Egyptian Journal of Biological Pest Control

REVIEW ARTICLE Open Access

Pathogenicity of entomopathogenic nematodes to dipteran leaf miners, house flies and mushroom flies

Mohamed Samir Tawfik Abbas*

Abstract

Background: The entomopathogenic nematodes (EPNs), especially in the 2 families Steinernematidae and Heterorhabditidae, are important biocontrol agents against insect pests. The leaf miners (Fam.: Agromyzidae) are cosmopolitan insect pests. There are more than 330 *Liriomyza* spp. including more than 20 species that have been reported as economically important pests of field crops, ornamentals and vegetables. The house flies are serious insect pests for human and animals. More than 100 human and animal diseases have been associated with house flies. Mushroom flies (phorid and sciarid families) are among the main arthropod pests affecting the cultivation of mushroom throughout the world.

Results: Virulence of EPNs differed clearly even on the same insect species and/or by the same nematode species. Such differences might be attributed to the method of treatment, the age of the stage of the insect as well as the concentrations of the tested nematodes. Laboratory studies revealed that the tested nematodes proved to be moderate to highly virulent to larvae as percentage of mortality reached 100%. As for pupae, some studies revealed their moderate or high susceptibility to nematodes, whereas others showed low susceptibility or resistance to infection. Treated adults, or those emerged from treated larvae or pupae, are also susceptible to infection.

Conclusion: Laboratory studies proved the virulence of EPNs to larvae of the 3 dipteran families. Semi-field and field trials indicated that they could successfully reduce the populations of some treated insects without affect the others.

Keywords: Entomopathogenic nematodes, Dipteran insects, Leaf miners, House flies, Mushroom flies, Pathogenicity, Virulence

Background

The free-living, non-feeding 3rd stage infective juveniles (IJs) of the entomopathogenic nematodes (EPNs) (Families Steinernematidae and Heterorhabditidae) possess attributes of both insect parasitoids or predators and microbial pathogens. Like parasitoids and predators, they have chemoreceptors and are motile, in soil, looking for suitable host. Like pathogens they are highly virulent, killing their hosts quickly and can be cultured easily in vivo and in vitro (Gaugler and Kaya 1990). The members of both families are associated with mutualistic bacteria of the genera *Xenorhabdus* (for Steinernematidae) and *Photorhabdus* (for Heterorhabditidae) (Poinar 1990). IJs can locate the host by detecting the insect excretory products, carbon dioxide levels, temperature gradients and movement of the host. IJs then penetrate the host through natural openings, mouth, anus or spiracles, and in addition, IJs in heterorhabditids possess a tooth that enable them to penetrate the host through the cuticle of certain insects. Once they enter the hemocoel, they release the bacteria which multiply and kill the host by cepticaemia (Georgis 1992). EPNs have positive characters including their broad host range, safety to

*Correspondence: samra_mst@hotmail.com

Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt

vertebrates, plants and nontarget organisms (Akhurst 1990), exempting from registration in many countries, easily applied using a standard spray equipment (Georgis 1990), compatible with many chemical and biopesticides and amenable to genetic selection (Kaya and Gaugler 1993). In field application, commercially, a concentration of $2.5–5 \times 10^9$ IJs/ha was recommended to give effective control comparable to chemical insecticides (Georgis and Hague 1991). ENPs have a great potential to be used in integrated pest management programs. They are more specific, proved to be safe and effective alternatives to chemical pesticides. The susceptibility of insect pests varies depending on the selectivity and applied rates of EPN species. Temperature, moisture, aeration and soil type, the species of EPN, age of target insects and soil fauna are important factors affecting the activity of EPNs. In this respect, Platt et al. (2020) mentioned that altering the time of nematode application to either late in the evening or early in the morning can play an important role in attaining efficacy as nematodes need only a few hours of optimum conditions to be able to infect the above ground insect pests.

The leaf miners (Fam.: Agromyzidae) are cosmopolitan insect pests and there are more than 330 *Liriomyza* spp. including more than 20 species that have been reported as economically important pests of field crops, ornamentals and vegetables. Six species, at least, are polyphagous: *L. sativa* Blanchard, *L. trifolii* (Burges), *L. huidobrensis* Blancard, *L. bryoniae* (Kaltenbach), *L. strigata* (Meig.) and *L. longei* Frick (Liu et al. 2009). Heavy infestation by leaf miners causes desiccation and premature fall of leaves. In addition, feeding punctures made by adult females in leaves may be invaded by fungi and bacteria (Noujeim et al. 2015).

The house fly, *Musca domestica* (L.) (Fam.: Muscidae), is a serious insect pest for human and animals. More than 100 human and animal diseases have been associated with house flies including protozoan, bacterial and viral pathogens that are threatening human, poultry and livestock industries (Khan et al. 2013). *Stomoxys calcitrans* (L.) Muscidae), known as "stable fly," presents in several regions in the world and can cause serious damages to cattle including losses of dairy and meat production (Taylor et al. 2012). The insect is able to transmit diseases caused by several microorganisms (Baldacchino et al. 2013).

The gray flesh fly, *Parasarcophaga aegyptiaca* (Salem) (Fam.: Sarcophagidae), is an external parasitoid that has veterinary importance due to its wide distribution in different regions in the world. The fly may cause serious diseases, such as myiasis, and invades various tissues of man and animals leading to serious consequences. The sheep blow fly (or sheep strike), *Lucilia sericata* (Mieg.) (Fam.: Calliphoridae), is found throughout the world and is widely distributed in the USA and Canada (El-Sadawy et al. 2006).

Mushroom flies (phorid and sciarid families) are among the main arthropod pests affecting the cultivation of mushroom throughout the world (Jess et al. (2007). Mushroom yield losses are either directly due to the larvae of mushroom flies feeding on the mycelia or carpophores or due to other pests and diseases vectored by these flies (Erler and Polat (2008). Phorid flies, especially *Megaselia halterata* (Wood), have been globally considered as a minor pest although they are very important problem in Spain mushroom farms. The populations of this phorid fly have recently increased and jumped from being a minor to a major pest in India, the UK and the USA where yield losses ranging between 10 and 40% were reported (Navarro et al. (2021). Mushroom sciarids of the genus *Lycoriella* were considered as the most significant pests of mushroom regardless where production occurs. Up to 5 times as many sciarids as phorids are frequently caught in mushroom cultivation (Jess et al. 2007).

Pathogenicity of EPNs to leaf miners

Laboratory experiments

Effect on larvae

Liu et al. (2009) stated that the IJs of EPNs enter the leaf mines via the punctures made by *Liriomyza* females during egg-laying or by larval feeding. The IJs then penetrate the insect via the anus rather than the mouth parts or spiracles and can kill 1st and 2nd larval instars soon (0.25–0.66 hrs.) postpenetration, whereas pre-pupae die after an average of 15 hrs.

Jacob and Mathew (2016) evaluated the pathogenicity of 3 EPN species against larvae of *L. trifolii* in infested leaves, in Petri dishes, using 5 concentrations: 10, 15, 20, 25 and 30 IJs/maggot. They found that *Steinernema carpocapsae, S. bicornutum* and *Heterorhabditis indica* caused 63–100, 43–93 and 16–67% mortality, respectively, at the 5 tested concentrations in the treated maggots. Similarly, Laleh et al. (2016) studied the efficacy of, *S. feltiae* against *L. sativa* in bean' leaves containing the insect eggs. The leaves were sprayed by *S. feltiae* suspension at the penetration sites of the hatched larvae in Petri dishes. Four concentrations were used: 700, 2500, 9000 and 12,500 IJs/ml. They stated that the IJs could enter the mines via holes made by the hatched larvae and the LC_{50} and LC_{80} for the larvae were 8345 and 74,598 IJs/ml, respectively. Gayatri and Duraimurugan (2019) tested *H. bacteriophora* against late instar larvae of *L. trifolii* at 5 concentrations: 10, 30, 50, 70 and 100 IJs/ larva. The highest mortality% reached 65.1 and 73.8% at 24 and 48 hrs, respectively, at the concentration of 100 IJs/larva. The respective values at 30 IJs/larva were 37.1 and 52%.

The LC_{50} values were found to be 54.1 and 37.8 IJs/larva at 24 and 48 hrs, respectively (Table 1).

Effect on pupae

Lebeck et al. (1993) reported that the early-formed pupae of *L. trifolii* (0.5–1 hrs. old) were found to be susceptible to *S. carpocapsae* infection as the IJs entered the host via the anus and possibly through the mouth parts as evidenced by video. Pupae more than 1 hrs. old were not susceptible to infection. Noujeim, et al. (2015) treated the pupae (the age was not defined) of *L. huidobrensis* by *H. indica* at a concentration of 1000 IJs/5 pupae in Petri dishes. An average of 53% of the treated pupae was found dead but no emerged IJs was noticed for 1 month post-treatment. Jacob and Mathew (2016) evaluated the pathogenicity of 3 EPN species (*S. carpocapsae, S. bicornutum* and *H. indica*) against *L. trifolii* pupae in infested leaves, in Petri dishes, using 5 concentrations, 10–30 IJs/mine, and found no mortality in the formed pupae (Table 1).

Greenhouse and field experiments

Broadbent and Althof (1995) stated that the humidity should be more than 90% in the treated greenhouse for the nematodes to kill the host. Garcia et al. (2018) mentioned that the susceptibility of *L. trifolii* to EPNs is related to the species or strain of the nematode tested.

Harris et al. (1990) carried out a field trial and stated that foliar application of *S. carpocapsae* could suppress the populations of *L. trifolii.* Likewise, Williams and Walters (2000) reported successful control (82%) against 2nd and 3rd larval instars of *L. huidobrensis, L. bryoniae* and *Chromatomyia syngenesiae* (Hardi) infesting vegetables in a greenhouse using foliar application of *S. carpocapsae* or *S. feltiae* at a concentration of 1×10^6 IJs/m^2. This finding was in agreement with that reported by Arthers et al. (2004). Head et al. (2002) carried out an experiment in a greenhouse planted with cabbage in 2 plots severely infested with *L. huidobrensis*. The insecticide deltamethrin (as Decis) was sprayed in the 2 plots and 7 days later one of the 2 plots was sprayed with *S. feltiae* at a concentration of 5000 IJs/ml. The experiment showed a reduction of 89% in the formed pupae in the plot which was treated with the insecticide and nematode than the one treated with the insecticide alone. In this respect, Devi (2019) stated that using *S. feltiae* with the insecticide dimethoate was synergistic against *L. huidobrensis* in IPM System.

However, Liu et al. (2009) reported that although nematodes can provide suppression of insect populations rapidly, their using against *Liriomyza* spp. proved impractical because of their sensitivity to humidity, high cost production and variable effectiveness on such insects in comparison with other control agents.

Pathogenicity of EPNs to house flies

Family Muscidae

Effect on larvae

Laboratory experiments Mahmoud et al. (2007) treated 2nd larval instar of *Musca domestica* (L.) and *Stomoxys calcitrans* (L.) in Petri dishes using 6 concentrations of *S. feltiae*: 50–500 IJs/ml (5 larvae/dish). The results showed that percent mortality in larvae ranged 0–58% and 0–41% in *M. domestica* and *S. calcitrans*, respectively. Also, Leal et al. (2017) evaluated the efficacy of *H. bacteriophora* (HP88) and *H. baujardi* (LPP7) against larvae of *S. calcitrans* in Petri dishes (5 larvae/dish) at 5 concentrations (25–200 IJs/larva). The results showed that *H. bacteriophora* caused 97% mortality in the treated larvae at all tested concentrations and the formed pupae did not give rise to adults. *H. baujardi* caused 33–93.3% mortality at the 5 concentrations. The LC_{50} and LC_{90} for *H. bacteriophora* were 0.36 and 29.1 IJs/larva, respectively, whereas

Table 1 Efficiency of entomopathogenic nematodes against leafminers

Hosts	EPN spp.	Methods of treatment	Concentrations	% mortality or LC_{50}	Authors
A. Larvae					
Liriomyza trifolii	*Steinernema carpocapsae* *S. bicornutum* *Heterorhabditis indica*	Petri dishes	5 conc 10–30 IJs/larva	63–100 43–93 16–46	Jacob and Mathew (2016)
L. trifolii	*H. bacteriophora*	Petri dishes	5 conc 10–50 IJs/larva	$LC_{50=}$37.8 IJs/larva	Gayatri and Duraimurugan (2019)
L. Sativa	*S. feltiae*	Infested leaves	4 conc 700–12,500 IJs/ml	$LC_{50=}$8345 IJs/ml	Laleh et al. (2016)
B. Pupae					
L. trifolii	*S. carpocapasae* *S. bicornutum* *H. indica*	Infested leaves in Petri dishes	5 conc 10–30 IJs/mine	0.0 0.0 0.0	Jacob and Mathew (2016)
L. huidobrensis	*H. indica*	Petri dishes	1000 IJs/5 pupae	53	Noujeim et al. (2015)

Table 2 Efficiency of entomopathogenic nematodes against houseflies

Hosts	EPN spp.	Methods of treatment	Concentrations	% Mortality or LC_{50}	Authors
1. Family Muscidae					
A. Larvae					
Musca domestica *Stomoxys calcitrans*	*Steinernema feltiae*	Petri dishes	6 conc. 50–500 IJs/larva	0.0–58 0.0–41	Mahmoud et al. (2007)
M. domestica	*Heterorhabditis bacteriophora*	Petri dishes (30 larvae/dish)	6 conc	37–100	Bream et al. (2018)
	H. indica		250–2500 IJs/ml	40–100	
	S. glaseri			47–97	
	S. carpocapsae			40–90	
M. domestica	*S. feltiae*	Petri dishes	7 conc. 50–3000 IJs/larva	$LC_{50=}$ 203 IJs/larva	Archana et al. (2017)
	S. glaseri			LC_{50} = 63 IJs/larva	
	S. abbasi			$LC_{50=}$ 309 IJs/larva	
	H. indica			LC_{50} = 29 IJs/larva	
S. calcitrans	*H. bacteriophora*	Petri dishes	5 conc. 25–200 IJs/larva	97% at all concentrations	Leal et al. (2017)
	H.baujard			33–93%	
B. Pupae					
M. domestica (1–2 days old) *S. calcitrans*	*S. feltiae*	Containers with soil	4 conc. 500–3000 IJs/cm^2	− 12% 5 − 52%	Mahmoud et al. (2007) Archana et al. (2017)
M. domestica	*S. feltiae* *S. glaseri*	Petri dishes	5000 IJs/pupa	0.0	
	S. abbasi			0.0	
	S. carpocapsae			0.0	
	H. indica			0.0	
M. domestica	*H. bacteriophora*	Plastic cups. 10 pupae/cup	6 conc. 250–2500 IJs/cup	6.7–83%	Bream et al. (2018)
	H. indica			26.7–70%	
	S. glaseri			10–70%	
	S. carpocapsae			0.0–70%	
S. calcitrans (3 days old)	*H. bacteriophora*	Petri dishes	3 conc. (100–200 IJs/pupa)	0.0	Leal et al. (2017)
2. Family sarcophagidae: larvae and pupae					
Parasarcophaga aegyptica larvae	*Steinernema riobrave*	Petri dishes	4 conc. (500–4000 IJs/5 larvae)	30–96%	El-Sadawy et al. (2006)
	Heterorhabditis bacteriophora	Petri dishes	4 conc. (500–4000 IJs/5 larvae)	26–86%	
P. aegyptica Pupae (8 days old)	*S. riobrave*	Petri dishes	4 conc. (500–4000 IJs/5 pupae)	42–86%	
	H. bacteriophora	Petri dishes	4 conc. (500–4000 IJs/5 larvae)	30–70%	
3. Family calliphoridae larvae and pupae					
Calliphora vicina larvae	*Steinernema feltiae*	Petri dishes	6 conc. (50–500 IJs/ml/larva	0.0–66%	Mahmoud et al. (2007)
Lucilia sericata larvae				16–100%	
C. vicina (1–2 days old) pupae)	*S. feltiae*	Petri dishes	4 conc. (500–3000 IJs/cm^2/5Pupae)	0.0–17%	
L. sericata (1–2 days old) pupae			4 conc. (500–3000 IJs/cm^2/5Pupae)	7–70%	

the respective values for *H. baujardi* were 39.85 and 239.18 IJs/larva, respectively (Table 2).

Archana et al. (2017) evaluated the efficacy of *S. feltiae, S. glaseri, S. abbasi,* and *H. indica* against larvae of *M. domestica* in Petri dishes using 7 concentrations (50–3000 IJs/larva). The LC_{50} values for 2nd instar larvae 3 days posttreatment were 203 for *S. feltiae*, 63 for *S. glaseri,* 309 for *S. abbasi* and 29 IJs/larva for *H. indica.* The respective LC_{90} values were 821, 724, 1561 and 119 IJs/larva. However, in poultry manure assay against 3rd instar larvae in Petri dishes, they found that *H. indica* and *S. carpocapsae* caused minimal mortality, while *S. feltiae, S. glaseri* and *S. abbasi* did not cause mortality in the treated larvae. The authors related this failure to the poor survival of IJs because of the ammonia produced in manure. Bream et al. (2018) evaluated 4 EPNs against 3rd larval instar of *M. domestica* at 6 concentrations (250–2500 IJs/ml) in Petri dishes (30 larvae/dish). The results showed that mortality in larvae ranged 36.7–100% by *H.bacteriophora*, 40–100% by *H. indica*, 46.7–96.7% by *S. glaseri* and 40–90% by *S. carpocapsae* at 3 days posttreatment. The respective LC_{50} values were 320, 390, 494 and 407 IJs/ml.

Arviga and Cortez-Madrigal (2018) evaluated *H. indica* against larvae and adults of *M. domestica* at 1200 and 1600 IJs/ml. The nematode caused the highest mortality (53.3%) in larvae when applied on peat moss. As for adults, the females were more susceptible to infection than males as the average mortality at 1600 IJs/ml was 79.2% for females and 35.5% for males (Table 2).

Semi-field experiments Belton et al.(1987) stated that application of *H. heliothidis* on manure in small barn could significantly reduce the number of emerged *M. domestica* flies. The larvae were susceptible to the nematode infection while the pupae were resistant. Ten weeks posttreatment in a large barn the numbers of emerged flies were 1487 from untreated manure compared to 317 from the treated one indicating 78.7% reduction in the fly population after treatment. Similarly, Taylor et al. (1998) reported that 2 strains of *S. feltiae* (SN and UNK-36) and 2 species of *Heterorhabditis, H. bacteriophora* and *H. megidis,* were tested in a fresh bovine manure substrate against larvae of *M. domestica*. All the 4 strains caused significant mortalities in the insect and the most promising strain, *S. feltiae* SN, gave LC_{50} and LC_{99} values of 4 and 82 IJs/maggot, respectively. These doses were equivalent to 5.1 and 104 IJs/cm^2 of surface area.

Effect on pupae

Laboratory experiments Mahmoud et al. (2007) treated the pupae of *M. domestica* and *S. calcitrans* (1–2 days old) with *S. feltiae* in plastic containers (500 cm^3 volume lined with soil) at 4 concentrations: 500–3000 IJs/cm^2. The results showed that% mortality in the treated pupae ranged 0.0–12 and 5–52%, respectively. However, Leal et al. (2017) reported that *H. bacteriophora* did not affect the viability of 3-day old pupae of *S. calcitrans* which reached 93–97% when treated at 3 concentrations (100, 150 and 200 IJs/pupa); the viability in the untreated pupae was 87.7%. Similarly, Archana et al. (2017) found that pupae of *M. domestica* were found to be resistant to infection by *S. feltiae, S. carpocapsae, S. glaseri, S. abbasi* and *H. indica* when treated at a concentration of 5000 IJs/pupa in Petri dishes. This result was in agreement with that reported by Belton et al. (1987) who found that application of *H. heliothidis* in manure in a small barn proved the resistance of *M. domestica* pupae to the nematode.

Bream et al. (2018) evaluated 4 EPNs against pupae of *M. domestica* (2 days old) at 6 concentrations (250–2500 IJs/ml) in plastic cups (10 pupae/cup). The results showed that mortality% ranged 6.7–83.3% by *H. bacteriophora*, 26.7–70.0% by *H.indica*, 10–66.7% by *S. glaseri* and 0.0–70% by *S. carpocaosae* four days posttreatment. The respective LC_{50} values were 1414, 1074, 1737 and 1718 IJs/ml.

Family Sarcophagidae

Effect on larvae

El-Sadawy et al. (2006) treated 3rd instar larvae of *Parasarcophaga aegyptiaca* (Salem) with *S. riobrave* and *H. bacteriophora* (in Petri dishes) at 4 concentrations: 500–4000 IJs/dish/5 larvae. The results showed that% mortality in larvae ranged 30–96% by *S. riobrave* and 26–86% by H. *bacteriophora*.

Effect on pupae

El-Sadawy et al. (2006) found that treatment the pupae of *P. aegyptiaca* (8 days old) by *S. riobrave* and *H. bacteriophora* at the concentrations of 500–4000 IJs/dish/5 pupae caused 42–86 and 30–70% mortality, respectively.

Family Calliphoridae

Effect on larvae

Toth et al. (2005) evaluated the pathogenicity of different strains of *H. bacteriophora, S. intermedia, S. glaseri, S. anomali, S. riobrave* and *S. feltiae* against 2nd instar larvae of *Lucilia sericata* (Mieg.). They found that all strains did not kill the larvae at 37 °C, whereas at 25°C only strains HU1 and HU2 of *S. feltia* showed significant mortality in the treated larvae. However, the IJs could not develop in the dead larvae. Mahmoud et al. (2007) treated 2nd larval instar of *Calliphora vicina* (Rob.) and *L. sericata* (in Petri dishes) at 6 concentrations of *S. feltiae*: 50–500 IJs/ml/ 5 larvae. The results showed that

percent mortality in larvae ranged 0–66% in *C. vicina* and 16–100% in *L. sericata.*

Effect on pupae

Mahmoud et al. (2007) treated the pupae of *C. vicina* and *L. sericata* (1–2 days old) in plastic containers 500 cm^3 volume lined with soil (5 pupae/container) at 4 concentrations: 500–3000 IJs/$cm^2$of soil. The results showed that % mortality in treated pupae were 0–17 and 7–70%, respectively.

Pathogenicity of EPNs to mushroom flies

Family Phoridae

Laboratory experiments

Scheepmaker et al. (1998) evaluated the susceptibility of 3rd instar larvae of the mushroom fly, *Megaselia halterata* (Wood) to 4 species of EPNs in Petri dishes at a concentration of 1500 IJs/30 larvae/dish). Percentages of mortality were 63, 69, 69 and 71% by *S. feltiae, S. carpocapsae, H. migidis and H. bacteriophora*, respectively. In another experiment, they evaluated *S. feltiae* against the 2nd instar larvae in 24-well tissue culture plates filled with compost (one larva/well) at 4 concentrations (30, 100, 300 and 1000 IJs/larva). The results showed that % mortality ranged 10–38% in the treated larvae. Lamba et al. (2008) treated 3-day old larvae of mushroom fly, *M. sandhui* (Disney) by 4 nematode species in plastic tubes (5 larvae/tube) using 2 ml of the nematode suspensions at 6 concentrations: 50–500 IJs/5 larvae. Two days post-treatment, % mortality was 0% by *Steinernema* sp. and ranged 0–13% by *S. abbasi*, 0–27% by *S. pakistanense* and 7–33% by *H. indica* (Table 3).

Field experiments

Grewal et al. (1993) did not find a significant control of phorid larvae by *S. feltiae* at a concentration of 3×10^6 IJs/m^2 of soil. Similarly, Koppenhofer et al. (2020) mentioned that larvae of the phoried flies have not been controlled effectively with EPNs in the field.

Navarro and Gea (2014) carried out a field experiment to evaluate the efficacy of *S. feltiae* against *M. halterata* at a concentration of 1×10^6 IJs/m^2. The results showed that application of the nematode 10 days after the beginning of infestation alone or with *S. carpocapsae* had no effect on the insect. Navarro et al. (2014) conducted 2 semi-field experiments, using S. *feltiae* and *S. carpocapsae* against *M. halterata*. They applied *S. feltiae* (Sf) at a rate of 1×10^6 IJs/m^2 in the 1st experiment and *S. feltiae* + *S. carpocapsae* (Sc) in the 2nd one at the rate of 0.5×10^6 of both/m^2. The mean number of the emerged *M. halterata*

Table 3 Efficiency of entomopathogenic nematodes against mushroom flies

Hosts	EPN spp.	Methods of treatment	Concentrations	% Mortality or LC_{50}	Authors
A. Family Phoridae larvae					
Megaselia halterata	*Steinernema feltiae*	Petri dishes	1500 IJs/30 larvae/dish	63%	Scheepmaker et al. (1998)
	S. carpocapsae			69%	
	Heterorhabditis migidis			69%	
	H.bacteriophora			71%	
M. halterata	*S. feltiae*	24-well plates filled with compost	4 conc. (30–1000 IJs/larva)	10–38%	
M. sandhui	*Steinernema* sp.	5 larvae/tube	6 conc. (50–500 IJs/5 larvae)	0.0	Lamba et al. (2008)
	S. abbasi			0.0–13%	
	S. pakistanense			0.0–27%	
	H. indica			7–33%	
B. Family Sciaridae larvae					
Lycoriella auripila	*S. feltiae*	24-well plates filled with compost	4 conc. (30–1000 IJs/larva)	91–100%	Scheepmaker et al. (1998)
Bradysia impatiens	*S. yitgalemense*	24-well plates with sand	100 IJs/larva	87%	Katumanyane (2017)
	S. feltiae			72	
	S. jeffreyense			0	
	S. khoisanae			0	
	Steinernema sp.			0	
	H. indica			84	
	H. zealandica			83	
	H. bacteriophora			52	
	H. noenieputensis			81	

adults in the 1st experiment was 233 for the infested control trays (IC) compared to 186.3 for the (SF) and 221.8 for the (Sf + Sc) with nonsignificant differences between them. Nonsignificant differences were observed among the numbers of flies captured in the treatments IC, Sf and Sf + Sc in the second experiment. In addition, there was also no effect of both experiments on % reduction of *M. halterata* adults.

Family Sciaridae

Laboratory experiments

Scheepmaker et al. (1998) tested the pathogenicity of *S. feltiae* to 4th instar larvae of *Lycoriella auripila* (Winn) in compost-filled 24-well tissue culture plates (one larva/well). Four concentrations of the nematode were used: 30, 100, 300 and 1000 IJs/larva. The results showed that % mortality in the treated larvae averaged 91, 100, 100 and 97%, respectively. Kim et al. (2004) studied the infectivity of *S. carpocapsae* to *Bradysia agrestis* Sasakawa and found that the highest mortality rate was achieved in the 3rd and 4th larval instars and the pupal stage (mortality% in 2nd larval instar ranged 23–35%). The egg and 1st instar larvae were not infected. Katumanyane (2017) tested 9 EPN species against 4th larval instar of *B. impatiens* in 24-well plates (one larva/well) at a concentration of 100 IJs/larva. Percentages of mortality in treated larvae were 87% by *S. yirgalemense* 72% by *S. feltiae* 81% by *H. noenieputensis* 84% by *H. indica* 83% by *H. zealandica,* and 52% by *H. bacteriophora.* However, *Steinernema* sp., *S. jeffreyense* and *S. khoisanae* did not cause mortality in the treated larvae. Two laboratory experiments were carried out by Anderson et al. (2021) to evaluate the efficacy of *Bacillus thuringiensis* (*Bt*) and *S. feltiae* against *Lycoriella* sp. larvae infesting mushroom in bioassay containers (3 × 8 cm). In the 1st experiment, they found that the reduction of the numbers of *Lycoriella* adults emerged from *Bt-* treated containers was 40%, whereas the reduction in *S. feltiae* treated ones was 10% compared to the control. The respective reductions in the 2nd experiment were 57% for *Bt* and 2% for *S. feltiae*. However, either *Bt* or *S. feltiae* did not embed the growth of mushroom population (Table 3).

Field experiments

Grewal et al. (1993) reported that *S. feltiae* proved to be successful in the control of *L. auripila* and *L. Mali* (Fitch) in field experiments. Similarly, Rinker et al. (1995) evaluated *S. feltiae* and *H. heliothidis* against *L. mali* in a series of small scale of mushroom crop. IJs were applied to the mushroom casing surface in the irrigation water at densities ranging from 28 to 1120 IJs/cm^2 of casing surface. The mortality of larvae ranged 52–100% by *H. heliothidis* and 38–100% by *S. feltiae*. In addition, Scheepmaker et al. (1997) applied *S. feltiae* against *L. auripila* on mushroom in growing rooms 1 day before and 1 day after casing on the compost at a concentration of 1×10^6 IJs/m^2. The treatment caused 97% control of the F1 generation of the females while the F2 generation was similarly controlled (95%) by an application 7 days after casing. Similarly, Navarro and Gea (2014) found that application of *S. feltiae* against *L. auripila* at a concentration of 1×10^6 IJs/$m^2$10 days after the beginning of infestation was efficient against the insect.

Kim et al. (2004) studied the infectivity of *S. carpocapsae* to *B. agrestis* in a propagation house of mushroom. When the watermelon seeds were treated with *S. carpocapsae* at sowing, the larval density of *B. agrestis* was significantly reduced to 4 and 8 in the nematode-treated plots on the 17th and 34th days posttreatment, respectively, compared to 26 and 30 in the control plots. In another experiment, they found insignificant difference in larval reduction at 7, 14 and 21 days postapplication of the nematode at concentrations of 5, 10 and 20 IJs/gm of soil. However, Leppla et al. (2018) reported that *S. feltiae* could be used for the control of *Bradysia* spp. Similarly, Koppenhofer et al. (2020) mentioned that *S. feltiae* is the only EPN species that is as effective as chemical insecticides against *Bradysia* spp. at the concentration of 2.5×10^6 IJs/m^2. Jess and Schweizer (2009) reported that lower emergence of *L. inginua* (Dufour) adults from mushroom with reduced activity was observed, following the application of *S. feltiae* (Filipjev) at 1.5×10^6IJs/m^2at casing but with no significant effect on mushroom yield.

Discussion

The present article proved the virulence of EPNs to larvae of leaf miners and houseflies under laboratory conditions as mortality may reach 100% (Bream et al. 2018). In case of mushroom flies, it was found that the larvae belong to family Sciaridae are mostly susceptible to EPN infection (Katomanyane 2017), while larvae of family Phoridae seemed to be resistant (Scheepmaker et al. 1998). As for the pupae, Lebeck et al. 1993 reported that the newly formed pupae of the leaf miner, *Liriomyza trifolii*, were found to be susceptible to nematode infection as the IJs entered the host via the anus and possibly through the mouth as evidenced by video. In the present review, some studies revealed low or moderate susceptibility of different ages of the pupae to nematode infection (Bream et al. 2018). Other studies, however, indicated the resistance of pupae to infection, especially the late-aged ones (Archana et al. 2017). It was suggested that the low susceptibility and/or resistance of dipteran pupae to nematode infection might be attributed to different reasons: (1) The completion of puparium and the closer of the anal and oral apertures (Lebeck et al 1993), (2) the toughness of

the puparium and the limited ability of IJs to penetrate through pupal spiracles (Toledo et al. 2005), and (3) the small size of spiracle openings that makes penetration of IJs difficult (Rhode et al. 2012). What supports such reasons is the finding of Kamali et al. (2013) who stated that the IJs of *S. carpocapsae* and *H. bacteriophora* were found to adhere to treated 1 day old pupae of *Dacus ciliates* at the natural openings but no evidence of entry via these openings was noticed. The relatively moderate or high susceptibility of more than 1 day old pupae to EPNs, as reported by some authors, may be attributed, partially, to injuries in pupae from handling, or pupae with incomplete integument that facilitates penetration of the IJs (Henneberry et al. 1995). It is necessary (as reported by Abbas,et al. 2016) to prove the mortality due to nematode infection of treated insects by dissecting the dead insects or by using White traps for migration of infective juveniles from the cadavers. In this respect, Noujeim et al. (2015) reported that treatment of pupae of the leaf miner, *L. huidobrensis* by *H. indica* at a concentration of 1000 IJs/5 pupae caused 53% mortality in such pupae but no emerged IJs was noticed for 1 month posttreatment.

Conclusion

The semi-field and field trials proved successful control achieved by applying EPNs against the populations of the leaf miners. Some field studies revealed the possibility of EPNs to control the sciarid insects of mushroom, while others indicated that they could not affect the populations of other sciarids as well as the phorid insects.

Abbreviations

EPNs: Entomopathogenic nematodes; ha: Hectare; hr: Hour; IJs: Infective juveniles.

Author contributions

The author read and approved the final manuscript.

Funding

Not applicable.

Availability of data and materials

Available when requested.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The author declares that he has no competing interests.

Received: 28 January 2022 Accepted: 6 June 2022
Published online: 22 June 2022

References

Abbas MST, Nouh GM, Abdel-Samad SSM, Negm AA (2016) Infectivity of entomopathogenic nematodes as bio-control agents to *Spodoptera littoralis*, *Ceratitis capitata* and *Bactrocera zonata*. Egypt J Biol Pest Control 26(3):609–613

Akhurst RJ (1990) Safety to non-target invertebrates of nematodes of economically important pests. In: Laird LA, Lacey EW, Davidson EW (eds) Safety of microbial insecticides. CRC Press, Baco Raton, pp 233–240

Anderson VM, Sward GFH, Ranger CM (2021) Microbial control agents for fungus gnats (Diptera, Sciaridae, *Lycoriella* sp.) affecting the production of Oyster mushrooms. Insects 12(9):786

Archana M, de Souza PE, Patil J (2017) Efficacy of entomopathogenic nematodes on developmental stages of house fly, *Musca domestica*. J Parasit Dis 41(3):782–794

Arthurs SK, Heinz M, Prasifka JR (2004) An analysis of using entomopathogenic nematodes against above-ground pests. Bull Entomol Res 94:297–306

Arviga AAM, Cortez-Madrigal S (2018) Susceptibility of *Musca domestica* larvae and adults to entomopathogenic nematodes, Heterorhabditidae and Steinernematidae native in Mexico. Vector Ecol 43(2):312–320

Baldacchino F, Muinworn V, Desquesnes M, Duvallet G (2013) Transmission of pathogens by Stomoxys flies: a review. Parasite 2013(20):26

Belton P, Rutherford TA, Trotter DB, Webster JM (1987) *Heterorhabditis heliothidis*: a potential biological control agent of houseflies in caged layer poultry barns. J Nematol 19:263–266

Bream AS, Fouda MA, Shehata IE, Ragab SH (2018) Evaluation of four entomopathogenic nematodes as biological control agents against the housefly, *Musca domestica* L. Egypt Acad J Biolog Sci 11(1):79–89

Broadbent AB, Althof THA (1995) Foliar application of *Steinernema carpocapsae* to control *Liriomyza trifolii* larvae in chrysanthemum plants. Environ Entomol 24:431–435

Devi G (2019) Compatibility of entomopathogenic nematodes with insecticides in IPM System. Int J Curr Res 11(11):8308–8317. https://doi.org/10.24941/ijer.37220.11.2019

El-Sadawy HA, Abou-Nour AA, Sobh HA (2006) Virulence of entomopathogenic nematodes to *Parasaecophagaa egyptiaca* and *Argas persicus*. In: Proceedings of 3rd international conference on veterinary research division. NRC, Cairo, pp 71–89

Erler F, Polat E (2008) Mushroom cultivation in Turkey as related to pest and management. Isr J Plant Sci 2008(56):303–308

Garcia-del-Pino F, Mortan A, Shapiro-Ilan D (2018) Entomopathogenic nematodes as biological control agents of tomato pests. In: Sustainable management of arthropod pests of tomato (2018): 269–282. https://doi.org/10.1016/B978-0-12-80441-6.00012.7

Gaugler R, Kaya HK (1990) Entomopathogenic nematodes in biological control. CRC Press Inc., Boca Raton, p 365

Gayatri B, Duraimurugan P (2019) Bio-efficacy of *Heterorhabditis bacteriophora* against serpentine leafminer, *Liriomyza trifolii* in oilseed crops. Biol Control. https://doi.org/10.18311/jbc/2019/23030

Georgis R (1990) Commercialization of Steinernematid and heterorhabditid entomopathgenic nematodes. In: Brighton crop protection conference—pests and diseases, vol 1, pp 275–280

Georgis R (1992) Present and future prospects for entomopathogenic nematode products. Biocontrol Sci Tech 2:83–99

Georgis R, Hague NGM (1991) Nematodes as biological insecticides. Pestic Outlook 2:29–32

Grewal PS, Tomalak M, Keil CB, Gaugler R (1993) Evaluation of *Steinernema feltiae* against mushroom sciarid, *Lycoriella mali*. Ann Appl Biol 123:695–702

Harris A, Begely JW, Warkentin DL (1990) *Liriomyza trifolii* suppression with foliar applications of *Steinernema carpocapsae* and abamectin. J Econ Entomol 83:2380–2384

Head J, Palmer LF, Walters KFA (2002) Augmentation biological control using entomopathogenic nematode, *Steinernema feltiae* against *Liriomyza huidobrensis*. In: Proceedings of 1st international symposium on biological control of arthropods, Honolulu, Hawaii, USA, January, 14–18, pp 136–140

Henneberry J, Lindegren E, Forlow J, Burke RA (1995) Pink bollworm, cabbage looper and beet armyworm pupal susceptibility to steinernematid nematodes. J Econ Entomol 88(2):835–839

Jacob YS, Mathew P (2016) Laboratory evaluation of EPNs against *Liriomyza trifolii*. J Biopestic 9(1):27–33

Jess S, Schweizer H (2009) Biological control of *Lycoriella inginua* cultivation: a comparison between *Hypoaspis miles* and *Steinernema feltiae*. Pest Manag Sci 65(11):1195–1200. https://doi.org/10.1002/ps.1809
Jess S, Murchie AK, Bingham JF (2007) Potential sources of sciarid and phorid infestations and implications for centralized phases I and II mushroom compost production. Crop Prot 2007(26):455–464
Kamali S, Karimi J, Hossini M, Herrera C, Duncan LW (2013) Biocontrol potential of entomopathogenic nematodes, *Heterorhabditis bacteriophora* and *Steinernema carpocapsae* on cucurbit fly, *Dacus ciliatus* (Dipt.:Tephritidae). Biocontrol Sci Technol 23(11):1307–1323
Katumanyane A (2017) Prospects for using EPNs as a bio-control agent against fungus gnats, *Bradysia* spp. in nursery and glasshouse crops. M.Sc. Thesis, Faculty of Agrisciences, Stellenbosch University, South Africa. Stellenbosch University https://scholar.sun.ac.za
Kaya HK, Gaugler R (1993) Entomopathogenic nematodes. Ann Rev Entomol 38:181–206
Khan HAA, Shad SA, Akram W (2013) Resistance to new chemical insecticide in the housefly, *Musca domestica* in Pakistan. Parasitol Resist 2013:18
Kim HH, Choo HY, Kaya HK, Lee DW, Lee SM, Jeon HY (2004) *Steinernema carpocapsae* as a biological control agent against the fungus gnat, *Bradysia agrestis* in propagation houses. Biocontrol Sci Technol 14:171–183
Koppenhofer AM, Shapiro-Ilan D, Hiltpold I (2020) Entomopathogenic nematodes in sustainable food production. https://doi.org/10.3389/fsufs.2020.00125/ful
Laleh E, Shiri MR, Dunphy GB (2016) Efficacy of EPN, *Steinernema feltiae* against *Liriomyza sativa*. Egypt J Biol Pest Control 26(3):583–586
Lamba JS, Walia KK, Mrig K, Walia RK (2008) Infectivity and virulence of indigenous entomopathogenic nematodes to mushroom phorid fly, *Megaselia sandhui*. J Biol Control 22(2):411–416
Leal LCS, de Monteiro CMO, de Mendonca AE, Bittencourt VRE, Bittencourt AJ (2017) Potential of EPNs of genus *Heterorhabditis* for the control of *Stomoxy scalcitrans* (Dipt.: Muscidae). Braz J Vet Parasitol 26(4):451–456
Lebeck LM, Gaugler R, Kaya HK, Hara AH, Johnson MW (1993) Host stage suitability of the leafminer, *Liriomyza trifolii* to the entomopathogenic nematode, *Steinernema carpocapsae*. J Invert Pathol 62:58–63
Leppla NC, Johnson MW, Merritt JL, Zalom FG (2018) Applications and trends in commercial biological control for arthropod pests of tomato. In: Sustainable management of arthropod pests of tomato, pp 283–303. https://doi.org/10.1016/B978-0-12-802441-6.00013.9
Liu T, Le Kang KM, Heinz K, Trimble J (2009) Biological control of *Liriomyza* leafminers: progress and prospective. In: CAB reviews: prospective in agriculture, veterinary science, nutrition and natural resources 2009. 4, No. 004. www.cababstractsplus.org/cabreviews
Mahmoud MF, Mandour NS, Romazkov YI (2007) Efficacy of the EPN, *Steinernema feltiae* against larvae and pupae of four fly species in laboratory. Nematol Medit 35:221–226
Navarro MJ, Gea FJ (2014) Entomopathogenic nematodes for the control of phorid and sciarid flies in mushroom crops. Pesq Agropec Bras 49(1):11–17
Navarro MJ, Carrasco J, Gea FJ (2014) Chemical and biological control of diptera in Spanish mushroom crops. In: Proceedings of the 8th international conference on mushroom biology and mushroom products. (ICMBMP8)
Navarro MJ, Carrasco J, Gea FJ (2021) Mushroom phorid flies. Agronomy 2021(11):1958
Noujeim E, Sakr J, El-Sayegh D, Nemer N (2015) In vitro susceptibility of the pea leafminer, *Liriomyza huidobrensis* pupae to EPNs, *Heterorhabditis indica* and the fungus *Beauveria bassiana*. Leban Sci J 16(2):19–26
Platt T, Stokwe NF, Malan AP (2020) A review of the potential use of entomopathogenic nematodes to control above-ground insect pests in South Africa. J Enol Vitic. https://doi.org/10.21548/41-1-2424
Poinar GO Jr (1990) Taxonomy and biology of Steinernematidae and Heterorhabditidae. In: Gaugler R, Kaya HK (eds) Entomopathogenic nematodes in biological control. CRC Press, Boca Raton, pp 23–61
Rhode C, Moino A, Carvalho F, da Silva P (2012) Selection of entomopathogenic nematodes for the control of the fruit fly, *Ceratitis capitata*. Agraria Ravista Brasileria de Celincias Agrilas (ISSN, Online, 1981-0997)
Rinker DL, Althof THA, Dano J, Alm G (1995) Effect of entomopathogenic nematodes on control of a mushroom sciarid fly and on mushroom production. Bio-Control Sci Technol 5:109–119
Scheepmarker JWA, Geels EP, Van Griensven LJ, Smits PH (1998) Susceptibility of larvae of the mushroom fly, *Megaselia halterata* to entomopathogenic nematode, *Steinernema feltiae* in bioassays. Biocontrol 43:201–214
Taylor DB, Szalanski AL, Adams BJ, Peterson RD (1998) Susceptibility of house fly larvae to entomopathogenic nematodes. Environ Entomol 27(6):1514–1519
Taylor DB, Moon RD, Mark DR (2012) Economic impact of stable flies on dairy and beef cattle production. J Med Entomol 49(1):198–209
Toledo J, Ibarra JE, Liedo P, Gomez A, Williams T (2005) Infection of *Anastrepha ludens* larvae by *Heterorhabditis bacteriophora* under laboratory and field conditions. Biocontrol Sci Technol 15(6):627–634
Toth EM, Marialigeti K, Fodor A, Lucska A, Farkas A (2005) Evaluation of entomopathogenic nematodes against larvae of *Lucilia sericata* (Dipt.:Calliphoridae). Acta Vet Hung 53:65–71. https://doi.org/10.1556/AVet.53.2005.1.7
Williams EC, Walters KFA (2000) Foliar application of the entomopathogenic nematode, *Steinernema feltiae* against leafminers on vegetables. Biocontrol Sci Technol 10:61–70

Publisher's Note