An Ecofriendly Root- Knot Nematode Pest Management strategy on sugar beet
3- Utilizing Certain Commercial Bioproducts of Microbial Origin

Maareg, M.F.*; A. Y.El- Gindi; K.M.Agami***and Abeer, S.Yassin***

* Department of Plant Protection, Sugar Crops, Research Institute, Agricultural Research Center, Giza, Egypt.

** Department of Agricultural Zoology and Nematology, Faculty of Agriculture, Cairo University, Giza, Egypt.

*** Department of Agricultural Treatments, Sugar Crops, Research Institute, Agricultural Research Center, Giza, Egypt.

Abstract: The potential of three commercial bioproducts, namely, Bioarc®, Bionematon®, and Biozeid® (containing, *Bacillus megaterium*, *Paecilomyces lilacinus Trichoderma album*, respectively) to control the nematode, *Meloidoyne incognita* infecting sugarbeet plants was evaluated under laboratory and field conditions. Additionally, their nematode potential was contrasted with that obtained by using the chemical compound Vydate®. The all tested material treatments significantly ($P \leq 0.05$) controlled nematode population, compared to the untreated treatment. In the laboratory test, all the tested materials were evaluated in different concentration rates and exposure periods. The mortality percentages of nematode, *M. incognita* varied according to the type, the concentration rate and the exposure period for bioproduct. All material treatments observed that the remarkable mortality in nematode when used at the highest concentration rate after 72 hours exposure period. The mortality of nematode, *M. incognita* were within the range of 86.78- 93.62% with the bioproduct treatments, compared to 98.94% with chemical product treatment (Vydate® 24% L.). In the field test, all the bioproducts and nematicide were evaluated at their recommended rates. The final nematode population decrease ranged from 72.25 to 85.70% in bioproduct-treated plots, and it was 84.15% in Vydate® 10% G-treated plots. Although the bioproducts appear to work well in the laboratory, their effectiveness may be reduced in the field due to dilution by irrigation water or interaction with the biotic and abiotic components of the surrounding environment. Also, the reproduction factor values of nematode was ranged between 5.00 and 9.70- fold with the bioproducts treated plots and those with Vydate® 10% G. treatment had an average value of 5.5- fold, while the untreated treatment had the highest average of 34.96- fold, compared to the initial nematode population. Generally, in sugarbeet field, the efficiency of *T. album* (85.7% reduction) as commercial bioproducts was found to be comparable to that of Vydate® (84.15%), followed by *P. lilacinus* (78.1%) and *B. megaterium* (72.25%). The all evaluated treatments also, enhanced sugarbeet productivity measurement for both roots yield and gross sugar yield/ fed. Compared to nonamended treatment, the maximum increase in roots and gross sugar yields were noticed when the soil was amended with Biozeid®, followed by Bionematon® and Bioarc®, respectively, while amendment with Vydate® 10% G. noticed the minimum increase in both yields. The results of this study confirmed that the nematicide, Vydate® 10% G. can be replaced by Biozeid® for the control of root- knot nematode, *M. incognita* disease of sugarbeet or application of Vydate and Bionematon® or Bioarc®, as components of integrated control programs may provide efficient pest management and increased sugarbeet productivity in the Nubaryia region.

Keywords: bioproducts, *Meloidoyne incognita*, Vydate®, *Trichoderma album*, *Bacillus megaterium*, *Paecilomyces lilacinus*, sugar beet, biological control, nematode mortality, exposure, nematicide.

1. Introduction

In Egypt, sugarbeet (*Beta vulgaris* L.) is considered the first source of sugar production, producing about 1.8 million tons (67.7%), corresponding to 0.9 million tons (33.3%) for sugarcane. Root- knot nematodes, *Meloidoyne incognita* and *M. Javanita* are major pests on sugarbeet where they cause considerable losses in yields. A reduction in roots yield and sugar yield in Egypt ranging from 0.7 to 50.8% and from 11.8 to 68.4%, respectively (**Gohar and Maareg, 2005**).

Chemical techniques have mostly been utilized to control nematodes. Chemical agents such as nematicides, Carbofuradan®, Codusafos®, Ethoprophos®, Fenamiphos®, Thionazin®, and Vydate®.... are successful in nematode control but are not ecofriendly and may pose a major danger to the ecological balance over time. Chemical pesticides cause reproductive harm and carcinogenesis in animals. Animals have died when given high dosages of these drugs. As a result, biological control agents (fungal and bacterial) are becoming increasingly important in the field of worm management. Another important function of these compounds is that they promote plant development.

Many researchers tested bacterial and fungal bio agents against nematode infection. **(Spieged *et al.*, 2005; Hammad and Zaid, 2007; Sahebani and Hadari, 2008; Oliveira *et al*, 2009; El- Nagdi *et al.*, 2011; Radwan *et al*, (2012); Raddy *et al.*, 2013; Maareg *et al.*, 2014; Al- Hazmi and Javeid, 2016; Mostafa *et al.*, 2018 and Yassin, 2018),** they found that, *Trichoderma* spp. and *Bacillus* spp. are potential nematode bio agents on many food, vegetables and cash crops**.**

Also, several research studies are available on biological nematode control, there continues to be a dearth of registered bio- nematicides. As a result, the current study was carried out in the laboratory and in the field to compare the performance of three commercial bioproducts of microbial origin (Bioarc®, Bionematon®, and Biozeid®) with the chemical nematicide, Vydate® for the management of root-knot nematode, *Meloidogyne incognita* infecting sugarbeet plants. In addition, the influence of all compounds examined on sugar beet productivity (as assessed by roots and gross sugar yields) was investigated.

2. Materials and methods

The tested commercial bioproducts in this study were Bioarc®, containing *Bacillus megaterium* at 2.5 $x10^{-6}$ colony forms unit g^{-1}, Bionematon®, containing *Paecilomyces lilacinus* at 10^{-6} spores ml^{-1} and Biozeid® containing *Trichoderma album* at 25 x 10^{-6} spores g^{-1} were obtained from the Agricultural Research Center (ARC) Giza, Egypt. The chemical nematicide, Vydate® (N-N-dimethyl-2methyl carbamoyloxmino- 2- (methylthio) acitamide was used as comparison treatment.

2.1. Laboratory test:

In this study, a direct- contact bioassay was used to evaluate the toxicity of Bioarc®, Bionematon® and Biozeid® bioproducts to freshly hatched second stage juneveles (J_{2s}) of *M. incognita* nematode. Aqueous J_{2s} suspension was obtained by incubating infected roots of egg plants (obtained from pure culture of *M. incognita* in a glass house) in water for 3 days at 28 ±5 c° and the hatched J_{2s} were collected and counted. Four aqueous concentration of each tested bioproduct (1.0, 1.5, 2.0, and 2.5%) were prepared by diluting the standard product concentration and separated test against J_{2s}. The same concentrations of Vydate® L 24% were also prepared, as comparison treatments. The assessment was carried out in 10 cm diam. Petri plates containing 9 ml different concentration of each tested product. Nematode suspension carrying one ml (100 juveniles, J_{2s}) with help of pipette was added. Petri plate containing J_{2s} in distilled water was kept as nematode only control. Each treatment was replicated five times. These dishes (plates) were recovered with lids and arranged in completely randomized design on a laboratory bench at 25±5 c°. Dead nematodes were counted under binocular microscope 24, 48 and 72 hours post-treatment, and corrected. Nematodes were considered to be dead if their bodies shopped straight shapes without movement when they were prodded with fine wooden dowels.

The corrected percentages of nematode mortality were calculated according to Abbott's formula (1925):

Mortality%= {(m-n)/ (100-n)} x 100

Were m and n indicating the number of J_{2s} mortality in treatment and control, respectively.

2.2. Field test:

This research was carried out on a field with sandy loam soil that was naturally infected with M. incognita and was watered with overhead sprinklers in the Nubariya province of Egypt. Bioarc, Bionematon, and Biozeid, as bioproducts, and Vydate® 10% G., as a chemical product, were used at the prescribed doses. 7 days before planting, all tested materials were scattered on the soil's surface and then integrated to a depth of 10- 20 cm using a hoe. The experimental field was split into four blocks, each with five plots measuring 3 m broad × 3.5 m long (= 10.5 m2 i.e. 1/400 Fadden). Each plot had six rows that were 50 cm apart. Each treatment had four duplicates, which were grouped in a randomized block design. Sugarbeet seeds, cv. Mammut, (the seeds were confirmed to be susceptible to nematode pest in previous study by **Maareg *et al.* 2018** were sown at a distance of 20 cm, (the normal density of 40,000 plants/ Fadden) in the last week of September. All treatments were managed throughout the growing season by standard agricultural practices and were irrigated as needed. The average soil nematode population density (Pi) was 2010/ 200 g soil.

2.3. At harvest:

Five soil samples were obtained from the rhizosphere of five sugarbeet plants (200 g soil/plant) from each plot, and the roots included in the samples were later used for additional studies. Each soil sample was thoroughly mixed, then a 200 g sub-sample was sieved and decanted to remove nematodes (Barker, 1985). According to Byrd et al (1983), the roots of each duplicate were sliced into small portions (0.5 cm long), put in Petri plates after staining with acid fuchsine Lactophenol, and then studied under a stereoscopic microscope for counting larval females and egg masses on the complete root system. The total number of nematodes in the soil and root system was utilized to calculate the final nematode population densities (Pfs) and reproduction factor RF (RF= Pf / Pi). Fresh roots weight of each plot was recorded and roots yield/ fed. Was determined. Sugar content was assessed according to Le- Docte as described by Mc Ginnis (1982) and gross sugar yield calculated as roots yield x sugar content. All data were subjected to analysis of variance by using Costat program (1988) and the comparison among means was portioned by

Duncan's (1955) multiple range tests at 5% level of probability.

3. Results and discussion

The nematicidal activity of commercial bioproducts Bioarc® (*Bacillus megaterium),* Bionematon® (*Paecilomyces lilacinus*), and Biozeid® (*Trichoderma album*) in comparison to chemical nematicide Vydate®. against root- knot nematode, *Meloidogyne incognita* infecting sugarbeet was evaluated under laboratory and field conditions. The effect of the investigated compounds on sugarbeet productivity was also assessed.

3.1. In laboratory test:

Under laboratory condition, the effect of exposure of the second stage juveniles stage (J_{2s}) of *M. incognita* nematode to the tested bioproducts in comparison with the nematicide (Vaydate L. 24%) was assessed. The results observed that J_{2s} was quite sensitive to all bioproducts and the tested nematicide. All the tested treatments showed variation in mortality percentages of J_{2s} as compared to the distilled water as control treatment. The mortality of J_{2s} treated with the bioproducts ranged from 61.25 to 93.62%, compared to that of 79.73 - 98.94% provided by nematicide, and those with control treatment (2.1 - 4.0%) as shown in Table 1.

The nematode's reaction varied depending on the type of bioproduct, the exposure length, and the concentration rate. The kind of bioproduct has a substantial impact on nematode mortality. Furthermore, fungi-based bioproducts had higher nematicidal activity than bacteria-based bioproducts. Bioarc® (*B. megaterium*) had the lowest mortality percentage in J2s (75.95%), while Biozeid® (*T. album*) had the greatest mortality percentage of infective J2s (83.3%), followed by Bionematon® *(P. lilacinus*), with a mortality percentage of (77.7%). There were significant variances between them. The nematicide, Vydate® L. 24%, on the other hand, reported 88.13% mortality in J2s, whereas the average mortality in the control treatment was 2.0%. Furthermore, as shown, there were significant differences among all tested treatments.

Time of exposure to bioproducts affected the mortality of J2s of root- knot nematode, M. incognita. The all tested bioproducts significantly increased the mortality percentage of J2s after 24, 48 and 72 hours of exposure periods, compared with control treatment.

Table 1: Nematicidal effect of three commercial bioproducts of *Bacillus megaterium, Paecilomyces lilacinus and Trichoderma album* on second stage juveniles (J_{2s}) of *Meloidogyne incognita* in comparison with nematicide, Vydate® L 24% under laboratory condition

Treatments	Concentration rate%	Juveniles (J_{2s}) mortality percentage Exposure periods			
		24 hours	48 hours	72 hours	Overall mean
Bioarc® (*Bacillus megaterium*)	1.0	61.25	63.01	64.54	62.92
	1.5	69.75	71.68	74.23	71.88
	2.0	81.50	82.40	85.42	83.11
	2.5	85.00	85.97	86.78	85.92
Mean		74.38	75.77	77.74	75.95
Bionematon® (*Paecilomyces lilacinus*)	1.0	65.00	66.58	68.62	66.72
	1.5	70.50	71.43	74.23	72.04
	2.0	82.50	85.20	86.48	84.71
	2.5	85.00	87.24	89.80	87.33
Mean		75.75	77.61	79.78	77.70
Biozeid® (*Trichoderma album*)	1.0	67.00	72.19	76.02	71.71
	1.5	76.75	83.42	85.46	81.84
	2.0	86.25	89.03	90.31	88.51
	2.5	88.50	91.33	93.62	91.13
Mean		79.63	83.99	86.35	83.30
Nematicide, Vydate	1.0	79.73	83.11	88.90	83.91
	1.5	83.00	84.64	92.68	86.77
	2.0	85.25	88.66	94.66	89.52
	2.5	85.75	92.94	98.94	92.54
Mean		83.43	93.61	93.61	88.13
Distilled water (control)		0.00	2.10	4.00	2.00
$L.S.D_{0.05}$, Exposure periods	0.47,	Treatments	1.00,	Concentration rate	0.80

The mortality percentage was already great after 24 hours of exposure and increased significantly only slightly up to 72 hours of exposure.

Also, the concentration rates of bioproduct significantly affected the nematode mortality. The mortality of J_{2s} of *M. incognita* increased significantly ($P \leq 0.05$) with increase of concentration rate of the bioproduct at the different exposure periods, compared to control treatment. Significant differences were recorded among the bioproducts at different

concentration rates and exposure periods with various levels of success (Table 1).

At the expiry of 72 hours of exposure, the results showed that the commercial bioproducts of *B. sutilis*, Bioarc®, *P lilacinus*, Bionematon® and *T. album*, Biozeid® as well as nematicide, Vydate® recorded the highest significant increase in mortality of J_{2s} when used at the highest concentration rate.

Among the tested bioproducts, Biozeid® had the highest nematicidal effect with 93.62% mortality, while, Bioarc® was relatively least effective causing 86.78% mortality in J_{2s} and Bionematon® ranked intermediate in descending order by 89.80% mortality. However, the mortality in J_{2s} was 98.94% and 4.0% with Vydate® and control treatments, respectively. There were significant differences among the all tested treatments (Table 1).

Based on these findings, the tested compounds were divided into two mean groups, i.e., 1- highest toxic (85- 90% mortality), consisting of Bioarc® and Bionematon® and 2- extremely toxic (> 90percentage), consisting of Vydate® and Biozeid® under laboratory conditions, as shown in Table 1.

3.2. In field test:

The population densities and reproduction factor values of root- knot nematode, *M. incognita* and sugarbeet productivity as influenced by the tested commercial bioproducts, Bioarc®, Bionematon® and Biozeid® in comparison with chemical nematicide, Vydate 10% G are present in Tables (2 and 3). All tested materials were applied at their recommended rates.

3.3. On the population densities and reproduction factor:

In the present study, application of all treatments tested caused a significant reduction in J_{2s} number in soil as compared to the untreated control. In addition, for all these tested treatments, a significantly lower of development stages per roots was observed. As mentioned above, the final population of *M. incognita* root- knot nematode corresponded with the reduction in the nematode damage parameters, J_{2s} number in soil and the number of development stages within plant roots.

After application of the bioproducts, Bioarc®, Bionematon® and Biozeid® as well as the tested nematicide, Vydate® the final nematode population number was significantly ($P \leq 0.05$) reduced, compared to the untreated control (Table 2). The number of final nematode population was within the range 10047 to 19497 with bioproduct treatments as compared to 11136 with nematicide and to 70269 in untreated control treatments. Among the bioproduct treatments, the highest reduction in final nematode population (85.70%) was recorded with the bioproduct containing *T. album*, followed by *P lilacinus*, (78.10%) and *B. megaterium*, (72.25%) treatments, compared with nematicide, Vydate (84.15%).

There were significant differences among the bioproduct treatments. However, the Biozeid® (*T. album*) treatment did not differ from Vydate® treatment as shown in Table (2).

The previous results show that Biozeid® (as commercial bioproduct) and nematicide, Vydate (as chemical product) have almost the same nematicidal activity (85.70 and 84.155% reduction, respectively) on *M. incognita* nematode in sugar beet field. Means in each column followed by the same letter are not significantly at $p \leq 0.05$.

Table 2: Nematicidal effect of three commercial bioproducts of *Bacillus megaterium, Paecilomyces lilacinus and Trichoderma album* on population densities and reproduction factor of *Meliodogen incognita* on sugarbeet in comparison with nematicide, Vydate® 10% G under field condition

Treatments	Second stage juveniles (J_{2s}) larvae in soil	Different stages in root system	Final population		Reproduction factor
			Number	Reduction %	
Bioarc® (*Bacillus megaterium*)	1830 b	17667 b	19497 b	72.25 c	9.70 b
Bionematon® (*Paecilomyces lilacinus*)	1631 c	13758 c	15389 c	78.10 b	7.66 c
Biozeid® (*Trichoderma album*)	1086e	8961 d	10047 d	85.70 a	5.00 d
Nematicide, Vydate	1300 d	9836 d	11136 d	84.15 a	5.50 d
Untreated	3411 a	66858 a	70269 a	-	34.96 a

Although, Based on these finding, the tested compounds were divided into two main groups, i.e., 1- toxic (70- < 80% reduction) consisting of bioproducts, Bioarc®, (*B. megaterium*) and Bionematon®, (*P lilacinus*) and 2- highly toxic (> 80% reduction) consisting of nematicide, Vydate® 10% G and commercial biological product, Biozeid® (*T. album*) as shown in Table 2.

The tested commercial bioproducts and nematicide seem to work well under laboratory conditions, their effect may decrease under field conditions due to dilution by irrigation water or

interaction with the biotic and abiotic components of the surrounding environment.

Also, the reproduction factor (RF) values of *M. incognita* root- knot nematode on sugarbeet were reduced significantly ($P \leq 0.05$) by application, all the tested treatments as compared with untreated one (Table 2). The average of RF values of bioproduct treatments were within the range 5.0- 9.70- fold; of nematicide, Vydate® treated plots was 5.54- fold, while in untreated treatment those was an average of 34.96- fold, compared with initial nematode population (Table 2).

The RF values with bioproduct treatments on sugarbeet plants as related to final population of *M. incognita* nematode, Bioarc® treated plots recorded the highest value (9.70- fold) of RF among the bioproducts, followed by Bionematon® (7.66- fold), while Biozeid® treated plots recorded the least value (5.00- fold) of RF among the bioproduct treatments. The RF values among the biological product treatments was significantly different from each other, but were not significantly different between Vydate® and bioproduct of *T. album* (Biozeid®) as shown in Table 2.

3.4.On sugar beet productivity:

Aside from reducing nematode infestation, the tested commercial bioproducts and nematicide increased sugarbeet productivity in terms of both root yield and gross sugar yield (Table 3). According to the table, all of the studied treatments produced significant (P 0.05) results for roots and gross sugar yields/fed sugarbeet. In general, the tested treatments of bioproducts and nematicide significantly ($P \leq 0.05$) increased the roots and gross sugar yields as compared to untreated treatment. All tested bioproduct treatments caused remarkable increase in this respect when compared to soil amended with nematicide treatment. The value of roots yield/ fed of sugarbeet plants were treated with bioproducts was within the range of 21.46- 27.25 tons/ fed and those with nematicide treatment had roots yield average of 18.0 tons, while untreated treatment had the least roots yield average of 12.75 tons/fed.

Among the bioproduct treatments, bioproduct of *T. album*, (Biozeid®) caused significantly ($P \leq 0.05$) the greatest roots yield (27.25 tons/ fed), followed by *P lilacinus* (Bionematon®) with an average of 22.92 tons, while, *B. megaterium* (Bioarc®), gave the least one (21.46 tons/ fed). There were significant differences among them. Compared to untreated treatment, the increase percentage in roots yield/ fed was about 113.73, 79.76 and 68.31% due to applying Biozeid®, Bionematon® and Bioarc® treatments, respectively, Table (3).

The gross sugar yield/ fed value of sugar beet plants with the tested bioproducts was in the range of 3.65 - 4.78 tons/ fed, compared to 2.97 and 1.91 tons/ fed for the plants treated with Vydate® and untreated treatments, respectively. In case the bioproducts treatments, significantly ($P \leq 0.05$) the highest gross sugar yield (4.78 tons/ fed) was obtained with application of bioproduct, Biozeid® followed by Bionematon® (3.95 tons/ fed) and Bioarc® (3.65 tons/ fed), respectively, without significant difference between Bionematon® and Bioarc® treatments. When sugar beet plants were received with Bioarc®, Bionematon® or Biozeid® bioproducts, the gross sugar yield increased by about 91.1, 105.76 or 150.26%, respectively, compared to untreated treatment.

The nematicide, Vydate® treatment resulted in significantly ($P \leq 0.05$) lowest increase percentage for both roots yield and gross sugar yield (41.18 and 55.5%, respectively), compared to that caused by bioproduct treatments as shown in Table 3.

Table 3: Effects of three commercial bioproducts of *Bacillus megaterium, Paecilomyces lilacinus and Trichoderma album* on roots and gross sugar yields of sugarbeet in comparison with nematicide, Vydate® 10% G under field condition

Treatments	Roots yield		Gross sugar	
	Tons/ fed	Increase %	Tons/ fed	Increase %
Bioarc® (*Bacillus megaterium*)	21.46 c	68.31	3.65 b	91.10
Bionematon® (*Paecilomyces lilacinus*)	22.92 b	79.76	3.93 b	105.76
Biozeid® (*Trichoderma album*)	27.25 a	113.73	4.78 a	150.26
Nematicide, Vydate	18.00 d	41.18	2.97 c	55.50
Untreated	12.75 e	-	1.91 d	-

Means in each column followed by the same letter are not significantly at $p \leq 0.05$.

In these studies, the obtained results observed that the tested commercial bioproducts, Bioarc®, Bionematon® and Biozeid® as well as nematicide, Vydate® revealed that suppressive effects against root-knot nematode, *M. incognita* in laboratory test and reduced the incidence of this nematode in sugarbeet field. In this respect, a few researchers evaluated the nematicidal activity of some different biological agents such as (*Aspergillus niger, A. tearus, Azosipirillum brasilens, Bacillus megaterium, B. Subtilis, Paecilomyces lilacinus, Pseudomonas*

fluorescens, Trichoderma harzianum and T. viride.), and showed that these biological agents could be suppress the damage caused by *M. incognita* and *M. javanica* root- knot nematodes on sugarbeet plants in vitro or in pots and field (**Maareg and Bdr, 2000 a & b; Gohar, 2003; Maareg *et al*, 2003, 2004, 2005 and 2014; Gohar *et al*., 2014; El- Nagdi *et al*, 2011; Mostafa *et al.*, 2018 and Yassin, 2018**).

Various fungal antagonists of nematodes have shown promising results. These mainly include endoparasitic fungi parasites of nematode egg, second stage juveniles and adult females as well as nematode trapping fungi.

The fungi, *P. lilacinus*, are egg parasitic fungi, which infects by direct hyphal penetration. The hyphae branch and grow across the eggshell (**Khan *et al*, 2006**). It has been suggested that its parasitism is associated with the enzyme serine protease, which is nematicidal in activity. It acts by degrading eggshell and prevents hatching (**Zareen *et al*, 2001**), Also, *P. lilacinus* is one of the potential biological agents, which can also colonize organic matter in soil and develop in the rhizosphere of plants.

Another the *Trichoderma* species parasitizes egg, juveniles and females of nematode. The hyphae penetrate the egg, juveniles and females cuticle by dissolving the chitin layer through enzymatic activity. They proliferate within the organism and produce toxic metabolites, (**Suarez *et al*, 2004** and **Yang *et al*, 2010**). Thus, the enzymes produced by *Trichoderma* spp. such as chitinases, glucamases and proteases seem to play an important role in parasitism. *Traichoderma* has not only been proved to parasitize nematode and inactivate pathogen enzymes but also help in inducing plant defense mechanisms leading to systemic resistance in plants by enhanced health of roots (**Sahebani and Halavi, 2008**). It participates in solubilization of inorganic nutrients. Thus *Trichoderma* colonized roots require lesser supply of manmade nitrogen fertilizers (**Harman, 2000**).

Species of *Bacillus megaterium* interrupts the root- knot nematode life cycle by producing toxic metabolites, which restrict their mobility and hinder the hatching and juveniles penetration into plant roots **(Sikora *et al*, 2007**). Bacteria many change nematode behavior, can function as competitors of nematodes for colonization sites and nutrients (**Khan *et al*, 2008**) and induced of systemic resistance (**Cartieaux *et al*, 2003**).

Also, previous results vouched that the nematicide, Vydate® surpassed all bioproducts treatments in laboratory test. However, in the field test, the Vydate® as well as, efficiently controlled the root- knot nematode on sugarbeet and came affect the bioproduct, Biozeid®, without significant difference between them. The compound is a carbonate systemic nematicide, which kills nematode inside plant tissues, thereupon, prevented the development of the invaded larvae and consequently stopped egg- deposition. Vydate® did not stop root penetration with juveniles presented in the rizosphere, some of which after the degradation of the compound can complete its life cycle after the protection period of nematicide. The role of this nematicide in reducing final nematode population and reproduction factor on sugarbeet is inclusively documented (**Maareg & Bader, 2000 a & b; Gohar, 2003 and Yassin, 2018).**

All the tested bioproducts treatments at the recommended rats significantly increased the sugarbeet productivity as measured by roots yield and gross sugar yield/ fed. Compared to those of untreated treatment. Previous investigators supported these results with the application of the different biological agents (**Maareg & Bader, 2000 a & b; Gohar, 2003; Maareg *et al*, 2004, 2005 and 2014; El- Nagdi *et al*, 2011; Mustafa *et al*., 2018 and Yassin, 2018).** The increases in yield components are depending on the vigor of plant growth which is due to (1) killing action of the parasitism of the bio agents on the majority of penetrated larvae. (2) Substances released from microorganisms that facilitate the uptake of nutrients (**Glick, *et al*, 1999**) and/ or facilitate the production of phytohormons such as auxins, cytokinins, gibberellins, (**Vessey, 2003**) and (3) includes productions of inhibitory substance or by increasing of natural resistance of the host (**Cartieaux *et al*, 2003**).

Also, nematicide, Vydate® significantly increased roots and gross sugar yields. This nematicide act by impairing nematode neuromuscular activity, thereby, reducing their movement, host invasion feeding, and consequentially the rate of development and reproduction (**Kher *et al*, 1983**). According the plant growth improves and the yield increases (So the sugar beet productivity was increased).

Our results suggested that the all tested bioproducts and the tested nematicide at the recommended rates were highly efficient in reducing *M. incognita* infection and increasing sugarbeet yields. The efficiency of bioproducts of *T. album* (Biozeid®) was found to be comparable to that of nematicide Vydate® 10% G, followed by *P. lilacinus* (Bionematon®), and *B. megaterium* (Bioarc®). The bioproduct, Biozeid® and Vydate® 10% G were classified as highly toxic compounds, while products, Bionematon® and Bioarc®were classified as toxic ones against *M. incognita* nematode. In addition, all the tested bioproduct treatments caused remarkable increase in yield components of sugar beet, compared to nematicide, Vydate® 10% G treatment. The highest and the lowest increase in both roots and gross sugar yields/ fed. Were recorded with Biozeid® and Vydate® 10% G, respectively. Because the chemical nematicide is environmental hazardous. Bioproducts which are bacterial and fungal were applied as safer and eco-friendly control alternative. Therefore, it could be concluded that nematicide can be replaced by bioproduct, Biozeid® for the control of *M. incognita* or application of nematicide and bioproducts, Bionematon® and Bioarc® as items of integrated

control program might provide an effective control of this pest and enhanced sugarbeet productivity.

REFERENCES

Abbott, W.S. (1925). A method of computing effectiveness of an insecticide. J. Econ. Entom., 18: 256- 267.

Al- Hazmi, A. S. and Javeed, M.T. (2016). Effects of different inoculum densities of *Trichoderma harzianum* and *Trichoderma viride* against *Meloidogyne javanica* on tomato. Saudi J. Biol. Sci., 23(2): 288- 292.

Barker, T. R. (1985). Nematode extraction and bioassays, pp. 19-35, In: An Advanced Treatise on *Meloidogyne*. Barker T. R., Cater T. R., Cater c. c. and Sasser, J. N. (eds). North Carolina State Univ, USA.

Byrd, D. W.; Kirkaptric, T; and Barker, K. (1983). An improved technique for clearing and staining plant tissue for detection of nematodes. J. Nematol., 15(3):pp 142- 143.

Cartieaux, F.; Thibaud, M. C; Zimmerli, L; Lessard, P. and Sarrobert, C. (2003). Transscriptome analysis of Arabidopsis colonized by a plant growth promoting rhizobacteria reveals a general effect on disease resistance. Plant Jounal, 36: pp 177-188.

Duncan, D. B. (1955). Multiple range and Multiple, F- test. Biometrices, 11: pp 10- 42.

El- Nagdi, W. M. A; Abd- El- Fattah, A.I. and Abd- El- Kair, H. (2011). Biological Control of *Meloidogyne incognita* and *Fusarium solani* in Sugarbeet. Nematol. Medit. 39: pp 59-71.

Glick, B. R.; Patten, C. L; Holguin, G. and Penrose, D. M. (1999). Biochemical and genetic mechanism used by plant growth promoting bacteria. Imperial college press, London, UK.

Gohar, I. M. A. (2003). The relationship between plant parasitic nematodes of sugarbeet and other soil fauna. Ph. D. Thesis, Fac. Agric. Moshtohor Zagazig Univ., Egypt,221 p.

Gohar, I. M. A. and M. F., Maareg (2005). Relationship between crop losses and initial population densities of root- knot nematode, *Meloidogyne incognita* in soil of sugarbeet grown in West Nubariya district. The Third International Coference of Plant Prodution Institute, 26- 29 November, Cairo, Egypt. J. Agric. Res., 83 (4): 1315- 1328.

Gohar, I. M. A.; Abo El- Ftooh, A. A. and Agami, K. M. (2014). Evaluation efficacy of nematicide seed treatments for the control of root- knot nematode, *Meloidogyne javanica* in sugarbeet production. Minufiya J. Agric. Res. Vol.39 No. 6: 1841-1854.

Hammad, E. A; Zaid, A. M. A. (2007). Biological control of root-knot nematode *Meloidogyne javanica* on sunflower plants by *Trichoderma album* and *Bacillus megaterium*. J. Agric. Sci., Mansoura University 32, pp 4747– 4756.

Harman, G.E. (2000). The myths and dogmas of bicontrol changes in percentages denved from research on *Trichorderma herzianum* strains T-22. Plt. Diseases. 84(4): 377- 393.

Khan, A; Sayed, M; Shaukat, S. S. and Handoo, Z. A. (2008). Efficacy of four plant extracts on nematodes associated with papaya in Sindh, Pakistan. Nematol. medit. (2008), 36: pp 93-98

Khan, A, Williams, K.L. and Nevalainen, H. K. M. (2006). Infection of plant parasitic nematodes by *Peacilomyces lilacinus* and *Monacrsporium lysipagum* Biol. Control 51(5): 659- 678.

Kheir, A. M; Osman, A. A. and Montasser, S. A. (1983). Efficacy of certain systemic nematicides in controlling *Meloidogyne javanica* infecting okra, Hibiscus esculentus. Pak. J. Nematol., 1(1): pp 49-55

Maareg, M. F. and Badr Sohir T. A. (2000a). Impact of three soil biofertilizers applied separately and in combination with a nematicide on *Meloidogyne incognita* infecting sugarbeet. Egyptian journal of Agronematology. 4 (1, 2): pp 71- 82.

Maareg, M. F. and Badr Sohir T. A. (2000b). The effect of certain biocontrol organisms, Oxamyl and their combinations on *Meloidogyne javanica* 158 infecting sugarbeet. Egyptian journal of Agronematology. 4(1, 2) : pp 95-104.

Maareg, M. F.; Aly, M. H; Manal. Y. Hussen and Eman A. Tantawy (2005). *Potential of Azospirillum brasilens, Bacillus megatherium, Glomus mosseae* and *Trichoderma viride* singly or concomitantly for control of root – knot nematode, *Meloidogyne javanica* infecting sugarbeet.Egypt. J. Agric Res., 83p (4).

Maareg, M. F.; EL- Gindi, A. Y; Gohar, I. M. A. and Agami, K. M. (2014). An Ecofriendly root- knot nematode pest management strategy on sugarbeet Utilizing microbial agents. Egypt. J. Agronematol., Vol, 13, No.1: pp 54- 74

Maareg, M. F.; El- Gindi, A. Y; Mona, E. El-Shalaby and Abeer. S. Yassin (2018). Evaluation of some sugarbeet varieties for their susceptibility to root- knot nematode, Meloidogyne incognita, according to modified host parasite index (MHPI) scale. Egypt. J. Agronematol. 17(1): pp 1-12

Maareg, M. F.; Gohar, I. M. A. and Manal, Y. Hussein (2004). Nematicidal activity of some soil microorganisms in controlling root-knot nematodes in sugarbeet fields. Science Conference, Sana, a (11-13) October. Abs.

Maareg, M.F.; M.A., Hassanein and Manal, Y. Hussein (2003). Inhibitory effect of certain soil fungal filtrates on *Meloidogyne incognita* on

sugarbeet. Annals of Agric. Sc. Moshtohor, 41 (2): 987- 998.

Mc Ginnis, R. A. (1982). Beet sugar technology 3rd ed. sugarbeet development foundation Fort Collins, 855 p.

Mostafa, Fatma, A. M.; Khalil A. E.; Nour El-Deen A. H. and Ibrahim Dina S. (2018). The role of *Bacillus megaterium* and otherbio-agents in controlling root- knot nematodes infecting sugarbeet under field condition. Egyption J. of Biological Pest Control 25: 66.

Oliveira, D. F.; Carvalho H.W.P.; Nunes A.S.; Campos V.P.; Silva G.H. and Campos V.A.C., (2009). Active substances against *Meloidogyne exigua* produced in a liquid medium by *Bacillus megaterium*. Nematologica Borasileira 33: 271- 277.

Opperman, C. H. and Chang, S. (1990). Plant-parasitic nematode acetylcholinesterase inhibition by carbamate and organophosphate nematicides. J. Nematol., 22: pp 481-488.

Raddy, H. M; Fouad, A. F. A; Montasser, S. A; Abdel- Lateef, M. F. and EL Samadisy, A. M. (2013). Efficacy of six nematicides and six commercial bioproducts against root-knot nematode, *Meloidogyne incognita* on tomato. J. Appl. Sci. Res., 9(7): pp 4410-4417.

Radwan M. A; Farrag, S. A. A; Abu- Elamayem, M. M. and Ahmed, N. S. (2012). Biological control of the root-knot nematode, *Meloidogyne incognita* on tomato using bioproducts of microbial origin. Applied Soil Ecology 56: pp 58-62.

Sahebani, N. and Hadavi, N. (2008). Biological control of the root- knot nematode *Meloidogyne javanica* by *Trichoderma harzianum*. Soil Biology and Biochemistry, 40: pp 2016-2020.

Sikora, R.A.; Schafer K. and Dababat A.A. (2007). Modes of action associated with microbially induce in planta suppression of plant parasitic nematodes. Australsian Plant Pathology 36: 124- 134.

Spiegel Y; Sharon, E. and Chet, I. (2005). Mechanisms and improved bio control of the root- knot nematodes by *Trichoderma* spp. Acta Horticulturae 698: pp 225-228.

Suárez, B; Rey, M; Castillo, P; Monte, E. and Llobell, A. (2004). Isolation and characterization of PRA1, a trypsin-like protease from the biocontrol agent *Trichoderma harzianum* CECT 2413 displaying nematicidal activity. Applied Microbiol. and Biotechnol., 65: pp 46-55.

Vassey, J. K. (2003). Plant growth promoting rhizobacteria as biofertilizers. Plant soil, 255: pp 571-586.

Yang, Z. S.; G. H. Li; P. J. Zhao; X. Zheng; S. L. Luo; L. Li; X. M. Niu and K. Q. Zhang (2010). Nematicidal activity of *Trichoderma* spp. and isolation of an active compound. Word J. Microbial & Biotechnol.,26: pp 2097-2102.

Zareen, A.; N.J., Khan and M.J. Zaki (2001). Biological control of *Meloidogyne javanica* (Treub) Chitwood, root- knot nematode of Okra (Abelmoschus esculenus L.) Moench. Pak. J. Biol. Sci. 4(8): 990- 994.

Yassin, Abeer S. (2018). Pathological effects of the root- knot nematode spp. On sugarbeet and its control in Nubariya Ph. D. Thesis. Fac. Agric. Cairo Unv. Egypt. 212 pp.

استراتيجية إدارة مكافحة نيماتودا تعقد الجذور على بنجر السكر بوسائل صديقة للبيئة ٣- إستخدام المركبات الحيوية الميكروبية التجارية

محمد فتحى معارج*، عبد المنعم ياسين الجندى، كمال محمد عجمى***، عبير صلاح ياسين***

* قسم الأمراض ووقاية النباتات- معهد بحوث المحاصيل السكرية- مركز البحوث الزراعية- جيزة- مصر، ** قسم الحيوان الزراعى والنيماتولوجى- كلية الزراعة- جامعة القاهرة- جيزة- مصر، *** قسم المعاملات الزراعية- معهد بحوث المحاصيل السكرية- مركز البحوث الزراعية- جيزة- مصر

الملخص العربى:

فى هذه الدراسة تم دراسة كفاءة ثلاث من المركبات الحيوية الميكروبية التجارية وهى بيوآرك وبيونيماتون وبيوزايد (التى تحتوى على البكتريا *Bacillus megaterium* والفطر *Paecilomyces lilacinus* والفطر *Trichoderma album* على التوالى) فى مكافحة نيماتودا تعقد الجذور *M. incognita* تحت ظروف المعمل والحقل مقارنة بكفاءة المبيد النيماتودى فايديت- وأظهرت النتائج أن كل المركبات الحيوية المختبرة والمبيد لها قدرة معنوية فى مكافحة هذه الآفة مقارنة بالكنترول الغير معامل تحت ظروف المعمل وظروف الحقل.

- تحت ظروف المعمل: وجد نسبة موت النيماتودا متغير تبعا لنوع المركب وتركيزه ومدة التعرض له، فوجد أعلى نسبة موت سجلت مع أعلى تركيز وأطول مدة تعرض لكل المركبات المختبرة وكذلك المبيد النيماتودى.

- النسبة المئوية لموت هذه النيماتودا إنحسر فى مدى من ٨٦,٨٧- ٩٣,٦٢% مع المركبات الحيوية المختبرة مقارنة مع نسبة موت ٩٨,٩٤% مسجلة مع المبيد "فايديت ٢٤% سائل".

- تحت ظروف الحقل: جميع المركبات الحيوية والمبيد طبقت بالمعدل الموصى به للفدان، وأظهرت النتائج أن الخفض فى التعداد النهائى للنيماتودا ينحسر فى مدى من ٧٢,٢٥ الى ٨٥,٧٠% فى المساحات المعاملة بالمركبات الحيوية أما المساحات المعاملة بالمبيد "فايديت ١٠% محبب" سجلت نسبة خفض ٨٤,١٥% فى التعداد النهائى للنيماتودا.

- تأثير المركبات المختبرة تحت ظروف الحقل كان أقل من تأثيرها تحت ظروف المعمل وهذا يرجع الى تخفيفها بالماء (ماء الرى) أو تفاعلها مع المكونات الحيوية والغير حيوية بالبيئة المحيطة- كما أظهرت النتائج قيم لمعامل التكاثر للنيماتودا فى المساحات المعاملة بالمركبات الحيوية كان فى مدى ٥ - ٩,٧، وفى المساحات المعاملة بالمبيد كان ٥,٥، أما فى المساحات غير المعاملة كان ٣٤,٩٦ ضعفا مقارنة بالتعداد الأولى فى التربة قبل الزراعة.

- عموما فى حقل بنجر السكر، المركب الحيوى بيوزيد الذى يحتوى على الفطر *T. album* يتكافأ فى تأثيره (٨٥,٧% خفض) مع تأثير المبيد "فايديت ١٠% محبب" (٨٤,١٥%) يليه المركب المحتوى على الفطر *P. lilacinus* (٧٨,١%) ثم المركب المحتوى على البكتريا *B. megaterium* (٧٢,٢٥%).

- كل المعاملات المختبرة حسنت من إنتاجية بنجر السكر الممثلة فى محصول الجذور ومحصول السكر الخام، فى مقارنة مع المساحات غير المعاملة، وجد أن المساحات المعاملة بالمركب الحيوي بيوزيد سجلت أعلى زيادة فى كل من محصول الجذور والسكر الخام يليه المركب بيونيماتون ثم المركب بيوآرك، بينما أقل زيادة سجلت مع المساحات المعاملة بالمبيد.

- من هذه النتائج يمكن للمركب الحيوى "بيوزيد" أن يحل محل المبيد "فايديت ١٠% محبب" فى مكافحة هذه النيماتودا على بنجر السكر أو يمكن مشاركة المركبان نيوماتون وبيوآرك مع المبيد فى برامج المكافحة المتكاملة لهذه الآفة ولتحسين إنتاجية بنجر السكر فى منطقة النوبارية.