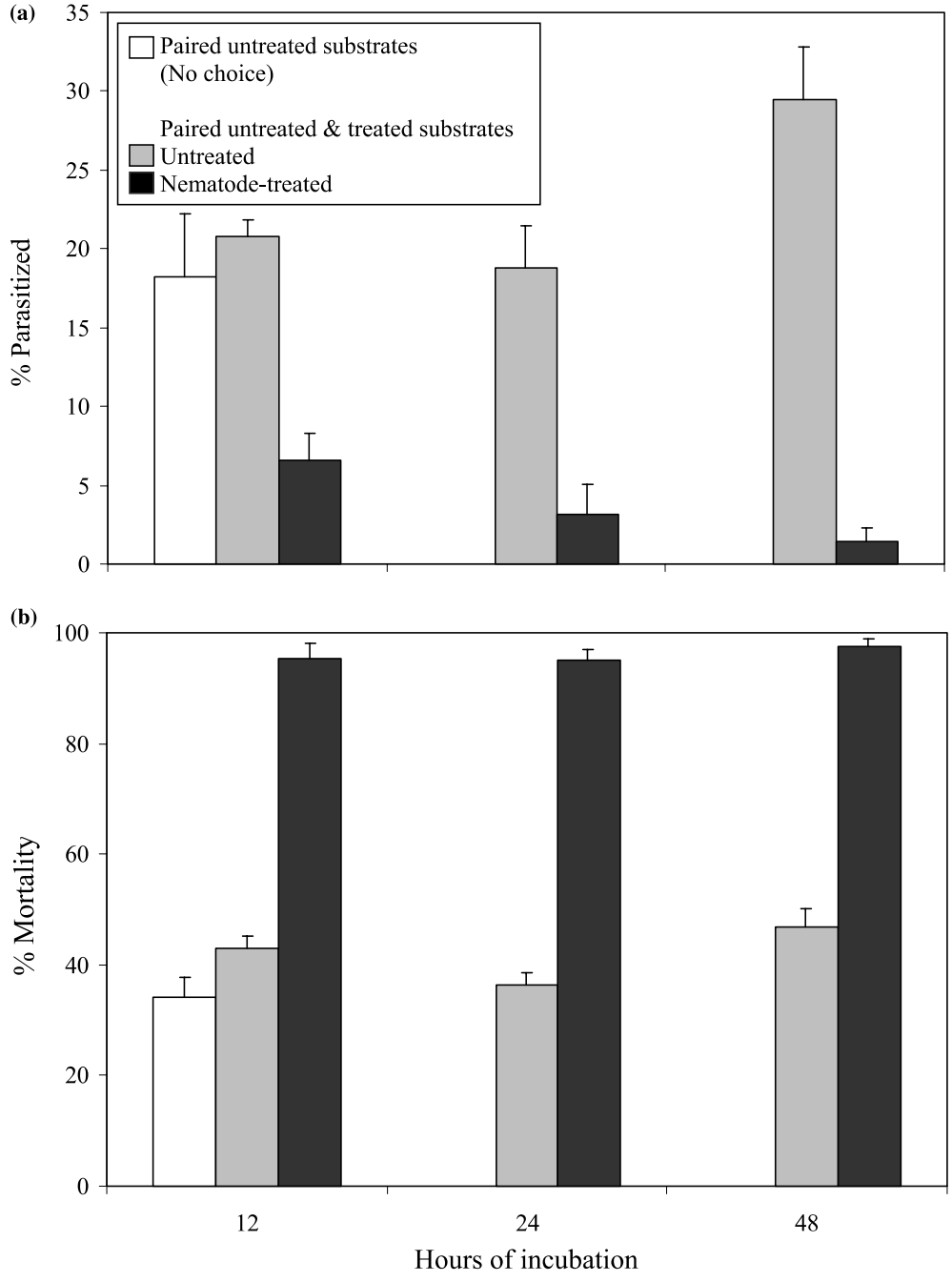

Fig. 1. (a) Percentage of codling moth larvae in untreated and nematode-treated cardboard substrates parasitized by four *Mastrus ridibundus* females 12, 24, or 48 h following application of infective juveniles of *Steinernema carpocapsae*. (b) Mortality of codling moth larvae due to *S. carpocapsae* and *M. ridibundus*. Mortality in substrates that were not treated with nematodes was due to parasitism and predation by *M. ridibundus*. Mortality in nematode-treated substrates was due primarily to infection by *S. carpocapsae* (≈95%) and wasp parasitism shown in (a).

the untreated test substrates (from the choice test) and the untreated control substrates (no choice) than in the nematode-treated substrates (12 h: $F_{2,6} = 1.94$, $P = 0.224$) (Fig. 3a). Comparisons of parasitism between treated and untreated test substrates at 24 and 48 h were highly significant (24 h: $F_{1,4} = 38.72$, $P = 0.0034$; 48 h: $F_{1,4} = 24.96$, $P = 0.008$). As in the studies with *M. ridibundus*, there was a clear decline in parasitism with increasing time after nematode-treatment (Fig. 3a). *L. caudatus* females demonstrated a 76-fold preference for untreated larvae over nematode-treated larvae by 48 h after application of nematodes. Mortality associated with *L. caudatus* was between 10 and 24% greater than parasitism alone in the untreated or control substrates (Fig. 3a vs b). Mortality in treatment effects mirrored that for parasitism but was consistently significant over time (12 h: $F_{2,6} = 79.15$, $P = 0.0001$; 24 h: $F_{1,4} = 26.25, P = 0.007$; and 48 h: $F_{1,4} = 18.08$, $P = 0.013$), (Fig. 3b).

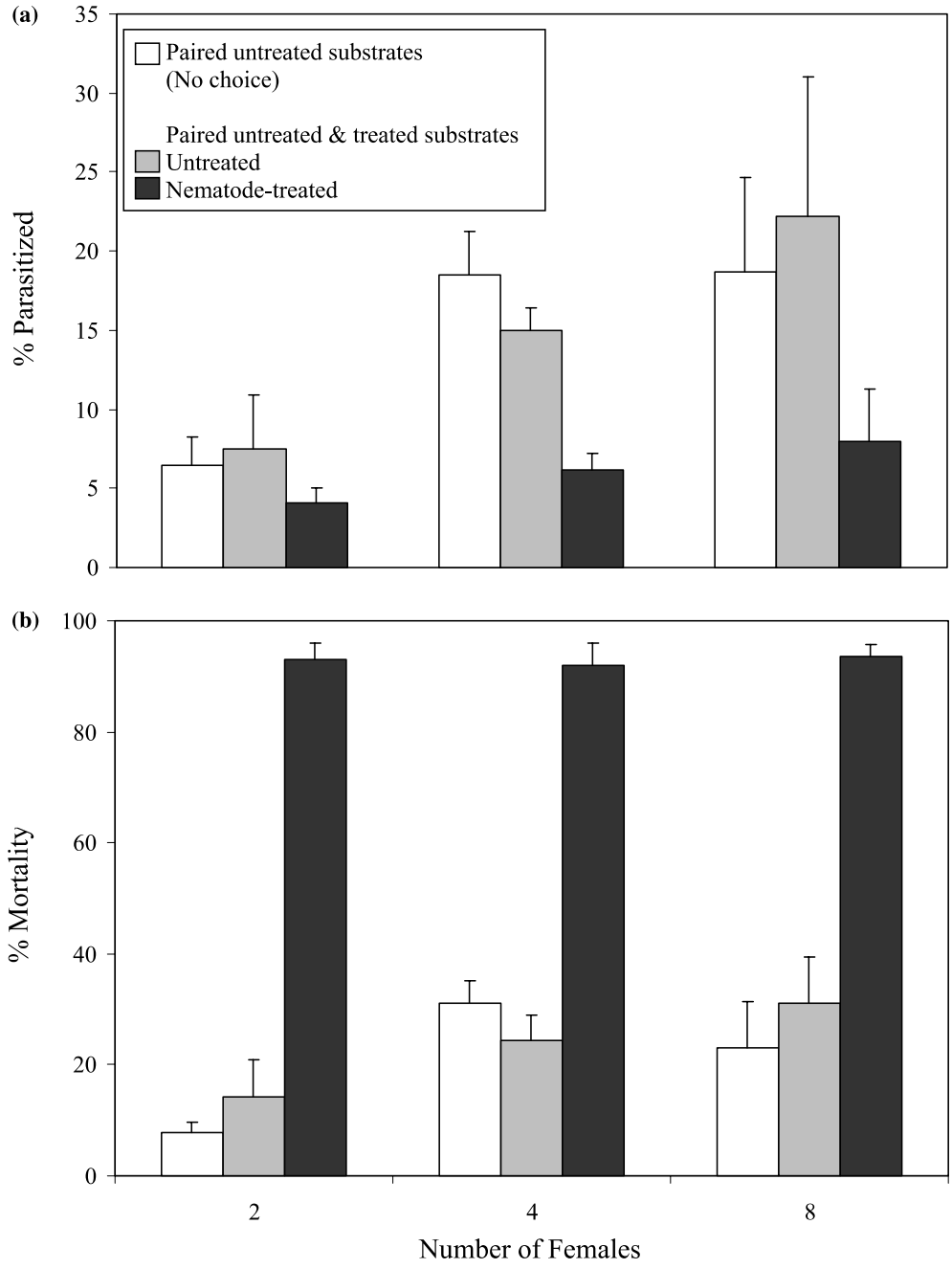

Fig. 2. (a) Percentage of codling moth larvae in untreated and nematode-treated cardboard substrates parasitized by 2, 4, or 8 *Mastrus ridibundus* females 24 h following application of infective juveniles of *Steinernema carpocapsae*. (b) Mortality of codling moth larvae due to *S. carpocapsae* and *M. ridibundus*. Mortality in substrates that were not treated with nematodes was due to parasitism and predation by *M. ridibundus*. Mortality in nematode-treated substrates was due primarily to infection by *S. carpocapsae* (≈95%) and wasp parasitism shown in (a).

Although, the extent of discrimination by wasps cannot be fully teased from these data, one comparison is illuminating. The no choice control substrates (two substrates per plate) showed similar levels of parasitism to that seen in the single untreated strip in the choice test at 12 h for both wasp species and at 24 h for *M. ridibundus*. By 48 h the parasitism rate per host was almost 50% higher in untreated test substrates for *M. ridibundus*, and just over 100% higher for *L. caudatus* compared to control (no choice substrates).

4. Discussion

The results of our studies clearly demonstrate the vulnerability of developing larvae of both *M. ridibundus* and *L. caudatus* to infection by *S. carpocapsae.* The

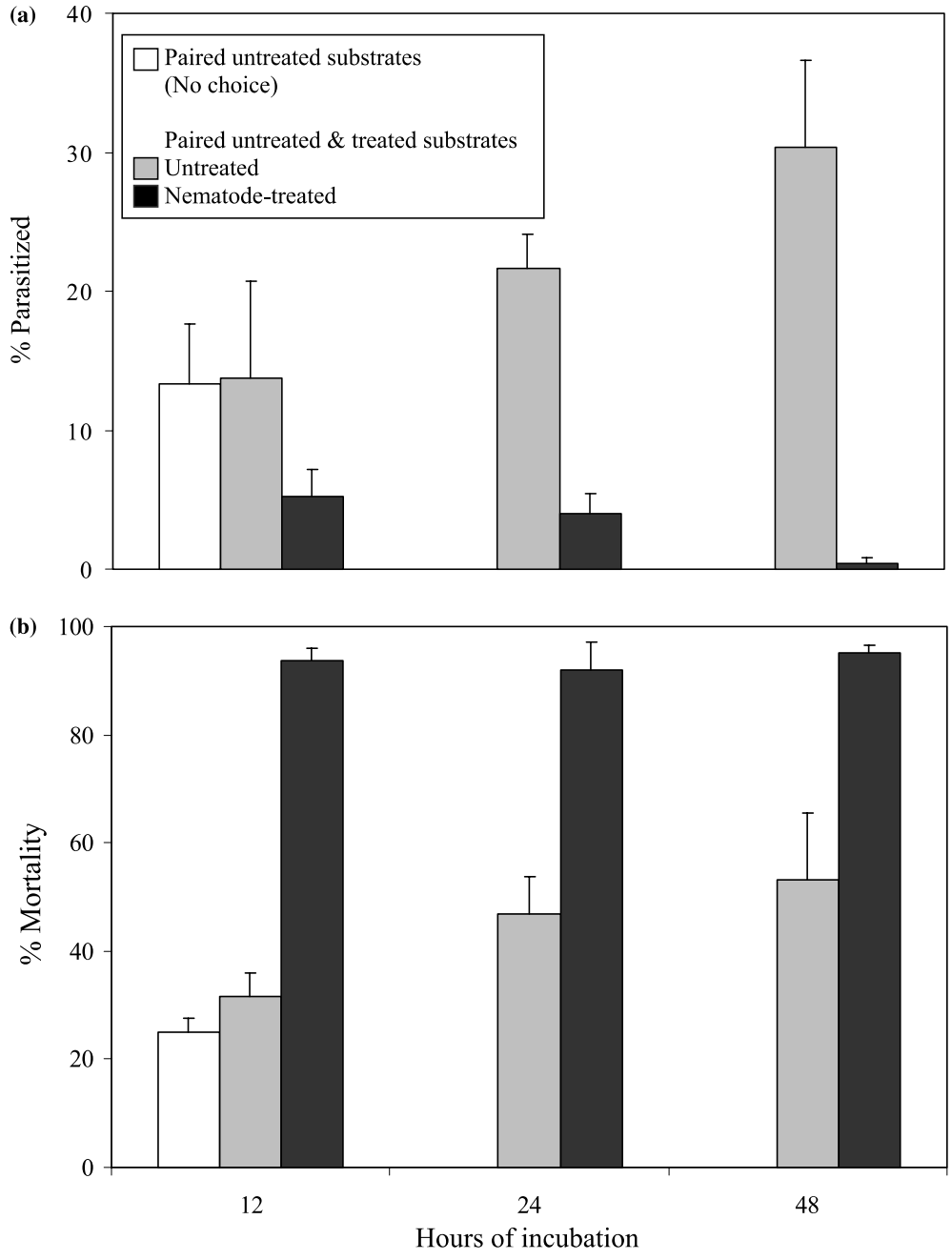

Fig. 3. (a) Percentage of codling moth larvae in untreated and nematode-treated cardboard substrates parasitized by four *Liotryphon caudatus* females 12, 24, or 48 h following application of infective juveniles of *Steinernema carpocapsae*. (b) Mortality of codling moth larvae due to *S. carpocapsae* and *L. caudatus*. Mortality in substrates that were not treated with nematodes was due to parasitism and predation by *L. caudatus*. Mortality in nematode-treated substrates was due to primarily to infection by *S. carpocapsae* (≈95%) and wasp parasitism shown in (a).

susceptibility of parasitic Hymenoptera to entomopathogenic nematodes has been reported by other researchers for both internal and external hymenopterous parasitoids (Battisti, 1994; Kaya, 1978a,b; Kaya and Hotchkin, 1981; Shannag and Capinera, 2000; Sher et al., 2000; Triggiani, 1985; Zaki et al., 1997). Premature death of the host with subsequent death of internal parasitoids is the most frequently reported consequence of host-parasitoid-pathogen interaction rather than direct infection of the parasitoid (Begon et al., 1997; Brooks, 1993). The outcome of competition between pathogens and parasitoids is often determined by the timing of infection or oviposition (Hochberg and Lawton, 1990). Kaya (1978b) observed death in the endoparasitic braconid, *Apanteles militaris* when the host armyworm was killed by *S. carpocapsae* before the parasitoid could complete development. However, when the host larvae were treated with the IJs late in the

development of the braconid (11 days old), 94% of the larvae emerged from the host and formed normal cocoons. Kaya (1978b) and Shannag and Capinera (2000) report the direct infection of internal hymenopterous parasitoids by *S. carpocapsae* after they have emerged from the host, but before completion of the pupal cocoon. In the case of the two ichneumonid external parasitoids in our study, they are directly invaded by the nematode and are as susceptible to infection as CM larvae. To minimize antagonistic interaction with the combined use entomopathogenic nematodes and *M. ridibundus* and *L. caudatus* careful timing of nematode applications will be required so as to avoid exposure of developing parasitoid larvae to IJs. The risk for nematode infection of nontarget organisms is a function of permanence of IJs in the environment (Barbercheck and Millar, 1999). CM chooses cryptic habitats in which to cocoon and unless these habitats are kept moist, IJs rapidly desiccate, thereby reducing their persistence and the potential risk for parasitoid species.

The reduction or lack of penetration of cocoons of diapausing *M. ridibundus* and *L. caudatus* by *S. carpocapsae* IJs provides an opportunity for application of entomopathogenic nematodes with minimal negative impact on the parasitoids. Fall application of nematodes when all or most of the CM larvae have left the fruit and when parasitoids have entered diapause should markedly reduce the risk of nematode infection to parasitoid larvae. Similar observations on the inability of *S. carpocapsae* to penetrate the cocoons of *Apanteles* spp. and other parasitoids were made by Kaya (1978b), Kaya and Hotchkin (1981), and Shannag and Capinera (2000).

Mastrus ridibundus and *L. caudatus* laid fewer eggs, as evidenced by parasitism, in arenas where one of the two host cardboard substrates were treated with nematodes than they laid when presented with two untreated substrates. However, as the degree of degradation of the host increased from nematode infection, parasitism in treated substrates (or time spent searching infected hosts) was replaced by more effective parasitism of the untreated strip. Because mortality also follows this pattern, our data suggest that the wasps also discriminate in favor of untreated hosts for probing/predatory behaviors. However, because nematode infection may partially mask wasp-induced mortality we cannot rule out that wasps are even more discriminating than shown here.

Soon after penetrating the insect host through a natural opening, the IJs of steinernematid nematodes release their symbiotic bacteria (*Xenorhabdus* spp.) which replicate in the insect hemocoel (Poinar, 1990) and kill most insect species within 48 h (Kaya and Stock, 1997). Twelve h after treatment of cocooned CM larvae with *S. carpocapsae* and its mutualistic bacterium, *Xenorhabdus nematophila*, over 98% of the CM larvae were still alive. Survival of the larvae was still over 80% 24 h following application of IJs, but nearly all of them had died within 48 h of treatment. Although the greatest avoidance of nematode-treated CM by both parasitoids was observed after 48 h incubation, significant host avoidance was evident as early as 12 h after treatment with *M. ridibundus* and 24 h with *L. caudatus.* Parasitic Hymenoptera use a complex set of semiochemical and physical cues to determine the suitability of hosts (Godfray, 1994; Vinson and Iwantsch, 1980; Vet and Dicke, 1992). These include both external and internal host cues. The change in host suitability shortly after penetration of IJs and before the CM larvae were killed, was ostensibly a function of bacterial activity within the host detected when the larvae were probed by the parasitoids. Similarly, the bacterial activity in infected hosts has been implicated in altering the infection behavior of IJs (Shapiro et al., 2000), and repelling other organisms such as ants (potential scavengers) (Zhou et al., 2002). The avoidance of nematode-infected CM larvae by ovipositing *M. ridibundus* and *L. caudatus* females enhances the complementarity of these parasitoids and entomopathogenic nematodes. Similar observations on the avoidance of oviposition by the eulophid *Digphus begini* in the leafminer *Liriomyza trifolii* infected with *S. carpocapsae* were made by Sher et al. (2000).

Environmental conditions that favor hymenopteran parasitoids or entomopathogenic nematodes will ultimately influence the type of interaction and compatibility or antagonism of these two groups of biological control agents. Parasitoids are better at exploiting uninfected hosts because of their abilities of search, whereas nematodes have limited search capacity and require proper environmental conditions, most notably, moisture. Based on the models of Begon et al. (1999) and Hochberg et al. (1990), coexistence and enhanced biological control are favored by complementarity between parasitoid and pathogens in terms of their extrinsic and intrinsic qualities. The compatibility of the two groups for CM control could be easily managed if mass releases of parasitoids are made 48 h or more after nematode applications, or, more likely, if applications are restricted to times when most parasitoid larvae are in diapause within cocoons. The ability of parasitoids to avoid nematode-treated larvae and to actively seek out and kill cocooned CM larvae that survived nematode treatments (escaped nematode infection) enhances the complementarity of entomopathogenic nematodes and *M. ridibundus* and *L. caudatus*.

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