Egyptian Journal of Biological Pest Control

RESEARCH

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Potential of different entomopathogenic nematode strains in controlling *Atrijuglans hetaohei* Yang (Lepidoptera: Heliodinidae)

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Abstract

Background: *Atrijuglans hetaohei* (Yang) (Lepidoptera, Heliodinidae) is a major pest of walnut in China. Larvae feed on the seeds of walnut fruit. Damaged fruits turned black and fallen prematurely. Current management relies upon broad-spectrum of chemical insecticides applied in the late spring or early summer. However, due to missed applications or poor application timing, high levels of *A. hetaohei* infestation may still occur. Entomopathogenic nematodes (EPNs) have been long used for suppressing the soil-dwelling and fruit-boring pest, but few were done on the control of *A. hetaohei*. The present study was conducted to determine the virulence of seven EPN strains against *A. hetaohei* under laboratory conditions.

Results: Among the seven EPN strains, *Steinernema glaseri* (Sgib strain) (Rhabditida: Steinernematidae) had good potentials in the management of *A. hetaohei* because of the better desiccation tolerance and pathogenicity than other strains. The cocooned and mature larvae of *A. hetaohei* all could be infected by the nematodes of Sgib strain. Sgib strain had the best efficiency to *A. hetaohei* under the infection condition of 24 °C and 100 IJs per larva. At 48-h exposure to 24 °C, the highest mortality rate of *A. hetaohei* treated with Sgib strain was 96.67%. Sgib strain of 100 IJs/larva caused 100% mortality after 72-h post-application.

Conclusion: Sgib strain from Steinernematidae was the favorable to control *A. hetaohei* larvae which inhabit in the soil surface effectively. Sgib strain may be a contribution to the biological control of *A. hetaohei* in China.

Highlights

- *Steinernema glaseri* (Sgib strain) could be used as biocontrol agent for *A. hetaohei* larvae because of the better anti-desiccation ability and pathogenicity.
- Sgib strain showed high infectivity to the cocooned larvae as well as the mature larval *A. hetaohei*.
- Sgib strain had the optimum infecting effect with 100 IJs per larva at 24 ℃ and still maintained high pathogenicity after spraying into the field for 96 h.
- The results suggested that some potential for Sgib strain from Steinernematidae as one tool in controlling *A.hetaohei* larvae, which is the main fruit-boring pest of walnut in China

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Keywords: *Atrijuglans hetaohei*, Entomopathogenic nematodes, Pathogenicity, Biological control

Background

Atrijuglans hetaohei (Yang) (Lepidoptera: Heliodinidae) is a major pest of walnut (*Juglans regia* Linnaeus, Juglandales: Juglandaceae) fruits. The pest is widely distributed in China (Wang et al. 2016). It has 1–2 generations a year and overwinters as mature larvae. The female moths deposit eggs on the outside of the walnut fruits after mating (Chen et al. 2017), once the eggs hatched, the larvae bore into the walnut fruits and results in the early fruit drop, which seriously affects the yield of walnut (Zhang et al. 2018). After completing their development inside the walnut fruits, the mature larvae come out of the fruits and overwinter within cocoon as larvae stage, in soil of a few centimeters deep. The difficulty of controlling this pest comes from its larvae hiding inside the fruits and its mature larvae and the cocoons are in the soil (Wang et al. 2016). At present, the control strategies of *A. hetaohei* largely depend on the chemical insecticides (Tian et al. 2010) and sex pheromone (Yin et al. 2014). Even now, few studies have been devoted to the other alternative control strategies of *A. hetaohei*.

Entomopathogenic nematodes (EPNs) are potent insect parasites, and they have been successfully used to control a variety of important economical insect pests, especially fruit-boring pests, *Cydia pomonella* (Linnaeus) (Lepidoptera: Olethreutidae) (Odendaal et al. 2016) and *Bactrocera zonata* (Saunders) (Diptera: Tephritidae) (Dolinski 2016).

EPNs also are potentially promising biological control agents for foliar pests. The infective juvenile (IJ) of EPNs is mutually symbiotic with the bacteria, *Xenorhabdus* (Enterobacteriales: Enterobacteriaceae) (in *Steinernema* and *Neosteinernema*) and *Photorhabdus* (in *Heterorhabditis*), which produce toxins after entering into the insect host body. The bacterial toxins are mainly responsible for the death of host which generally occurs within 48 h (Kaya and Gaugler 1993). Then, the IJs fed on the nutrients and multiplied. The new IJs left the cadaver in search for a new host (Georgis et al. 2006). EPNs are amenable to mass produce and do not require specialized application equipment as they are compatible with standard agrochemical equipment, including various sprayed and irrigative systems (Askary and Abd-Elgawad 2021).

In this study, virulence of different strains of EPNs for infectivity of *A. hetaohei* larvae was compared and evaluated the optimum infective conditions of the selected EPN strain in laboratory experiments. This study may provide more information in using EPNs as an effective control method for *A. hetaohei*.

Methods

Materials

Seven EPN strains of *Steinernema glaseri* (HBgy and Sgib strains) (Rhabditida: Steinernematidae), *S. carpocapsae* (HB310 and All strains) (Rhabditida: Steinernematidae), *S. longicaudum* (CBZB strain) (Rhabditida: Steinernematidae), and *Heterorhabditis bacteriophora* (HB8 and HB140 strains) (Rhabditida: Heterorhabditidae) were maintained in Pest Bio-control Laboratory, Hebei Agricultural University, China. The nematodes were cultured in the larvae of the greater wax moth, *Galleria mellonella* (Linnaeus) (Lepidoptera: Pyralidae). Harvested IJs were kept in sterilized water at 10 ± 1 °C in the fridge until they were used in the experiments.

The blackened walnuts were collected from Baoding district (114°50′–115°17′E, 38°45′–39°07′N and 50–1006.7 m a.s.l.), which is located in Hebei Province, China. All the black walnuts were placed on a sandy plate. Matures emerged from the fruit and cocooned in the sand. Under natural conditions, the matures overwinter within cocoon as larval stage in the soil. In this study, the mature larvae and cocoon were selected for the experiments. *G. mellonella* larvae were obtained from the Pest Bio-control Laboratory, Hebei Agricultural University, China. Larvae were fed on artificial diet (22% maize meal, 22% wheat germ, 11% dried milk, 5.5% dry yeast, 17.5% bee wax, 11% honey, and 11% glycerin) (Mukherjee et al. 2009) and reared in insectary (28 ± 1 °C, RH 80 ± 5%, and a photoperiod of 14L: 10D).

Larvicidal activity of seven EPN strains against *A. hetaohei* larvae

Bioassays were conducted to compare the infectivity of seven EPN strains against the mature larvae of *A. hetaohei*. Twenty mature larvae were placed in Petri dish (90-mm in diameter), filled with two filter papers, which was added with 1 ml IJs suspension (1000 IJs/ml). Petri dishes were sealed with para-film and maintained in a climatic chamber of 25 ± 1 °C and RH 80 ± 5%. Controls received water only. The experiments were carried out three times under the same conditions on different dates with 60 larvae distributed in three replicates (each of 20 larvae). Corrected mortality of the larvae was recorded every 24 h for 120 h, respectively. Dead cadavers were dissected to check for nematodes presence or not under the stereomicroscope.

Determination of desiccation tolerance of seven EPN strains

Desiccation tolerance of nematodes was evaluated under the relative humidity of 50% as described by Chen et al (1999). Each Petri dish filled with two filter papers was provided by 1 ml IJs suspension (1000 IJs/ml). The Petri dishes were kept into the incubator at 25 ± 1 °C, RH 50 ± 5% for 7 h. After then, each Petri dish was added 5 ml distilled water and nematode mortality rate was assessed from three subsamples (100 µl per sample) from each Petri dish under a stereo-zoom microscope. Nematodes were considered dead if they did not respond to prodding.

Determination of the optimal EPN strain against cocooned and mature larvae of *A. hetaohei*

One optimal EPN strain was gained on the basis of the above experimental results. The infectivity of the optimal EPN to the cocooned and mature larvae of *A. hetaohei* was tested, respectively. The methods and assessment were done as described above. Larval mortality of each treatment was recorded 48 h post-inoculation. The experiments were carried out three times under the same conditions on different dates with 60 larvae distributed in three replicates (each of 20 larvae).

Influence of temperature and nematode concentrations on virulence of EPNs

Virulence of the optimal EPN strain to *A. hetaohei* was tested at different temperatures of 16, 20, 24, 28 and 30 °C. Petri dish (90-mm) with two layers of filter paper was added with 20 mature larvae and 1 ml nematode suspension (1000 IJs/ml). Petri dishes were sealed with a para-film and maintained in a climatic chamber at above condition, respectively. Larval mortality of each treatment was recorded after 24, 48, 72 and 96 h post-application. The experiments were carried out three times under the same conditions on different dates with 60 larvae distributed in three replicates (each of 20 larvae).

The virulence of the optimal EPN strain to *A. hetaohei* larvae was assessed at different nematode concentrations. Bioassay was carried out in 24-well micro-plates. Each well with two layers filter papers was added with one mature larvae and different nematode suspension of 5, 10, 20, 50 and 100 IJs per larva, respectively. All the plates were maintained in a humidity controller at 25 ± 1 °C and RH 80 ± 5%. Larval mortality of each treatment was recorded after 24, 48, 72 and 96 h post-application. Each cadaver was dissected under a stereomicroscope to ensure nematode presence. The experiments were carried out three times under the same conditions on different dates with 60 larvae distributed in three replicates (each of 20 larvae).

Data analysis

All experimental data in the virulence assay were subjected to analysis of variance with treatment as fixed effect (ANOVA). The significant differences between the treatments were determined by using Tukey's test ($p < 0.05$). All data analyses were performed using SPSS software (SPSS Inc. 2009).

Results

Selection of the optimal EPN strain on *A. hetaohei* larvae

Corrected mortality rates of *A. hetaohei* larvae treated by seven EPNs are shown in Fig. 1A. The larvae of *A. hetaohei* were sensitive to the majority of EPN strains tested. There was a positive correlation between the exposure time for EPNs and larval mortality of *A. hetaohei*. HBgy, Sgib, HB310, HB8 and HB140 strains all caused 100% larval mortality within 96 h. According to Fig. 1B, Sgib strain was showed the best anti-desiccation ability with the corrected mortality of 23.83%. The anti-desiccation abilities of HBgy, CBZB, HB8 and HB140 strains were very poor with the corrected mortalities ranged from 98 to 100%. There was no mortality in untreated controls. In a word, Sgib strain was selected as the optimal EPN strain against *A. hetaohei* based on the results of the pathogenicity and desiccation tolerance.

The normal walnut fruits (Fig. 2a) blackened gradually (Fig. 2b) after boring by *A. hetaohei* larvae. At the end, the damaged walnut fruits became completely black, sunken and dry (Fig. 2c). So, it is always called "walnut black." The mature larva of *A. hetaohei* was 7.5–9.0 mm in length with creamy white body and tawny head (Fig. 2d). The larvae of *A. hetaohei* became moribund and the whole body gradually presented different characteristics after infected by the steinernematids and heterorhabditis. The larval cadaver was similar to the normal larvae after infected by steinernematid (Fig. 2e), while the dead one infected by heterorhabditid turned purplish-red (Fig. 2f).

Virulence of Sgib strain on cocooned and mature larvae of *A. hetaohei*

In the field, matures of *A. hetaohei* overwinter within cocoon as larval stage (Fig. 3B). Infectivity of Sgib strain against the cocooned larvae was tested as well as the mature larvae in this study (Fig. 3A). The results showed that the cocooned larvae of *A. hetaohei* also could be infected by IJs of Sgib strain. The corrected mortality of cocooned and mature larvae of *A. hetaohei* was 91.67% and 100%, respectively. This result indicated that Sgib

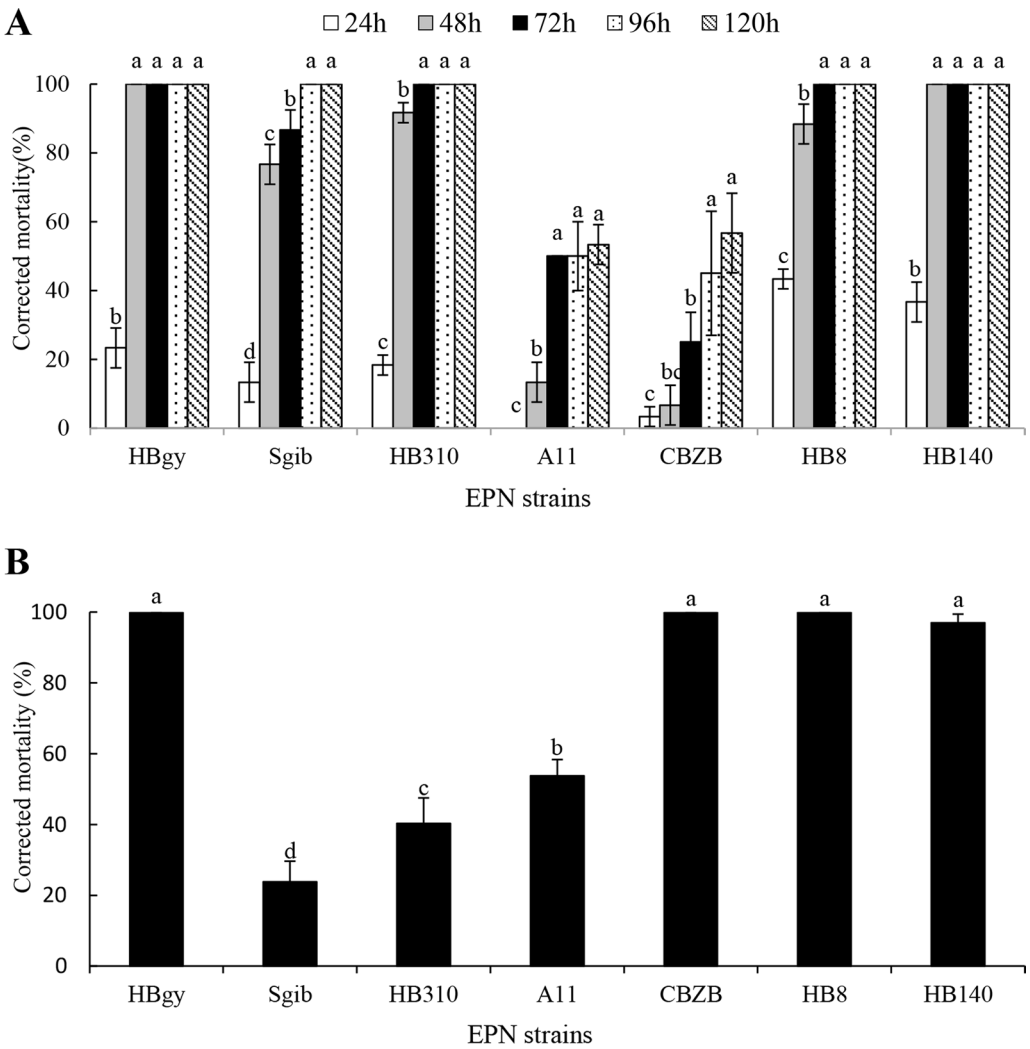

Fig. 1 Pathogenicity and desiccation tolerance of seven EPN strains on *Atrijuglans hetaohei*. **A** Corrected mortality of *A. hetaohei* larvae caused by seven EPN strains. **B** Corrected mortality of different EPN strains in anti-desiccation assays. HBgy = *Steinernema glaseri*, Sgib = *Steinernema glaseri*, HB310 = *Steinernema carpocapsae*, All = *Steinernema carpocapsae*, CBZB = *Steinernema longicaudum*, HB8 = *Heterorhabditis bacteriophora*, HB140 = *Heterorhabditis bacteriophora*. Differing letters indicate values significantly different from one another at the $p < 0.05$ level (Tukey's HSD test)

strain could infect both mature and cocoon stages of *A. hetaohei*.

Influence of temperature and nematode concentrations on virulence of Sgib strain

Infective juveniles of Sgib strain was affected by different infection temperatures (Fig. 4A). Exposing the treated *A. hetaohei* larvae to 16 or 30 °C resulted in a weak infection activity of Sgib strain obviously. At 48-h exposure to 24 °C, the highest mortality rate of *A. hetaohei* treated with Sgib strain was 96.67%. But there were insignificant differences among the different treatments at temperatures 20, 24 and 28 °C ($P > 0.05$).

The virulence of Sgib strain was also significantly affected by nematode concentrations. At the highest concentration of 50 IJs/larva and 100 IJs/larva, Sgib strain caused higher mortality than treatments of 5 IJs/larva and 10 IJs/larva for 24, 48, 72, and 96 h (Fig. 4B), showing significant differences among the treatments tested ($P < 0.01$). But there was non-significant differences between the treatments of 50 and 100 IJs/larva ($P > 0.05$).

Discussion

Application timing of EPNs with reference to the life cycle of the target insect is a key factor to increase efficacy. In order to achieve the best control result, the nematodes should be applied at the most susceptible

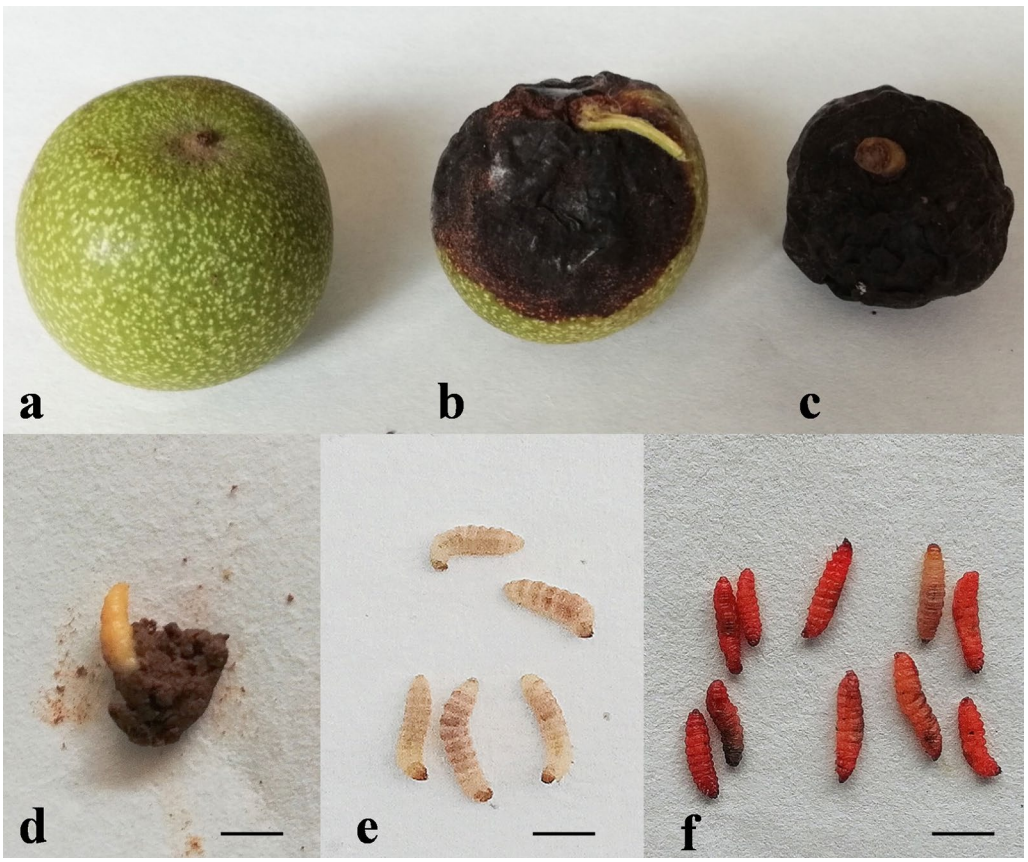

Fig. 2 Symptoms of the damaged walnut fruit and the dead larvae *Atrijuglans hetaohei* infected with EPNs. **a** Healthy walnut fruit; **b**, **c** Walnut fruit bored by *A. hetaohei* larvae; **d**: Healthy *A. hetaohei* larvae; **e**, **f** Infected larvae of *A. hetaohei* with Steinernematidae and Heterorhabditidae, respectively

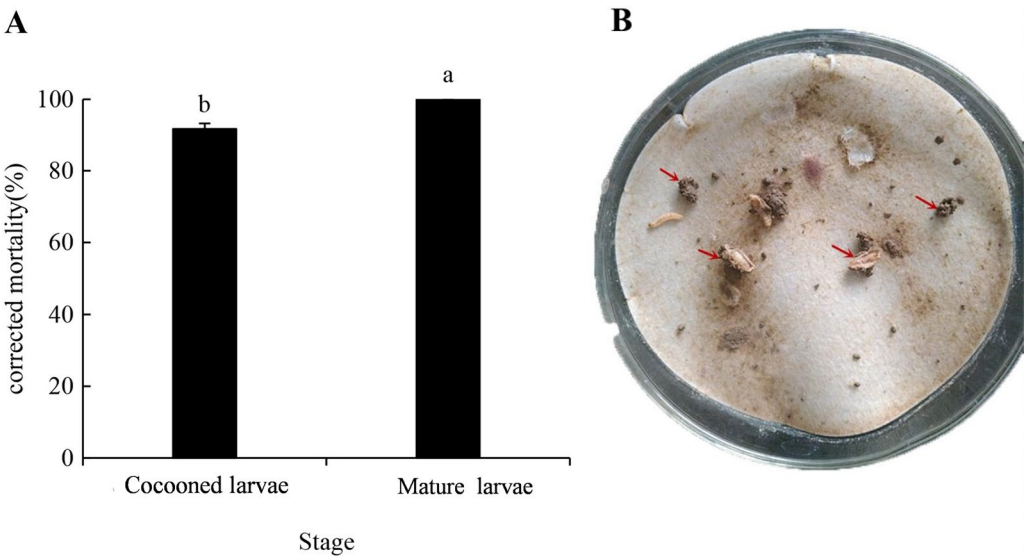

Fig. 3 Efficacy of Sgib nematodes on the cocoons and mature larvae of *Atrijuglans hetaohei*. **A** Corrected mortality of cocooned and mature larvae of *A. hetaohei*; **B** The cocooned and mature larvae of *A. hetaohei*. The arrow indicates the cocooned larvae. Differing letters indicate values significantly different from one another at the $p < 0.05$ level (Tukey's HSD test)

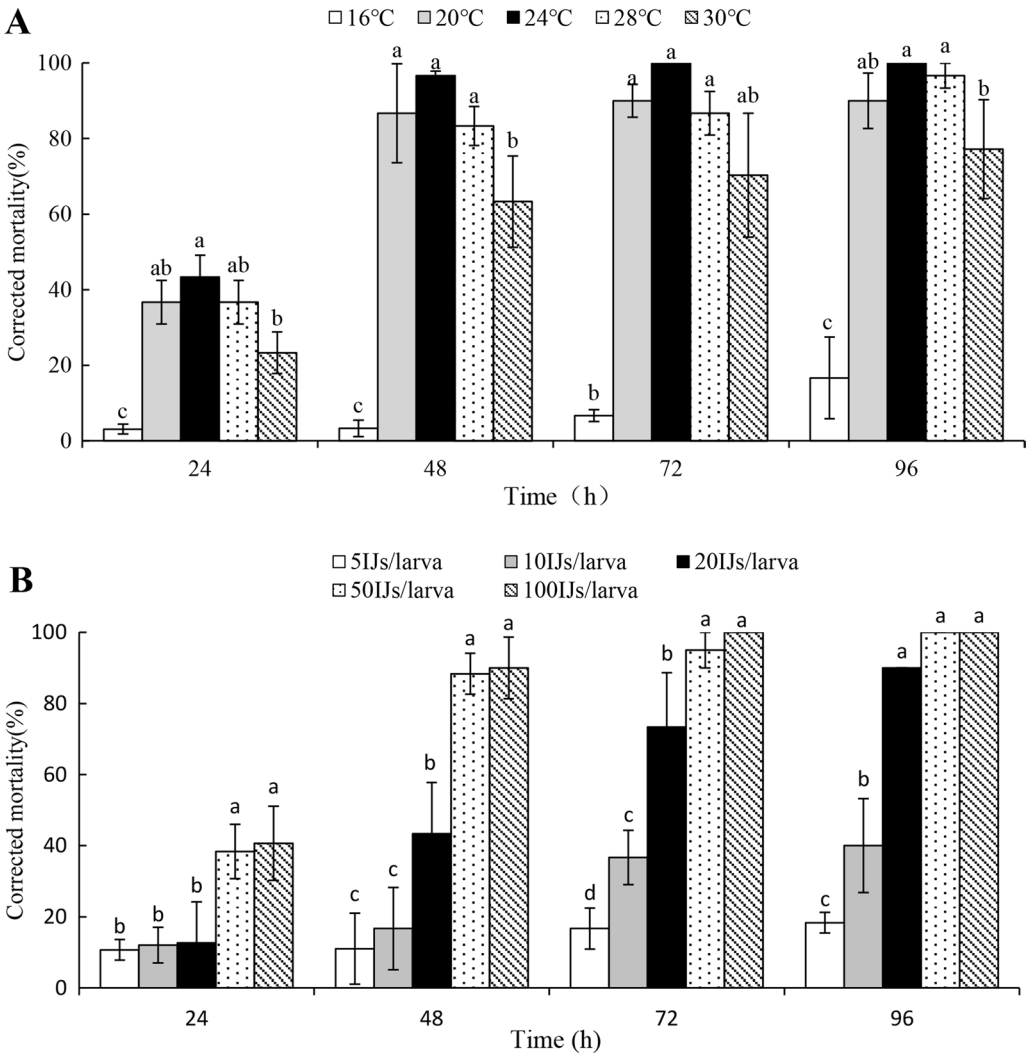

Fig. 4 Screening the optimal infection conditions of Sgib nematodes against *Atrijuglans hetaohei*. **A** Corrected mortality of *A. hetaohei* larvae after exposure to different temperature. **B** Corrected mortality of *A. hetaohei* larvae after exposure to different nematodes concentration. Differing letters indicate values significantly different from one another at the $p < 0.05$ level (Tukey's HSD test)

stage of different host (Nikdel et al. 2010). Saffari et al. (2013) observed a great mortality of prepupae and pupae of *Thrips tabaci* (Lindeman) (Thysanoptera: Thripidae) compared to 2nd instar larvae when applying *Steinernema feltiae* (Rhabditida: Steinernematidae), *H. bacteriophora*, and *S. carpocapsae*. The pupae of *Frankliniella fusca* (Hinds) (Thysanoptera: Thripidae) also were found to be susceptible to eight EPN species according to Gulzar et al. (2021). The highest susceptibility of pupae may be due to their lack of mobility (Saffari et al. 2013). In this study, the newly cocooned mature larvae of *A. hetaohei* could be infected by nematodes as well as mature larvae because of their loose cocoon structure. The cocooned mature larvae of *A. hetaohei* as the target stage were selected, because the cocooned mature larvae are a more realistic option in practical applications.

Factors such as desiccation tolerance and temperature are reported to affect the survival and activity pathogenicity of the nematodes (Van Damme et al. 2016). In the present study, *S. glaseri* Sgib strain can be used as the potential biological control agent against *A. hetaohei* with the high efficacy of desiccation tolerance. As the same EPN species, *S. glaseri* HBgy strain had the poor desiccation tolerance. This is an interesting experimental result. We speculated that it was related to the ecological environment of the EPNs soil sample. The soil samples of *S. glaseri* Sgib strain are collected in natural surroundings with drier microenvironment. Nematodes have evolved

strong anti-desiccation in such environments. It has been reported that nematodes have significant differences in desiccation tolerance among different genus (Husin and Port 2021), and obtained results further indicate that this difference may also exist among different nematode strains of the same species.

The result in this paper suggested that non-statistical difference was observed among 20, 24 and 28 °C for the efficacy of Sgib strain against *A. hetaohei* larvae. Exposing the treated *A. hetaohei* larvae at 16 °C resulted in the weak infection activity of Sgib strain obviously. Previous studies also showed that EPNs will enter the "diapause" state with the decrease of the enzymatic activity, mobility and metabolic expenditures at low temperature (Cagnolo and Campos 2008) or become inactivate at 30–40 °C (Saleh et al. 2018). As the temperature in Baoding during May to June is at 19–29 °C, the environment temperature was suitable for the EPNs performance in biological control programs of *A. hetaohei* larvae. However, the biological factors during the field use of EPNs should also be considered. Mcgraw and Schlossberg (2017) suggested that the biotic factors may be more important than soil moisture in improving the post-application persistence and ultimately EPNs performance in biological control programs in turf grass (Mcgraw and Schlossberg 2017).

EPNs can spontaneously search for host insect lived in the soil or cryptic habitats and often were used to control a variety of target pests (Mcgraw and Schlossberg 2017). EPNs infection required for efficiently release the pathogenic bacteria they carry along with toxin proteins that function in killing the host or modulating its immune response (Maurizio and Maristella 2018). According to the research of Saleh et al. (2018), applying *Heterorhabditis marelatus* D1 (Rhabditida: Heterorhabditidae) at the two rates (5 and 15 IJs/cm^2 of soil surface) on sandy soil containing old pupae of *Bactrocera zonata* (Saunders) (Diptera: Tephritidae), resulted in different mortality percentages of adults, according to the rate of nematode application rather than the nematode species. Obtained results in this paper showed that it is economical to use 50 IJs/larva and 100 IJs/larva under laboratory. When EPNs are sprayed in the field, the spray concentration should be higher than 100 IJs/larva due to the different soil types, temperature and ultraviolet in the environment. It was also noted that the nematodes concentration used as well as the time exposed to the nematodes are all important for improving the control performance. The foraging strategy of IJs can give rise to the differences in pathogenicity among different EPNs species (Erdou 2021). Heterorhabditidae nematodes display cruiser strategy and suit to infect insects lived under the soil, while steinernematid nematodes are more effective to insects near the soil surface because of their ambusher strategy (Mahmoud, 2017). Obtained results indicated that Sgib strain from steinernematids was favorable to control *A. hetaohei* larvae which inhabit in the soil surface effectively.

Successful control of pests using EPNs is not easy because nematode efficacy is closely related to a number of adverse environment factors, but also directly related to EPN species. As we know, the main cause of death of the host infected with the EPNs is related to the symbiotic bacteria. Compared to EPNs, symbiotic bacteria are easier to be cultured in vitro on a large scale and sprayed against the foliar pests in the field. For example, *Xenorhabdus szentirmaii* and *X. nematophila* (Enterobacterales: Morganellaceae) supernatants killed over 90% of *Tetranychus urticae* (Acari: Tetranychidae) adults (Eroglu et al. 2019). The insecticidal effect of different symbiotic bacteria was obviously different due to the different EPNs. For *Steinernema glaseri*, co-operation is of more importance for the killing of insects in the *S. glaseri–Xenorhabdus poinarii* couple. *Xenorhabdus poinarii* alone is a virulent or only weakly virulent against several insects (Ansari et al. 2003). Jean-Claude et al (2014) provided insight into the mechanisms underlying genomic erosion in *X. poinarii*. Further studies are required to determine the possible role of such complementation in insect virulence.

Atrijuglans hetaohei has one generation per year in Baoding district, China. The mature larvae overwinter within cocoon as larval stage, in soil of a few centimeters deep. It is difficult to be controlled by the tradition chemicals due to the confining behavior of their larvae inside the fruits or their pupae in the soil. It was being confirmed that this study was one of the beneficial explorations of EPNs on this walnut boring pest, even though field trials are necessary. Next step, field bioassays over more than two consecutive years was planned to evaluate the efficacy of EPNs against *A. hetaohei* in a variety of abiotic and biotic soil properties, including texture, compaction, and abundance of several common soil microarthropod groups, to determine which properties were most strongly associated with EPNs efficacy. The result of the study laid the solid foundation for controlling *A. hetaohei* larvae by EPNs in the future.

Conclusion

The present study was conducted to determine the virulence of seven EPN strains against *A. hetaohei* under laboratory conditions. *Steinernema glaseri* (Sgib strain) had good potentials in the management of *A. hetaohei* because of the better desiccation tolerance and pathogenicity than other strains. The cocooned and mature larvae of *A. hetaohei* all could be infected by the nematodes of Sgib strain. Sgib strain had the best efficiency

to *A. hetaohei* under the infection conditions of 24 °C and 100 IJs per larva. **S**teinernematid Sgib strain seems as a promising result for favorable control of *A. hetaohei* larvae.

Abbreviations
EPNs: Entomopathogenic nematodes; IJs: Infective juveniles; RH: Relative humidity.

Acknowledgements
Authors would like to thank the Research Fund of Hebei Agricultural University which supported experimental facilities to carry out experiments.

Author contributions
ZNG conceived research and wrote the manuscript. WC and AZ conducted experiments with WG. PS analyzed the virulence assays. QW was responsible for writing reviews and edits. All authors read and approved the final manuscript.

Funding
This work was supported by the Research Fund of Hebei Agricultural University.

Availability of data and materials
All data generated or analyzed during this study are included in this published article.

Declarations

Ethics approval and consent to participate
This article does not contain any studies with human participants or animals performed by any of the authors. All participants have given oral informed consent.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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Received: 19 April 2022 Accepted: 30 July 2022
Published online: 17 September 2022

References

Ansari MA, Tirry L, Moens M (2003) Entomopathogenic nematodes and their symbiotic bacteria for the biological control of *Hoplia philanthus* (Coleoptera: Scarabaeidae). Biol Control 28:111–117. https://doi.org/10.1016/S1049-9644(03)00032-X

Askary TH, Abd-Elgawad M (2021) Opportunities and challenges of entomopathogenic nematodes as biocontrol agents in their tripartite interactions. Egypt J Biol Pest Control 31:42. https://doi.org/10.1186/s41938-021-00391-9

Cagnolo S, Campos V (2008) Effect of storage temperature on survival and infectivity of *Steinernema rarum* (OLI strain) (Rhabditida: Steinernematidae). J Invertebr Pathol 98:114–115. https://doi.org/10.1016/j.jip.2008.02.013

Chen SB, Yang HW, Luan XQ, Yang XF, Jian H (1999) The antidesiccation ability of entomopathogenous nematodes. Chin J Biol Control 15:100–103 (**in Chinese with English abstract**)

Chen DD, Li TF, Wang QQ, Tang GH (2017) The ultrastructure of *Atrijuglans hetaohei* (Lepidoptera: Oecophoridae) moth antennae. Scientia Silvae Sinicae 53:118–132. https://doi.org/10.11707/j.1001-7488.20171013 (**in Chinese with English abstract**)

Dolinski C (2016) Entomopathogenic nematodes against the main guava insect pests. Biocontrol 61:325–335. https://doi.org/10.1007/s10526-015-9695-y

Erdou FD (2021) On the efficiency of entomopathogenic nematodes (Rhabditida: Heterorhabditidae and Steinernematidae) on rust red flour beetle, Tribolium castaneum (Herbst.) (Coleoptera: Tenebrionidae). Egypt J Biol Pest Control 31:116. https://doi.org/10.1186/s41938-021-00461-y

Eroglu C, Cimen H, Ulug D, Karagoz M, Hazir S, Cakmak I (2019) Acaricidal effect of cell-free supernatants from *Xenorhabdus* and *Photorhabdus* bacteria against *Tetranychus urticae* (Acari: Tetranychidae). J Invertebr Pathol 160:61–66. https://doi.org/10.1016/j.jip.2018.12.004

Georgis R, Koppenhöfer AM, Lacey LA, Bélair G, Duncan LW, Grewal PS, Samish M, Tan L, Torr P, van Tol R (2006) Successes and failures in the use of parasitic nematodes for pest control. Biol Control 38:103–123. https://doi.org/10.1016/j.biocontrol.2005.11.005

Gulzar S, Usman M, Wakil W, Wu S, Shapiro-Ilan D (2021) Virulence of entomopathogenic nematodes to pupae of *Frankliniella fusca* (Thysanoptera: Thripidae). J Econ Entomol 114:2018–2023. https://doi.org/10.1093/jee/toab132

Husin T, Port GR (2021) Efficacy of entomopathogenic nematodes against *Tuta absoluta*. Biol Control 160:104699. https://doi.org/10.1016/j.biocontrol.2021.104699

Jean-Claude O, Sylvie P, Gaëlle B, Hélène C, Claudine M, Zoé R, Corinne T, Stéphanie V, Patrick T, Alain G, Sophie G (2014) Attenuated virulence and genomic reductive evolution in the entomopathogenic bacterial symbiont species, *Xenorhabdus poinarii*. Genome Biol Evol 6:1495–1513. https://doi.org/10.1093/gbe/evu119

Kaya H, Gaugler R (1993) Entomopathogenic nematodes. Annu Rev Entomol 38:181–206. https://doi.org/10.1146/annurev.en.38.010193.001145

Mahmoud MF (2017) Biology and use of entomopathogenic nematodes in insect pests biocontrol, a generic view. Cercetari Agronomice in Moldova 49:85–105. https://doi.org/10.1515/cerce-2016-0039

Maurizio B, Maristella M (2018) Nematobacterial complexes and insect hosts: different weapons for the same war. Insects 9:117. https://doi.org/10.3390/insects9030117

Mcgraw BA, Schlossberg MJ (2017) Fine-scale spatial analysis of soil moisture and entomopathogenic nematode distribution following release in wetting agent-treated turf. Appl Soil Ecol 114:52–61. https://doi.org/10.1016/j.apsoil.2017.02.016

Mukherjee K, Altincicek B, Hain T, Domann E, Vilcinskas A, Chakraborty T (2009) *Galleria mellonella* as a model system for studying listeria pathogenesis. Appl Environ Microbiol 76:310–317. https://doi.org/10.1128/AEM.01301-09

Nikdel M, Niknam G, Dordaei AA (2010) Evaluation of susceptibility of the brown tail moth, *Euproctis chrysorrhoea* (L.), to entomopathogenic nematodes under laboratory conditions. Nematol Mediterranea 38:3–6

Odendaal D, Addison MF, Malan AP (2016) Evaluation of above-ground application of entomopathogenic nematodes for the control of diapausing codling moth (*Cydia pomonella* L.) under natural conditions. Afr Entomol 24:61–74. https://doi.org/10.4001/003.024.0061

Saffari T, Madadi H, Karimi J (2013) Pathogenicity of three entomopathogenic nematodes against the onion thrips, *Thrips tabaci* Lind. (Thys.; Thripidae). Arc Phytopathol Plant Prot 46:2459–2468. https://doi.org/10.1080/03235408.2013.797158

Saleh MME, Metwally HMS, Mahmoud YA (2018) Potential of the entomopathogenic nematode, *Heterorhabditis marelatus*, isolate in controlling the peach fruit fly, *Bactrocera zonata*, (Saunders) (Diptera: Tiphritidae). Egypt J Biol Pest Control 28:22–27. https://doi.org/10.1186/s41938-018-0029-0

Tian MJ, Liu FL, Dong JQ (2010) Outbreaking regularity of *Atrijuglans hetaohei* and its comprehensive control techniques. J Northwest for Univ 25:127–129. https://doi.org/10.3724/SP.J.1238.2010.00474 (**in Chinese with English abstract**)

Van Damme VM, Beck BK, Berckmoes E, Moerkens R, Wittemans L, De Vis R, Nuyttens D, Casteels HF, Maes M, Tirry L, Clercq PD (2016) Efficacy of entomopathogenic nematodes against larvae of *Tuta absoluta* in the laboratory. Pest Manag Sci 72:1702–1709. https://doi.org/10.1002/ps.4195

Wang QQ, Zhang ZQ, Tang GH (2016) The mitochondrial genome of *Atrijuglans hetaohei* Yang (Lepidoptera: Gelechiidae) and related phylogenetic analyses. Gene 581:66–74. https://doi.org/10.1016/j.gene.2016.01.027

Yin WH, Yu ZD, Tang GH, Peng SB, Zhai MZ (2014) Control of fruit pests and diseases of walnut with trunk injection. J Fruit Sci 31:175–180 (**in Chinese with English abstract**)

Zhang ZQ, Li TF, Tang GH (2018) Identification and characterization of conserved and novel miRNAs in different development stages of *Atrijuglans hetaohei* Yang (Lepidoptera: Gelechiidae). J Asia-Pac Entomol 21:9–18. https://doi.org/10.1016/j.aspen.2017.10.014

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