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# Effectiveness of different entomopathogenic nematode species against the variegated cutworm, *Peridroma saucia* (Hubner) (Lepidoptera: Noctuidae)

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## Abstract

*Peridroma saucia* (Hubner) (Lepidoptera: Noctuidae) is a polyphagous pest that attacks nearly all vegetable fields in Turkey. Entomopathogenic nematodes (EPNs) are successfully used as a biological control agent. The efficacy of four EPNs, *Steinernema carpocapsae*, *S. feltiae*, *Heterorhabditis bacteriophora* and *H. indica* against the last instar larvae of the pest, was tested under laboratory conditions. Suspensions of nematodes were applied at four concentrations (10, 50, 100 and 200 infective juveniles/larva) at 25 ± 1 °C. Mortality percent was evaluated 48 and 96 h post application. Mortality rate increased with increasing concentrations. The highest effect caused 70% mortality by *H. bacteriophora* and *H. indica* species after 48 h exposure time and the lowest mortality was 33% for *Steinernema carpocapsae*. There was no significant difference between virulence of *Heterorhabditis* spp. and *Steinernema* spp. when tested against the larvae of *P. saucia* after 96 h exposure time. These results showed that EPNs have a significant potential in the biological control of *P. saucia* under controlled conditions.

**Keywords:** *Peridroma saucia*, Variegated cutworm, Entomopathogenic nematode, Biological control

## Background

The variegated cutworm (VCW), *Peridroma saucia* (Hubner, 1808) (Lepidoptera: Noctuidae), is a common polyphagous pest of many vegetable and field crops and found in many areas of the world (Rings et al., 1976 and Klein Koch and Waterhouse, 2000). VCW was first recorded in Europe in 1790 and then caused serious outbreaks in many countries throughout the Americas in 1841 (Capinera et al., 1988). The adults of these cutworms were discovered in 1967 in Turkey. At the present time, VCW is widely distributed in Turkey's agricultural areas and it is one of the most abundant and damaging cutworm species of Turkey (Akdagcik and Ulusoy, 2007). VCW has a wide range of host plants which includes economic crops such as potato, tomato, corn, lettuce, carrot and sugar beet. VCW larvae do much damage to crops and cause considerable mortality to seedlings in the early growing season by cutting off the plant at the soil surface and feeding on the

foliage of these crops (Capinera et al., 1988). Due to the high tolerance of VCW, repeated applications of conventional insecticides are widely used. Development of resistance and concerns over the destructive effects of chemicals to environmental and human safety have accelerated the development of alternative control methods for this pest (Yoshida, 2010).

Entomopathogenic nematodes (EPNs) are highly effective biological control agents against many agricultural pests particularly soil-inhabiting lepidopterous larvae because of their presence in larval stages below ground (Vashisth et al., 2013). EPNs have searching ability on hosts and the potential to survive in the soil environment. They possess free-living third-stage infective juvenile (IJ) that can survive a long time without feeding (Koppenhöfer et al., 2000). The IJs invade their hosts via natural body openings, such as the mouth, the anus and the spiracles. Once they enter to haemocoel, the mutualistic bacteria *Xenorhabdus* in *Steinernema* and *Photorhabdus* in *Heterorhabditis* are released to kill the host within 2 days (Gaugler, 2002; Griffin, et al., 2005; Kaya et al., 2006).

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EPNs that infect insects have received considerable attention by scientists for their potential in the biological management of many agricultural pests (Gaugler, 1981; Gaugler and Kaya, 1990; Georgis et al., 2006; Koppenhöfer et al., 2000; Smart Jr, 1995). Many studies have been started for testing the pathogenicity of these indigenous EPN species all over the world (Ozer et al., 1995; Kepenekci, 2002; Hazir et al., 2003; Unlu et al., 2007 and Erbas et al., 2013). There are some differences known in terms of survival, pathogenicity and host range between indigenous and non-indigenous EPN species (Lacey and Georgis, 2012). Indigenous species of EPNs may be more successful in biocontrol as a consequence of compatibility to native habitats (Goudarzi et al., 2015).

The aim of this study is to evaluate the effect of four Turkish species of EPNs for the biological control of the variegated cutworm (VCW), *P. saucia*, under laboratory conditions.

## Materials and methods

### Insect culture

Healthy VCW larvae were collected on a regular basis from different vegetable fields throughout the vicinity of Central Anatolia and Mediterranean Region, Turkey. For the experiment, larvae were reared on lettuce. *P. saucia* was established in a growth chamber at a temperature of  $25 \pm 1$  °C, relative humidity of 60% and a photoperiod of 16:8 (L:D) (Scott-Dupree et al., 2008). Healthy last instar larvae were selected to be used for testing the virulent effects of the nematodes in a dose-response experiment.

### Entomopathogenic nematodes

Four indigenous nematode isolates of *Heterorhabditis bacteriophora* FLH-3-H, *H. indica* 216-H, *Steinernema feltiae* Y29-S and *S. carpocapsae* E76-S were collected during the surveys conducted in Adana and Kahramanmaraş provinces in the Mediterranean region of Turkey.

### Pathogenicity test

Nematode pathogenicity was tested against last instar larvae of *P. saucia* in a Petri dish arena. In order to avoid cannibalism, only one last instar larva was placed on two moist filter papers in each  $100 \times 15$  mm Petri dish inoculated with 1 ml of water containing different concentrations of IJs (10, 50, 100 and 200 IJs/larva). Each nematode concentration was tested against ten *P. saucia* larvae and replicated for three times. Control plates were treated with distilled water only. Petri dishes were kept at  $25 \pm 1$  °C. The number of dead larvae was recorded at two different exposure times (48 and 96 h) and dead insects were dissected to determine whether nematodes were present or not.

### Statistical analysis

Data was evaluated without being regulated by the Abbott formula because there was no mortality in control plates (Abbott, 1925). Statistical analyses were carried out by SAS software (Version 9.1.3; SAS Institute, Cary, NC (1990)). The experiment was established in a completely randomized design with a factorial treatment arrangement consisting of four nematode species and four application rates. Mean values were separated using Tukey Multiple Range Test ( $P < 0.05$ ).

## Results and discussion

The virulence of four indigenous nematode species against last larval instar of *P. saucia* was evaluated in a laboratory experiment. Results revealed that all EPN species had the ability to kill and reproduce in *P. saucia* larvae by sickening them. All EPN species tested and application concentrations significantly affected the mortality rates ( $F: 27.85$ ,  $df: 3$ ,  $P < 0.001$  for EPN species and  $F: 18.12$ ,  $df: 3$ ,  $P < 0.001$  for application of the tested concentrations after 48 h;  $F: 5.14$ ,  $df: 3$ ,  $P < 0.05$  for EPN species and  $F: 1.30$ ,  $df: 3$ ,  $P < 0.05$  for application of concentrations after 96 h). No significant differences were noted statistically in mortality rates caused by the concentrations-nematode interaction.

Mortality rates have increased generally with increasing concentrations. *H. bacteriophora* and *H. indica* strains showed higher effects at all application concentrations than *S. feltiae* and *S. carpocapsae* strains at the first exposure time (48 h) except *H. indica* and *S. carpocapsae* at 10 IJs/larva concentrations. The highest larval mortality was achieved when EPNs were applied at the concentrations of 200 IJs/larva after 48 h (Table 1). However, after the second exposure time (96 h), generally no differences were found among strains except 10 IJs/larva concentrations.

All EPN species tested showed a great mortality at the lowest concentration of 10 IJs/larva after 96 h of exposure time and at least 80% of *P. saucia* were killed by *S. feltiae* Y29-S strain. The difference in mortality between *H. bacteriophora* FLH-3-H (100%) and *S. feltiae* Y29-S (80%) was statistically significant. At the exposure time of 48 h, the highest mortality rate was induced by *H. indica* isolate, while the lowest one was determined by *S. carpocapsae* isolate at the concentrations of 10 and 50 IJs/larva. Only the mortality produced by *S. carpocapsae* E76-S was different than the one by *H. indica* 216-H significantly at the concentration of 10 IJs/larva, but at 50 IJs/larva concentrations, they were divided into two groups which statistically differed when compared to each other. The interaction effect of the different entomopathogenic species and the concentrations on

**Table 1** The effect of different concentrations of EPNs on the mortality of last instar larvae of *P. saucia* for 48 and 96 h post application at 25 ± 1 °C

EPNs	Mortality rates of <i>P. saucia</i> larvae <sup>a</sup>							
	10 <sup>b</sup>		50 <sup>b</sup>		100 <sup>b</sup>		200 <sup>b</sup>	
	48 h	96 h	48 h	96 h	48 h	96 h	48 h	96 h
H.b.FLH-3-H	26.66ab	100.00a	66.66a	100.00a	86.66a	100.00a	100.00a	100.00a
S.f. Y29-S	20.00ab	80.00b	26.66b	100.00a	33.33b	100.00a	66.66b	100.00a
S.c. E76-S	13.33b	93.33ab	13.33b	100.00a	40.00b	100.00a	66.66b	100.00a
H. i. 216-H	33.33a	93.33ab	73.33a	100.00a	73.33a	100.00a	100.00a	100.00a

H.b.FLH-3-H, *Heterorhabditis bacteriophora* FLH-3-H isolate; S.f. Y29-S, *Steinerinema feltiae* Y29-S isolate; S.c. E76-S, *S. carpocapsae* E76-S isolate; H. i. 216-H, *H. indica* 216-H isolate

<sup>a</sup>Mean values followed by different lowercase letters in the same column are significantly different according to Tukey's test ( $P < 0.05$ )

<sup>b</sup>The number of infective juveniles for each larva

larval infection were not significant after exposure for 96 h at 50, 100 and 200 IJs/larva. The most virulent species was *H. bacteriophora* at the lowest IJ concentration of 100% mortality after 96 h of exposure time followed by *S. carpocapsae* and *H. indica* with 93.3% mortality (Table 1).

*S. carpocapsae* and *S. feltiae* caused equal mortality in the last instar larvae which was less virulent than *H. bacteriophora* and *H. indica*, at a concentration of 200 IJs/larva over the exposure time of 48 h (Table 1).

In a laboratory experiment, *Heterorhabditis* spp. proved to be more efficient in suppressing *P. saucia*. Increase in the time of exposure caused more mortality to last instar *P. saucia* larvae by all the tested nematode species (Table 1). Similar studies have been carried out to evaluate the effectiveness of EPNs against *P. saucia* larvae where it was revealed that EPNs have great potential for the management of *P. saucia* larvae (Yoshida, 2010; Morris and Converse, 2012). The pathogenicity of steirnematid and heterorhabditid nematode species against *P. saucia* larvae was variable in previous studies. Morris and Converse (2012) exposed *P. saucia* larvae to six strains of steirnematid and two species of heterorhabditid nematodes in the soil surface. *S. feltiae* was found as the most virulent nematodes while the heterorhabditid nematodes were the best performing species in our study. The pathogenicity of 17 Japanese isolates of EPNs was tested against the middle instar larvae of *P. saucia* in a laboratory experiment at different temperatures. Three isolates belonging to *S. feltiae* caused 70% average mortality at 25 °C which is similar to our study (Yoshida, 2010).

## Conclusions

Under laboratory conditions in the petri dish experiment, the heterorhabditid nematodes proved a more effective control of *P. saucia* larvae and increase in time of exposure led to more mortality of the tested nematode species. More studies are needed to determine the real

potential of these four indigenous EPNs both in the open field and greenhouse environments to be included in biological control programs of *P. saucia*.

## Authors' contributions

EY carried out the laboratory studies and drafted the manuscript. All authors read and approved the final manuscript.

## Competing interests

The authors declare that they have no competing interests.

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