Biological control of *Temnorhynchus baal* Reice and Saulcy (Coleoptera: Scarabaeidae) attacking Egyptian golf courses using entomopathogenic nematodes

A. A. El Halafawy [1], Mona A. Hussein [2,*], H. M. S. Khalifa [1], and H. A. Mohamed [1].

[1] *Plant Protection Department, Faculty of Agriculture, Al-Azhar University, Cairo, Egypt.*
[2] *Pests and Plant Protection Department, Agricultural and Biological Research Institute, National Research Centre, Cairo, Egypt*

[*] Corresponding author E-mail: **ma.hussein@sci.nrc.eg** (M. Hussein)

ABSTRACT:

Background and aim: This study is the first to report the use of entomopathogenic nematodes (EPNs) against Temnorhynchus baal, a significant pest of golf courses in Egypt. Golf tourism has been recently introduced to Egypt, and this sport represents a valuable addition to the country's tourism portfolio, aligning with the growing global demand. EPNs have been successfully commercialized as biocontrol agents for many Scarabaeidae species. Methodology: Four different concentrations of nine different species and strains of EPNs were used against young and grown larvae of T. baal under laboratory conditions.Results: Heterorhabditis marilatus and H. bacteriophora were notably effective in managing T. baal larvae. Moreover, data showed that Steinernema glaseri, S. riobravae, and S. carpocapsae (BA2) were more effective than S. carpocapsae (ALL) in controlling the larvae of T. baal. Each strain demonstrated the capability of providing greater than 90% mortality, The range of mortality for S. glaseri and S. riobravae was 80-100% and 60-90% for S. carpocapsue (All) for young larvae of T. baal while the mortality % ranged between 60-90% and 50-80% for the same strains, for grown larvae of T. baal. Conclusion: This study provides a broad understanding for the ability of selected native and foreign species of EPNs in attacking a serious pest of golf course. EPNs could play a promising role in controlling golf course pests successfully and could be incorporated into an IPM program.

Keywords: Golf course pests; *Temorhynchus*; *Heterorhabditis*; *Steinernema*; Biological control.

INTRODUCTION

Turfgrass is essential for environmental sustainability and human well-being, serving multiple purposes such as erosion control, air purification, water filtration, aesthetic enhancement, providing a cooling effect, and supporting wildlife habitats (Potter and Braman, 1991; Mathew, 2021). Globally, turfgrass is a fundamental component of golf courses and various landscapes. In environments like golf courses and sod farms, where turfgrass is grown primarily for its utility and attractiveness, any discoloration is unacceptable (Dupuy and Ramirez, 2016). Golf clubs play a significant role in supporting local economies by creating jobs, attracting tourism, and stimulating various sectors of the economy (Donaldson et al., 2011).

There are several insects damaging turf grass including: Various species of white grubs in the genera *Popillia*, *Phyllophaga*, and *Amphimallon*; Chinch bugs (mainly *Blissus* species, such as *Blissus leucopterus* and *Blissus insularis*); Sod webworms species (such as *Crambus* and *Herpetogramma*); Billbugs (primarily *Sphenophorus* species); larvae of armyworms moth species (such as: *Spodoptera frugiperda* and *Pseudaletia unipuncta*); and the white grubs, *Temnorhynchus baal* Reice and Saulcy (Coleoptera: Scarabaeidae) (Gireesh and Joseph, 2020).

Larvae of the white grubs, *T. baal* are usually found throughout Cyprus, Greece, Lebanon, Saudia Arabia, Yemen, tropical Africa, Algeria, Libya, and Egypt as severe pests of Strawberry

(Endrodi, 1985). Larvae attack both the roots and the underground stems. The primary cause of these pests' economic significance is the 3rd larval instar grubs' feeding activity (Veeresh, 1988; Chandel et al., 2015). *Temnorhynchus* larvae were first recorded in 2006 as a serious insect pest of sugarcane crop in Upper Egypt (Abd-Rabou and Abd-el-Samea, 2006). Recently, *T. baal* was found to attack and induce severe damage to golf courses at El-Katamiya Heights Resorts, New Cairo, Egypt (This study).

Research on creating safe and alternative control methods is vitally needed because chemical pesticides pose health risks and cause environmental damage (Jagodič et al., 2019). Entomopathogenic nematodes (FPNs) in the families of Heterorhabditidae and Steinernematidae along with their symbiotic bacteria *Photorhabdus* and *Xenorhabdus*

(Enterobacteriales: Enterobacteriaceae), respectively are effective microscopic bioagents successfully applied against many insect pests both in soil and in cryptic environments worldwide (Akhurst et al., 1992; Ehlers, 2007; Saleh et al., 2009; Nouh and Hussein, 2014; Hussein, 2021). The eco-friendly EPNs are considered unique bioinsecticides for having many positive criteria such as, their high virulence against a wide range of pests, their ability for host-seeking, the ease of production and application, the exemption from pesticides registration in many countries, and their environmental safety. In addition to that, EPNs are compatible with a wide range of chemicals and non-chemical control techniques (Gorgis et al., 1991; Koppenhofer and Kaya, 1998; Lacey and Georgis, 2012; Akhurst and Smith, 2002; Abate et al., 2017). The increased demand for organic food and turf grass opened a new market for commercial products of EPNs. Application of EPNs against Coleopteran insect pests was reported by several authors and EPNs were found to be as effective as insecticides (Kajuga et al., 2018; Abdel-Razek et al., 2018, Torrini et al., 2020).

Once the only free-living infective juvenile (IJs) stage locates an insect host; it penetrates the hemocoel through its natural openings and release their associated bacteria (Kaya and Stock, 1997). The bacteria start multiplying and secrete toxic metabolites which lead to the death of their host within 24-48 h due to septicemia (Poinar, 1979). The nematode begins to feed on both the bacteria and host tissues, develop, mate, and multiply for several generations. The newly produced IJs retain some bacteria in their guts and leave the cadaver searching again for a new host.

Parasitic nematodes were recovered and isolated (Kaya et al., 2006) from different soil types all around the world. The native EPNs are expected to be more tolerant and more adapted to climatic changes such as sun radiation; drought and high temperatures than the foreign nematodes (Atwa and Hassan, 2014; Hussein, 2021& 2022). The Egyptian soil contains several treasures and finds to be rich in EPNs different species which were recovered and described (Hussein, 2004; Hussein and Abou El-Soud, 2006; El-Khonezy, 2007; Abdel-Razek et al., 2018; Machado et al., 2021; Hussein, 2022). Native EPNs are more appreciated because they are likely more adaptive and safer to be applied for the Egyptian fauna and flora (Atwa and Hassan, 2014; Abdel-Razek et al., 2018), therefore, laboratory testing of additional species or new strains may lead to identification of EPNS with superior potential traits for controlling *T. baal*.

In this study, we examined nine different species and strains of EPNs (BA1, HP88, HBE, Mar and new field strain from Heterorhabtids and BA2, All, S.G. and Rio from Steinernematids) against the young and grown larvae of *T. baal* under laboratory conditions.

MATERIAL AND METHOD

Organisms:

Galleria mellonella (Lepidoptera: Pyralidae):

The culture of the greater wax moth, *Galleria mellonella* L. was maintained on the artificial diet modified by Hussein et al. (2022) and Metwally et al. (2012). The insect diet consisted of 22% corn groats, 22% wheat flour, 11% milk powder, 11% bee honey, 11% glycerol, 5.5% yeast powder and 17.5% bee wax. The honey and glycerol were combined in 1L beaker. The honey-glycerol mixture was then added to the dry ingredients and mixed properly. The resulting mixture was stored in a refrigerator and transferred to the rearing containers of *G. mellonella* larvae. The larvae were obtained from a permanent culture of a susceptible strain reared in the Pests and Plant Protection laboratory at NRC, Dokki, Giza, and were transferred to transparent glass rearing jars (15 cm in diameter and 25 cm in height) containing 250g of the previously prepared diet. The containers were closed with a filter paper disc and a metal screen and incubated at 28± 2°C. When the larvae grew to pupae and then to moths, the adult females laid eggs on the filter paper discs, which were collected and transferred to new rearing jars containing fresh media.

Temnorhynchus baal (Coleoptera: Scarabaeidae):

The larvae of the white grub, *T. baal* were collected from golf courses turfgrass at El-Katamyia Heights resorts, New Cairo, Egypt during the seasonal activity for three successive seasons (2021:2024). The larvae of *T. baal* were identified for the first time from golf course in Egypt at the Taxonomy Unit, Plant Protection Research Institute, Agricultural Research Centre in 2022. All white grubs have three C-Shaped larval instars based on Wilson (1969). According to their head capsules and their feeding activity, larvae were divided into young (1st and 2nd instars) and grown (3rd instar) larvae. The larval collection site has not been treated with EPNs or insecticides during or before experiments. White grub larvae were

gathered by digging out turfgrass root and soil in 0.4x0.4X0.4 m pits. After that, the collected larvae were placed in plastic cups 5 kg capacity, 25 cm height, and 20 cm in diameter), half filled with moistened sterile sand soil, covered with cotton clots. Larvae were fed for a week on the stem and roots of turf grass. The young and gown larvae of the white grub were selected for running the experiments.

Entomopathgenic nematode strains and sources.

The infective juveniles (IJs) of the tested EPNs were reared in vivo, on the full-grown larvae of the greater wax moth, *G. mellonella* at 25 ± 2 °C. The *Galleria* culture was reared and maintained on an artificial media developed by Hussein et al. (2022). Two hundreds of *G. mellonella* last instar larvae were placed on a Petri dish (20 cm diameter) padded with two filter paper discs. About 10,000 IJs in 5 ml distilled water were moistened but not wet. Fourteen days after the larval infection, new IJs were collected from the insect cadavers using the White trap procedure White **(**1927**).** Culturing, production, and harvesting of the IJs followed the methods of Kaya and Stock (1997). The list of tested heterorhabtid and steinernematid EPNs used in the study are represented in Table (1). The choice of the tested EPNs strains were based on preliminary experiments conducted with different species and strains. Four native and five foreign EPNs species and strains were selected. The IJs were reared for 5 successive generations in vivo before application. Field strain (EGHB isolate) EPNs isolated from the golf course and identified based on 16SRNA at the Physiology Department, Plant Protection Research Institute, Agricultural Research Centre. The strain was deposited at the GenBank with accession No. PP446814.

Laboratory screening:

The efficacy of nine different species and strains of native and foreign EPNs against *T. baal* larvae were conducted at the Nematology Lab., Pests & Plant Protection Dept., Agricultural & Biological Research Institute, National Research Centre. The 24-well bioassay protocol was used to test the different nematode species and strain (Table 1) for their ability to infect young and grown larvae under laboratory conditions. The EPNs were suspended in distilled water and the concentration of the suspension was adjusted and determined according to Kaya and Stock (1997). The suspended EPNs were acclimatized for at least 6 h at room temperature before application. Five young and grown larvae of *T. baal* were placed in plastic cup (15x9x7 cm) half-filled with sterile sandy soil (the same component of sandy soil at the golf course) at a depth of 1 cm from the surface with the roots and stem of the grass plant for their feeding.

The larvae were infected with the tested EPNs, *H. bacteriophora* (HP88, BA1, and HBE), *H. marilatus* (Mar), *S. carpocapsae* (BA2 and All), *S. riobravae* (Rio), *S. glaseri* (SG), and the new nematode strain,

H. bacteriophora (EGHP isolate), which was isolated from infected *T. baal* larvae cadavers obtained from golf courses turfgrass at El-Katamyia Heights Resorts, New Cairo, Egypt and identified based on the 16S RNA at the Physiology Department, Plant protection research institute, Agricultural Research Centre and deposited in the GenBank Database with accession No. PP446814.

Since the white grubs were collected from golf course and the appearance of grass is very critical to the playground, The number of sample size collected was restricted. The plastic cups were covered with plastic perforated lids and each cup received 2 ml of the four tested EPNs concentrations (500, 1000, 2000, and 4000 IJs/ larva). For each concentration 25 larvae/ 5 replicates were conducted for each EPNs strain. The control plates were inoculated with distilled water only. The experiments were conducted under controlled laboratory conditions, maintaining at a temperature of 25 ± 2 °C and 55-65 % R.H. The water content of the soil was kept constant (20%) during the experiment using water sensor from Handan Yantai Import and Export Co., Ltd., China Mortality percentages were recorded daily for a week approximately and the LC_{50}, LC_{90}, LT_{50}, and LT_{50} for each EPNs strain were estimated. Experiments were repeated three times for each strain.

Statistical analysis

The obtained normally distributed data were subjected to analyses of variance (ANOVA) by using CoHort software program (2005) and significant differences among the tested factors portioned by LSD and F test at probability level of $P < 0.05$. The $LC_{50,90}$ or $LT_{50,90}$ values were estimated using log-probit software program Ldp line® model" Ehabsoft" (Bakr, 2000).

RESULTS AND DISCUSSIONS: -

Heterorhabtids nematodes against *T. baal*:

Data in Table (2) showed that the efficiency of different strains of two species (*H. bacteriophora* and *H. marilatus*) on young larvae of the scarabaeied beetle, *T. baal* under laboratory conditions. The data indicated that *H. marilatus* (mar) caused a 100% mortality of young larvae of *T. baal* three days post-treatment with a concentration of 4000 IJS/ml. However, 100% mortality induced to the same instars of *T. baal* 5 days after treatment by the two strains of *H. bacteriophora* (HP88 and BA1) at the highest concatenation 4000 IJs/ml. On the other hand, the new isolated field strain, *H. bacteriophora* (EGHP isolate) caused 93.32% larval mortality after 5 days when applied against young larvae at the same concentration (400 IJS/ml).

The LC_{50} and LC_{90} values were 34.2 and 1573 IJs/ml when *T. baal* young larvae treated by *H. marilatus* (mar.) followed by *H. bacteriophora* (BA1) which recorded LC_{50} and LC_{90} values of 44.25 and 777 IJs/ml, respectively, when applied against young larvae of *T. baal*. Moreover, data in Table (2) showed that the LT_{50} and LT_{90} values were 1.25 and 3.33 days when young larvae of *T. baal* treated by *H. marilatus* (mar) followed by new isolated strains, *H. bacteriophora* (EGHP isolate) which recorded LT_{50} and LT_{90} of 1.4 and 3.5 days, respectively.

From the results in Table (2), there are significant differences between the strains (F= 8.050 – P = 0.001 and LSD = 5.0712) when *Heterorhabtids* nematodes used against young larvae of *T. Baal*. Also, there are significant differences between the concentrations in all tested Heterorhabtid strains (F= 35.456 – P= 0.000 – LSD = 4.535). While no significant differences between the strains and concentrations (F= 1.375 – P = 0.217 – LSD = 10. 142).

Data in Table (3) indicated the efficiency of different strains of the two species of *H. bacteriophora* and *H. marilatus* on grown larvae of the scarabaied beetles *T. baal* under laboratory conditions. The mortality percentage of the grown larvae of *T. baal* recorded 100% 4 days after treatment with *H. marilatus* (mar), while induced 90% and 86% mortality 5 days post-treatment by *H. bacteriophora* (HP88) and new filed strain *H. bacteriophora* (EGHP isolate), respectively. The LC_{50} and LC_{90} for grown larvae of *T. baal* were 171 and 272.6 IJs, 304.2 and 6992 IJs and 450 and 655 IJs/ml after treatment by *H. bacteriophora* (HBE), *H. bacteriophora* (HP88) *and H. marilatus*, respectively.

According to data in table (3), the values of LT_{50} and LT_{90} for grown larvae of *T. baal* after treated with *H. bacteriophora* (EGHP isolate), *H. bacteriophora* (HBE), *H. bacteriophora* (BA1), and *H. marilatus* (mar) were 2.36 and 6.6 days, 2.4 and 5.13 days, 2.55 and 6.93 days and 2.56 and 3.92 days, respectively.

Based on results recorded in Table (3), there are significant differences between the strains (F= 21.752 – P = 0.000 and LSD= 5.500) when *Heterorhabtids* nematodes used against grown larvae of *T. baal*. There are significant differences also between the concentrations in all tested Heterorhabtids strains (F= 42.237– P= 0.000 – LSD = 4.919). In addition, there are significant differences between the strains and concentrations (F= 3.800 – P = 0.0007 – LSD = 11. 001.

Data in Table (2 and 3), the EPNs *H. marilatus* (mar) at concentration of 4000 IJs/ml was more effective than all other tested nematode strains, when applied against young and grown larvae of *T. baal* while the *H. bacteriophora* (HBE) more effective against grown larvae of *T. baal*. Moreover, the young larvae of *T. baal* were more susceptible to most tested nematodes than the grown larvae. Nematode dosage at higher concentrations yielded highest larval mortality as compared to lower concentrations.

Steinernematid nematodes against *T. baal*:

The results for the efficiency of different strains of three species of steinernematids EPNs (*S. carpocapsae, S. riobravae,* and *S. glaseri*) on young larvae of the scarab beetle, *T. baal* under laboratory conditions are represented in Table (4). Results showed a gradual increase in the larval mortality with the increase of the concentration for each nematode strain. The mortality rate of *T. baal* due to application of EPNs ranged between 60 to 100% after 5 days pot-treatment.

Data in Table (4) indicated that the mortality % of young larvae of *T. baal* were 100, 100, 100, and 90% after treatment by *S. carpocapsae* (BA2), *S. riobravae* (Rio), *S. glaseri* (S.G.) and *S. carpocapsae* (ALL), respectively when treated with the highest concentration (4000IJs/ml) 5 days post-treatment.

The LC_{50} values for the young larvae of *T. baal* were 38, 194, 194, and 315 IJs/ml after treatment by *S. glaseri* (S.G.), *S. riobravae* (Rio), *S. carpocapsae* (BA2), and *S. carpocapsae* (ALL), respectively. The LC_{50} data in Table (4) cleared that the EPNs, *S. glaseri* was more effective

than other tested steinernematids against young larvae of scarab beetles, *T. baal.*

The LT_{50} of *T. baal* young larvae were 1.72, 1.9, 2.0, and 2.1 days after being treated with *S. carpocapsae* (BA2), *S. carpocapsae* (ALL), *S. riobravae* (Rio) and *S. glaseri* (S.G.). The statistical analysis showed that there were significant differences between different strains and between different concentrations, and no significant differences interactions found between different strains concentrations.

Data in Table (5) revealed that, the efficiency of different steinernematid strains of EPNs on grown larvae of *T. baal* under laboratory conditions, The total mortality % at higher concentrations (4000 IJs/ml) were 100, 90, 90, and 80% after 5 days of treatment by *S. glaseri* (S.G.), *S. riobravae* (Rio), *S. carpocapsae* (BA2), and *S. carpocapsae* (ALL), respectively.

The LC_{50} values for the *T. baal* grown larvae after treatment by the same species were 280, 315, 399, and 517 IJs/ml, respectively. Data in Table (5) cleared that, *S. glaseri* (S.G.) was more effective than other tested steinernematids against grown larvae of scarab beetles, *T. baal* followed by other strains depending on the mortality % and LC_{50} values.

The results in Table (5) stated that *S. carpocapsae* (ALL) was the lowest effective on *T. baal* grown larvae depending on the mortality %, LC_{50}, LC_{90}, LT_{50}, and LT_{90} than other tested steinernematids strains.

The LT_{50} values recorded with *T. baal* after being treated with *S. carpocapsae* (BA2), *S. riobravae* (Rio), *S. glaseri* (S.G.), and *S. carpocapsae* (ALL) were 1.92, 2.1, 2.51 and 2.64 days, respectively. Based on the statistical analysis there is a significant difference between different strains and between different concentrations, and no significant differences in interactions between strains and concentrations.

The native nematode, *H. bacteriophora* (BA1), was proved to be more effective nematode in controlling the grown larvae of scarab beetle attacking golf course in Egypt. However, the young larvae were more susceptible to the endemic *H. marilatus* (Mar). On the other hand, and based on data recorded in Tables (4) and (5), the EPNs *S. glaseri* (S.G.) was more effective in controlling both young and grown larvae of *T. baal* at higher concentrations than the other tested species of steinernematid nematodes.

This study is the first report on using EPNs against one of the most serious golf course insect pests namely, *T. baal* in Egypt. Golf tourism has recently been introduced to the Egyptian community (López-Bonilla et al., 2020). This sport represents a valuable addition to Egypt's tourism offerings, aligning with the growing global demand for diverse tourist activities. Recent advances carried out on the bio-control of scarab beetles in the United States of America, Australia, Africa, and Egypt (on strawberry) showed their susceptibility to fungi, bacteria, and EPNs (Koppenhöfer et al., 2000).

Our results show that EPNs can control young and grown larvae population of *T. baal*, at levels comparable to chemical insecticides. New isolates of EPNs species may prove to be more effective for controlling certain insect groups, for example, *S. kushidae* appears more specific and effective against scarabaeids than other species (Mamiya, 1988, 1989). Kaya (1990) stated that laboratory bioassays showing efficacy against pests cannot be applied under field conditions, where a high level of control is required. However, laboratory experiments provide directions to better use of biological control agents. In our laboratory studies, *H. marilatus* (mar) and the newly Egyptian isolated strain *H. bacteriophora* (EGHP isolate), induced higher mortality than the foreign strain *H. bacteriophora* (HP88) on young and grown larvae of *T. baal* under laboratory conditions. The low larval mortality by some different species and strains of heterorhabtids EPNs against the larvae of the scarab beetles, *T. baal* at low concentration may have resulted from the failure of its associated *Photorhabdus* bacteria to be established in the larvae. Saunders and Webster (1999) and Shapiro-Ilan et al. (2000) suggested that the rate of EPNs infection and pest mortality varies due to the differences in the rate and time of EPNs penetration.

Results revealed that *H. marilatus* (mar) and *H. bacteriophora* (EGHP isolate) demonstrated significant control of *T. baal* larvae and these data are consistent with McGraw and Koppenhöfer (2008). Moreover, several studies also agree with our results. Atwa (2009) studied the efficiency of *S. glaseri* (NJ) against *T. baal* attacking Strawberry under laboratory conditions. The mortality rates of *T. baal* were 100, 100, 94, and 96% for the first, second, third larval instars, and adult stage 5 days post-treatment, respectively. Additionally, EPNs are tolerant with wide range of chemicals and amendment commonly used in turfgrass management (Krishnayya and Grewal, 2002; Koppenhofer and Grewal, 2005; Alumai et al.,

2006). Several research has demonstrated that EPNs can effectively manage white grub populations just as well as other pesticides when the proper circumstances are met (Grewal et al., 2004; Guo et al., 2013; Patil et al., 2020). The nematode can find, attack, and kill white grub larvae deep in soil which pesticides cannot. Moreover, EPNs have a second chance of new parasitism by producing a new generation of thousands of IJs inside grubs' cadever. The emerged juveniles move back in soil, searching, and attacking new insect pests.

The use of EPNs could be increased by the development of new control approaches, e.g. their combination with synergistic such as the chloronicotinyl insecticides, imidacloprid or BTs (Koppenhöfer and Kaya, 1998; Koppenhöfer et al., 1999) on golf courses.

CONCLUSION

Chemical insecticides can cause more problems than benefit in turf grass pest management and based on the research and this current study, EPNs have a huge potential to reduce the pest populations in golf course. This study provides a broad understanding of the ability of selected native and foreign species of EPNs in attacking a serious pest of golf course. Our findings suggest that both the native nematode, *H. bacteriophora* (BA1) and the foreign *S. glaseri* were better than other tested EPNs species in controlling the scarab beetle larvae. Although the researchers stated that the young instar larvae are more susceptible to the EPNs than the grown larvae, the native EPNs (BA1) were more efficient in controlling grown larvae than all other tested nematode strains. However, the young larvae were more susceptible to the foreign *H. marilatus* (Mar). On the other hand, *S. glaseri* achieved the lowest LC_{50} values in controlling both young and grown larvae of *T. baal.* The EPNs showed better control potential against grown larvae than chemical pesticides. Further research is required to reveal the applicability of the tested EPNs species and strains to control the scarab beetle, *T. baal* under field conditions and their persistence in field as well. This study provides a broad understanding for the ability of EPNs in attacking larvae of the scrab beetles. Finally, our findings suggested that EPNs should be applied on a large scale, under field conditions, along with other biological control elements, such as botanical insecticides to cut down on chemical pesticide application.

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Table 1: List of entomopathogenic nematode genera, species, strain, entomopathogenic bacteria, Genbank accession number, and source.

EPN*	EPN species	Strain	Origin	EPB**	Accession No.	Source
Heterorhabditis	*H. bacteriophora*	BA1	Native	*Photorhabdus aegyptia*	MT355495	Mona A. Hussein
	H. bacteriophora	HP88	Foreign	*P. luminescence*	DSM 15139	H. Kaya
	H. bacteriophora	HBE	Native	*P. luminescence*	FJ755891	Mona A. Hussein
	H. marelatus	Mar	Foreign	*P. luminescence*	AY321479	R.-U. Ehlers
	H. bacteriophora	New***	Native	*P. luminescence*	PP446814	This study****
Steinernema	*S. carpocapsae*	BA2	Native	*Xenorhabdus nematohilus*	88772603	Mona A. Hussein
	S. carpocapsae	ALL		*X. nematophilus*	CM016762.1	R.-U. Ehlers
	S. glaseri	S.G.	Foreign	*X. poinarii*	40577	R. Georgis
	S. riobravae	Rio		*X. nematophilus*	AF331905	H. Kaya

*Entomopathogenic nematode genera ** Entomopathogenic bacteria associated with EPNs

***Field strain isolated from golf course.

****The strain (EGHP isolate) was identified at the Physiology Department, Plant Protection Research Institute, Agricultural Research Centre

Table 2: Efficiency of some different species of heterorhabtid nematodes on the young larvae of *T. baal* under laboratory conditions.

EPNs species	EPNs strain	Origin	Conc. IJs/ml	Mortality % after					Total mortality %	LC_{50}	LC_{90}	LT_{50}	LT_{90}
				D1	D2	D3	D4	D5					
H. bacteriophora	HP88	Exotic	4000	30.0	20.0	20.0	10.0	20.0	100a	194.7	2111	1.8	6.6
			2000	20.0	30.0	20.0	0.0	10.0	80b				
			1000	10.0	20.0	30.0	20.0	0.0	80b				
			500	0.0	20.0	10.0	30.0	10.0	70c				
	BA1	Native	4000	20.0	30.0	20.0	10.0	20.0	100a	44.25	777	2	5.66
			2000	20.0	20.0	30.0	10.0	0.0	80b				
			1000	10.0	20.0	20.0	10.0	20.0	80b				
			500	0.0	20.0	10.0	20.0	20.0	70c				
	HBE		4000	30.0	30.0	20.0	10.0	0.0	90a	154	5809	1.55	4.22
			2000	20.0	20.0	30.0	10.0	0.0	80ab				
			1000	0.0	0.0	30.0	10.0	10.0	70b				
			500	10.0	10.0	30.0	0.0	20.0	70c				
	NEW		4000	33.33	33.33	20.0	6.66	0.0	93.32	41.75	1633.71	1.4	3.50
			2000	13.33	6.6	53.0	13.32	6.66	92.87				
			1000	33.33	26.66	27.0	0.0	0.00	86.99				
			500	20.0	13.33	20.0	20.0	6.66	79.99				
H, marilatus	mar	Exotic	4000	40.0	30.0	30.0	0.0	0.0	100a	34.21	1573	1.25	3.33
			2000	20.0	10.0	30.0	20.0	10.0	90b				
			1000	30.0	0.0	40.0	10.0	10.0	90b				
			500	0.0	10.0	30.0	40.0	0.0	80c				

* Untreated control: all insects were alive

D1: Day 1

M.: Mortality

Main factors	F	P	LSD at 5%
Strain(s)	8.05029	.0001 ***	5.071219
Concentration (C)	35.45617	.0000 ***	4.535836
S*C	1.375162	.2178 ns	10.14244

Table 3: Efficiency of some different species of heterorhabtid nematodes on the grown larvae of *T. baal* under laboratory conditions.

EPNs species	EPNs strain	Origin	Conc. IJs/ml	Mortality % after					Total mortality %	LC_{50}	LC_{90}	LT_{50}	LT_{90}
				D1	D2	D3	D4	D5					
H. bacteriophora	HP88	Exotic	4000	10.0	10.0	10.0	20.0	40.0	90a	304.2	6992	4.6	18.81
			2000	0.0	0.0	20.0	40.0	10.0	70b				
			1000	10.0	0.0	10.0	40.0	10.0	70b				
			500	0.0	20.0	10.0	30.0	0.0	60c				
	BA1	Native	4000	10.0	30.0	20.0	10.0	10.0	80a	17074	27260	2.55	6.93
			2000	10.0	20.0	20.0	10.0	10.0	70b				
			1000	0.0	20.0	20.0	20.0	10.0	70b				
			500	0.0	10.0	10.0	20.0	20.0	60c				
	HBE		4000	10.0	20.0	40.0	10.0	10.0	90a	171	27260	2.4	5.13
			2000	10.0	30.0	10.0	20.0	0.0	70ab				
			1000	0.0	20.0	30.0	10.0	10.0	70ab				
			500	0.0	10.0	20.0	10.0	20.0	60b				
	NEW		4000	20.0	13.33	20.0	26.66	6.66	86.65	738.69	6458.88	2.36	6.6
			2000	6.66	6.66	20.0	20.0	13.33	66.65				
			1000	20.0	0.00	13.33	13.33	13.33	59.99				
			500	20.0	0.00	13.33	13.33	13.33	59.99				
H, marilatus	mar	Exotic	4000	0.0	30.0	20.0	50.0	0.0	100a	450	655	2.56	3.92
			2000	30.0	0.0	40.0	30.0	0.0	100b				
			1000	0.0	20.0	70.0	10.0	0.0	100b				
			500	20.0	10.0	30.0	0.0	0.0	60c				

* Untreated control: all insects were alive
D1: Day 1
M.: Mortality

Main factors	F	P	LSD at 5%
Strain(s)	21.7524	.0000 ***	5.500533
Concentration (C)	42.23746	.0000 ***	4.919826
S*C	3.800103	.0007 ***	11.00107

Table 4: Efficiency of some different species of steinernematid nematodes on the young larvae of *T. baal* under laboratory conditions.

EPNs species	EPNs strain	Origin	Conc. (IJs/ml)	Mortality % after					Total mortality %	LC_{50}	LC_{90}	LT_{50}	LT_{90}
				D1	D2	D3	D4	D5					
S, carpocapsae	All	Exotic	4000	30.0	10.0	30.0	10.0	10.0	90a	315	4543	1.9	6.0
			2000	20.0	0.0	30.0	20.0	10.0	80ab				
			1000	10.0	20.0	20.0	10.0	10.0	70bc				
			500	10.0	20.0	10.0	0.0	20.0	60c				
	BA2	Native	4000	20.0	40.0	20.0	10.0	10.0	100a	194	2111	1.72	4.1
			2000	20.0	30.0	30.0	10.0	0.0	90b				
			1000	10.0	20.0	20.0	20.0	10.0	80c				
			500	20.0	20.0	20.0	10.0	0.0	70d				
S. riobravae	Rio	Exotic	4000	20.0	20.0	30.0	20.0	10.0	100a	194	2111	2.0	4.84
			2000	20.0	10.0	10.0	20.0	20.0	80b				
			1000	10.0	20.0	20.0	20.0	10.0	80b				
			500	10.0	20.0	10.0	30.0	10.0	80b				
S. glaseri	S.G.	Exotic	4000	30.0	10.0	20.0	20.0	20.0	100a	38	2746	2.1	8.23
			2000	20.0	20.0	30.0	10.0	10.0	90b				
			1000	0.0	30.0	20.0	30.0	0.0	80c				
			500	10.0	10.0	30.0	20.0	10.0	80c				

* Untreated control: all insects were alive
D1: Day 1
M.: Mortality

Main factors	F	P	LSD at 5%
Strain(s)	6.555556	.0014 **	6.236809
Concentration I	25.22222	.0000 ***	6.236809
S*C	0.925926	.5162 ns	12.47362

Table 5: Efficiency of some different species of steinernematid nematodes on the grown larvae of *T. baal* under laboratory conditions.

EPNs species	EPNs strain	Origin	Conc. (IJs/ml)	Mortality % after					Total mortality %	LC_{50}	LC_{90}	LT_{50}	LT_{90}
				D1	D2	D3	D4	D5					
S, carpocapsae	All	Exotic	4000	20.0	20.0	10.0	10.0	20.0	80a	517	12605	2.64	10.32
			2000	20.0	30.0	10.0	0.0	10.0	70ab				
			1000	10.0	0.0	20.0	10.0	20.0	60bc				
			500	0.0	10.0	10.0	10.0	20.0	50c				
	BA2	Native	4000	20.0	20.0	40.0	10.0	0.0	90a	399	3514	1.92	4.34
			2000	10.0	20.0	20.0	30.0	10.0	90ab				
			1000	0.0	20.0	10.0	20.0	10.0	60b				
			500	0.0	10.0	30.0	10.0	10.0	60b				
S. riobravae	Rio	Exotic	4000	10.0	40.0	20.0	20.0	0.0	90a	315	4542	2.1	4.28
			2000	10.0	20.0	30.0	10.0	10.0	80b				
			1000	10.0	20.0	10.0	20.0	10.0	70c				
			500	0.0	10.0	20.0	20.0	10.0	60d				
S. glaseri	S.G.	Exotic	4000	20.0	10.0	10.0	50.0	10.0	100a	280	5798	2.51	4.36
			2000	0.0	10.0	50.0	0.0	20.0	80b				
			1000	10.0	30.0	0.0	10.0	20.0	70c				
			500	0.0	0.0	30.0	30.0	0.0	60d				

* Untreated control: all insects were alive
D1: Day 1
M.: Mortality

Main factors	F	P	LSD at 5%
Strain(s)	7.375	.0007 ***	5.88012
Concentration (C)	51.375	.0000 ***	5.88012
S*C	1.375	.2402 ns	11.76024

المكافحة الحيوية للنباشات *Temnorhynchus baal* (Coleoptera: Scarabaeidae) التي تصيب ملاعب الجولف المصرية باستخدام النيماتودا الممرضة للحشرات

عادل أنور الحلفاوى [1]، منى أحمد حسين [2,*]، حسن محمد صبحى خليفة [1]، حمدى أحمد محمد [1]

[1] قسم وقاية النبات، كلية الزراعة، جامعة الأزهر، القاهرة، مصر
[2] قسم آفات وقاية النبات، معهد البحوث الزراعية والبيولوجية، المركز القومى للحوث، القاهرة، مصر

[*] البريد الإليكتروني للباحث الرئيسي: ma.hussein@sci.nrc.eg

الملخص العربي:

الخلفية العلمية والهدف: هذه الدراسة هي الأولى التي تتحدث عن استخدام النيماتودا الممرضة للحشرات ضد نباشات *الجولف Temnorhynchus baal*، وهي آفة خطيرة تصيب ملاعب الجولف في مصر. وقد تم إدخال سياحة الجولف مؤخرًا إلى مصر، وتمثل هذه الرياضة إضافة قيمة إلى عائدات السياحة في البلاد، بما يتماشى مع الطلب العالمي المتزايد. وقد تم تسويق النيماتودا الممرضة للحشرات بنجاح كعوامل مكافحة بيولوجية للعديد من أنواع Scarabaeidae. الطرق والأدوات المستخدمة: تم استخدام أربعة تركيزات مختلفة من تسعة أنواع وسلالات مختلفة من النيماتودا الممرضة للحشرات ضد يرقات *T. baal* الصغيرة والبالغة في ظل ظروف المختبر. النتائج: كانت *Heterorhabditis marilatus* و*H. bacteriophora* فعالين بشكل ملحوظ في مكافحة يرقات *T. baal*. وعلاوة على ذلك، أظهرت النتائج أن *Steinernema glaseri* و*S. riobravae* و*S. carpocapsae* (BA2) كانت أكثر فعالية من *S. carpocapsae* (ALL) في السيطرة على يرقات *T. baal*. وقد أظهرت كل سلالة قدرتها على توفير معدل وفيات يزيد عن 90%، وتراوحت نسبة الوفيات لـ *S. glaseri* و80-100% و*S. riobravae* و60-90% لـ *S. carpocapsue* (All) ليرقات *T. baal* الصغيرة بينما تراوحت نسبة الوفيات بين 60-90% و50-80% لنفس السلالات، ليرقات *T. baal* البالغة. الاستنتاج: توفر هذه الدراسة فهمًا واسعًا لقدرة الأنواع المحلية والأجنبية المختارة من النيماتودا الممرضة للحشرات في مكافحة آفة خطيرة في ملاعب الجولف. ويمكن أن تلعب هذه النيماتودا دورًا واعدًا في السيطرة على آفات ملاعب الجولف بنجاح ويمكن دمجها في برنامج إدارة الآفات المتكاملة.

الكلمات الاسترشادية: آفات ملاعب الجولف؛ النباشات؛ هتيرورابديتس؛ شتينرنيما؛ المكافحة الحيوية.