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Field Control of *Synanthedon myopaeformis* Borkh and *Zeuzera pyrina* L. Using Entomopathogenic Nematodes, Insecticides and Microbial Agents

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ABSTRACT

Three field trials were carried out to evaluate the efficacy of two local strains of entomopathogenic nematode, *Heterorhabditis bacteriophora* (Ar-4 and Ht strains) isolated from EL-Arish and Giza in Egypt and two commercially available nematode species, *Steinernema carpocapsae* (All strain) and *H. bacteriophora* (HP88 strain) for the control of fruit tree borers, *Synanthedon myopaeformis* and *Zeuzera pyrina* on apple trees. The entomopathogenic nematodes (EPNs) were used alone or combined with recommended dose of the microbial pesticide, *Bacillus thuringiensis* (Diple2X), and insecticides, diazinon and phenthoate. The obtained results showed that the combined EPNs with insecticides were more effective as injection technique than spray method. Mortality percentages of *S. myopaeformis* ranged between 47.61 to 88.88 % in the injection treatments while in the spraying one it ranged between 37.14 to 74.07 %. In comparison between the efficacy of native and imported entomopathogenic nematodes in controlling *Z. pyrina* using spray technique, percentage mortality resulted from the combination of native nematodes i.e. *H.bacteriophora* (Ht strain) and *H. bacteriophora* (Ar-4 strain) and insecticides either penthoate or diazinon reached 69.23; 64.00 and 22.72; 64.18%, respectively. Whereas, percentages mortality were 44.44; 50.00 and 52.94; 54.16% with *S. carpocapsae* (All strain) and*H. bacteriophora* (HP88 strain), respectively. The susceptibility of *Z. pyrina* to the nematode species differed greatly according to the nematode species used either alone or in combination with the recommended dose of *B. thuringiensis*. The entomopathogenic nematode *H. bacteriophora* (HP88 strain) was more virulent than *S. carpocapsae* (All strain) in controlling *Z. pyrina* with percentage mortality reached 77.5 and 62.1%, respectively.

Key words: *Synanthedon myopaeformis,* Pathogenicity, *Zeuzera pyrina*, *Steinernema, Heterorhabditis,* Compatibility.

INTRODUCTION

Borer insects (*Synanthedon myopaeformis* Borkh and *Zeuzera pyrina* L.) are most important pests since they attack a large number of shrubs and deciduous tree species including apple, pear, peach, cherry, olive as well as walnut trees in Egypt (Abdel- Kawy et al.,1988). Control of the leopard moth has many difficulties, since the larvae escape from direct exposure to pesticides because they live inside branches and trunks (Shamseldean et al., 2009). Chemical pesticides are widely used to combat pests. However, increasing awareness of environmental and human health associated with chemical pesticides are forcing scientists to search for less toxic pest management methods.

Finding safer alternatives to chemical pesticides is especially urgent for borer insects (*S. myopaeformis* and *Z. pyrina*) of fruit trees. In the cryptic habitats, biological control agents especially, the genera *Steinernema* and *Heterorhabditis* of entomopathogenic nematodes (EPNs) could be considered appropriate candidates against these pests. Since, *Z. pyrina* larvae were susceptible to infection by *S. carpocapsae* and *H. bacteriophora* under laboratory and field conditions (Abdel-Kawy and El-Bishry, 1992). Using low impact pesticides or other bioagents like *Bacillus thuringiensis* reduced rates of pesticides and could achieve adequate control through minimizing the adverse effects of pesticides (Mannion et al ., 2000).

The objective of the present study was to determine the relationship between application methods of native and commercial entomopathogenic nematode, diazinon and phenthoate as insecticides as well as *B. thuringiensis* either alone or mixed in controlling the clear-wing moth, *S. myopaeformis* and the leopard moth, *Z. pyrina* on apple trees.

MATERIALS AND METHODS

1-Rearing Technique:

a- Rearing of the greater wax moth, *Galleria mellonella*L.

Last instar larvae of *G. mellonela* (Lepidoptera: Pyralidae) were separated from bee hives for nematode culture, leaving small sized larvae for moth emergence and egg laying. Fresh laid eggs were transferred to modified artificial medium contained honey, wheat bran, glycerol, soy flour, milk powder, dry yeast, and honey bee wax in glass jars kept at 27 ºC.(Ekmenet al., 2010).After reaching last instars, they were taken out from the diet and used for storage and nematode isolation/multiplication according to Kaya and Stock (1997).

Heterorhabditis bacteriophora (HP88 strain), *Steinernema carpocapsae* (All strain) and two local strains isolated from EL-Arish and Giza, Egypt. *H. bacteriophora* (Ar-4) and *H. bacteriophora* (Ht), respectively were multiplied and harvested from greater wax moth larvae (Woodring and Kaya, 1988) and infective juveniles of these nematodes were washed in three changes of distilled water (Dutky et al.,1964). Nematodes were stored at13°C. for up to two weeks prior to use in experiments. All experiments were conducted at 25 ± 4 °C.

b-Isolation of native entomopathogenic nematodes:

Baiting technique of *G. mellonella* modified after Bedding and Akhurst (1975) was followed. Soil samples infested with nematodes were placed in plastic bags and transferred to the laboratory. From each bag a soil sample was thoroughly mixed and then divided into 4 portions. Fifty grams from each were put in 14-cm Petri-dish. Five *G. mellonella* late instar larvae, were placed onto the soil surface and the dishes were kept at 25°C. As mature wax larvae tended to move upward the Petri-dishes were turned upside down every 24 hours, so that the confined larvae were also allowed to pass through the soil in order to increase the possibility of their encounter with the nematodes. Dishes were examined 5 days later, and the dead larvae, which were suspected to be nematode-infected, were picked up carefully, rinsed several times with distilled water and incubated at 25°C.in an extraction plates. Each plate contained only one dead larva. The original method described by Dutky et al. (1964) was used with some modifications, where the dead larvae were placed on a special fabric muslin cloth to avoid the disintegration of the filter papers. Nematode juveniles were maintained in 0.1% formaldehyde until identification. Two isolates designated as *H.bacteriophora* (Ar-4 strain) from EL-Arish and *H. bacteriophora* (Ht strain) from Giza were exracted from soil samples.

c-The biocontrol agent *Bacillus thuringiensis*:

The Diple 2X *Bacillus thuringiensis* var.*kurstaki*; 32000 IU/ mg was used in the present study with the recommended dose of 1g/liter. The mixtures of the nematodes and the bacterium were performed by mixing the nematode strains of *H.bacteriophora* (HP88 strain), *S. carpocapsae* (All strain) and *H.bacteriophora* (Ht strain)(2000 IJs/ml for each) with the recommended dose of the bacterium.

2- Field Experiments:

Field Experiments were carried out in four different farms during three consecutive years of 2014, 2015 and 2016 to determine the efficiency of certain native and imported strains of entomopathogenic nematodes with two application methods against two species of fruit tree borers, the leopard moth *Zeuzera pyrina* and the clear-wing moth *Synanthedon myopaeformis* on apple trees. Also, these strains of nematodes were used at two concentration (1500 and 3000 IJs), separately or 1000 IJs when combined with two insecticides (Diazinon and Phenthoate; 3ml/liter water) or the microbial insecticide *B. thuringiensis* (Diple2X at concentration of 32000 IU/mg for evaluating the efficacy of these combinations against these borers. The field experiments were conducted as follows:

A-The first experiment took place in two districts, EL-Mansouria locality in Monofyia Governorate and EL-Marwa& EL-Zeitoun farms in Giza Governorate. Heavily *S. myopaeformis* infested apple trees were sprayed or injected by water suspensions of two strains of imported entomopathogenic nematodes, *S. carpocapsae* (All), *H. bacteriophora* (HP88) and two local strains isolated from EL-Arish and Giza in Egypt, *H.bacteriophora* (Ar-4) and *H. bacteriophora* (Ht) at two concentrations of each (approximately 1500 and 3000 IJs/ml distilled water). Also, combinations of entomopathogenic nematodes and either Diazinon (basudin) or Phenthoate (cedial L60) at a concentration of 3ml/liter of water were undertaken on infested apple trees. For spray and injection techniques, one concentration of nematodes (1000IJs/ml) was used spraying technique using a handle sprayer of 2.5 liter was directed towards the active galleries, whereas the injection was carried out using 50cc syringe.

B-The second experiment was carried out in both previous areas on heavily *Z. pyrina* infested apple trees. The infested apple trees were sprayed only by 1500 IJs water suspensions of two strains of imported entomopathogenic nematodes, *S. carpocapsae* (All) , *H. bacteriophora* (HP88) and two native strains, *H.bacteriophora* (Ar-4) and *H. bacteriophora* (Ht) at concentrations of 3000 IJs/ml distilled water only. Also, combinations of abovementioned nematodes and either diazinon or phenthoate at a concentration of 3ml/liter of water were undertaken on infested apple trees.

C-The third experiment was conducted at EL-Marwa & EL-Zeitoun farms in Giza Governorate on apple trees that were heavily infested with *Z. pyrina.* In such independent experiment, *S. carpocapsae* (All strain), *H.bacteriophora* (HP88) and *H.bacteriophora* (Ht) in combinations with *B. thuringiensis* (Diple 2X), were evaluated by spray technique against *Z. pyrina* at the beginning of May 2013. The same treatments were repeated in the preultimate week of November 2014.

For all experiments the check or control trees, were treated with water and all treatments were carried out before sunset to avoid excessive evaporation and direct sunlight (UV).The treatments were replicated 3 times, since injection technique was conducted only once but spraying was repeated 3 times with one week interval.The treated trees were weekly inspected after application, where the galleries that had new exploded frasses, were considered active beside the actively moving larvae. Also, some randomly galleries in the trunk and main branches, were inspected by using a sharp knife and (about 50 cm long) a flexible wire to about 50 cm long. Mortality percentages were calculated according the following formula:

$$\frac{\text{No. of active galleries before treatment - No. of active galleries after treatment}}{\text{No. of active galleries before treatment}} \times 100$$

3- Statistical analysis:

The experiments were carried out in a completely randomized design with 3 replications for each treatment. Data were subjected to analysis of variance (ANOVA) using MSTAT version 4 (1987). probability. Means were compared by Duncan's multiple range test (Duncan, 1955) at $P \leq 0.05$

RESULTS AND DISCUSSION

Data in Table (1) showed the impact of entomopathogenic nematodes viz. *S. carpocapsae* (All strain), *H. bacteriophora* (HP88 strain), *H. bacteriophora* (Ar-4 strain) and *H. bacteriophora* (Ht strain) alone or in combination with insecticides either diazinon or phenthoate on *S. myopaeformis* infesting apple trees. Two different methods of nematode application were used (spray and injection). It was found that combination of *S. carpocapsae* and diazinon gave the highest effect with average mortality of 72.17 % followed by *S. carpocapsae* and phenthoate with average 64.70 % whereas; *S. carpocapsae* spray alone had the lowest effect with average mortality 50.55%. Injection technique proved to be more effective than spray, *S. carpocapsae* and phenthoate gave the highest effect followed by *S. carpocapsae* and diazinon with percentage mortality of 76.47 and 74.07 %, respectively. Concerning heterorhabditid nematodes, *H. bacteriophora* (HP88 strain), *H. bacteriophora* (Ar-4 strain) and *H. bacteriophora* (Ht strain) used to control larvae of *S. myopaeformis* on apple trees, mortalities showed that *H. bacteriophora* (HP88 strain) alone or in combination with the used insecticides were more effective when compared with *H.bacteriophora* (Ar-4 strain) and *H. bacteriophora* (Ht strain) alone or with insecticides, diazinon and phenthoate. Average percentages mortality for application of *H.bacteriophora* (HP88 strain) were 46.02 and 46.76 when used alone as spray and injection techniques, respectively. In combination between *H.bacteriophora* (HP88 strain) and insecticides, average percentages mortality were 65.66 and 74.69 with diazinon and phenthoate, respectively. Whereas the parallel values with *H. bacteriophora* (Ar-4 strain) were 36.23,43.66 when used alone by spray and injection techniques, respectively and 65.17; 51.57 in combination with the two insecticides, respectively. As injection treatments, among the four nematode strains, *S. carpocapsae* (All strain) was the most effective followed by *H.bacteriophora* (Ht strain), *H. bacteriophora* (HP88 strain) and *H. bacteriophora* (Ar-4 strain) with average mortalities 56.47, 54.65, 46.76 and 43.66, respectively. Whereas the two insecticides caused 67.65 and 41.15 average mortality and phenthoate was the least effective insecticide.

Table 1. Comparison of entomopathogenic nematodes and insecticides applied with two different methods for controlling *Synanthedon myopaeformis* Borkh on apple trees.

Treatments	Applicatin Method	Conc.	No. of active galleries		% Mortality	Average
			Before treatment	After treatment		
S. carpocapsae (All strain)	Spray	3000	15 d*	9 bc	40.00	50.55abc
		1500	21 cd	8 bc	61.90	
S. carpocapsae (All strain)	Injection	3000	17 d	8 bc	52.94	56.47abc
		1500	15 d	6 cd	60.00	
S. carpocapsae (All strain) +Diazinon	Spray	1000	37 a	11 b	70.27	72.17ab
	Injection	1000	27 bc	7 cd	74.07	
S. carpocapsae (All strain)+Phenthoate	Spray	1000	34 ab	16 a	52.94	64.70 ab
	Injection	1000	17 d	4 d	76.47	
H.bacteriophora (HP88 strain)	Spray	3000	34 c	18 a	47.05	46.02 cd
		1500	20 d	11 c	45.00	
H.bacteriophora (HP88 strain)	Injection	3000	38 b	18 a	52.63	46.76cd
		1500	22 d	13 b	40.90	
H.bacteriophora (HP88 strain) +Diazinon	Spray	1000	37 b	15 b	59.45	65.66 ab
	Injection	1000	32 c	9 c	71.87	
H.bacteriophora (HP88 strain) +Phenthoate	Spray	1000	44 a	13 b	70.45	74.69 a
	Injection	1000	19 d	4 d	78.94	
H.bacteriophora (Ar-4 strain)	Spray	3000	22 e	13 bc	40.90	36.23 d
		1500	19 f	13 bc	31.57	
H.bacteriophora (Ar-4 strain)	Injection	3000	17 f	10 d	41.17	43.66 c
		1500	26 d	14 bc	46.15	
H.bacteriophora (Ar-4 strain) +Diazinon	Spray	1000	41a	24 a	41.46	65.17 a
	Injection	1000	18 f	2 bc	88.88	
H.bacteriophora (Ar-4 strain) + Phenthoate	Spray	1000	35 b	17 b	51.42	51.57 b
	Injection	1000	29 c	14 bc	51.72	
H.bacteriophora (Ht strain)	Spray	3000	23cd	9 d	60.68	47.09bc
		1500	33 b	22 a	33.33	
H.bacteriophora (Ht strain)	Injection	3000	19 d	7 d	63.15	54.65 a
		1500	26 c	14 bc	46.15	
H.bacteriophora (Ht strain)+ Diazinon	Spray	1000	35 b	22 a	37.14	44.88bcd
	Injection	1000	38 a	18 b	52.63	
H.bacteriophora (Ht strain)+ Phenthoate	Spray	1000	26 c	16 bc	38.46	43.03bcd
	Injection	1000	21cd	11 d	47.61	
Diazinon (3ml/liter)	Spray	3 ml/ l	36 a	14 b	61.11	67.65 ab
	Injection	3 ml/l	31 c	8 d	74.19	
Phenthoate (3ml/liter)	Spray	3 ml/l	25 d	12 c	52.00	41.15 bc
	Injection	3 ml/l	33 b	23 a	30.30	

*Each value is a mean of three replicates.
The same letter (s) in columns indicates no significant differences at $P \leq 0.05$ according to Duncan's multiple range test.

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Data in Table (2) revealed that applications of heterorhabditid nematodes, *H. bacteriophora* (HP88 strain), *H. bacteriophora* (Ht strain) and *H. bacteriophora* (Ar-4 strain) alone were more effective when compared with insecticides, penthoate and diazinon with percentage mortalities in *Z. pyrina* reached 60.86 , 38.46 , 66.67, 27.27 and 34.37, consequently. *S. carpocapsae* (All strain) was the least effective against larvae of *Z. pyrina* when used alone with percentage mortality amounted to 23.07%.

In dual application larval mortality reached 44.44 & 50.00; 52.94 & 54.16 and 69.23 & 64.00 with *S. carpocapsae* (All strain), *H. bacteriophora* (HP88 strain) and *H. bacteriophora* (Ht strain) with penthoate and diazinon, respectively. The native nematode, *H. bacteriophora* (Ar-4 strain) and penthoate decreased larval mortality by 22.72 %. In general it could be stated that, integrating entomopathogenic nematodes with insecticides yielded high percentage of mortalities in *Z. pyrina* than using nematodes or insecticides alone.

Table 2. Comparison between native and imported entomopathogenic nematodes applied using spray method alone or in combination with certain insecticides for controlling *Zeuzera pyrina* L. on apple trees.

Treatments	Active galleries before treatment	Active galleries after treatment	% Mortality
S.carpocapsae (All strain)	13 c	10 bc	23.07 e
S.carpocapsae (All strain) +Diazinon	16 c	8 c	50.00 bc
S.carpocapsae (All strain) +Penthoate	18 c	10 bc	44.44 c
H.bacteriophora (HP88 strain)	23 bc	9 c	60.86 ab
H.bacteriophora (HP88 strain)+Diazinon	24 bc	11 bc	54.16 bc
H.bacteriophora (HP88strain) +Penthoate	17 c	8 c	52.94 a
H. bacteriophora (Ar-4 strain)	12 c	4 d	66.67 a
H. bacteriophora (Ar-4 strain)+ Diazinon	54 a	19 a	64.18 a
H. bacteriophora (Ar-4 strain)+Penthoate	22 bc	17 ab	22.72 e
H. bacteriophora (Ht strain)	26 bc	16 a	38.46 cd
H.bacteriophora (Ht strain)+ Diazinon	25 bc	9 c	64.00 a
H.bacteriophora (Ht strain)+ Penthoate	26 bc	8 c	69.23 a
Penthoate	11 c	8 c	27.20 e
Diazinon	32 b	21 a	34.37 d

*Each value is a mean of three replicates.
The same letter (s) in columns indicate no significant differences at $P \leq 0.05$ according to Duncan's multiple range test.

Data presented in Table (3) indicate that the degree of *Z. pyrina* susceptibility to the nematode species differed greatly according to the nematode species used either alone or in combination with the recommended dose of *B. thuringiensis* in autumn and spring. In spring (26.1°C) it was evident that, nematode species of *H. bacteriophora* (HP88) was more virulent than *S. carpocapsae* (All strain) to *Z. pyrina* with percentage mortality 77.5 and 62.1, respectively. Nematode species were more virulent to *Z.pyrina* than *B. thuringiensis*. It was also evident that using entomopathogenic nematode strains against *Z. pyrina* individually resulted in higher percentages of mortality than the combinations of nematodes and bacterium and /or the bacterium alone except in the case of *S. carpocapsae* with *B. thuringiensis* in spring. On the other hand, the strains of *H. bacteriophora* were more effective in high degrees of temperatures. Data revealed that there was no any synergistic or additive interaction between the nematodes and the bacterium except in the case of *S. carpocapsae*. In spring, no significant differences were found between *H.bacteriophora* strains (HP88 &Ht), although some degree of significance was found between the nematode strains and the bacterium.

Table 3. Infectivity of three entomopathogenic nematodes alone or combined with *Bacillus thuringensis* on *Zeuzera pyrina* L.in two different seasons.

Treatments	% Mortality			
	Spring (26.1 °C)		Autumn (20.9 °C)	
	Nematode or Bt alone	Nematode +Bt	Nematode or Bt alone	Nematode +Bt
S. carpocapsae (All strain)	62.1 b*	73.4 a	79.8 a	69.1 a
H.bacteriophora(HP88 strain)	77.5 a	62.2 b	73.5 b	51.7 b
H.bacteriophora(Ht strain)	76.8 a	58.3 b	62.6 c	55.4 b
B. thuringensis	30.3 c	-	10.0 d	-

*Each value is a mean of three replicates.
The same letter in columns indicates no significant differences at $P \geq 0.05$ according to Duncan's multiple range test.

Our previous results indicate that curative applications of entomopathogenic nematodes, *S. carpocapsae* or *H. bacteriophora* to reduce populations of *S. myopaeformis* and *Z. pyrina*, caused significant mortality and suppressed damage when applied alone or in combination with chemical and biopesticides under field conditions. Such curative treatments with recommended insecticides and EPNs may contribute to protectapple trees and reducing subsequent populations and substantial damage from larval feeding occurred in the spring and autumn. El-Kholy et al. (2014) reported that application of entomopathogenic nematodes in controlling larvae or pupae of *Z pyrina* infested olive branches either via spraying or injection caused significant control inside the branches. By comparing nematodes and recommended insecticides for controlling *S. myopaeformis* on apple trees, our findings agree with that of Bedding and Miller (1981); Deseo et al. (1984a); Deseo and Docci (1985); Solomon (1985); Kaya and Brown (1986); Abdel-Kawy et al. (1988); Yanget al.(1990); Abdel-Kawy & EL-Bishry (1992); Deborah et al. (1996); Shamseldean (2000) and Shamseldean et al. (2009). Rovesti et al. (1988) approved that the greater concentration of pesticide might prevent the compatibility use of the chemical with *H. bacteriophora* nematode. Shapiro-Ilan et al. (2009) indicated that nematodes applied in control of the peach tree borer, *S.exitiosa* reduced damage to levels similar to what is achieved with recommended chemical insecticide treatments. Bari and Kaya (1984); Deseo et al. (1984b); EL-Bishry & Bekheit (1994); Gill &Michael (1994); Azazy (1996) and Pasqualini et al. (1996) indicated that combination between entomopathogenic nematodes and recommended dose of *B. thuringiensis* did not result insignificantly greater control than that achieved by the nematode alone. Due to the cost of product and in some cases a change in the environment, microbial control agents are usually used to achieve economic pest control.

Our data revealed that infective juveniles (IJs) of native EPN isolates were more sensitive to tested pesticides than commercially available nematode species and *H. bacteriophora* (HP88) was the most tolerant treatment as compared to *H. bacteriophora* (Ar-4 strain) and *Steinernema carpocapsae* (All strain). Likewise, similar findings were reported by Radova (2011) and Laznik &Trdan (2013).Grewal et al. (1998) suggested that incompatible nematicides and insecticides can be used by wait 2 and 1 weeks after application, respectively.

In conclusion, effectiveness of entomopathogenic nematodes varies depending upon the application method and concentration. However, further studies are needed under field conditions to evaluate new application methods and better compatibility of entomopathogenic nematodes with other chemicals or biopesticides may be considered as a strong option for usage in IPM.

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الملخص العربي

المكافحة الحقلية لكل من حفار ساق الحلويات رائقة الأجنح Borkh *Synanthedo myopaeformis* وحفار ساق التفاح .*Zeuzera pyrina* L باستخدام النيماتودا الممرضة للحشرات والمبيدات الحشرية والعوامل الميكروبية.

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أجريت ثلاث تجارب حقلية لتقييم كفاءة سلالتين محلتين من النيماتودا الممرضة للحشرات المعزولتين من العريش والجيزة فى مصر هما (Ar-4&Ht)والتابعتين للنوع *H. bacteriophora* ونوعين من النيماتودا المتوفرة بشكل تجارى هما(All strain) *Steinernema carpocapsae* و *Heterorhabditis bacteriophora* (HP88 strain) لمكافحة يرقات اثنين من ناخرات أخشاب أشجار الفاكهة هماحفار ساق التفاح .Zeuzera pyrina L. وحفار ساق الحلويات رائقة الأجنحة *Synanthedon myopaeformis* Borkh على أشجار التفاح. استخدمت النيماتودا الممرضة للحشرات منفردة أو مخلوطة مع التركيز الموصى به لكل من المبيد الميكروبى (Diple2X) *Bacillus thuringiensis* والمبيدين الحشريين diazinon وphenthoate. أوضحت النتائج المتحصل عليها أن النيماتودا الممرضة للحشرات المخلوطة مع المبيدات الحشرية والمستخدمة بطريقة الحقن كانت أكثر فاعلية مقارنة بطريقة الرش. وتراوحت نسبة الموت في يرقات*S.myopaeformis* أما بين ٤٧.٦١ إلى ٨٨.٨٨ % في طريقة الحقن بينما تراوحت هذه النسبة بين ٣٧.١٤ إلى ٧٤.٠٧ % في تقنية الرش.

وعند مقارنة فعالية النيماتودا الممرضة للحشرات المعزولة محليا وبين تلك الأنواع المستوردة والتي استخدمت فى مكافحة.*Z. pyrina* L بتقنية الرش ، كانت نسبة الموت الناتجة من خلط النيماتودا المحلية *H.bacteriophora* (Ht strain) و(Ar-4 strain) *H. bacteriophora* مع المبيدين الحشريين Diazinon وPenthoate هي (٦٩.٢٣ &٦٤.٠٠%) و (٢٢.٧٢ & ٦٤.١٨ %) على التوالي . بينما كانت نسبة الموت مع نوعى النيماتودا المستوردة (All strain) *S. carpocapsae* و *H. bacteriophora* (HP88 strain) هي (٤٤.٤٤ & ٥٠.٠٠ %) و (٥٢.٩٤ &٥٤.١٦ %) على الترتيب.

اختلفت قابلية يرقات *Z. pyrina* للإصابة تبعا لنوع النيماتودا المستخدمة إما منفردة أو بالخلط مع التركيز الموصى به من بكتيريا *B. thuringiensis* وكانت نيماتودا (HP88 strain) *H. bacteriophora* أكثر تأثيرا من نيماتودا(All strain) *S. carpocapsae* في قتل يرقات*Z. pyrina* وأحدثت نسبة موت قدرها ٧٧.٥ و ٦٢.١ على التوالي.