

# **RESULTS AND DISCUSSION**

## RESULTS AND DISCUSSION

### I- Laboratory studies:

#### 1-Effect of combination of pathogenic nematodes and nematicides on some stages of *Spodopetra littoralis* and *Agrotis ipsilon*:

These series of experiments were carried out in the laboratory, in order to study the effect of combination of the nematode strains *Heterorhabditis bacteriophora* (HP88), *H. bacteriophora* (H t) and *Steinernema carpocapsae* with some nematicides (Rugby, Temik, Nema-cur and Vydate) on the 6<sup>th</sup> instar larvae, prepupae and pupae of both *S. littoralis* and *A. ipsilon* in soil. Nematode concentration was 10,000 infective stages /100 gm soil for each, which was combined with 200,400 and 800 ppm of every tested nematicide. Each nematode strain was applied alone against every insect stage, also the nematicides were solely tested against the insect stages. The treatments were inspected after 4 days for the 6<sup>th</sup> instar and 6 days for prepupae and pupae, mortality percentages, were calculated and recorded.

##### a)- Effect of combination on 6<sup>th</sup> instar larvae:

Data in Table (1) show the effect of three species of entomopathogenic nematodes combined with certain pesticides against the 6<sup>th</sup> larval instar of *S. littoralis*. The three tested concentrations of Cadusafos (Rugby) alone or combined with *S. carpocapsae* (All) or *H. bacteriophora* (HP88) gave 100%

mortality, whereas combinations with *H. bacteriophora* (Ht) gave only 93.3%. However, nematodes alone, gave similar results.

The tested three species of nematodes combined with Oxamyl (Vydate) gave higher mortalities than Vydate alone, but the higher concentrations of Vydate in the mixtures resulted in an increase in mortality rates in the case of Sc. and HP88 strains. In this respect, it is worth to note that there was no effect in case of *H. bacteriophora* (Ht). When the entomopathogenic nematodes were mixed with Fenamiphos (Nemacur) an enhancement in mortalities occurred with the three tested concentrations of Nemacur mixed with Ht and HP88, while vice versa was shown with *S. carpocapsae*.

Mortalities of the three tested concentrations of Aldicarb (Temik), evidently differed when they were combined with nematodes.

Statistical analysis showed no significant differences between the concentrations of the tested pesticides combined with Ht and HP88 strains, whereas there were significant differences in mortalities in case of pesticides alone or combined with *S. carpocapsae*.

Table (2) shows that all concentrations of Rugby combined with *H. bacteriophora* (Ht), *S. carpocapsae* and *H. bacteriophora* (HP88) gave mortalities reached 100% that were more than either the pesticide or each species of nematodes alone against *A. ipsilon*. Combinations of Vydate or Nemacur with

**Table (1)** Comparison between the nematode and the nematicides and their mixtures *S. littoralis* 6<sup>th</sup> instar larvae.

Treatment	Concentration	% Mortality $\pm$ SE			
		Pesticide Alone	+ H T	+ S c	+ HP88
Nematodes	Alone		93.3 $\pm$ 6.7 a	100 a	100 a
Cadusafos	200 ppm	100 a	93.3 $\pm$ 6.7 a	100 a	100 a
	400 ppm	100 a	93.3 $\pm$ 6.7 a	100 a	100 a
	800 ppm	100 a	93.3 $\pm$ 6.7 a	100 a	100 a
Oxamyl	200 ppm	56.7 $\pm$ 20.1 cd	85 $\pm$ 9.3 ab	65 $\pm$ 11.9 c	76.7 $\pm$ 16.3 a
	400 ppm	56.7 $\pm$ 16.2 cd	85 $\pm$ 9.3 ab	71.7 $\pm$ 8.1bc	78.3 $\pm$ 8.97 a
	800 ppm	45 $\pm$ 14.3 d	85 $\pm$ 9.3 ab	70 $\pm$ 20.7bc	85.3 $\pm$ 9.3 a
Fenamiphos	200 ppm	63.3 $\pm$ 19b cd	71.7 $\pm$ 7.3 b	100 a	91.7 $\pm$ 8.3 a
	400 ppm	71.7 $\pm$ 8.1 abcd	100 a	100 a	91.7 $\pm$ 8.34 a
	800 ppm	91.7 $\pm$ 9.3 ab	100 a	85.2 $\pm$ 10.4 abc	100 a
Aldicarb	200 ppm	91.7 $\pm$ 9.3 ab	91.7 $\pm$ 8.3 a	75 $\pm$ 11.4 abc	100 a
	400 ppm	91.7 $\pm$ 9.3 ab	100 a	91.7 $\pm$ 9.3ab	91.7 $\pm$ 8.3 a
	800 ppm	83.3 $\pm$ 11.4 abc	100 a	83.3 $\pm$ 11.4 abc	83.3 $\pm$ 11.6 a
*L.S.D		33.09	18.9	23.23	21.7

**One way ANOVA Completely Randomized**

Main Effect	df	F value	P
Pesticides	11	2.8	.0014 **
H t	12	1.63	.113 ns
S.c	12	2.72	.0063**
HP88	12	1.4	.197ns

**\*Dunca's multiple range test**

Values in the same column followed by the same letter are not significant different (P 0.05)

Table (2) Comparison between the nematode and the nematicides and their mixtures *A. ipsilon* 6<sup>th</sup> instar larvae.

Treatment	Concentration	% Mortality $\pm$ SE			
		Pesticide Alone	+ H T	+ S c	+HP88
Nematodes	Alone		85 $\pm$ 9.3 abc	91.7 $\pm$ 8.3 ab	91.7 $\pm$ 8.3
Cadusafos		91.7 $\pm$ 8.3 ab	100 a	100 a	100
	200 ppm	85 $\pm$ 9.3 ab	100 a	100 a	100
	400 ppm	100 a	100 a	100 a	100
	800 ppm	43.3 $\pm$ 14.5 cd	60 $\pm$ 12.5 c	71.7 $\pm$ 7.3 abc	30 $\pm$ 3.3 c
Oxamyl	200 ppm	23.3 $\pm$ 6.7 d	66.7 $\pm$ 10.5 bc	63.3 $\pm$ 13.3 bc	31.7 $\pm$ 14 c
	400 ppm	51.6 $\pm$ 13 bcd	66.7 $\pm$ 10.5 bc	50 $\pm$ 10.5 c	51.67 $\pm$ 7.6 b
	800 ppm	36.7 $\pm$ 17 cd	85 $\pm$ 9.3 abc	70 $\pm$ 18.6 abc	38.3 $\pm$ 11.7 c
	200 ppm	63.3 $\pm$ 17 abc	91.7 $\pm$ 8.3 ab	63.3 $\pm$ 13.3 bc	45 $\pm$ 7.3 bc
Fenamiphos	400 ppm	71.7 $\pm$ 7.3 abc	85 $\pm$ 9.3 abc	71.7 $\pm$ 7.3 abc	16.7 $\pm$ 7.5
	800 ppm	85.2 $\pm$ 9.3 ab	93.3 $\pm$ 6.7 ab	91.7 $\pm$ 8.3 ab	63.3 $\pm$ 13.3
	200 ppm	83.3 $\pm$ 16.7 ab	60 $\pm$ 12.5 c	85.9 $\pm$ 9.3 ab	71.7 $\pm$ 12.8
	400 ppm	56.7 $\pm$ 20.8 bcd	66.7 $\pm$ 10.5 bc	91.7 $\pm$ 8.3 ab	71.7 $\pm$ 12.8
Aldicarb	800 ppm	27.4	25.1	26.04	21.7
*L.S.D					

One way ANOVA Completely Randomized

Main Effect	df	F value	P
Pesticides	11	2.8	.0032* **
Ht	12	3.08	.0024**
Sc	12	10.4	.0000***
HP88	12	10.33	.0000***

\*Dunca's multiple range test

Values in the same column followed by the same letter are not significant different (P 0.05)

both Ht and S.c (All) increased mortalities than either Vydate alone or with HP88. The same results were obtained with the lowest concentration of Temik (200ppm) mixed with the three tested species of nematodes, but in case of 400 and 800 ppm, mortalities ranged between 60-85.9 and 56.7- 91.7 %, respectively.

Statistical analysis proved that there were highly significant differences between the tested nematicides with Ht, S.c, HP88 strains and the nematicides alone.

**b) – Effect of combination on prepupae:**

Both toxicity of the nematicides Rugby, Vydate, Nemacur and Temik, alone and / or combined with three strains of entomopathogenic nematodes Ht, S.c and HP88, were studied against the prepupal stage of *S. littoralis* and the obtained data are shown in Table (3).

In spite of that the combined nematicides with nematodes were more effective than nematicides alone but their efficiency was less or equal to those of nematodes alone, which reached 100% mortality for each strain. However toxicity of the used nematicides alone ranged between 40-83.3 %, whereas those of the combinations ranged between 54.2-100 % (with Ht), 76.7-100% (with S.c) and 70-100% (with HP88).

Statistical analysis demonstrated that there were no significant differences were observed between the concentrations of the tested nematicides with nematode strains, but there were significant differences between the nematicides alone.

Table (3) Comparison between nematodes, nematicides and their mixtures on *S. littoralis* prepupae stage.

Treatment	Concentration	% Mortality $\pm$ SE			
		Pesticide Alone	+ H T	+ Sc	+ HP88
Nematodes Cadusafos	Alone		100 a	100 a	100 a
	200 ppm	70 $\pm$ 7.7 ab	100 a	91.7 $\pm$ 8.3 a	100 a
	400 ppm	70 $\pm$ 7.7 ab	89.6 $\pm$ 10.4 a	100 a	91.7 $\pm$ 8.3 ab
	800 ppm	83.3 $\pm$ 10.2 a	100 a	76.7 $\pm$ 9.7 a	100 a
Oxamyl	200 ppm	40 $\pm$ 11.3 bc	83.3 $\pm$ 16.7 ab	91.7 $\pm$ 8.3 a	91.7 $\pm$ 8.3 ab
	400 ppm	70 $\pm$ 7.7 ab	83.3 $\pm$ 16.7 ab	83.3 $\pm$ 10.2 a	91.7 $\pm$ 8.3 ab
	800 ppm	76.7 $\pm$ 9.6 a	83.3 $\pm$ 16.7 ab	91.7 $\pm$ 8.3 a	83.3 $\pm$ 10.2 ab
Fenamiphos	200 ppm	76.7 $\pm$ 9.6 a	100 a	100 a	100 a
	400 ppm	33.4 $\pm$ 17.5 c	89.6 $\pm$ 10.4 a	100 a	83.3 $\pm$ 10.2 ab
	800 ppm	83.3 $\pm$ 10.2 a	89.6 $\pm$ 10.4 a	100 a	70 $\pm$ 7.7
Aldicarb	200 ppm	70 $\pm$ 7.7 ab	54.2 $\pm$ 20.6 b	91.7 $\pm$ 8.3 a	83.3 $\pm$ 10.2
	400 ppm	76.7 $\pm$ 9.6 a	100 a	83.3 $\pm$ 10.2 a	91.7 $\pm$ 8.3
	800 ppm	83.3 $\pm$ 10.2 a	100 a	100 a	83.3 $\pm$ 10.2
*L.S.D		29.2	28.2	20.6	12.6

One way ANOVA Completely Randomized

Main Effect	df	F value	P
Pesticides	11	2.51	014 *
Ht	12	1.7	.093 ns
Sc	12	1.24	.28 ns
HP88	12	1.42	.187 ns

\*Dunca's multiple range test

Values in the same column followed by the same letter are not significant different (P 0.05)

Regarding the effect of nematicides and nematode strains and their combinations against *A. ipsilon* prepupae, it was found that nematode strains yielded the most higher percentage mortalities namely 100, 100 and 91.7% for Ht, S.c and HP88, respectively (Table 4). On the other hand the combinations of either S.c or HP88 strains with the four tested nematicides (Rugby, Vydate, Nematicur and Temik) resulted in an increase in the mortality percentage of prepupae of *A. ipsilon* than did pesticides alone. The corresponding mortalities ranged between 83.3-100 and 41.7-91.7 %, respectively. Data revealed also that the mixtures of Ht nematode strain with the four tested pesticides resulted in lower mortality rates (31.3-72.9%) than those of pesticides alone (36.7-100%).

Statistical analysis demonstrated that there were high significant differences in the case of combination between nematicides and Ht. Also, significant differences were found in the case of pesticides alone or when combined with HP88. But in case of combined nematicides and S.c no significant difference were found.

#### **c)- Effect of combinations on pupae:**

Toxicity of the pesticides alone or combined with nematode strains against pupae of *S. littoralis* had been shown in Table (5). It was found that Rugby alone was the most potent pesticide, giving mortalities between 83.3 -97.7%, while insect mortalities of the other pesticides ranged between 3.3-25.5% only. On the other hand the potency of the four tested pesticides combined with the entomopathogenic nematodes increased



Table (4) Comparison between nematodes, nematicides and their mixtures on *A. epsilon* prepupae stage.

Treatment	Concentration	% Mortality $\pm$ SE			
		Pesticide Alone	+ H T	+ S c	+ HP88
Nematodes	Alone		100 a	100 a	91.7 $\pm$ 8.3
Cadusafos		53.3 $\pm$ 15.8 bc	54.2 $\pm$ 7.2 bcd	91.7 $\pm$ 8.3 a	91.7 $\pm$ 8.3
	200 ppm				
		76.7 $\pm$ 9.7 ab	43.8 $\pm$ 11.5 cd	91.7 $\pm$ 8.3 a	83.3 $\pm$ 10
	400 ppm				
Oxamyl		100 a	62.5 $\pm$ 13.8 bc	100 a	91.7 $\pm$ 8.3
	800 ppm				
		53.3 $\pm$ 15.8 bc	70.8 $\pm$ 9.9 bc	83.3 $\pm$ 10 a	91.7 $\pm$ 8.3
	200 ppm				
Fenamiphos		76.7 $\pm$ 9.6 ab	62.5 $\pm$ 2.4 bc	83.3 $\pm$ 10 a	61.7 $\pm$ 13.3
	400 ppm				
		60 $\pm$ 13.3 bc	68.8 $\pm$ 19.9 bc	83.3 $\pm$ 10 a	91.7 $\pm$ 8.3
	800 ppm				
Aldicarb		48.3 $\pm$ 10 bc	54.2 $\pm$ 7.2 bcd	100 a	83.3 $\pm$ 16.7
	200 ppm				
		63.3 $\pm$ 10.7 bc	43.8 $\pm$ 11.5 cd	100 a	91.7 $\pm$ 8.3
	400 ppm				
HP88		70 $\pm$ 13.1 abc	33.4 $\pm$ 11.8 d	83.3 $\pm$ 10 a	83.3 $\pm$ 10
	800 ppm				
		36.7 $\pm$ 10.7 c	52.1 $\pm$ 12 bcd	100 a	76.7 $\pm$ 9.7
	200 ppm				
LSD		36.7 $\pm$ 15 c	72.9 $\pm$ 9.2 b	83.3 $\pm$ 10 a	76.9 $\pm$ 9.7
	400 ppm				
		50 $\pm$ 6.9 bc	31.3 $\pm$ 9.8 d	100 a	41.7 $\pm$ 9.7
	800 ppm				
*L.S.D		32.8	23.8	20.2	28

ONE way ANOVA Completely Randomized

Main Effect	df	F value	P
Pesticides	11	2.51	.0139 *
Ht	12	4.85	.000***
Sc	12	1.24	.279 ns
HP88	12	2.13	.0304*

\*Dunca's multiple range test

Values in the same column followed by the same letter are not significant different (P 0.05)

Table (5) Comparison between the nematodes and the nematicides and their mixtures on *S. littoralis* pupal stage.

Treatment	Concentration	% Mortality $\pm$ SE			
		Pesticide Alone	+ H T	+ S c	+ HP88
Nematodes	Alone		100 a	100 a	83.3 $\pm$ 10.2 abc
Cadusafos		83.3 $\pm$ 10.2 a	79.2 $\pm$ 12 abc	83.3 $\pm$ 10.2 ab	100 a
	200 ppm	91.7 $\pm$ 8.3 a	100 a	83.3 $\pm$ 10.2 ab	83.3 $\pm$ 10.2 abc
	400 ppm	83.3 $\pm$ 10.2 a	89.6 $\pm$ 10.4 ab	41.7 $\pm$ 14 d	91.7 $\pm$ 8.3 ab
	800 ppm	3.3 $\pm$ 3.4 b	62.5 $\pm$ 13.8 bcd	16.7 $\pm$ 6.8 c	56.7 $\pm$ 12.2 bcd
Oxamyl	200 ppm	6.7 $\pm$ 4.1 b	79.2 $\pm$ 12 abc	79.2 $\pm$ 12 abc	41.7 $\pm$ 9.1 cd
	400 ppm	16.7 $\pm$ 6.8 b	68.8 $\pm$ 19.9 abc	41.7 $\pm$ 14 d	46.7 $\pm$ 16.2 cd
	800 ppm	13.3 $\pm$ 6.2 b	35.4 $\pm$ 8.6 d	62.5 $\pm$ 2.4 bcd	83.3 $\pm$ 16.7 abc
Fenamiphos	200 ppm	6.7 $\pm$ 4.1 b	79.2 $\pm$ 12 abc	54.2 $\pm$ 7.2 cd	66.7 $\pm$ 20 abcd
	400 ppm	13.3 $\pm$ 6.3 b	68.8 $\pm$ 19.9 abc	62.5 $\pm$ 2.4 bcd	53.3 $\pm$ 21.7 bcd
	800 ppm	25 $\pm$ 14.4 b	62.5 $\pm$ 2.4 bcd	62.5 $\pm$ 2.4 bcd	68.8 $\pm$ 20 abcd
Aldicarb	200 ppm	16.7 $\pm$ 6.8 b	60.4 $\pm$ 17.1 bcd	70.8 $\pm$ 9.9 bc	37.5 $\pm$ 17 d
	400 ppm	25 $\pm$ 4.8 b	52.1 $\pm$ 18.1 cd	70.8 $\pm$ 9.9 bc	62.5 $\pm$ 2.4 abcd
	800 ppm				
*L.S.D		20.01	21.1	22.3	35.9

One way ANOVA Completely Randomized

Main Effect	df	F value	P
Pesticides	11	22.4	.0000***
H t	12	3.3	.0013**
Sc	12	7.7	.0000***
HP88	12	2.5	.013*

\*Dunca's multiple range test

Values in the same column followed by the same letter are not significant different (P 0.05)

mortality rates to 35.4 - 100, 41.7- 83.3 and 41.7- 100 % in case of combination with Ht, S.c and HP88, respectively.

Statistical analysis proved that there were high significant differences between the concentrations of the tested pesticides either alone or combined with Ht and S.c nematode strains, but in case of combination with HP88 there was significant correlation.

The sensitivity of *A. ipsilon* pupae in treated sandy soil differed according to the used pesticide either alone or combined with the three nematode strains (Table 6). Rugby alone or combined with Ht, S.c or HP88 gave the highest mortality rates which ranged between 70.8-100%, that seemed to be nearly similar to those of nematodes alone (89.6 for Ht, 100 for S.c and 91.7 for HP88). With respect to other three tested pesticides (Nemacur, Temik and Vydate), it was found that their combinations with entomopathogenic nematodes raised their effectiveness to levels more than the pesticides alone. Their mortalities ranged between 8.4-18.8% (pesticides alone), 29.2-79.2 % (with Ht) 27.1-72.9 % (with S.c) and 4.2-54.2 % (with HP88).

Statistical analysis demonstrated that, there were high significant differences between the tested concentrations of either pesticides alone, or combined with all tested nematode strains.

Entomopathogenic nematodes infectivity to insects, differed considerably among the different developmental stages

Table (6) Comparison between nematodes, nematicides and their mixtures on *A. ipsilon* pupal stage.

Treatment	Concentration	% Mortality $\pm$ SE			
		Pesticide Alone	+ H T	+ S c	+ HP88
Nematodes	Alone		89.6 $\pm$ 10.4 ab	100 a	91.7 $\pm$ 8.3 a
Cadusafos		83.3 $\pm$ 11.4 a	70.8 $\pm$ 9.9 abc	100 a	100 a
	200 ppm				
	400 ppm	91.7 $\pm$ 8.3 a	79.2 $\pm$ 12 abc	100 a	100 a
	800 ppm	83.3 $\pm$ 11 a	100 a	100 a	100 a
Oxamyl		16.7 $\pm$ 6.8 b	56.7 $\pm$ 6.8 cde	60.4 $\pm$ 17 bcd	8.4 $\pm$ 4.8 d
	200 ppm				
	400 ppm	8.4 $\pm$ 4.8 b	60.4 $\pm$ 17.1 bcd	68.8 $\pm$ 20 bc	20.8 $\pm$ 7.9 cd
	800 ppm	8.4 $\pm$ 4.8 b	79.2 $\pm$ 12 abc	70.8 $\pm$ 10 bc	8.4 $\pm$ 4.8 d
Fenamiphos		8.4 $\pm$ 4.8 b	52.1 $\pm$ 18.1 cde	54.2 $\pm$ 7.2 bcde	18.8 $\pm$ 14 cd
	200 ppm				
	400 ppm	8.4 $\pm$ 4.8 b	62.6 $\pm$ 13.8 bcd	58.4 $\pm$ 24 bcd	54.2 $\pm$ 7.2 b
	800 ppm	18.8 $\pm$ 13.8 b	70.8 $\pm$ 9.9 abc	27.1 $\pm$ 12.4 c	52.1 $\pm$ 18 b
Aldicarb		4.2 $\pm$ 4.2 b	29.2 $\pm$ 16.8 c	35.4 $\pm$ 8.6 de	4.2 $\pm$ 4.2 d
	200 ppm				
	400 ppm	14.6 $\pm$ 14.6 b	52.1 $\pm$ 12 cde	43.8 $\pm$ 12 cde	41.8 $\pm$ 14.4 bc
	800 ppm	14.6 $\pm$ 14.6 b	33.4 $\pm$ 12 de	72.9 $\pm$ 16 ab	50 $\pm$ 20.4 b
*L.S.D		22.3	27.9	25.3	22.2

One way ANOVA Completely Randomized

Main Effect	df	F value	P
Pesticides	11	18.9	.0000***
H t	12	4.43	.0001***
Sc	12	7.4	.0000***
HP88	12	21	.0000***

\*Dunca's multiple range test

Values in the same column followed by the same letter are not significant different (P 0.05)

of the insects. Generally, the obtained data revealed that 6<sup>th</sup> instar larval and prepupal stages of both two tested insects, were more vulnerable to nematode infection than the pupal stage. The path of entry for infective stages is throughout the natural openings (mouth, anus and spiracles). Entry of entomopathogenic nematodes via spiracles is considered to be a main infection route into insect pupae, which have neither mouth nor anus. (Hara and Kaya 1983b), (Kaya and Grieve 1982) and (Poinar 1979). Susceptibility of pupae to nematodes considerably varies with different insect species. Soil-or-litter-pupating insects are generally less susceptible than those, pupate above ground Kaya and Hara (1981). They believe that the insect susceptibility is, closely related to the level of the cuticle sclerotization because *P. unipunctata* pupae (least susceptible) are more sclerotized than *S. exigua* ones (moderately susceptible), while *G. mellonella* pupae (most susceptible) are the least sclerotized. These results indicate that structure and hardness of spiracles are clearly responsible for pupal susceptibility to the entomopathogenic nematodes (Ahmed 1982, Azazy 1996) and this may interprets the more susceptibility of *S. littoralis* pupae to nematode infection than *A. ipsilon* ones in our case. Compared to the infection in active *S. litura* larvae, *S. feltiae* infection occurred more easily in insects, which were weakened by some treatments, as with formalin solution or with a sublethal dose of pesticides (Hatsukade and Yamanaka 1988) and (Kaya and Burlando, 1989). The obtained results are in full harmony with the findings of Hara and Kaya (1982), Ishabashi *et al* (1987) and Kamionek (1978).

They said that, the efficiency of *S. flitiae* could be improved in compatible with pesticides in an integrated pest management program. **Ishabashi et al (1987)** found that, in field trials, a mixed application of *S. carpocapsae* with insecticides has provided more effective insect control than using each of them, separately. Also, **Strak and Shanks (1993)**, **Ishabashi (1993)** and **franceschini (1999)** gained similar results in this field.

## 2-Effect of some pesticides on the activity of *H. bacteriophora* dauer stages:

The effect of nematicides (Cadusafos, Fenamiphos, Oxamyl) and insecticides (Diazinon, Phenthoate) on activity of the infective stages of *H. bactreiophora* (Ht) and *H. bacteriophora* (HP88) was investigated. It was observed that the level of toxicity of the tested pesticides in aqueous solution varied, especially on the (Ht) infective stages (Table7). Results showed that, the observed mortality, after 48h exposure to Fenamiphos ranged between 3.3 to 15.7%. Fenamiphos gave mortalities of 4.7, 3.3, 3.8, 8.5, 7.3 and 15.7% for concentrations of (25,50,100,200,400,800 ppm), respectively. However, mortality of the nematicide (Oxamyl) ranged between 1.8 to 19.4%. That of Diazinon was 5.3, 4.6 and 2.97% for 400,800,1600 ppm whereas, Phenthoate gave 4.7, 5.3, and 4.6% mortalities due to concentrations of 375, 750, 1500 ppm, respectively.

**Table (7)** Mortality percent of the infective stages of *II. bacteriophora* (Ht) after 48h exposure to five pesticides.

Pesticides Concentration	% Mortality of IJs (Mean $\pm$ SE)					
	Nematicides			Insecticides		
	Cadusafos (R.)	Fenamiphos (N.)	Oxamyl (V.)	Diazinon (B.)	Phenthoate (C.)	
25 ppm	5.9 $\pm$ 2	4.7 $\pm$ 2.3	b	1.8 $\pm$ 0.9	b	-
50 ppm	6.7 $\pm$ 1.5	3.3 $\pm$ 0.4	b	3.5 $\pm$ 1.9	b	-
100 ppm	3.6 $\pm$ 2.2	3.8 $\pm$ 1.9	b	4.4 $\pm$ 0.9	b	-
200 ppm	5.1 $\pm$ 2.6	8.5 $\pm$ 5.2	ab	7.4 $\pm$ 0.8	b	-
325 ppm	-	-	-	5.3 $\pm$ 1	a	-
400 ppm	10.1 $\pm$ 5.1	7.3 $\pm$ 2.9	ab	4.4 $\pm$ 0.6	b	4.7 $\pm$ 1.1
750 ppm	-	-	-	4.6 $\pm$ 0.4	a	-
800 ppm	12.8 $\pm$ 1.8	15.7 $\pm$ 3.3	a	19.4 $\pm$ 4.3	a	5.3 $\pm$ 0.8
1500 ppm	-	-	-	3 $\pm$ 0.6	a	-
1600 ppm	-	-	-	-	-	4.6 $\pm$ 0.1
* L.S.D	8.7	9.4	6.3	2.5	4.3	

**One way ANOVA Completely Randomized**

Main Effect	df	F value	P
Cadusafos	5	1.5	.26 ns
Fenamiphos	5	2.33	.10 ns
Oxamyl	5	9.9	.0006***
Diazinon	2	2.7	.15 ns
Phenthoate	2	0.11	.9 ns

**\*Duncan's multiple range test**

Values in the same column followed by the same letter are not significant different (P 0.05)

Generally the average mortalities were 4.3, 4.9, 6.8, 7.2 and 7.4% of Diazinon, Phenthoate, Oxamyl, Cadusafos, Fenamiphos, respectively. Statistical analysis revealed that there were no significant differences between the tested concentrations of pesticides, except of the nematicide (Oxamyl), which showed highly significant differences between its concentrations.

Data in Table (8) showed the toxicity effect of organophosphorus compounds Cadusafos, Diazinon, Fenamiophos and Phenthoate on *H. bacteriophora* (HP88) infective stages during 48h of exposure. These pesticides achieved nematode mortalities ranged between 1.7-9.6, 1.4-13.8, 1.1- 6.8 and 1.4- 13.4% for each, successively. The carbamate compound (Oxamyl) was more toxic at high concentration of 800 ppm to *H. bacteriophora* infective stage giving 34.4% mortality.

Statistical analysis proved that there were significant differences between concentrations of (Cadusafos, Diazinon and Phenthoate). The nematicide (Oxamyl) showed highly significant differences between concentrations, while Fenamiophos revealed that there were no significant differences between its concentrations. These results encourage us to suggest that most of the pesticides used for controlling pest maintenance can be successfully used with entomopathogenic nematode applications. The nematicides can, probably be used at concentrations of no more than 800 ppm, but insecticides could be used at concentrations more than 1600 ppm with nematodes.



**Table (8)** Mortality percent of the infective stages of *H. bacteriophora* (HP88) after 48h exposure to five pesticides.

Pesticides Concentration	% Mortality of IJs (Mean $\pm$ SE)				
	Nematicides		Insecticides		
	Cadusafos (R.)	Fenamiphos (N.)	Oxamyl (V.)	Diazinon (B.)	Phenthoate (C.)
25 ppm	1.7 $\pm$ 0.9 c	1.1 $\pm$ 0.6 b	0.8 $\pm$ 0.8 c	-	-
50 ppm	2.0 $\pm$ 1.0 bc	3.7 $\pm$ 0.3 ab	5.7 $\pm$ .7 c	-	-
100 ppm	2.0 $\pm$ 1.0 bc	3.8 $\pm$ 0.6 ab	15.2 $\pm$ 1.1 b	-	-
200 ppm	9.2 $\pm$ 0.9 a	6.8 $\pm$ 3.5 a	14.7 $\pm$ 4.9 b	-	-
325 ppm	-	-	-	1.4 $\pm$ 1.0 b	-
400 ppm	9.6 $\pm$ 0.6 a	5.4 $\pm$ 0.9 ab	15.4 $\pm$ 4.1 b	-	1.4 $\pm$ 1.0 b
750 ppm	-	-	-	7.1 $\pm$ 1.4 ab	-
800 ppm	6.3 $\pm$ 2.7 ab	6.5 $\pm$ 1.7 ab	34.4 $\pm$ 1.4 a	-	7.1 $\pm$ 1.4 ab
1500 ppm	-	-	-	13.8 $\pm$ 3.4 a	-
1600 ppm	-	-	-	-	13.4 $\pm$ 3.8 a
*L.D.S	4.2	5.04	7.8	8.4	7.7

One way ANOVA Completely Randomized

Main Effect	df	F value	P
Cadusafos	5	7.03	.0027 **
Fenamiphos	5	1.6	.22 ns
Oxamyl	5	20.8	.000 ***
Diazinon	2	6.2	.034 *
Phenthoate	2	7.8	.02 *

\*Dunca's multiple range test

Values in the same column followed by the same letter are not significant different (P 0.05)

Overall, results indicate the feasibility of an IPM use of these nematode species and chemical pesticides in crop protection (Heungens & Buysse (1987), Jarowska (1990), Rovesti *et al* (1990), Rovesti & Doeso (1990)).

### **3-Dispersal and migration of nematodes:**

Some factors affecting dispersal and migration of five nematode infective stages had been studied in laboratory in column of sandy soil. These factors included the presence or absence of the insect host, insect feces, the nematicide (Nemacur), and host species.

#### **3. 1-Comparison between the effect of *G. mellonella* and Nemacur:**

Twenty-four hours post nematode inoculation, majority of IJs moved away from the point of application in all tested nematode strains. The proportion of migration (%m) was not the same for the different nematode species. In the case of host presence, the migration of IJs of *S. glaseri* was the highest recording 89.28% (Fig.2). The other strains achieved migration rates of 79.7, 76.48, 29.85 and 21.45% for Ht, HP88, S.c (All) and S.c (*agriotis*), respectively. However, in case of host absence, results showed that the strain Ht was the best (26% migration rate), whereas S.c (All) was the lowest one (2.32%).

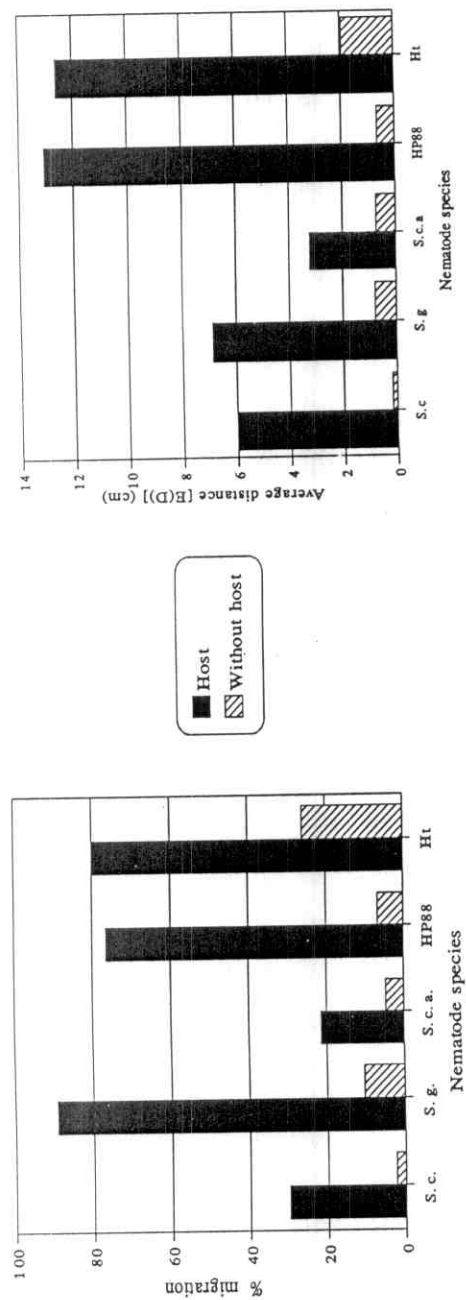


Fig. (2): Effect of presence or absence of *G. mellonella* on dispersal and migration of five nematodes strains

S.c (All) = *S. carpocapsae*  
 S.g = *S. glaseri*  
 S.c.a = *S. carpocapsae agriotis*  
 HP88 = *H. bacteriophora*  
 Ht = *H. bacteriophora* (local)

Regarding the "average net distance" [E (D)], IJs of HP88 was found to move an "average net" longer than that of the other strains reaching the maximum distance of 13.07cm. The [E (D)] of the other tested strains ranged between 3.23 to 12.62 cm.

Studying the effect of nematicide (Nemacur, 400 ppm), compared with that of *G. mellonella* host upon the migration of the tested strains, it was found that the nematicide improved the performance of the nematode migration, especially in the case of HP88, Ht. S.c (*agriotis*), S.c (All) showing 93.02, 81.13, 68 and 30.86%. On the other hand the nematicide caused clear inhibition in S.g migration (1.08%). Also, strains S.c, S.c.a and HP88 combined with Nemacur, moved longer distances [E (D)] of 7.6, 11.9 and 17.15 cm, successively, while those of S.g and Ht were the least, showing 0.1 and 10.16 cm, respectively (Fig.3).

### 3. 2-Comparison between the effect of *S. littoralis* and Nemacur:

The presence of *S. littoralis* as a bait, increased the migration rates of all the tested strains (Fig.4). S.g showed the highest values (from 10.43 to 91.12%), followed by HP88 (from 16.1 to 80.06%), Ht (from 25.54 to 34.2%), S.c.a (from 21.5 to 27.3%), S.c (from 9.38 to 22.14). Accordingly, as a result of the increase in % m, an increase in [E (D)] values was evident in all strains.

The addition of Nemacur to nematode solutions activated the migration of S.c.a, H t, and S.c strains showing percent migration of 100, 84.1 and 54.55%, respectively.

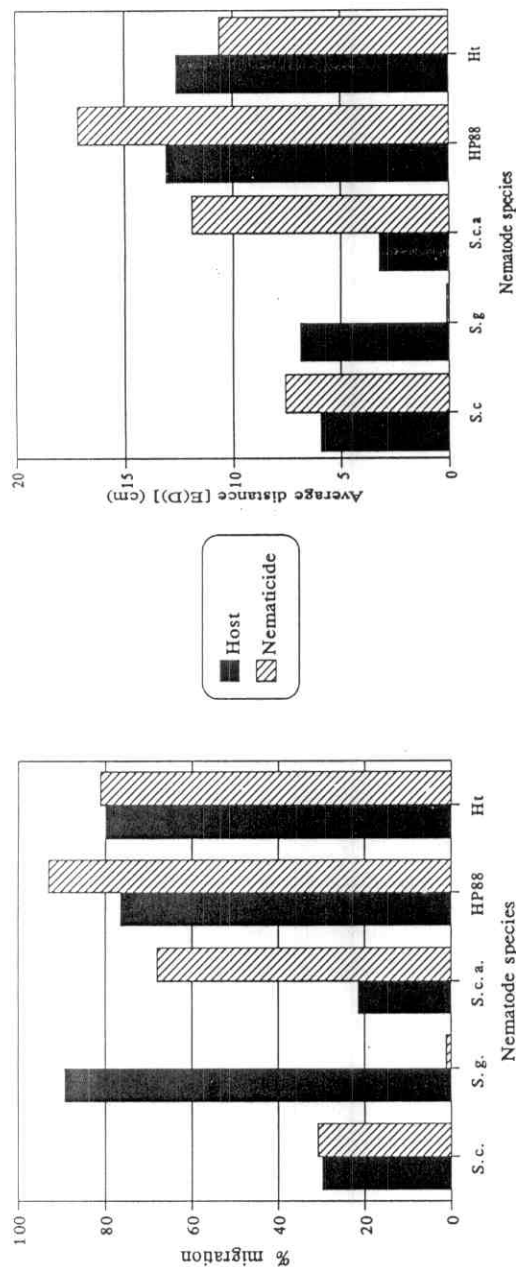


Fig. (3): Effect of NemaCur on dispersal and migration of five nematode strains.

S.c (All) = *S. carpocapsae*  
 S.g = *S. glaseri*  
 S.c.a = *S. carpocapsae agriotis*  
 HP88 = *H. bacteriophora*  
 Ht = *H. bacteriophora* (Local)

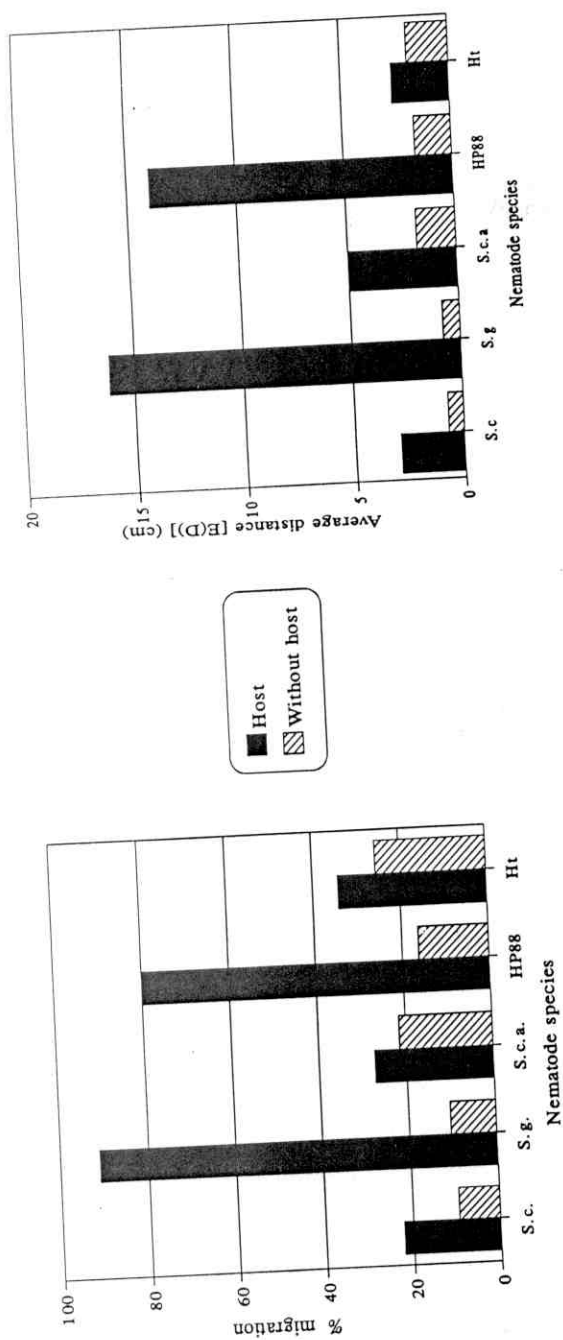


Fig. (4): Effect of presence or absence of *S. littoralis* on dispersal and migration of five nematode strains

S.c (All) = *S. carpocapsae*  
 S.g = *S. glaseri*  
 S.c.a = *S. carpocapsae agriotis*  
 HP88 = *H. bacteriophora*  
 Ht = *H. bacteriophora* (Local)

However, relative inhibition in migration rate, occurred in the case of other two, strains recording 4.06 and 42.19 % for S.g and HP88, consecutively (Fig.5).

Accordingly, as a result of the increase of % m, an increase in average net distance [E (D)] value was evident in all nematode strains.

### 3. 3-Effect of *S. littoralis* feces:

As shown in Fig. (6), the migration rate (%m) of all strains (except S.g). in presence of feces were better than those of the presence of the host. These values, were 83.64, 63.89, 52.3 and 42.39% for HP88, S.c.a, S.c and Ht respectively. In contrasts S.g was the least responded strain to *S. littoralis* feces, achieving 19.48% of migration rate against 91.12 % migration towards the host itself.

The presence of feces increased values of E (D) of S.c, S.c.a and Ht (8.21,10.14 and 3.96 cm), but those values were decreased, remarkably in the strains of S.g and HP88 than those of the host.

### 4. 3 -The effect of Host species:

Results shown in Fig (7) revealed that, the nematode strains varied in their attraction towards the two hosts. Two strains (Ht and S.c) were attracted more by *G. mellonella* larvae, while the other three strains (S.g, S.c.a and HP88) were attracted by *S. littoralis* more than *G. mellonella*. Eighty percent of Ht populations were directed towards *G. mellonella* while only 35%

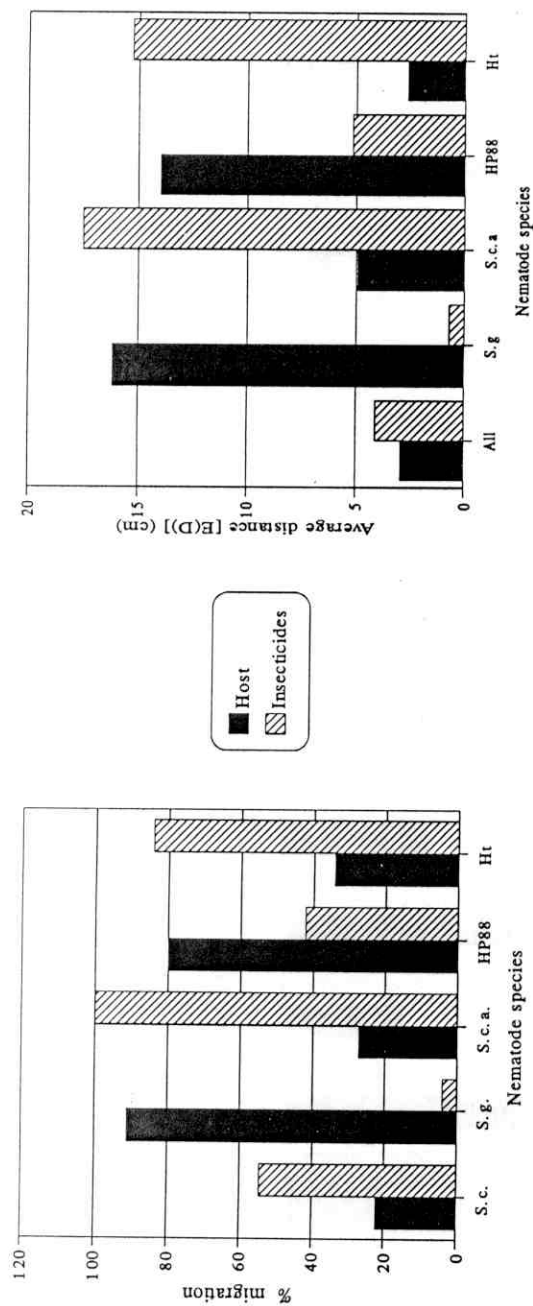


Fig. (5): Effect of NemaCur on dispersal and migration of nematode strains

S.c (All) = *S. carpocapsae*  
 S.g = *S. glaseri*  
 S.c.a = *S. carpocapsae agriotis*  
 HP88 = *H. bacteriophora*  
 Ht = *H. bacteriophora* (Local)



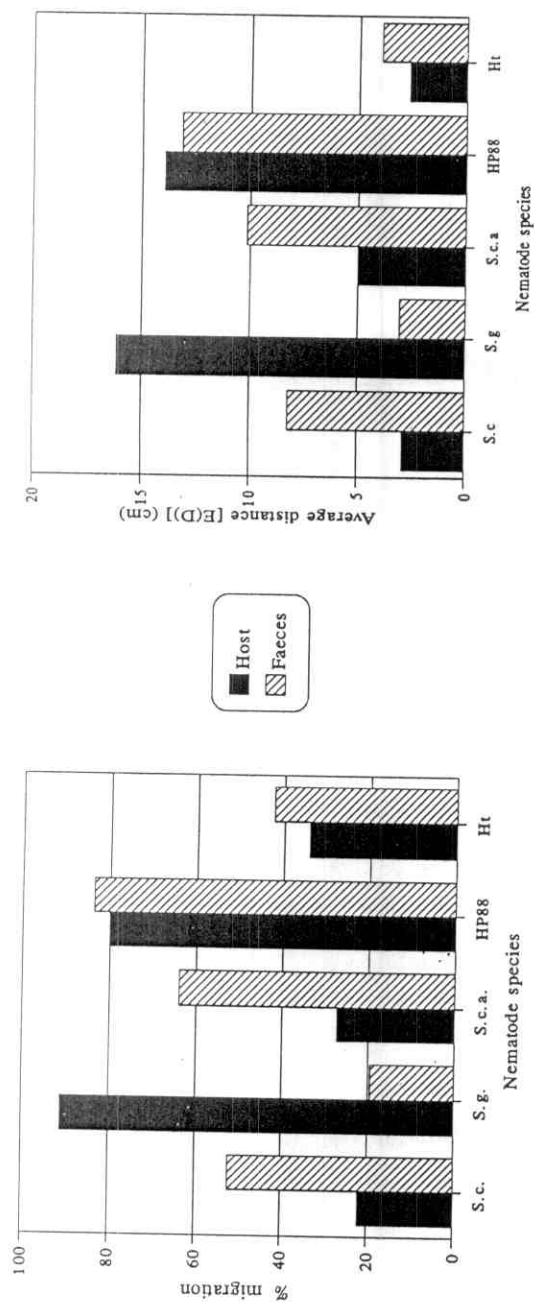


Fig. (6): Effect of *S. littoralis* feces on migration and dispersal of five nematode strains

S.c (All) = *S. carpocapsae*  
 S.g = *S. glaseri*  
 S.c.a = *S. carpocapsae agriotis*  
 HP88 = *H. bacteriophora*  
 Ht = *H. bacteriophora* (Local)

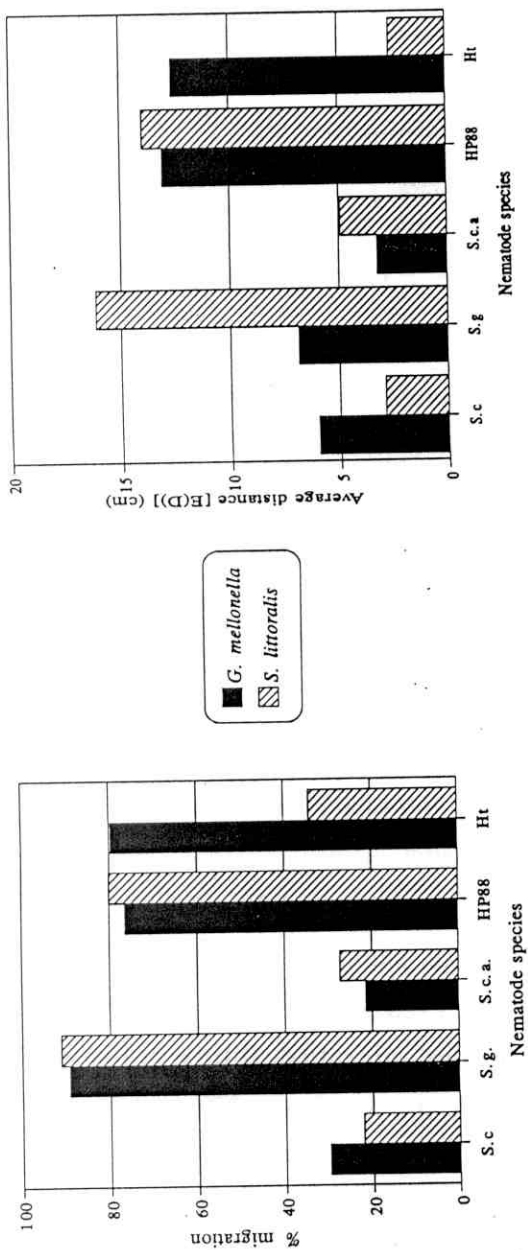


Fig. (7): Comparison between two host species on migration and dispersal of five nematode strains

S.c (All) = *S. carpocapsae*  
 S.g = *S. glaseri*  
 S.c.a = *S. carpocapsae agriotis*  
 HP88 = *H. bacteriophora*  
 Ht = *H. bacteriophora* (Local)

were attracted by *S. littoralis* larvae. The variations in the other strains were not too high between the two hosts as in the aforementioned strains. Regarding E (D) values, it is clear that the strain S.g which was attracted by, *S. littoralis* was the superior (16.15cm) followed by the strain HP88 of both hosts, then Ht. which was attracted to *G. mellonella* and the other two strains (S.c and S.c.a), were the last.

Entomopathogenic nematodes of the Heterorhabditidae and Steinernematidae appear to be capable of long- distance dispersal and local migration. Their transmission strategies include both, highly active seek- and destroy behaviours and ambusher strategies, and they may be sensitive to sex related factors in their own populations (Downes & Griffin, 1996). Although nematode dispersal is necessary for host -finding, they may disperse passively (Epsky *et al.*, 1988; Timper *et al.*, 1988), active dispersal, particularly by cruiser nematodes, appears to be the primary means for host finding, since it increases the likelihood of encountering sedentary host larvae. On the other hand, high mobility without sensitivity to host cues leads to quickly depleted food reserves (Molyneux, 1984 & Vänninen, 1990). Both locomotion and migration ability of the nematodes are thus critical factors in the control of sedentary insect soil. The sand column technique used in the present investigation enabled us to study the dispersal and migration of the tested nematode species, as well as some factors that may affect them. Also, complying with the situation of sessile root pests in the field, and host larvae in the assay unit were immobile (Fig.1),

allowed the formation of chemical gradient around them (Steiner, 1996).

In the present investigation, it was found that dispersal and migration differ among the tested strains. These differences could be related to the inherited characters of each species, in addition to the reaction between these features and the other factors. Factors affecting active nematode dispersal and host finding in soil, include small pore spaces (Blackshaw & Senthamizheselvan, 1991; Georgis & Poinar, 1983 a, b & c; Molyneux & Bedding, 1984), moisture, temperature and plant roots (Choo *et al.*, 1989).

Active dispersal of entomopathogenic nematodes is short range and may be influenced by host cues. Host derived compounds such as CO<sub>2</sub> (Gaugler *et al.*, 1980) and fecal components (Schmidt & All, 1979) have been shown to be attractive to these nematodes. Migration rate may be directly related to the host finding strategy of tested nematode species. The low migration rate of *S. carpocapsae* (All) and *S. carpocapsae* (*agriotis*) in most experiments typical of an ambusher or a sit and wait strategy (Kaya & Gaugler 1993) and they tended to remain near the point of application limits contact with sedentary hosts (Georgis & Poinar, 1983; Gaugler *et al.*, 1989). As shown in Fig (2) the presence of the host increased the dispersal and migration of the tested nematode species. This finding is in a full agreement with that of Georgis & Poinar (1983c) who stated that the presence of the host increased dispersal of *H. bacteriophora*, but the majority were still found

near the placement site. The pattern of dispersal and migration in *S. carpocapsae* (agriotis) and *S. glaseri* respectively were not affected by changing the host. Also the present results are in accordance with the finding of **Grewal et al. (1995)** who reported that entomopathogenic nematode species are different in their response to host chemical cues.

The present work proved that the nematicide (Nemacur) is an important factor affecting migration and dispersal of nematode species. In previous studies, combination between the nematodes and nematicide increased migration rate and average net distance E(D) which were better than using the nematodes alone except in some cases. This finding agree with the present study, since Nemacur caused a remarkable inhibition in the migration and E (D) of both S.g and HP88 strains (Figs, 3 and 5). **Ishibashi and Shingi, 1993** stated that the insecticides stimulated the entomopathogenic nematodes to move actively. On other hand (**Kamionek 1979**) reported that the compatibility between some herbicides and insecticides can diminish host seeking ability and diminish reproductive potential without causing significant mortality in exposed nematode populations.

*S. littoralis* feces attracted more portions of the tested nematode populations than the host it self and accordingly caused an increase in the average net distances except in the case of *S. glaseri* strain , since the host attracted the nematode more than feces . This finding confirmed by the work of **Schmidt and All (1978,1979)**. In the laboratories they found

that *S. feltiae* positively responds various stimuli such as CO<sub>2</sub>, thermal gradient, and excretory products in insect feces.

Comparing between *S. littoralis* larvae and *G. mellonella* larvae in attracting various nematode strains, it was found that *G. mellonella* attracted about 80% of Ht population while 35% were directed towards *S. littoralis* larvae. Also the infective stages of the strain S.c preferred the same host with a less degree. On the other hand the strains S.g, S.c.a and HP88, somewhat preferred larvae of *S. littoralis* than the other host. Klein-Beekman *et al.* (1994) found that dispersal of *S. glaseri* juveniles was enhanced in the presence of *Melolontha melolontha* larvae, clear response direction towards the host was not observed. Steiner (1996) reported that unidentified *Steinernema* species and *S. kraussei* exhibited negative migration when using *G. mellonella* larvae as a host. He suggested that *G. mellonella* was repellent to those nematodes. Also, he found that the host finding ability of a strain of *S. feltiae* was smaller for *M. melolontha* than for *G. mellonella*. He attributed this decrease, to that *M. melolontha* feces were repellent to the nematode juveniles.

## II-Filed studies dealing with Biological control of *Zeuzera pyrina* and *Synanthedon myopaeformis* by nematodes:

a)-Effect of nematode concentration and application method on insect morality:

### 1-Effect of Nematode concentration:

This experiment was carried out to evaluate the efficiency of spraying and injection methods of application. Two infested fields of *Z. pyrina* and *S. myopaeformis* were used in this work. The first orchard was in the EL-Monofya Governorate (*Z. pyrina* infested pear orchard) and the second was in EL-Mansoria region. Three nematode strains were applied either by spraying three times in 7-day intervals or by injection one time. The applied nematode species were *S. carpocapsae* and two local *H. bacteriophora* strains D1 and D2. Two concentrations were used (3000, 1500 IJs/ ml distilled water) in each treatment. The obtained results are given in tables (9&10). It could be mentioned that the used two methods, were effective against both tested insects. Mortality of *Z. pyrina* (Table, 9) ranged between 70.7 to 74.5 % in the spraying technique while in the injection method, it ranged between 69.4 to 80.03%. On the other hand, mortality rates of *S. myopaeformis* (Table, 10) ranged between 45.9 to 59.3% in the spraying technique but in the injection method ranged between 53.8 to 76%.

Statistical analysis showed that there were significant differences between the two strains and concentrations in case of *Z. pyrina* but there were no any significant differences between the tested strains and concentrations of *S. myopaeformis*. This finding is in coincidence with that of Bedding and Miller (1981), Deseo *et al.* (1984), Deseo and Docci (1985), Kaya and Brown (1986), Abed EL-Kawy *et al.* (1988), Yang *et al.* (1990), Azazy (1996).

**Table (9)** Effect of three nematode strains on *Z. pyrina* infesting pear trees.

Treatment	Techniques	Active galleries pre treatment	Active galleries post treatment	Concentration		% Mortality	Average
				3000	1500		
<i>S. carpocapse</i> (All)	Spray	119	30	75.3±4.3	73.6±3.8	74.45	77.2 a
	Injection	110	24	82.1±4.3	77.97±4.4	80.03	
<i>H. bacteriophora</i> (D1)	Spray	97	27	75.2±4.7	71.5±7.3	73.4	71.4 a
	Injection	118	25	77.1±1.9	61.7±7	69.4	
<i>H. bacteriophora</i> (D2)	Spray	104	30	75.5±3.4	65.9±3.6	70.7	61.95 b
	Injection	78	41	57.6±7.5	48.8±9.8	53.2	
Average				73.8 a	66.6 b		

Three way ANOVA Completely Randomized

Main Effect	df	F value	P
Technique	1	2.42	.12 ns
Nematode	2	7.00	.0003 **
Concentrations	1	4.6	.033 *
Interactions			
Tech X Nema	2	3.96	.0213 *
Tech X Cons	1	0.43	.514 ns
NemaX Cons	2	0.41	.66 ns
TechX Nema X Cons	2	0.3	.73 ns



Table (10) Effect of three nematode strains on *S. myopaeformis* infesting apple trees.

Treatment	Techniques	Active galleries pre treatment	Active galleries post treatment	Concentration		% Mortality	Average
				3000	1500		
<i>S. carpocapse</i> (All)	Spray	36	17	44.7 ± 16.8	73.8 ± 16.3	59.3	67.7 a
	Injection	48	15	75 ± 13.9	77 ± 15.4	76	
<i>b. bacteriophora</i> (D1)	Spray	60	31	50.7 ± 4.7	51.99 ± 16.8	51.3	52.6 a
	Injection	74	35	59.4 ± 5.9	48.2 ± 13.5	53.8	
<i>b. bacteriophora</i> (D2)	Spray	41	26	31.7 ± 9.3	60.1 ± 20	45.9	58.7 a
	Injection	76	26	76.4 ± 10.5	66.7 ± 10.6	71.6	
Average				56.3 a	62.7 a		

## Three way ANOVA Completely Randomized

Main Effect	df	F value	P
Technique	1	3.92	.05 ns
Nematode	2	1.4	.255 ns
Concentrations	1	0.74	.394 ns
Interactions			
Tech X Nema	2	0.8	.466 ns
Tech X Cons	1	3.22	.079 ns
NemaX Cons	2	0.7	.52 ns
TechX Nema X Cons	2	0.3	.751 ns

Another treatment was carried out at EL-Mansoria region in order to compare the effect of the previous three strains of nematodes on *Z. pyrina* and *S. myopaeformis* on apple trees. In this experiment, two application spraying methods, and injection using two nematode concentrations (3000, 1500 IJs/ ml ds.water) were applied. The obtained data are shown in (Tables 11 & 12). In the spraying method *Z. pyrina* mortality rates ranged between 65.9 to 75.5% but in *S. myopaeformis* mortality percentages ranged between 31.7 to 73.8% (Table, 11). In injection method (Table, 12) the mortality of *Z. pyrina* ranged between 48.8 to 82.2% while the *S. myopaeformis* mortality was 48.2 to 77%.

Statistical analysis showed highly significant differences among the two tested insects in the spraying treatment. On the other hand, no significant differences were found among insects in the case of injection. It was found also that all interaction effects among the treatments were insignificant except in the case of insects and concentration (Table 11), and in case of insects and nematodes (Table 12). Such results are in a full coincidence with the work of **Abed EL-Kawy & EL-Bishry (1992)**, **Azazy (1996)**. There are several reasons which may cause such differences in the control of borers by the steinernematid and heterorhabditis nematodes. The nematode strain may not be as efficacious against the borers or the sesiid larval size may have affected the nematode ability to seek out its host or infect it. It is known that the smaller insects are less susceptible to nematode infection (**Gaugler & Molloy, 1981**, **Kaya, 1985**).

**Table (11)** Comparative studies for controlling *Z. pyrina* & *S. myopaeformis* by entomopathogenic nematodes using spray technique.

Treatment	Concentration	<i>S. myopaeformis</i>				<i>Z. pyrina</i>		
		Active galleries pre treatment	Active galleries post treatment	% Mortality	Active galleries pre treatment	Active galleries post treatment	% Mortality	Average
<i>S. carpocapse</i> (All)	3000	15	9	44.7	46	13	75.3	66.9 a
	1500	21	8	73.8	74	18	73.6	
<i>H. bacteriophora</i> (D1)	3000	38	18	50.7	46	14	75.2	62.4 ab
	1500	22	13	52.2	51	13	71.5	
<i>H. bacteriophora</i> (D2)	3000	19	13	31.7	45	10	75.5	58.3 b
	1500	22	13	60.12	63	20	65.9	
Average				52.2 b			72.8 a	

### Three way ANOVA Completely Randomized

Main Effect	df	F value	P
Insect	1	46.9	.0000 ***
Nematode	2	2.67	.073 ns
Concentrations	1	5.83	.017 *
<b>Interactions</b>			
InsectX Nema	2	0.89	.412 ns
InsectX Cons	1	16.67	.0001 ***
NemaX Cons	2	2.15	.12 ns
InsectX Nema X Cons	2	2.7	.067 ns

**Table (12)** Comparative study for controlling *Z.pyrina* & *S. myopaeformis* by injection entomopathogenic nematodes Using injection technique

Treatment	Concentration	<i>S. myopaeformis</i>			<i>Z. Pyrina</i>			Average
		Active galleries pre treatment	Active galleries post treatment	% Mortality	Active galleries pre treatment	Active galleries post treatment	% Mortality	
<i>S. carpocapse</i> (All)	3000	24	8	75	40	8	82.2	78.07 a
	1500	24	7	77	70	16	77.9 7	
<i>H. bacteriophora</i> (D1)	3000	43	22	59.4	71	15	77.7	61.6 b
	1500	31	17	48.2	41	14	61.7	
<i>H. bacteriophora</i> (D2)	3000	35	10	76.4	46	26	57.6	62.4 b
	1500	41	16	66.7	32	15	48.8	
Average				67.1 a			67.7 a	
Three way ANOVA Completely Randomized								
Main Effect		df	F value	P				
Insect		1	0.022	.882 ns				
Nematode		2	10.96	.0000 ***				
Concentrations		1	5.90	.016 *				
Interactions				.0001 ***				
InsectX Nema		2	9.43	.629 ns				
InsectX Cons		1	0.23	.291 ns				
NemaX Cons		2	1.244	.905				
InsectX Nema X Cons		2	0.099					

## 2-Effect of Application method:

Comparing between four entomopathogenic nematodes, three methods of application and one concentration of nematode suspension (1000 IJs/ml ds.water) were tried in controlling *S. myopaeformis* and *Z. pyrina* on apple trees in EL-Mansoryia region. Two species and four strains of nematodes were used. These strains were *S. carpocapsae* (All), *H. bacteriophora* (HP88), *H. bacteriophora* (AR-4) and *H. bacteriophora* (Ht). Three application techniques were applied; namely spraying, injection and the cotton plug technique. Data in table (13) showed that the average mortality rates ranged between 47.7 to 69.3% of *S. myopaeformis*.

Statistical analysis showed significant differences among the employed techniques, but no significant differences were found among nematode species. All interaction effects among the treatments were not significant. The strains of *Heterorabditis* sp. were more effective than the *S. carpocapsae*. Spray and injection techniques, were statistically equal in their effect, giving larval mortalities of 46.4 and 59.6%, respectively. On the other hand, the cotton plugs technique induced 68.4% larval mortality.

Regarding to *Z. pyrina*, it was found that mortality rates ranged between 71.39 to 94.13% depending on the nematode species and application method.

**Table (13)** Comparison between application techniques for controlling *Z. pyrina* and *S. myopaeformis* on apple tree with four strains

Treatment	Mortality $\pm$ SE							
	<i>Z. pyrina</i>			<i>S. myopaeformis</i>				
	Spray	Injection	Cotton	Average	Spray	Injection	Cotton	Average
<i>H. bacteriophora</i> (AR-4)	89.1 $\pm$ 9.8	88.2 $\pm$ 3.2	87.5 $\pm$ 6.2	88.3 a	63.6	70.7 $\pm$ 6.6	73.8 $\pm$ 8.7	69.3 a
<i>H. bacteriophora</i> (HP88)	91.7 $\pm$ 6.5	96.4 $\pm$ 3.6	91.7 $\pm$ 4.6	94.1 a	59.5 $\pm$ 13.7	71.9 $\pm$ 8.1	68 $\pm$ 9.7	66.5 ab
<i>S. carpocapsae</i> (All)	87.8 $\pm$ 9.7	62.1 $\pm$ 16.8	95.2 $\pm$ 2.6	81.7 ab	20 $\pm$ 12.2	44.3 $\pm$ 19.7	82.2 $\pm$ 5.3	48.8 ab
<i>H. bacteriophora</i> (H t)	83.3 $\pm$ 9.1	61.1 $\pm$ 6	69.8 $\pm$ 5.8	71.4 b	42.3 $\pm$ 14.3	51.4 $\pm$ 4.3	49.5 $\pm$ 21.2	47.7 ab
Average	87.98 a	76.95 b	86.1 ab		46.4 b	59.6 a b	68.4 a	

**Tow way ANOVA Completely Randomized**

Main Effect	df	<i>Z. pyrina</i>		<i>S. myopaeformis</i>	
		F value	P	F value	P
Technique	2	2.54	.089 ns	3.34	.044 *
Nematode	3	4.71	.006 **	2.65	.059 ns
Interactions					
Tech X Nema	6	1.52	.191 ns	1.32	.266 ns

Statistical analysis showed that both of the cotton plug and the spray techniques, were the superiors, while injection came in the last. Also it was found that Ar-4 and HP88 strains of *H. bacteriophora* gave higher mortality rates than (Ht) strain of *H. bacteriophora* and *S. carpocapsae* (All). All interaction effects also, among the different treatments were insignificant. However, cotton plugs and the spray techniques, were statistically equal in their effect giving larval mortalities of 86.1 and 87.98%, respectively. On the other hand, the injection technique induced 76.95% larval mortality.

#### **b- Comparative effect of both entomopathogenic nematodes and pesticides:**

Comparing nematodes and recommended insecticides for controlling *S. myopaeformis*, on apple trees, in EL-Mansoria region, in 1997 season, two concentrations of three species of entomopathogenic nematodes were evaluated. Data in Table (14) showed that combinations of the insecticide (Phenthoate) and the three tested species of nematodes were the best, giving the greatest values of mortality. Average percentage of mortalities were 75.03, 79.9 and 68.7. Combinations of Phenthoate (Cedial L60) with *S. carpocapsae* and *H. bacteriophora* (D1, D2) gave high mortality rates than using either nematodes or insecticides alone. Both nematodes and insecticides alone achieved least mortalities, which ranged between 45.9 to 59.23%. Combination of Diazinon (Bazodin) and nematode *S. carpocapsae* caused 69.7% mortality, while this rate was 49.63, when the same

**Table (14)** Comparison between entomopathogenic nematodes & insecticides on controlling *S. myopaeformis* on apple trees.

Treatment	Insecticides	Concentration	Active galleries pre treatment	Active galleries post treatment	% Mortality $\pm$ SE	Average
<i>S. carpocapsae</i> (All)	Alone	3000	15	9	44.7 $\pm$ 16.8 bc	59.23
		1500	21	8	73.8 $\pm$ 16.3 ab	
	Phenthoate	3000	27	7	73.7 $\pm$ 8.8 ab	75.03
		1500	37	11	76.4 $\pm$ 11.2 ab	
	Diazinon	3000	34	16	53.1 $\pm$ 1.7 abc	69.7
		1500	17	4	86.4 $\pm$ 8.8 a	
<i>H. bacteriophora</i> (D1)	Alone	3000	38	18	50.7 $\pm$ 4.7 abc	51.4
		1500	22	13	52 $\pm$ 16.8 abc	
	Phenthoate	3000	44	13	75.9 $\pm$ 6.9 ab	79.9
		1500	19	4	84 $\pm$ 11.7 ab	
	Alone	3000	19	13	31.7 $\pm$ 9.3c	45.9
		1500	22	13	60.1 $\pm$ 12abc	
<i>H. bacteriophora</i> (D2)	Phenthoate	3000	36	14	63.8 $\pm$ 10.6abc	68.7
		1500	31	8	73.7 $\pm$ 8.8ab	
	Diazinon	3000	25	12	65.9 $\pm$ 16abc	49.6
		1500	33	23	33.3 $\pm$ 5.6c	
	Alone		29	15	34.3 $\pm$ 2.8 c	51
			28	10	48.3 $\pm$ 9.3 bc	

One way ANOVA Completely Randomized

Main Effect      **df**      **F value**      **P**  
 Treatment      17      2.36      .0061 \*\*  
 L.S.D (.05)= 31.93



insecticide was combined with the D2 strain of *H. bacteriophora*. In general it could be stated that, mixing of nematodes and insecticides yielded high percentage of mortalities, than using nematodes or insecticides alone.

Statistical analysis showed significant differences among the treatments.

Comparing the performance of both the entomopathogenic nematodes, *S. carpocapsae* (All), *H. bacteriophora* (HP88), *H. bacteriophora* (AR-4) and *H. bacteriophora* (Ht) alone or combined with conventional insecticides had been evaluated in another experiment against *S. myopaeformis* and *Z. pyrina* on apple trees in 1998 season. Data of this experiment are shown in Tables (15,16). *S. myopaeformis* mortality rates resulted by nematode strains ranged between (20-63.6%) and when combined with insecticides achieved mortality rates ranged between 34.3 and 64.3%, while the insecticides alone achieved 30 to 31.3% control of *S. myopaeformis*. Generally, nematode strains induced percentage average reductions in *S. myopaeformis* of 27.3,55.1,37 and 49.3% for (AR-4), HP88, *S. carpocapsae*, and (Ht), respectively (Table 15).

Statistical analysis showed no significant differences among treatments. It could be mentioned that there were no clear differences between using entomopathogenic nematodes alone or combined with the insecticides on mortality rates of this pest.

**Table (15)** Comparison between entomopathogenic nematodes and insecticides in controlling *S. myopaeformis* on apple trees.

Nematodes	Insecticides	Active galleries pre treatment	Active galleries post treatment	% Mortality $\pm$ SE	Average
<i>H. bacteriophora</i> (AR-4)	Alone	12	12	63.6 b	27.3
	Phenthoate	22	17	18.3 $\pm$ 7.6 ab	
	Diazinon	54	19	63.6 $\pm$ 7.6 a	
<i>H. bacteriophora</i> (HP88)	Alone	23	9	59.5 $\pm$ 13.7 a	55.1
	Phenthoate	17	8	64.3 $\pm$ 18.4 a	
	Diazinon	24	11	41.5 $\pm$ 22.3 ab	
<i>S. carpocapse</i> (All)	Alone	13	10	20 $\pm$ 12.2 ab	37
	Phenthoate	18	10	34.3 $\pm$ 11.3 ab	
	Diazinon	16	8	56.7 $\pm$ 19.4 a	
<i>H. bacteriophora</i> (Ht)	Alone	26	16	42.3 $\pm$ 14.3 ab	49.3
	Phenthoate	26	8	60.8 $\pm$ 17.5 a	
	Diazinon	25	12	44.7 $\pm$ 13.5 ab	
Phenthoate	Alone	11	8	31.3 $\pm$ 13.4 ab	30.7
Diazinon	Alone	32	15	30 $\pm$ 13.3 ab	

One way ANOVA Completely Randomized

Main Effect	df	F value	P
Treatment	13	1.32	.231 ns

L. S. D ( 0.05 ) = 40.8

**Table (16)** Comparison between entomopathogenic nematodes and insecticides in controlling *Z. pyrina* on apple trees.

Nematodes	Insecticides	Active galleries pre treatment	Active galleries post treatment	% Mortality $\pm$ SE	Average
<i>H. bacteriophora</i> (AR-4)	Alone	10	1	90 $\pm$ 10 a	81.7
	Phenthoate	66	12	79.3 $\pm$ 5.1 a	
	Diazinon	53	14	75.7 $\pm$ 4.2 a	
<i>H. bacteriophora</i> (HP88)	Alone	10	1	94.3 $\pm$ 3.9 a	85.6
	Phenthoate	40	10	79.8 $\pm$ 7.2 a	
	Diazinon	43	16	82.8 $\pm$ 10.6 a	
<i>S. carpocapse</i> (All)	Alone	31	4	87.8 $\pm$ 9.7 a	77.6
	Phenthoate	65	14	77.4 $\pm$ 7.2 a	
	Diazinon	35	12	67.7 $\pm$ 5.8 a	
<i>H. bacteriophora</i> (Ht)	Alone	13	1	83.3 $\pm$ 9.1 a	64.8
	Phenthoate	15	8	36.7 $\pm$ 15.3 b	
	Diazinon	22	4	74.3 $\pm$ 11.2 a	
Phenthoate	Alone	39	34	31.3 $\pm$ 13.4 b	30.7
Diazinon	Alone	41	39	30 $\pm$ 13.3 b	

One way ANOVA Completely Randomized

Main Effect df 13

Treatment

F value 4.879

P

.0000 \*\*\*

L. S. D ( 0.05 ) = 27.6

The same nematode strains yielded contradictory percentages of mortality against *Z.pyrina* when combined with the same pesticides. It is clear that using nematodes alone achieved higher mortality rates than that gained by combination of nematodes and insecticides. As shown in (Table 16) high mortality rates ranged between 83.3 and 94.3% when the nematodes used alone. Combination between the nematodes and insecticides achieved mortality rates ranged between 36.7 to 79.3% but using insecticides alone achieved 30 to 31.3% control of *Z. pyrina*.

Statistical analysis showed highly significant differences among treatments. It could be mentioned that the tested entomopathogenic nematodes alone was more effective than the insecticides alone or when combined with the entomopathogenic nematodes. This finding is in a full agreement with those found by Solomon (1985), Deborah *et al* (1996). The greater persistence of the nematode than pesticide was attributed to nematode propagation inside the tree. It can be that some larvae die within the tree and the nematodes propagate within them, producing new generation(s) which are able to survive and locate and then infect cossid larvae, that escaped from infection of the initial treatment Yang *et al* (1989).

**c)- Comparative effect of both entomopathogenic nematodes and *Bacillus thuringiensis* in controlling *Z.pyrina*:**

This experiment was carried out in two different seasons (autumn and spring of, 1999) in the farm of EL-Marowa

(EL-Giza Governorate) on apple trees. Three strains of entomopathogenic nematodes were applied using injection technique either alone or in combination with the recommended dose of *B. thuringiensis*. Another treatment was carried out using the bacterium individually. The mean of temperature degrees was 20.9 and 26.1°C in autumn and spring, respectively. As shown in (Table 17), it could be stated that using the pathogenic 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* + *B. thuringiensis* in the spring. It could also stated that the strain *S. carpocapsae* (all) was more effective against the borer in autumn than in spring. On other hand, the strain of *H. bacteriophora* was 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*.

Statistical analysis revealed that there was no any significant difference between treatments in the autumn. In spring, no significant differences were found between nematode strains, although some degree of significance was found between the nematode strains and the bacterium. On the other hand, high degree of significance was found between the nematode strains and combinations of nematodes and bacterium. This finding is in harmony with that of **Bari and Kaya (1984)** who indicated that combination of nematode and bacterium did not result in

**Table (17)** Infectivity of some entomopathogenic nematodes alone or combined with *B. thuringiensis* on *Z. pyrina* in two different seasons.

Treatment	% Mortality $\pm$ SE				
	Spring		Average	Autumn	
	Alone	+B.t		Alone	+B.t
<i>S. carpocapsae</i> (All)	62.1 $\pm$ 4.9	73.4 $\pm$ 6.2	67.7 a	79.8 $\pm$ 8.7	69.1 $\pm$ 3.2
<i>H. bacteriophora</i> (HP88)	77.5 $\pm$ 3.4	62.2 $\pm$ 4.5	70.01 a	73.5 $\pm$ 8.3	51.7 $\pm$ 13.9
<i>S. carpocapsae</i> (agrotis)	76.8 $\pm$ 6	58.3 $\pm$ 5.1	67.5 a	62.6 $\pm$ 4.9	55.4 $\pm$ 6.6
<i>B. thuringiensis</i>	30.3 $\pm$ 7.5			10 $\pm$ 4.4	
<b>Average</b>	<b>72.1 a</b>	<b>64.7 b</b>		<b>72 a</b>	<b>58.7 a</b>

**Tow way ANOVA Completely Randomized**

Main Effect	df	Spring F value	P	Autumn F value	P
Nematode +B.t	1	5.2	.0253 *	3.81	.063 ns
Nematode	2	0.38	.789 ns	1.99	.273 ns
<b>Interactions</b>					
Comb X Nema	2	8.3	.0005 ***	0.417	.664 ns

significantly greater control than that achieved by the nematode alone Deseo *et al* (1984), EL-Bishry and Bekheit (1994), Gill and Michael (1994), Azazy (1996), Pasqualimi *et al* (1996) gained similar results.

**d)- Influence of temperature on infectivity of some pathogenic nematode strains :**

This experiment was carried out in two different seasons, autumn and spring of 1999, in EL-Marwa farm (EL-Giza governorate). Mean temperature during experimental period, was 21.1°C at November and 26.1°C at April. Three nematode strains were applied against leopard moth with two techniques (injection and cotton plugs).

Data in Table (18) shows that the strain, *S. carpocapsae* (All) in November (21°C) tended to give better control than in May (26.1°C) because of the sensitivity of such nematode species to high temperatures. However, the two strains of *H. bacteriophora* (HP88) and *H. bacteriophora* achieved mortality rates of 67.95% and 74.8%, respectively in November. However, in high temperatures of May *Heterhabditis* sp were the better in killing *Z. pyrina* larvae which achieved mortalities of 77.4 and 78.1 against 62.3 % for *S. carpocapsae*. Results indicated that, there was no significant differences between the two applied techniques (injection and cotton plugs) in the two seasons (autumn and spring) in insect mortality rates. On the other hand significant differences were found among the nematode strains. However, it could be recommended to use the

**Table (18)** Effect of tow different temperature degrees on the infectivity of some entomopathogenic nematodes on *Z. pyrina*.

Nematodes	% Mortality $\pm$ SE					
	26.1°C			21.1°C		
	Cotton	Injection	Average	Cotton	Injection	Average
<i>S. carpocapsae</i> (All)	62.5 $\pm$ 4.7	62.1 $\pm$ 5	62.3 b	91.1 $\pm$ 2.7	82.9 $\pm$ 3.1	87 a
<i>H. bacteriophora</i> (HP88)	77.3 $\pm$ 4.9	77.5 $\pm$ 3.4	77.4 a	66.3 $\pm$ 3.1	69.6 $\pm$ 7.6	67.95b
<i>H. bacteriophora</i> (Ht)	76.1 $\pm$ 4.6	80.1 $\pm$ 5.2	78.1 a	79.6 $\pm$ 4.1	70 $\pm$ 5.2	74.8 b
Average	72.3 a	73.3 a		79 a	73 a	

**Tow way ANOVA Completely Randomized**

Main Effect	df	26.1°C		21.1°C	
		F value	P	F value	P
Technique	1	0.05	.81 ns	2.2	.144 ns
Nematode	2	5.95	.005 **	7.6	.0015 **
Interactions					
Tech X Nema	2	0.2	.85 ns	1.4	.25 ns



strains of *S. carpocapsae* against the pest in winter, and autumn and the strains of *H. bacteriophora* in spring. This finding is in a full coincidence with that of nematodes at each temperature. **Kung et al. (1991)** found that pathogenicity and survival of *S. carpocapsae* was, significantly greater at lower temperatures (5-25°C) than at the highest temperatures (15-35°C). **Zervos et al. (1991)** reported that *H. heliothidis* activity against *G. mellonella* larvae was inhibited below 10°C and significantly decreased at 30°C.