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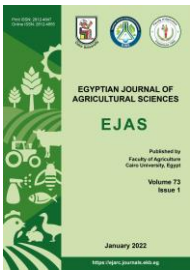

Biocontrol of *Meloidogyne incognita* Using Entomopathogenic Nematodes and their Symbiotic Bacteria under Greenhouse Conditions

By

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ABSTRACT

Ten entomopathogenic nematode (EPN) strains, five different types of bacteria, oils and nematicide (Vydate) were tested in pots for their antagonistic effects against *Meloidogyne incognita* (galling, egg masses production and soil population) on eggplant plants. One strain of *Steinernema abbasi* (S.3), *S. arenarium* (S.RIO), and three strains of *S. carpocapsae* were used in greenhouse studies. Additionally, pots containing 2000 RKN J2s were treated with three *Heterorhabditis indica* strains, two *H. bacteriophora* strains, and the associated bacterium (*Photorhabdus* sp.). There was a significant reduction in *M. incognita* densities in all EPN's treatments when compared to the check. EPNs and *Photorhabdus* bacterium decreased soil population up to 99.6 % when treated with *S. arenarium* (S.RIO) and 99.2 % reduction occurred by *H. indica* (3 MANGO) treatment and up to 99.5 % soil population reduction in MANGO3 BACTERIA (*Photorhabdus* sp.) compared to the check. The findings also showed a significant reduction in the number of galls (-46.5% reduction) and egg masses (-65.2% reduction) as compared to the check for the S.3 (*S. abbasi*) strain.

Regarding the plant growth criteria, a decrease was seen in all *Steinernema* treatments compared to the check, with the exception of the shoot fresh weight and plant length of ATS and all plant growth criteria of S.3, S.RIO isolates, and Vydate when compared with the nematode alone. Except for CITRUS 5 isolates, which showed insignificant count reduction in shoot fresh and dry weights but the contrary was seen in root fresh weight. *Heterohabditis* treatments enhanced shoot fresh and dry weights.

Key words: *Meloidogyne incognita, eggplant, entomopathogenic nematodes, Photorhabdus* sp**.**

1. INTRODUCTION

In Egypt, one of the most common vegetable crops is eggplant (*Solanum melongena*). It is plagued by a variety of pests, of them, the root-knot nematode (RKN) *Meloidogyne* spp. This main group of plant-parasitic nematodes is responsible for significant economic losses globally, particularly in Egypt (Elkelany *et al.*, 2020; Heflish *et al.*, 2021; Tauseef *et al.*, 2021; Ahmad *et al.*, 2021).

Microscopically small roundworms known as entomopathogenic nematodes (EPNs) are members of the Heterorhabditidae and Steinernematidae families of the phylum Nematoda. Beneficial

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nematodes known as EPNs display a parasitic method of survival (Bhat *et al.* 2020).

The bacto-helminth parasites known as entomopathogenic nematodes (EPNs) exhibit classic mutualism with the genera *Xenorhabdus* and *Photorhabdus*. The nematodes and the bacteria have an endosymbiotic relationship which have the potential to reduce the use of chemical pesticides. The use of bioagents significantly reduced the rat of infections. These bioagents are also simple to be used, eco-friendly, and biodegradable, protecting crops from a variety of harmful organisms. Tomar *et al.(*2022).

Entomopathogenic bacteria (EPB) have been successfully employed to regulate nematodes without endangering the environment. The symbiotic bacteria linked to the entomopathogenic nematodes of the genera *Steinernema* and *Heterorhabditis*, respectively, include *Xenorhabdus* spp. and *Photorhabdus* spp. The potential of bacterial symbionts to be used for the management of agriculturally important pests has been the focus of studies of the virulence mechanisms and secondary metabolic features of *Xenorhabdus* and *Photorhabdus* bacteria (Hinchliffe *et al.* 2010; Zhang *et al.* 2012; Kumari *et al.* 2015; Stock *et al.* 2017 ;Lulamba *et al.,* 2021; Tomar *et al.,* 2022). EPB can preserve ecological changes while reducing the usage of chemicals for plant protection (Migunova and Sasanelli, 2021). Our research aimed to evaluate different strains of *Steinernema* and *Heterorhabditis*, as well as bacteria associated to *Heterorhabditis*, as potential biocontrol agents for the root-knot nematode *Meloidogyne incognita* compared with oils and Vydate as a nematicide.

2. MATERIALS AND METHODS

2.1. Nematodes

Pure culture of root-knot nematode, *M. incognita* was obtained from isolates belonging to branch of Nematology, Zoology and Agricultural Nematology Department, Faculty of Agriculture, Cairo University (https://goo.gl/maps/JwVSt8W7ATJ9VvDJ6) propagated on Tomato cv. Super strain B.

2.2. Treatments and doses

2.2.1. Entomopathogenic nematodes and isolation of *Photorhabdus* sp. bacteria

The entomopathogenic nematodes of genera *Steinernema* and *Heterorhabditis* were isolated and mass cultured at the Applied Center for Entomonematodes (ACE), Department of Zoology and agricultural Nematology, Faculty of Agriculture, University of Cairo, Giza, Egypt.

For each EPN isolate, bacteria were extracted from a pool of 500 freshly emerged IJs which were disinfected by immersing them in a 10% sodium hypochlorite solution for 10 min. Bacteria were grown on NBTA (bromothymol blue) agar following procedures described by Akhurst (1980). Isolates were examined for the main phenotypic characteristics of the genus *Xenorhabdus*, using the methods of Boemare and Akhurst (1988). DNA extraction of bacterial symbionts was performed according to Tailliez *et al.* (2006). The small subunit (16S) of rDNA was amplified with primers and PCR conditions that followed procedures described by Tailliez *et al*. (2006).

Ten treatments of entomopathogenic nematodes and 5 types of bacteria isolated from Heterorabtitis nematode & oils and nematicide (Vydate) as a check listed in Table (1).

2.3. Greenhouse experiments

2.3.1. Plant preparation

One month old clean seedlings of eggplant cultivar (Hanen) with uniform size were obtained from Horticulture Research Institute, Agriculture Research Center, Giza, Egypt (https://goo.gl/maps/8FYWyMnvbEFmGfYM9) and cultivated singly in 15 cm diameter earthen pots filled with steam-sterilized sandy loam soil (1:1, v/v).

2.3.2. Experiment

The effect of 17 treatments of Entomopathogenic nematodes and of bacteria isolated from *Heterorabtitis* nematode and oils as well as nematicide (Vydate) were tested against the root-knot nematode, *M. incognita*. Each Seedling in pot 15cm diameter was inoculated with 2000 J2 of *M. incognita*. One week after inoculation, each seedling was treated as previously mentioned as soil drench. All treatments were replicated 4 times. Two

Table (1): List of entomopathogenic nematodes and associated bacteria applied as soil drenches and their doses.

Treatments	Dose
S.3 (*Steinernema abbasi*)	2000 IJs/ ml
ATS (*Steinernema carpocapsae*)	2000 IJs/ ml
AT4 (*Steinernema carpocapsae*)	2000 IJs/ ml
AT5 (*Steinernema carpocapsae*)	2000 IJs/ ml
S.RIO (*Steinernema arenarium)*	2000 IJs/ ml
3 MANGO (*Heterorhabditis indica*)	2500 IJs/ ml
2MANGO (*Heterorhabditis indica*)	2500 IJs/ ml
4MANGO(*Heterorhabditis indica*)	2500 IJs/ ml
HB88 (*Heterorhabditis bacteriophora*)	2500 IJs/ ml
CITRUS5 (*Heterorhabditis bacteriophora*)	2500 IJs/ ml
MANGO3 BACTERIA (*Photorhabdus* sp.)	5000 cell/ ml
CITRUS 3 BACTERIA (*Photorhabdus* sp.)	5000 cell/ ml
HB88 BACTERIA (*Photorhabdus* sp.)	5000 cell/ ml
MANGO2 BACTERIA (*Photorhabdus* sp.)	5000 cell/ ml
MANGO4 BACTERIA (*Photorhabdus* sp.)	5000 cell/ ml
Oils mixtures (Mineral oil 50%, Citronella 20%, Jasmine oil 20%, emulsifier 10%)	100 ml/ L
Vydate 24% SL Nematicide (Oxamyl 24%)	0.4ml/plant as a soil drench

treatments, healthy (without either nematode or treatment) and infected (with nematode only) were kept as checks. Pots were labeled and arranged in a complete randomized design on a clean bench, receiving similar horticulture treatments.

After 45 days from inoculation, plant growth criteria were recorded and nematode population in soil pots were extracted by means of Hooper *et al.,* 2005. Nematodes on stained roots were counted according to Goody 1957. The nematode final population (Egg-masses + nematodes in soil) was calculated.

The percentage change in each nematode population and plant growth criteria was measured using the formula:

$$\% \text{ Change} = \frac{\text{Treatment} - \text{Check (nematode only)}}{\text{Check (Nematode only)}} \times 100$$

2.3.3. Statistical analysis

Differences among treatments were determined with Analysis of Variance using SPSS (2015) statistical package. Whenever significant differences were detected, means were separated using least significant Difference test (LSD) at 5% level of significance.

3. RESULTS

The influence of the EPNs isolates *(Steinernema*) on the root knot nematode soil and root population is shown in Table 2. The number of *M. incognita* soil population (J2) was significantly reduced across the different treatments when compared to the check with no significantl differences. *Steinernema abbasi (*S.3) , Oils and Vydate signifcantly reduced the number of galls and egg-masses with no differences between them (Fig. 1).

Table 3 and Fig. 2 showed the effect of *Heterorhabditis* isolats on the nematode population. The highly root population observed on CITRUS5 (*Heterorhabditis bacteriophora*) but the highly reduction were seen in soil population in all treatments up to 96 % reduction compared to nematode only.

All isolates of *Heterorhabditis* increased the numbers of galls and eggmasses in the root, except in 3 MANGO (*Heterorhabditis indica*) and 4MANGO (*Heterorhabditis indica*) isolate were slightly reduced (18.8 and 25.7).

The isolated bacteria (*Photorhabdus* sp.) were tested on the number of *Meloidogyne incognita.* Numbers of galls, egg-masses and soil population are illustrated in Table 4 and Fig. 3. Data showed that all isolates reduced the number of egg-masses and soil population but the highly reductions were seen in oils and vydate treatments. The opposite was observed in the number of galls as the highly numbers were shown in (MANGO3 BACTERIA (*Photorhabdus* sp.), HB88 BACTERIA (*Photorhabdus* sp.), MANGO2 BACTERIA *(Photorhabdus* sp.) and nematode only treatment with no significant differences among them.

Table (2): Influence of *Steinernema* isolats on root-knot nematode infecting eggplant under greenhouse conditions after 45 days from inoculation.

Treatments	No.of galls/root	% change	No. of egg masses/root	% change	Soil population	% change
S.3 (*Steinernema abbasi*)	310.5 d	-46.5	189.5 d	-65.2	1332.0 b	-96.9
ATS (*Steinernema carpocapsae*)	506.0 c	-12.8	250.0 d	-54.1	1628.0 b	-96.2
AT4 (*Steinernema carpocapsae*)	577.0 c	-0.5	659.0 b	21.0	555.5 b	-98.7
AT5 (*Steinernema carpocapsae*)	1055.5 a	82.0	826.5 a	51.8	1137.8 b	-97.3
S.RIO (*Steinernema arenarium)*	886.0 b	52.8	476.5 c	-12.5	156.0 b	-99.6
OILS	299.3 d	-48.4	271.3 d	-50.2	134.5 b	-99.7
Vydate	242.5 d	-58.2	210.5 d	-61.3	95.5 b	-99.8
NEMATODE ONLY	580.0 c	0.0	544.5 c	0.0	42730 .5 a	0.0

Means followed by the same letter(s) within a column are not significantly different ($p \leq 0.05$) according to Duncan's' multiple range test.

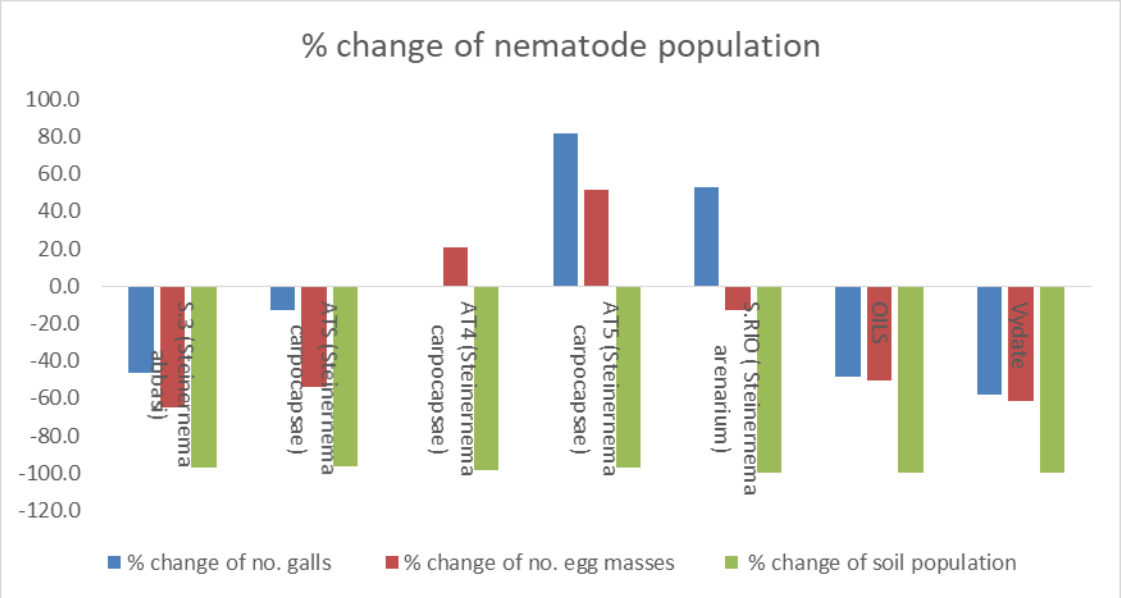

Fig. (1): Reduction percentage of root knot nematode (galls, egg masses and soil population) affected with *Steinernema* isolates under greenhouse conditions.

Table (3): Influence of *Heterorhabditis* sp. isolates on root-knot nematode populations infecting eggplant under greenhouse conditions after 45 days inoculation.

Treatments	No.of galls/root	% change	No. of egg masses/root	% change	Soil population	% change
3 MANGO (*Heterorhabditis indica*)	747.0 c	28.8	442.0 c	-18.8	337.8 b	-99.2
2MANGO (*Heterorhabditis indica*)	941.5 b	62.3	585.0 b	7.4	901.5 b	-97.9
4MANGO(*Heterorhabditis indica*)	785.0 c	35.3	404.5 c	-25.7	829.0 b	-98.1
HB88 (*Heterorhabditis bacteriophora*)	769.0 c	32.6	547.5 b	0.6	696.8 b	-98.4
CITRUS5 (*Heterorhabditis bacteriophora*)	1576.0 a	171.7	1370.0 a	151.6	1713.0 b	-96.0
OILS	299.3 e	-48.4	271.3 d	-50.2	134.5 b	-99.7
Vydate	242.5 e	-58.2	210.5 d	-61.3	95.5 b	-99.8
NEMATODE ONLY	580.0 d	0.0	544.5 b	0.0	42730 .5 a	0

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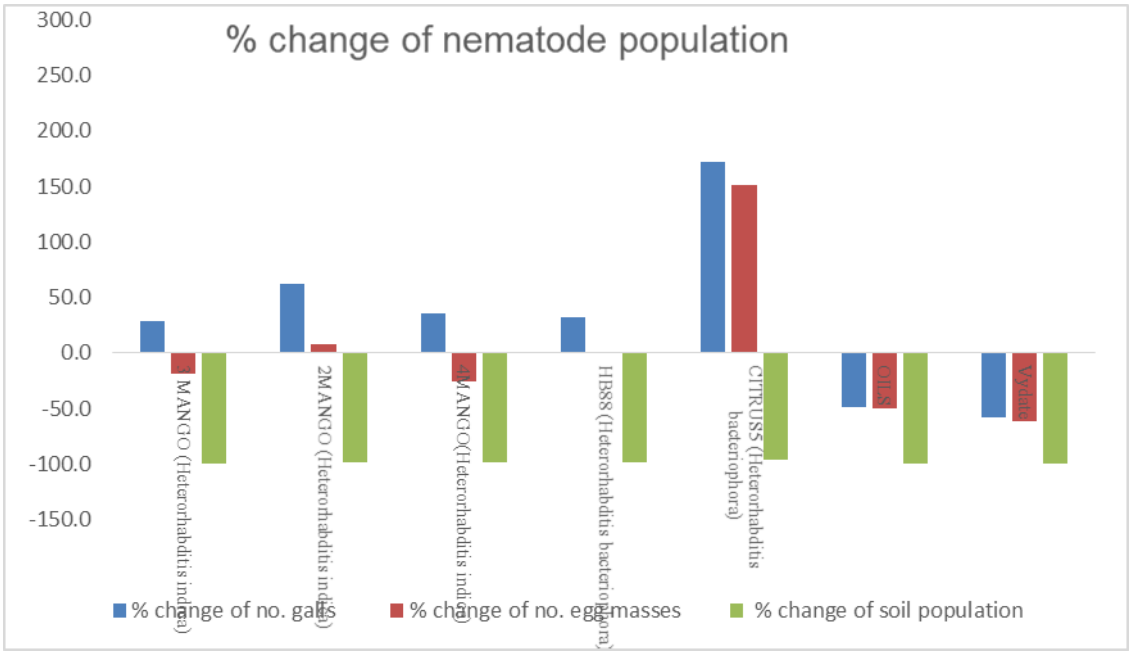

Fig. (2): Reduction percentage of root knot nematode (galls, egg masses and soil population) influenced with *Heterorhabditis* isolates in greenhouse conditions.

Table (4): Influence of *Photorhabdus* sp. bacteria isolated from *Heterorhabditis* sp. on root-knot nematode populations infecting eggplant in greenhouse conditions after 45 days inoculation.

Treatments	No.of galls/root	% change	No. of egg masses/root	% change	Soil population	% change
MANGO3 BACTERIA (*Photorhabdus* sp.)	598.0 ab	3.1	520.5 a	-4.4	226.0 c	-99.5
CITRUS 3 BACTERIA (*Photorhabdus* sp.)	543.0 b	-6.4	392.5 bc	-27.9	5037.0 b	-88.2
HB88 BACTERIA (*Photorhabdus* sp.)	667.0 a	15.0	398.0 bc	-26.9	506.0 c	-98.8
MANGO2 BACTERIA (*Photorhabdus* sp.)	578.5 ab	-0.3	473.0 ab	-13.1	3686.0 b	-91.4
MANGO4 BACTERIA (*Photorhabdus* sp.)	408.5 c	-29.6	322.5 de	-40.8	435.3 c	-99.0
OILS	299.3 d	-48.4	271.3 e	-50.2	134.5 c	-99.7
Vydate	242.5 d	-58.2	210.5 de	-61.3	95.5 c	-99.8
NEMATODE ONLT	580.0 ab	0.0	544.5 a	0.0	42730.5 a	0.0

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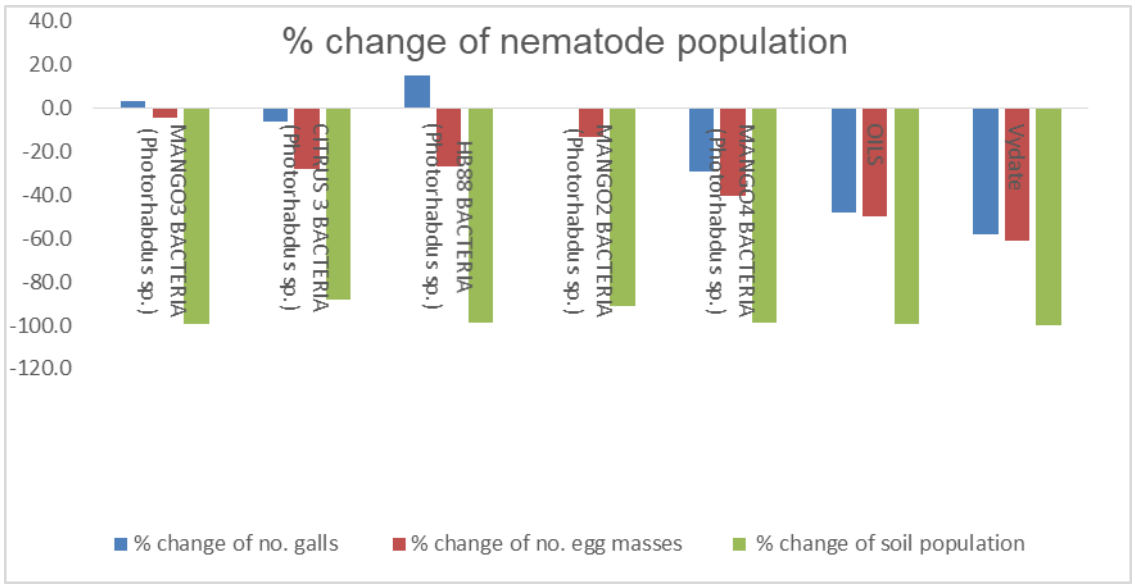

Fig. (3): Reduction percentage of root knot nematode (galls, egg masses and soil population) influenced with *Photorhabdus* bacteria in greenhouse conditions.

3.1. Plant growth criteria

Concerning the plant growth criteria, a reduction in all *steinernema* isolates treatments was showed when compared with check except shoot fresh weight and plant length of ATS and all plant growth criteria of S.3 , S.RIO isolates and Vydate compared with the nematode only (Table 5 and Fig. 4). *Heterorhabditis* isolates treatments (Table 6 and Fig. 5) increased shoot fresh and dry weight except CITRUS 5 isolates showed slight reduction but the opposite was observed in root fresh weight. The oils reduced all plant growth criteria but Vydate increased them. Regarding the effect of the isolated bacteria on parameters

Table (5): Effect of *Steinernema* isolates on the growth of eggplant infected with *M. incognita* after 45 days inoculation.

Treatments	Shoot fresh weight (g)	% change	shoot dry weight (g)	% change	root fresh weight (g)	% change	Plant length (cm)	% change
S.3 (*Steinernema abbasi*)	17.6 b	21.4	3.3 ab	17.9	7.1 ab	0.0	59.7 ab	2.1
ATS (*Steinernema carpocapsae*)	17.8 b	22.8	2.6 bcd	-7.1	4.0 c	-43.7	64.8 a	10.8
AT4 (*Steinernema carpocapsae*)	14.3 b	-1.4	2.3 cd	-17.9	5.6 bc	-21.1	52.3 b	-10.6
AT5 (*Steinernema carpocapsae*)	21.3 a	46.9	2.7 bc	-3.6	5.6 bc	-21.1	58.2 ab	-0.5
S.RIO (*Steinernema arenarium)*	19.5 a	34.5	3.4 ab	21.4	7.2 ab	1.4	65.8 a	12.5
OILS	10.5 c	-27.6	1.7 d	-39.3	5.7 bc	-19.7	54.3 b	-7.2
Vydate	20.4 a	40.7	3.7 ab	32.1	8.4 a	18.3	60.5 ab	3.4
NEMATODE ONLY	14.5 b	0.0	2.8 bc	0.0	7.1 ab	0.0	58.5 ab	0.0

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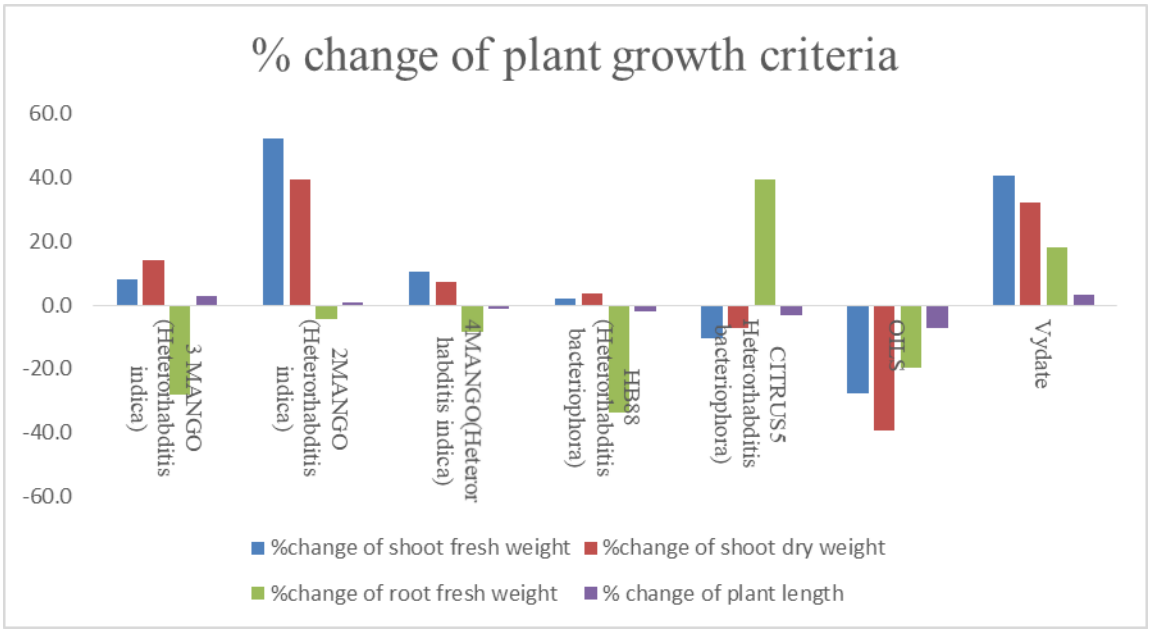

Fig. (4): Percentage reduction of plant growth infected with root knot nematode and treated with *Heterorhabditis* isolates

Table (6): Effect of *Heterorhabditis* isolats on the growth of eggplant infected with *M. incognita* after 45 days inoculation.

Treatments	Shoot fresh weight (g)	% change	shoot dry weight (g)	% change	root fresh weight (g)	% change	plant length (cm)	% change
3 MANGO (*Heterorhabditis indica)*	15.7 b	8.3	3.2 abc	14.3	5.1 e	-28.2	60.1 a	2.7
2MANGO (*Heterorhabditis indica)*	22.1 a	52.4	3.9 a	39.3	6.8 cd	-4.2	59.0 a	0.9
4MANGO(*Heterorhabditis indica)*	16.0 b	10.3	3.0 abc	7.1	6.5 cd	-8.5	57.9 a	-1.0
HB88 (*Heterorhabditis bacteriophora*)	14.8 b	2.1	2.9 bc	3.6	4.7 e	-33.8	57.3 a	-2.1
CITRUS5 *Heterorhabditis bacteriophora*)	13.0 bc	-10.3	2.6 c	-7.1	9.9 a	39.4	56.6 a	-3.2
OILS	10.5 c	-27.6	1.7 d	-39.3	5.7 de	-19.7	54.3 a	-7.2
Vydate	20.4 a	40.7	3.7 ab	32.1	8.4 b	18.3	60.5 a	3.4
NEMATODE ONLY	14.5 b	0.0	2.8 bc	0.0	7.1 bc	0.0	58.5 a	0.0

Means followed by the same letter(s) within a column are not significantly different ($p \leq 0.05$) according to Duncan's' multiple range test.

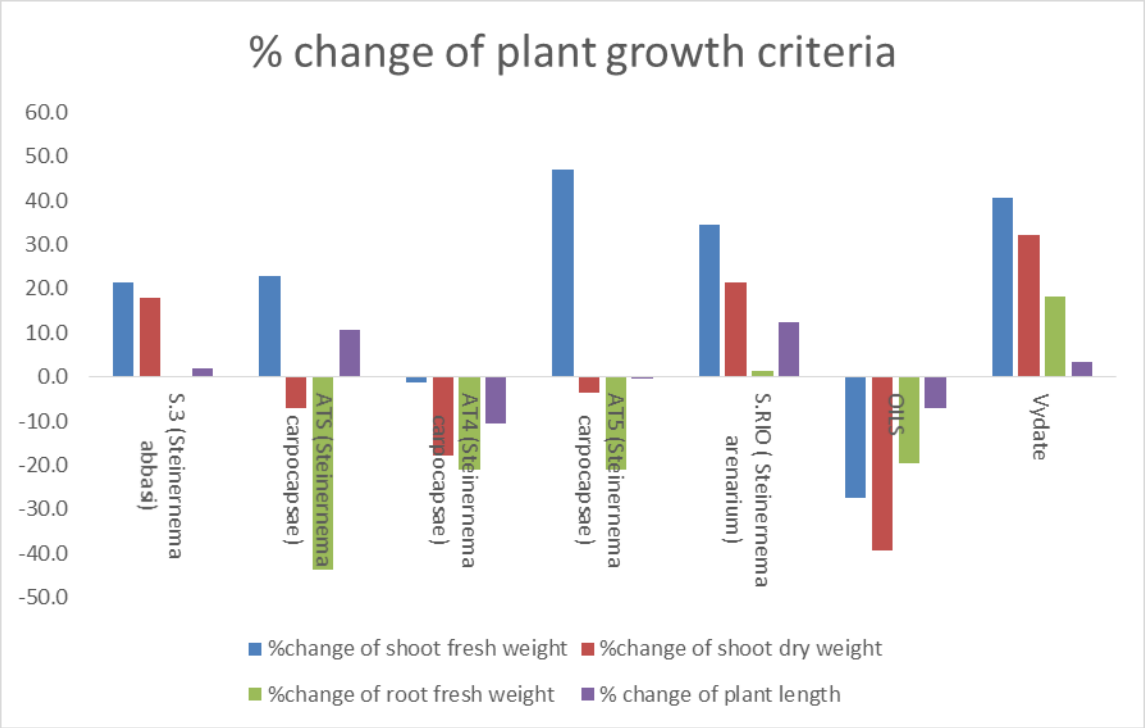

Fig. (5): Reduction percentage of plant growth infected by root knot nematode and treated by *Steinernema* isolates

, the results indicated to an overall increase in all growth criteria. However, the strain MANGO3 caused a reduction in shoot fresh weight, root fresh weight, and plant height, while MANGO4 led to a decrease in root fresh weight only (Table 7 and Fig. 6).

4. DISCUSSION

Ten isolates of entomo-pathogenic nematodes

Table (7): Effect of *Photorhabdus* isolats on the growth of eggplant infected with *M. incognita*.

Treatments	Shoot fresh weight (g)	% change of shoot fresh weight	Shoot dry weight (g)	% change of shoot dry weight	Root fresh weight (g)	% change of root fresh weight	plant length (cm)	% change of plant length
MANGO3 BACTERIA (*Photorhabdus* sp.)	12.6 bc	-13.1	3.2 ab	14.3	6.0 c	-15.5	53.1 b	-9.2
CITRUS 3 BACTERIA (*Photorhabdus* sp.)	21.7 a	49.7	4.3 a	53.6	7.8 ab	9.9	59.9 ab	2.4
HB88 BACTERIA (*Photorhabdus* sp.)	15.8 b	9.0	4.1 a	46.4	7.3 abc	2.8	63.0 a	7.7
MANGO2 BACTERIA (*Photorhabdus* sp.)	23.7 a	63.4	3.2 ab	14.3	8.8 a	23.9	60.4 ab	3.2
MANGO4 BACTERIA (*Photorhabdus* sp.)	15.9 b	9.7	3.2 ab	14.3	6.9 bc	-2.8	58.5 ab	0.0
OILS	10.5 c	-27.6	1.7 c	-39.3	5.7 c	-19.7	54.3 b	-7.2
Vydate	20.4 a	40.7	3.7 ab	32.1	8.4 ab	18.3	60.5 ab	3.4
NEMATODE ONLT	14.5 b	0.0	2.8 bc	0.0	7.1 abc	0.0	58.5 ab	0.0

Means followed by the same letter(s) within a column are not significantly different ($p \leq 0.05$) according to Duncan's' multiple range test.

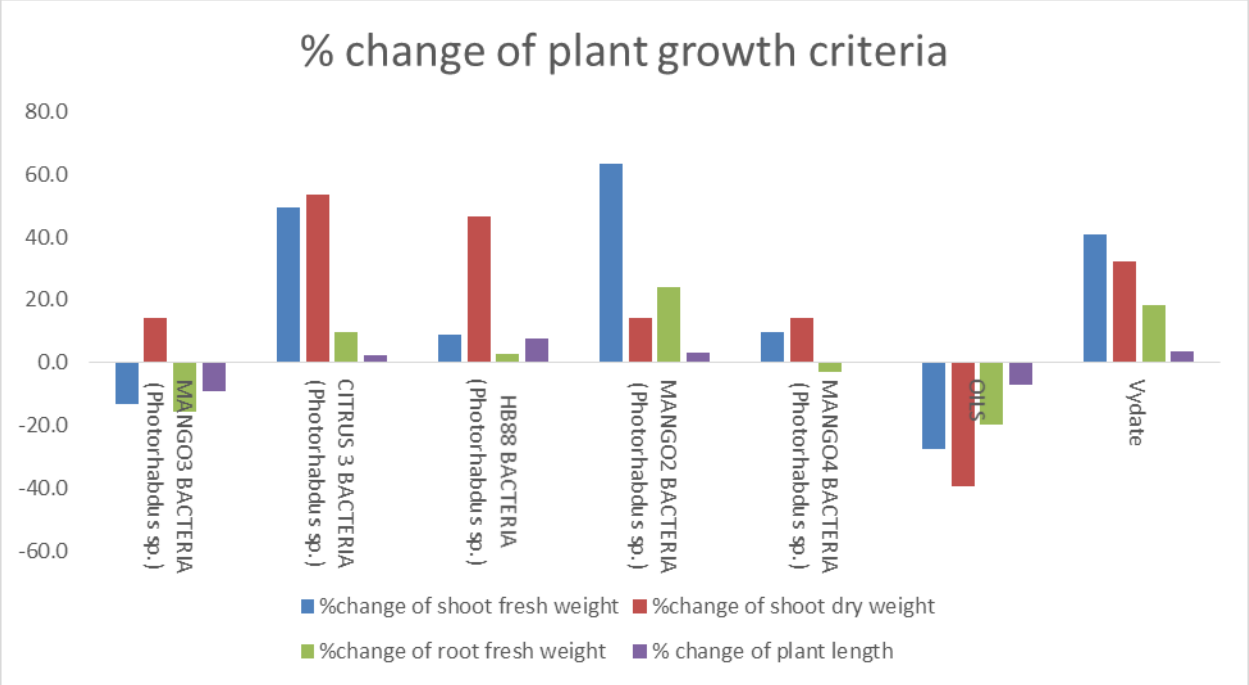

Fig. (6): Reduction percentage of plant growth infected by root knot nematode and treated with *Photorhabdus* isolates.

and 5 isolated bacteria (*Photorhabdus* sp.) were evaluated against the root-knot nematode *M. incognita* on eggplant under greenhouse conditions. Our research proved that EPNs have antagonistic and/or repellent effects on plant-parasitic nematodes *Meloidogyne* spp. (Grewal *et al.* 1999; Sayedain *et al.* 2021 and Dai *et al.*, 2022), served as the base for the current study. Our results showed that all treatments are capable of reducing nematode soil population up to 99.8 % reduction compared to positive control. Previous studies indicated that the direct application of EPN IJs has shown an antagonistic effect on different PPN species (Maru *et al.*, 2013 and Aatif *et al.*, 2012). Additionally, El Aimani *et al.*, (2022) reported that the antagonistic effects of the various EPN treatments applied to the soil had a moderate to significant impact on the J2 *M. javanica* densities in the soil and roots. According to Grewal *et al.* (1999), Hu *et al.* (1999) and Jagdale *et al.* (2002), alellelopathic substances produced by live or dead IJ may be toxic and/or repellent to PPN, thus reducing their population density. EPN associated bacteria, *Xenorhabdus* spp. or *Photorhabdus* spp., produce endotoxins composed of lipopolysacarides that are toxic and could kill or affect in another way the evaluated stages (Dunphy and Webster, 1988). The dead IJ caused infection reduction when both eggs and J2 were used. Jagdale *et al.* (2002) stated that live and dead *S. carpocapsae* IJ reduced PPN populations 15 and 30 days after the application by more than 50%. They also suggested a chemical disturbance instead of a physical one. Our study added more evidences that bacterial compound like- toxins are responsible for inhibiting egg hatching or J2 penetration.

The results also revealed that S.3 (*Steinernema abbasi*) strain's showed a significant decrease in the number of galls (-46.5% reduction) and egg masses (-65.2% reduction) as compared to the positive control. The same outcomes were found by (El Aimani *et al.*, 2022) who found that EL45 and MOR9 strains of *Steinernema feltiae* were significantly more effective at lessening nematode impact than strains of *H. bacteriophora* and other *Steinernema* sp., where different processes may have interfered.

Additionally, treatment with the isolated bacterium *Photorhabdus* sp. from *Heterorhabditis* sp. reduced the number of galls up to 29.6% and the number of egg masses (40.8%) which was more effectively than treatment with the enomopathogenic nematode *Heterorhabditis* sp. According to Zakaria *et al.* (2013), under simulated field conditions, the symbiotic bacterium *Photorhabdus luminescens* significantly reduced gall formation and other criteria on cucumber roots whether used singly or in combination. Naturally occurring symbionts of *Heterorhabditis* worms, Entomopathogenic *Photorhabdus* bacteria are a valuable source for the discovery of biologically active secondary metabolites (Kusakabe *et al.*, 2022). A range of secondary metabolites are produced by the bacteria *Photorhabdus*, which coexist as endosymbionts. Only two secondary metabolite substances indole and a stilbene derivative called 3,5-dihydroxy-4-isopropylstilbene were found to have nematicidal action. These metabolites, which also include different kinds of antibiotics, proteases, adhesions, lipases, and hemolysins, are used as biocontrol agents for eliminating parasitic nematodes, especially those belonging to the genus *Meloidogyne* (Lulamba *et al.*, 2021; Tomar *et al.*, 2022). Two phenylpropanoid and one alkaloid secondary metabolites of *Photorhabdus* l. sonorensis strain Caborca's culture filtrates were isolated and identified. The root-knot nematode (*Meloidogyne incognita*) and the citrus nematode (*Tylenchulus semipenetrans*), were both targets of the three discovered compounds' selective nematicidal and/or nematistatic actions (Kusakabe *et al.*, 2022). Furthermore, a study done in vitro by Srivastava and Chaubey (2022) on the nematicidal activity of entomopathogenic bacteria *Photorhabdus* spp. and *Xenorhabdus* spp. against the root knot nematode *Meloidogyne incognita* shown that 100% mortality was attained after 48 hours with a 10% filtrate of *H. indica* isolate DH3.

According to plant growth criteria, all *Steinernema* isolate treatments compared to the check exhibited a decrease in the shoot fresh weight and plant length of ATS, as well as all plant growth parameters of S.3, S.RIO isolates, and Vydate. With the exception of CITRUS 5 isolates, all *Heterohabditis* isolate treatments raised shoot fresh and dry weight while having the reverse effect on root fresh weight. *S. feltiae* strain (EL45) considerably increased plant height and root length, whereas *H. bacteriophora* strain (HB-MOR7) only increased root fresh weight, according to El Aimani et al. (2022). Zakaria *et*

al. (2013) also found that, when applied *to Meloidogyne incognita* on cucumber plants, symbiotic bacterium *Photorhabdus luminescens* significantly enhanced plant growth, including length of shoot and root, fresh and dry weight of shoot and root, number of leaves, flowers, fruits, and weight of fruits per each plant, when compared to check under simulated field conditions.

In addition, when *Steinernema carpocapsae*, *S. feltiae*, and *Heterorhabditis bacteriophora* three species of entomopathogenic nematodes (EPNs) were used as carriers of biocontrol agents on *M. javanica* infected cucumber under growth chamber and greenhouse conditions, the pathogenicity indices were significantly reduced. For all treatments except *S. feltiae*, a significant increase in plant growth indices (such as fresh/dry weight of shoots/roots) was observed. (Sayedain *et al.*, 2021).

Conclusion

The study shows the potentiality of entomopathogenic nematodes (EPNs) and the bacteria that they are associated with, especially *Photorhabdus* sp., as biocontrol agents against *Meloidogyne incognita*. A number of EPN strains, including *Heterorhabditis indica* (MANGO strains), *Steinernema abbasi* (S.3), and *Steinernema arenarium* (S.RIO), successfully decreased *M. incognita* soil population densities as well as galls and egg masses counts, with reductions up to 99.6%. Among the treatments, *Photorhabdus* sp. bacteria also demonstrated encouraging outcomes, boosting plant growth indices and dramatically lowering galls and egg masses. Some EPN treatments inhibited plant growth, but strains such as S.3 and S.RIO, as well as Vydate, had little effect on plant growth. EPNs and related bacteria, particularly *Photorhabdus* sp., may be useful agents for managing *M. incognita* infestations in eggplant crop in a sustainable manner, according to these findings.

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المكافحة الحيوية لنيماتودا تعقد الجذور *Meloidgyne incognita* باستخدام النيماتودا الممرضة للحشرات و البكتريا المصاحبة لها فى الصوب الزراعية

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ملخص

تم عمل اختبارات باستخدام 10 عزلات من النيماتودا الممرضة للحشرات و 5 سلالات من البكتريا المعايشة لنيماتودا الحشرات و استخدام خليط من الزيوت الطبيعية و مبيد نيماتودى vydate على نباتات باذنجان مزروعة فى أصص تحت ظروف الصوبة مصابة بنيماتودا تعقد الجذورلقياس تأثيرهم على تكوين العقد الجذرية و تعداد افراد وكتل بيض نيماتودا تعقد الجذور فيها. تم استخدام عزلة من *Steinernema abbasi* (S.3) و*S. arenarium* (S.RIO) و 3 عزلات من *S. carpocapsae* بعدة تركيزات على الأصص المزروعة شتلات باذنجان مصابة بتركيزات 2000 فرد نيماتودي/ اصيص. كذلك تمت المعاملة باستخدام 3 عزلات نيماتودا ممرضة للحشرات من*Heterorhabditis indica* و عزلتان من *H. bacteriophora* و 5 عزلات من البكتريا المصاحبة لهذا الجنس (*Photorhabdus* sp.).

أظهرت النتائج انخفاض فى كثافة تعداد أفراد *M. icognita* فى كل المعاملات التى تمت باستخدام النيماتودا الممرضة للحشرات. النيماتودا الممرضة للحشرات و البكتريا المصاحبة لها خفضت أعداد النيماتودا فى التربة لنسبة تصل إلى 99.6 %عند المعاملة بالنيماتودا *Steinernema arenarium* (S. Rio) و بنسبة 99.2 % عند المعاملة بالنيماتودا *Heterorhabditis indica* (3 Mango) و تخفيض أعداد النيماتودا فى التربة بنسبة تصل إلى 99.2% عند المعاملة بالبكتريا *Photorhabdus* sp. والتي تم عزلها من النيماتودا *Heterorhabdits indica*.

أظهرت النتائج أيضا انخفاضا واضحا فى أعداد العقد الجذرية الناتجة عن الإصابة بنيماتودا تعقد الجذور بنسبة 46.5% و انخفاضا فى أعداد كتل البيض بنسبة 65.2% عند المعاملة بنيماتودا *Steinernema abbasi* .

عند دراسة وملاحظة نمو نباتات الباذنجان فى وقت المعاملات كان هناك انخفاضا واضحا معدل نمو النباتات فى كل المعاملات التى تمت باستخدام نيماتودا من جنس *Steinernema* فيما عدا وزن الجذر الطازج و طول النباتات و كل معدلات نمو و طول النبات عند المعاملة ب S.3, S.RIO و مبيد Vydate عند المقارنة بالنباتات الغير معاملة. كان هناك تخيضا واضحا فى وزن الجذر الطازج و الجاف فى حالة استخدام نيماتودا citrus5 بالمقارنة بتحسن الوزن الجاف و الطازج للجذور عند المعاملة بنيماتودا من جنس *Heterorhabditis* التي شهدت تحسنا كبيرا فى نمو النباتات.

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