J. Plant Protection and Pathology, Mansoura Univ. Vol. 1 (7): 539- 558, 2010

NEW TREND FOR *Meloidogyne javanica* MANAGEMENT BY *Myrothecium verrucaria* (DITERA) AS PROMISING BIOLOGICAL AGENT.

Khalil, A. E.*; A.G. El-Sherif; M. A. Bekhiet* and A. M. Kella***

*** Nematology Division, Plant Pathology, Res. Inst., ARC. Giza Egypt.**

**** Nematology Res. Unit, Agric. Zoology Dept., Fac. of Agric., Mansoura Univ., Egypt.**

ABSTRACT

Evaluation of DiTera a commercial formulation of *Myrothecium verrucaria* against *Meloidogyne javanica* was examined in-vitro for mortality, greenhouse as well as field trials on tomato and grape using various rates and method of applications. In-vitro test revealed a positive correlation between *M. javanica* J_2 mortality and biocide compound rate and concentration at various exposure periods. Larval mortality significantly increased with the increase of rates and concentrations from 10 up to 30%, as well, whereas, a slightly increase in *M. javanica* J_2 mortality was insignificantly noticed according to the tested two temperatures. With respect to tomato greenhouse trial, all tested DiTera rates and type of applications obviously caused remarkable increase in tomato growth cv. Castle Rock. Among DiTera rates, treatment at 0.3 g / seedling ranked first in increasing percentage of fresh weight of the whole plant with values of 56.73, 41.70 and 77.32% for the three type of applications i.e. at planting, post planting and pre planting respectively. As the rates of DiTera increased from 0.1 up to 0.3 g/ seedling, the percentage increase values of such plant growth parameter increased from 21.26 to 41.70%; 36.6 to 56.73%; and 57.5 to 77.32% for post planting, at planting and pre- planting application, respectively. Similar trend was noticed for suppressing nematode parameters, where the pre-planting application ranked first with values of 85.2, 89.2 and 94.1%, for the rate of 0.1, 0.2 and 0.3 g/ seedling, respectively. Moreover, It is worthy to note that there was a positive correlation between the rate of nematode build-up and DiTera compound rates, whereas this nematode criterion recorded to be the least value (0.26) by oxamyl treatment. Meanwhile, in tomato field trial, the rate of 0.3 g/ tomato seedling significantly decreased the number of galls on tomato roots with reduction percentages of 85.8, 71.0 and 59.7% for the pre-, at- and post- planting applications, respectively. All tested DiTera rates along with application times reduced tested nematode parameters. Among the three application times, the pre-planting method of adding DiTera compound accomplished the highest reduction percentages of nematode population densities with values of 74.3, 82.2 and 85.2 for 0.1, 0.2 and 0.3 g /plant respectively. Regarding grape greenhouse and field trials, all tested DiTera applications i.e. 2 g/plant 4 , 5, and 6 times at one week interval in both greenhouse and field trials obviously showed protection performance in grape plants cv. Thompson against nematode infestation of *M. javanica* in terms of reduction percentages of tested nematode parameters on grape roots as well. Likewise, as the addition of 2 g/ plant raised from four up to six times at one week interval, nematode criteria significantly decreased. Plants receiving DiTera treatments six times accomplished the highest reduction percentages in nematode parameters followed by that of five times and then four times applications. Moreover, oxamyl treatment twice at one month interval surpassed all tested DiTera application in suppressing nematode population, number of galls and eggmasses in both trials.

Keywords*:* Bio-control agent, DiTera compound (*Myrothecium verrucaria)*, grape, tomato, *Meloidogyne javanica.* Oxamyl.

INTRODUCTION

Tomato, *Lycopersicon esculentum* Mill. is considered to be one of the most important commercial and dietary vegetable crops all over the world including Egypt. Tomatoes are grown annually in three seasons - winter, summer and autumn - on about 3 percent of Egypt's total cultivated area. In 2005, Egypt ranked the fourth among the five top producers of tomatoes in the world, where 7600000 tons are produced according to FAOSTAT. Furthermore, the cultivated area of tomato in Egypt was approximately 464286 feddans.

Moreover, grape, *Vitis vinifera* is grown perennially. Its total cultivated area about 160.000 feddans in Egypt, where most of this area is located within the new reclaimed land.

Plant parasitic nematodes caused significant damage and losses to most agricultural crops in the tropical and sub-tropics (Luc *et al.*, 2005).

The root-knot nematodes, *Meloidogyne* spp. is among the most economically important parasites of tomato and grape cultivars in Egypt. When plants are severely infected by *Meloidogyne* spp. the normal root system is reduced to a limited number of severely galled roots with a completely disorganized vascular system. Rootlets are almost completely absent. The roots are seriously hampered in their main functions of uptake and transport of water and nutrients (Netscher and Sikora, 1990). Of the root-knot nematodes, *M. incognita* (Kofoid&White) Chitwood, *M. javanica* (Treub) Chitwood, *M. arenaria* (Neal) Chitwood and *M. hapla* Chitwood are considered to be the most popular species which affect major field and vegetable and fruit crops and cause more than 90% of the estimated damages.

Today, plant parasitic nematodes are successfully controlled by chemical nematicides. With an increase awareness of the harmful effects of chemical pesticides and the changing public attitude towards environmental pollution, chemical nematicides are losing their popularity among farmers for protecting their crops from nematode infestations. It has now become essential to search for alternative cheap and environmentally friendly ways for the management of phytonematodes in various agricultural crops. Some safe procedures for nematode control have been developed based on biological control agents and organic amendments; however, there is still a need for alternative compounds for effective nematode control to be developed (Noling& Becker, 1994).

DiTera, a commercial formulation of *Myrothecium verrucaria* (Valent, U.S.A) which is known to be produced commercially as new biological nematicides (Warrier *et al.,* 1999). DiTera used as biological control of root-knot nematode, *Meloidogyne* spp. on tomato by using different formulation from fungal antagonists (Hagag, 2009)

Shawky *et al*., (2010) reported that using Nemathoren (10%) and *Myrothecium verrucaria* (DiTera) especially at the highest concentration (1.5 g/pot) performed the highest decrease of *M. incognita* parameters in both soil and root of tested cassava cultivars, comparing with the other treatments , as well as, greatly improved the fresh weight of the whole plant where the

percentage of increase reached 87.2 and 72.1%, respectively. Literature revealed no enough information about DiTera as a biological control agent against such plant parasitic nematodes parasitizing economic plant crops. Therefore, the objective of the present research was study the following points:

I. Effect of DiTera compound at various rates and concentrations in comparison with distilled water on *Meloidogyne javanica* J_2 mortality at three period of exposure under two temperature regimes in-vitro,
II. Impact of DiTera compound *(Myrothecium verrucaria)* at three rates in comparison with oxamyl on controlling *Meloidogyne javanica* infesting tomato seedling cv. Castle-Rock under greenhouse conditions (25±2 C°),
III. Management of *Meloidogyne javanica* on tomato plants grown in a naturally infested sandy loam soil treated by DiTera at three rates through three types of application under field conditions, and
IV. Management of *Meloidogyne javanica* on grape plant cv. Thompson grown in naturally contaminated sandy loam soil by DiTera compound in comparison with oxamyl under greenhouse as well as field conditions.

MATERIALS AND METHODS

I. Source and Identification of *Meloidogyne javanica* under study:

To identify the root-knot nematode, *Meloidogyne javanica* used in this work, several young females were isolated from the infested cultivated cucumber roots field which was located at Omar Makrim village, Badr County, South Tahrir province, Behera governorate, Egypt, fixed in 4% formalin and kept in a test tube for perineal patterns examination. Following the procedure recorded by Taylor and Sasser (1978) in this respect, characteristics of their perineal patterns indicated the relation of young females of root-knot nematodes to *M. javanica*. Moreover, the second-stage juveniles of this nematode were then extracted from soil by sieving and modified Baermann technique (Goodey, 1957) according to design of each experiment of this investigation where was needed.

II. Source and preparation of DiTera tested rates and concentrations of each rate in nematode mortality test in-vitro

DiTera compound was brought by the authors, its tested rates was 0.1, 0.2, and 0.3 g that were separately added to 100 ml distilled water in a flask (250 ml each), then the concentration of each rate was separately obtained by adding 10 , 20 and 30 ml to 90, 80 and 70 ml distilled water to give the concentration of 10 , 20 and 30% for each one in flask (250 ml) separately.

A. Effect of DiTera compound at various rates and three concentrations in comparison with distilled water on *Meloidogyne javanica* J_2 mortality at three period of exposure under two temperature regimes in-vitro.

To study the mortality of *M. javanica* J_2 as influenced by DiTera compound at three rates i.e 0.1 , 0.2 and 0.3 g with three concentrations each i.e 10, 20 and 30% at 24, 48 and 72 hrs of exposure incubated in Petri dishes at 20 and 30 C° in-vitro test , 2 ml of each concentration of each DiTera rate

was added (as previously prepared) to half ml nematode suspension contained 100 second stage juveniles of *M. javanica* in Petri dish. As control treatment, two and half ml distilled water was also added to half ml nematode suspension (100 J_2). Each treatment was replicated four times. The nematode mortality counts were recorded according to the design of this experiment and any nematode recovery was observed and counted immediately. The exposure time required for 100% nematode mortality was recorded for each concentration of such tested DiTera rates as well as distilled water treatment. Data were statistically subjected to analysis of variance (ANOVA) (Gomez and Gomez, 1984), followed by Duncan's multiple-range test to compare means (Duncan, 1955).

B. Impact of DiTera compound (*Myrothecium verrucaria)* at three rates in comparison with Oxamyl on controlling *Meloidogyne javanica* infesting tomato seedling cv. Castle-Rock under greenhouse conditions (25±2 C°).

In order to study the impact of DiTera compound at three rates i.e. 0.1,0.2 and 0.3 g/pot comparison with oxamyl at the recommended dose (0.1 ml / pot on controlling *M. javanica* infesting tomato seedling cv. Castle-Rock under greenhouse conditions (25±2 C°) , thirty six earth pots, 20-cm-diam. containing 2kg each of sandy loam soil (90 parts sand : 8 parts clay : 2 parts silt)(V:V:V) was used in this experiment. This soil was brought from cucumber soil of Omar Makrim village, south Tahrir province, Behera governorate. Moreover, this soil was naturally contaminated with juveniles (J_2) of *M. javanica* at the level of one J_2 per one cm soil (2000 J_2 / pot). Soil of three earth pots was separately steam-sterilized before tomato seedling planting in order to serve as a control treatment free of nematode and any DiTera rates. Nine earth pots containing the contaminated soil with nematode separately received rates of DiTera compound i.e 0.1 , 0.2 and 0.3 g /pot (three pots each) without tomato seedling one week before planting that served as pre-planting application. Similar DiTera rates were also added to nine earth pots (three pots each) with one tomato seedling each at planting time. Another nine earth pots received these rates one week after tomato seedlings planting as post-planting treatment. Oxamyl as a nematicide was added twice to soil of three earth-pots with one seedling each at planting time and one month later, while the remaining three earth pots with soil and one seedling each was left without any DiTera rates under study to serve as nematode alone. Each DiTera rate was replicate three times within each method of application. Treatments were as follows:

a. pre- planting tomato-seedling application (nine pots):

1. N+ 0.1 g DiTera / pot
2. N + 0.2 DiTera / pot
3. N + 0.3 g DiTera / pot

b. At planting tomato-seedling application (nine pots):

1. N+ 0.1 g DiTera / pot
2. N + 0.2 DiTera / pot
3. N + 0.3 g DiTera / pot

c. post- planting tomato-seedling application (nine pots):

1. N+ 0.1 g DiTera / pot
2. N + 0.2 DiTera / pot
3. N + 0.3 g DiTera / pot

d. 1. N + oxamyl (0.1 ml) tomato seedling (3 pots):
e. 1. N alone (3 pots):
f. 1. Plant free of N and untreated (control) (3 pots):

Plants received water and protected conventional pesticide against mites as needed. After 60 days from the beginning of the experiment, plants were removed. Data dealing with lengths and weights of fresh shoot and roots were recorded. Number of *M. javanica* second stage juveniles in 250g soil/pot were extracted by sieving and modified Baermann- technique (Goodey, 1957), then calculated for the soil of each pot (2000 g soil / pot)counted by Hawksely counting slide under x 10 magnification and recorded. Rate of nematode build-up was measured and recorded for each DiTera rate and method of application. Infected roots of ten grams from each plant were washed with tap water, fixed in 4% formalin for 24 hr and stained in 0.01 lactic acid-fuchsin (Byrd *et al*., 1983) and then examined for the number of galls, developmental stages, females and egg-masses. Number of eggs per eggmass was also determined and recorded (Hussey and Baker 1975). Data were subjected to analysis of variance (ANOVA) as previously mentioned.

C. Management of *Meloidogyne javanica* on tomato plants grown in naturally infested sandy loam soil treated by DiTera at three rates through three types of application under field conditions.

In order to study the impact of DiTera at three rates i.e. 0.1, 0.2 and 0.3 g /tomato plant that was grown in naturally infested sandy loam soil (90 : 8:2 parts sand : clay and slit) *by Meloidogyne javanica* at the level of 2000 J2 /kg soil in comparison with oxamyl at the recommended dose (Vydate 24 % L)(0.1 ml /plant, twice at one month interval) under field conditions at Omar Makrim village, South Tahrir province, Behera governorate, Egypt, plots were cultivated by 40 tomato seedlings cv. Castle Rock at-15-days old each in experiment evaluating DiTera were 10 m. long with plants spaced at 25 cm. Each design of application i.e pre -, at- and post planting represented 3 plots where one plot was treated by one rate i.e 0.1 or 0.2 or 0.3 g /plant. Oxamyl as a nematicide treatment has only one plot (10m long) while one plot (10m) without any type of treatment as nematode alone (control). Treatments were as follows:

a. pre- planting application (3 plots):
1. DiTera compound 0.1 g /plant (4g/plot)
2. DiTera compound 0.2 g /plant (8g/ plot)
3. DiTera compound 0.3 g /plant (12g /plot)

b. At planting application (3 plots)
1. DiTera compound 0.1 g /plant (4g /plot)
2. DiTera compound 0.2 g /plant (8g /plot)
3. DiTera compound 0.3 g /plant (12g /plot)

c. post- planting application (3 plots)
1. DiTera compound 0.1 g /plant (4 g/ plot)
2. DiTera compound 0.2 g /plant (8g/ plot)
3. DiTera compound 0.3 g /plant (12g /plot)

d. Oxamyl treatment (one plot) at 0.1 ml/ plant (0.1ml / plant twice at one moth interval).

e. Nematode alone without any type of treatment (one plot)

Plants received water and protected by conventional pesticide against mites and insects as needed. After 60 days from starting the experiment, plants were up-rooted. Data dealing with either plant growth or nematode parameters was determined and recorded as previously mentioned .Root gall index (RGI) and eggmasses index (EI) were determined according to the scale gavin by Taylor and Sasser (1978) as follows: 0 =0 galls or eggmasses, 1=1-2 galls or eggmasses , 2=3-10 galls or eggmasses, 4=31-100 galls or eggmasses, 5= more than100 galls or eggmasses. Data were statistically subjected to analysis of variance (ANOVA) as previously mentioned.

D. Management of *Meloidogyne javanica* on grape plant cv. Thompson grown in naturally infested sandy loam soil by DiTera compound in comparison with oxamyl under greenhouse as well as field conditions.

1. **Greenhouse experiments**:

In order to study the impact of DiTera compound in comparison with oxamyl on controlling *Meloidogyne javanica* (J2) infesting soil with grape seedling cv. Thompson under greenhouse conditions, fifteen clay pots 25-cm-diam filled with sand loam soil (90 : 8:2 parts sand : clay and slit)(v:v:v)(2 kg soil) naturally infested by *M. javanica* (J2) at the level of 2000 J2/pot were divided into five groups, in the first , second, and third groups each pot/seedling separately received DiTera rates of 2 g /plant four or five or six at one week interval, while 5 ml of oxamyl was added twice to each seedling/ pot for the fourth group at one month interval, Meanwhile, the fifth group of pots/seedlings grown within that contaminated soil was left without any treatment as nematode alone to serve as a check (control). Each treatment was replicated three times. Treatments were as follows:

1. 1st group of pots /plants received 2 g DiTera/plant 4times at one week interval.
2. 2nd group of pots /plants received 2 g DiTera/plant 5 times at one week interval.
3. 3rd group of pots /plants received 2 g DiTera/plant 6times at one week interval.
4. 4th group of pots /plants received 5 ml / plant oxamyl twice at one month interval.
5. 5th group of pots or plants left without thing added as nematode alone (Control).

Plants received water and protected by conventional pesticide against mites and insects as needed. After 60 days from starting the experiment, plants were removed. Data dealing with nematode parameters was determined and recorded as previously mentioned. Data were also statistically subjected to analysis of variance as previously mentioned.

2. Field trials

In this experiment, one hundred grape trees cv. Thompson grown with the same contaminated soil by *Meloidogyne javanica* (J2) at the level of 2000 J2 /Kg soil. These hundred grape trees were divided into five groups where each group consisting of twenty trees with four replicates for each

treatment. Treatments of this field trial were carried out according to the previously design experiment for grape seedlings. After 60 days from the beginning of the experiment, number of *M. javanica* second stage juveniles in 250g soil/pot were extracted by sieving and modified Baermann- technique (Goodey, 1957), then calculated for the soil of each one kg soil, counted by Hawksely counting slide under x 10 magnification and recorded. Data dealing with nematode parameters was determined and recorded as previously mentioned. Data were subjected to analysis of variance (ANOVA) as previously mentioned.

The present study was undertaken in laboratory and greenhouse of Nematology Division, Plant Pathology Res. Inst., Giza and a naturally infested field with *M. javanica* on either cucumber or grape plants in Omar Makrim, Bader County, South Tahrir province, Behera governorate, Egypt.

RESULTS AND DISCUSSION

Nematicidial activity of DiTera compound, *Myrothecium verrucaria* against *Meloidogyne javanica* (J_2) in-vitro:

Impact of DiTera compound , *M. verrucaria* at three rates i.e. 0.1 , 0.2 and 0.3 g with three concentrations each i.e. 10 ,20 and 30% in comparison with distilled water singly on mortality of newly hatched juveniles of *M. javanica* under two temperature regimes (20&30 °C) in-vitro test are depicted in Table (1). In general, a positive correlation had observed between *M. javanica* J_2 mortality and fungus compound rates and concentrations at the three different exposure periods. Meanwhile, larval mortality significantly increased with the increase of fungal rates from 10 up to 30%, as well, whereas, a slightly increase in *M. javanica* J_2 mortality was insignificantly noticed according to the two temperature regimes within each fungal concentrations rate and time of exposure (Table 1) It is worthy to note that nematode mortality was recorded to be either 57.3 or 59.3%; 60.0 or 62.0% ; and 69.0 or 71.0% at 24 hr at 20 or 30 °C at the rate of 0.1 g DiTera compound with 10, 20 and 30%, respectively. Similar trend was obtained with 0.2 and 0.3 g DiTera compound with the same concentrations with values of 66.3 or 68.3% ; 75.3 or 76.3% ; 81.0 or 83.3% ; and 77.3 or 79.3% 81.7 or 83.7; and 92.0 or 94.9% at 24 hr at 20 or 30°C, respectively. Meanwhile, the highest values of nematode mortality were significantly obtained by the rates 0.1, 0.2 and 0.3 DiTera compound at 30% after 72 hr which were recorded to be either 73.7 or 75.7%; 90.0 or 92.0%; and 99.0 or 100.0% for 20 or 30 °C, respectively, comparing to distilled water as control treatment. (Table 1).

Table (1): Impact of DiTera compound (*Myrothecium verrucaria*) as a new biological agent at three rates with three concentrations each on mortality of *Meloidogyne javanica* J_2 at three exposure periods under two temperature regimes in-vitro

Rate of DiTera (g)	Concentration	Time of exposure (hr) / Temperature (C°)	*Nematode Mortality (n)		
			24	48	72
0.1	10%	20C°	57.3 f	59.7 ef	62.0 bcde
		30C°	59.3 ef	61.7 cde	64.0 z
	20%	20C°	60.0 def	64.3 z	67.7 xyz
		30°	62.0 bcde	66.3 yz	69.7 uvwxy
	30%	20C°	69.0 vwxy	61.6 cde	73.7 qrstu
		30°	71.0 ef	72.0 stuvw	75.7 abcd
0.2	10%	20C°	66.3 yz	70.7 tuvwx	72.0 stuvw
		30C°	68.3 wxyz	70.7 tuvwx	72.7 rstuv
	20%	20C°	75.3 pqrs	77.3 nopq	81.3 klmn
		30C°	76.3 qrst	79.7 lmno	81.7 klm
	30%	20C°	81.0 lmn	86.7 ghij	90.0 efg
		30C°	83.3 opqr	88.7 fgh	92.0 def
0.3	10%	20C°	77.3 nopq	79.0 mnop	81.0 lmn
		30C°	79.3 mnop	81.0 lmn	83.0 jklm
	20%	20C°	81.7 klm	85.3 hijk	88.0 fgh
		30C°	83.7 ijkl	87.0 ghi	90.0 efg
	30%	20C°	92.0 def	95.7 bcd	99.0 ab
		30C°	94.9 cde	97.7 ab	100.0a
Distilled water Control		20C°	0	0	0.67
		30C°	0	0	0.67

N=100 J2 of *M. javanica*
****Each value is a mean of four replicates.**
Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.
LSD (0.05) : 4.311

Data in Table (2) revealed the effect of DiTera compound rates i.e. 0.1 , 0.2 and 0.3 g/ seedling at three types of application i.e. pre planting, at planting and post planting in comparison with oxamyl at the recommended dose 0.1 ml/ seedling on tomato growth cv. Castle Rock infested with *M. javanica* under greenhouse at 25±2°C. Results indicated that all tested DiTera rates and type of their application obviously caused remarkable increase in tomato growth with various degrees. Among DiTera rates tested, rate treatment at 0.3 g / seedling ranked first in increasing percentage of fresh weight of the whole plant with values of 56.73 , 41.70 and 77.32% for the three type of applications i.e. at planting , post planting and pre planting respectively, comparing to nematode alone (Table 2). As the rates of DiTera increased from 0.1 up to 0.3 g/ seedling, the percentage increase values of such plant growth parameters increased from 21.26 to 41.70%; 36.6 to 56.73% ; and 57.5 to 77.32% for post planting, at planting and pre- planting application, respectively. Oxamyl as a nematicide ranked first over DiTera

other DiTera treatments tested in improving plant growth parameters i.e. the fresh weight of whole plant with percentage increase value of 82.20%, whereas, plant free of nematode and without any DiTera rates for the same criteria gave the highest percentage increase value of 85.08 % , comparing to nematode alone, respectively. (Table 2).

Table (2): Plant growth response of tomato cv. Castle Rock infested with *Meloidogyne javanica* as affected by three rates of Di-Tera compound (*Myrothecium verrucaria*) as a new biological nematicides and three types of application under greenhouse conditions at 25± 2C°.

Treatments	Time of application	DiTera Rate		*Plant growth response			
				Fresh weight (g)			
		pot (g)	Fed (kg)	Shoot	Root	Fresh wt. of the whole plant (g)	Increase %
DiTera	one week before planting	0.1	2	75.43 bc	26.33 e	101.76 c	57.50
		0.2	4	79.50 ab	30.73 bc	110.23 b	70.70
		0.3	6	81.50 a	33.03 b	114.53 ab	77.32
	at the same time of planting	0.1	2	63.79 fg	24.43 ef	88.22 d	36.60
		0.2	4	69.20 de	26.70 e	95.90 cd	48.50
		0.3	6	71.43 cd	29.80 cd	101.23 c	56.73
	one week after planting	0.1	2	55.18 h	23.14 f	78.32 f	21.26
		0.2	4	58.90 gh	25.93 ef	84.83 e	31.34
		0.3	6	63.40 fg	28.10 cde	91.50 d	41.70
Oxamyl 24% L		0.1 ml	3 lt+3 lt	81.71 a	36.00 a	117.7 a	82.2
Control				47.53 i	17.06 g	64.59 g	-
Plant free of nematode or any treatments				82.84 a	36.70 a	119.54 a	85.08

***Each value is a mean of three replicates.**

***Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.**

Data in Table (3) showed the impact of DiTera compound at three rates i.e. 0.1 , 0.2 and 0.3 g/ seedling added at three types of application i.e. pre –, at – and post –planting singly in comparison with oxamyl on number of developmental stages ; females and eggmasses on root system as well as number of juveniles of *M. javanica* per 2000 g soil infesting tomato under greenhouse conditions at 25±2°C. Data indicated that all tested materials obviously reduced such nematode parameters. Among tested DiTera rates at the three types of application, the pre-planting application ranked first in suppressing nematode population density with values of 85.2, 89.2 and 94.1%, followed by at planting application with values of 76.6, 82.9 and 87.9% and then that of post-planting application with values of 63.8, 75.9 and 82.3% for the rate of 0.1, 0.2 and 0.3 g/ seedling comparing to nematode alone, respectively. (Table 3). On the other hand, oxamyl treatment gave the highest percentage reduction of the same nematode criterion with value of

95.4% as comparing with nematode alone. Moreover, It is worthy to note that as there was a positive correlation between the rate of nematode build-up and DiTera compound rates, since as the rate raised from 0.1 up to 0.3 g/seedling, the rate of nematode reproduction decreased from 7.6 to 3.7; 4.9 to 2.5 ; and 3.1 to 1.22 for the post- , at- and pre- planting applications respectively , whereas this nematode criterion recorded to be the least value (0.96) by oxamyl treatment comparing to nematode alone. Among all tested rates and their application types, 0.3 g/ tomato seedling significantly decreased the number of galls on tomato roots with reduction percentages of 85.8, 71.0 and 59.7% for the pre- , at- and post- planting applications respectively. Concerning eggmass number, *M. javanica* reproduced less on plant treated with oxamyl or 0.3 g/seedling as pre-planting treatments with reduction percentage values of 89.4 and 88.6%. Likewise, significant results were noticed between eggmass indices of all DiTera rates irrespective of their application types and that of nematode alone (Fig 1).

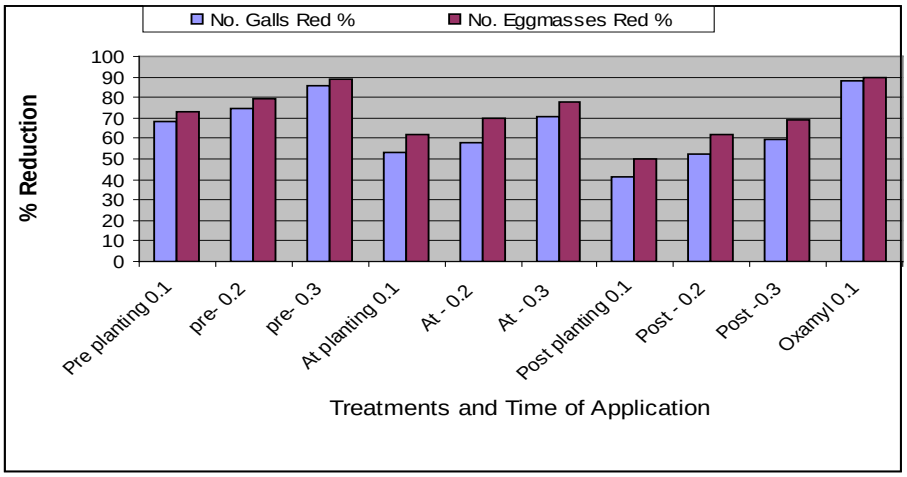

Fig. 1: Percent reduction of galls and eggmasses number of *M. javanica* on tomato cv. Castle Rock as influenced by DiTera compound rates 0.1, 0.2 and 0.3 g/plant added at pre- , at – and post planting applications in comparison with oxamyl (0.1 ml/plant) under greenhouse conditions.

Data presented in Table (4) revealed the impact of DiTera compound rates i.e 0.1, 0.2 and 0.3 g/ plant with three application methods pre-, at- and post- planting application in comparison with oxamyl at the recommended dose 0.6 ml/plant n controlling *M. javanica* J_2 on tomato cv. Castle Rock grown in naturally infested sandy loam soil at the level of 2000 *M. javanica* J_2 per 1 kg soil under field conditions. Results indicated that all tested DiTera rates along with application times reduced nematode population percentages, number of root galls, eggmasses as well as rate of nematode build up comparing to nematode alone (Table 4).

3

For each application time, as the Ditera rates increased from 0.1 up to 0.3 g reduction percentages of nematode parameters significantly increased comparing to nematode alone (Table 4). Among the three application times, the pre-planting method of adding DiTera compound accomplished the highest reduction percentages of nematode population densities with values of 74.3 , 82.2 and 85.2 for 0.1 , 0.2 and 0.3 g /plant respectively, comparing to nematode alone. Similar trend was observed in the case of root gall and eggmass number with reduction percentages values of 68.3, 71.9 and 80.0% and 64.4, 73.8 and 76.2% for the same application time, whereas the least values for the same nematode parameters were recorded by post planting application that amounted to 51.5, 56.2 and 71.2% and 58.8, 60.7 and 66.4% for 0.1, 0.2 and 0.3 g/plant comparing to nematode alone, respectively (Fig 2). Moreover, oxamyl as a nematicide rank first over DiTera compound treatments tested in suppressing nematode population, number of galls and eggmasses with values of 88.8, 83.2 and 81.4% followed by those of pre-planting application at 0.3 g/plant with values of 85.2 , 80.0 and 76.2% respectively, comparing to nematode alone. Meanwhile, among three application times of DiTera compound tested, the pre-planting application (one week before planting) achieved the lowest rate of nematode reproduction with values of 9.1 , 6.3 and 5.2 for 0.1 , 0.2 and 0.3 g/ plant, whereas, oxamyl treatment gave the least value of this nematode criterion (3.9) comparing to nematode alone, repectivily. It was worthy to note that DiTera rate of 0.3 gave moderately values of nematode reproduction that amounted to 8.0 and 8.7 for at – and post-planting applications comparing to nematode alone. Likewise, oxamyl treatment gave an equal values of galls and eggmasses indices (4) with the three DiTera rates of the pre- planting application as well as that of at planting time with 0.2 and 0.3 g/ plant, according to Taylor and Sasser (1978).

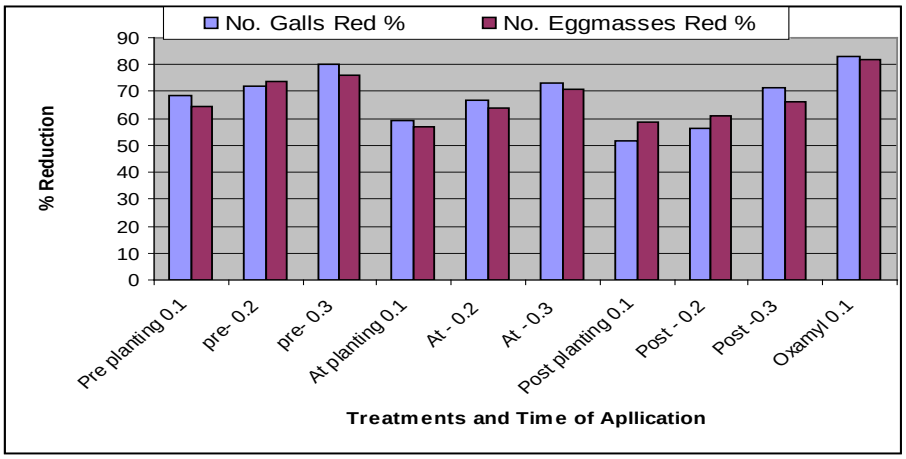

Fig. 2: Percent reduction of galls and eggmasses number of *M. javanica* on tomato cv. Castle Rock as influenced by DiTera compound rates 0.1, 0.2 and 0.3 g/plant added at pre- , at – and post planting applications in comparison with oxamyl (0.1 ml/plant) under field conditions

Data presented in Tables (5&6) revealed that all tested DiTera applications i.e. 2 g/plant 4 , 5, and 6 times at one week interval in both greenhouse and field trials obviously showed protection performance in grape plants cv. Thompson against nematode infestation of *M. javanica* in terms of reduction percentages of nematode population densities in soil and roots, numbers of galls and eggmasses and rate of nematode build-up on grape roots as well. It was clear that as the addition of 2 g/ palnt raised from four up to six times at one week interval, nematode parameters significantly decreased , especially between the four and six times of DiTera applications comparing to nematode alone (Table 5&6). Plants receiving DiTera treatments six times accomplished the highest reduction percentages in nematode population and number of galls and eggmasses followed by that of five times and then four times applications comparing to nematode alone with values of 83.23, 79.0 and 68.7% ; 68.7 , 62.6 and 57.1% ; 75.5 , 70.3 and 56.3 % in the greenhouse trial (Table 5 and Fig 3). Likewise, similar trend was noticed in the case of field trial where the same nematode parameters showed reduction percentage values of 92.9 , 87.2 and 78.5% ; 87.3 , 81.7 and 76.9 % ; and 90.8 , 85.5 and 77.2% , comparing to nematode alone, respectively. Moreover, oxamyl treatment surpassed all tested DiTera application in suppressing nematode population, number of galls and eggmasses in both trials, with values of 90.0 and 95.2% ; 73.2 and 89.5% ; and 81.6 and 93.2% for greenhouse and field trials comparing to nematode alone, respectively (Table 6 and Fig 4). Similar results were also recorded with the rate of nematode build-up where oxamyl treatment also ranked first in this nematode criterion, followed by DiTera compound at six time application with values of 1.6 and 1.1; and 2.7 and 1.7 for greenhouse and field trials, respectively.

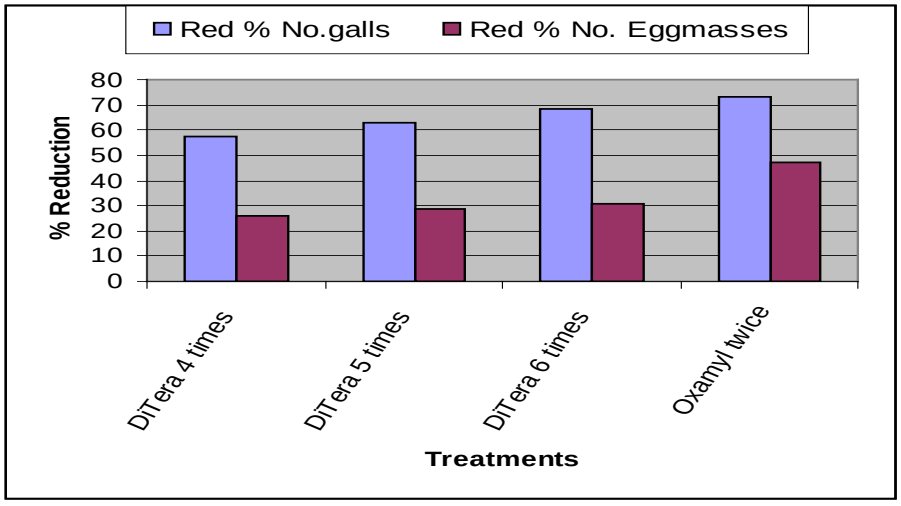

Fig. 3: Percent reduction of galls and eggmasses number of *M. javanica* on grape cv. Thompson as influenced by DiTera compound at 2g / plant with different times in comparison with oxamyl (5 ml/plant) under greenhouse conditions.

5

6

Promising results were also observed among tested DiTera applications especially with the indices of root galls as well as egmasses number where the lowest indices for those two nematode criteria was achieved by that of six time application with equal values (4&3 and 3&3) for greenhouse and field trials comparing to nematode alone, respectively (Table 5&6).

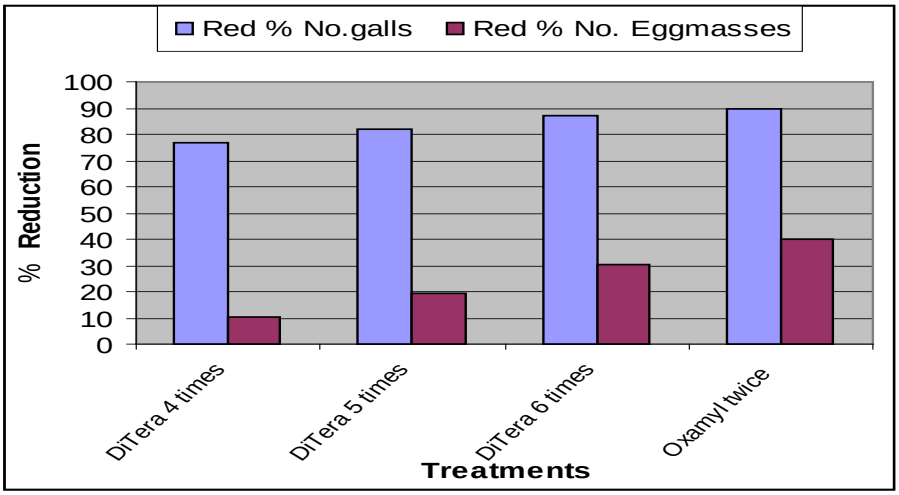

Fig 4: Percent reduction of galls and eggmasses number of *M. javanica* on grape cv. Thompson as influenced by DiTera compound with different times in comparison with oxamyl (5 ml/plant) under field conditions.

Apparently, results of the present study revealed the nematicidal properties of DiTera compound against the root-knot nematode, *M. javanica* in-vitro as well as under greenhouse and field trials on tomato and grape as well. The degree of nematode control varied due to certain factors. Of these factors, rate of the tested compound concentrations and methods of application. However, DiTera as a biological nematicide is based on a naturally occurring fungus, *Myrothecium verrucaria* . the fermentation process that yields DiTera produces multiple components, including water soluble molecules that either kill or impair nematodes. Properly timed DiTera applications protect roots from nematodes during key root flushes, when it matters the hosts. In addition, studies have demonstrated that DiTera increase beneficial microbial populations in the soil, contributing to over all root health. As for the rate and method of DiTera applications in tomato trials in the present investigation, the pre-planting application along with the rate of DiTera i.e. 0.1, 0.2 and 0.3 g/ seedling showed protection performance in tomato plants cv. Castle Rock against nematode infestation of *M. javanica* in terms of reduction percentage of nematode criteria and improving plant growth parameters. This is not surprising since the nematicidial activity of this DiTera compound occurs on contact, while the nematostatic activity slow nematode movement, inhibit egg hatch, interferes with sensory perception

and reduce feeding. Similar trend was observed with grape cv. Thompson trial under greenhouse and field trial as well, where as the addition of 2 g/ plant raised from four up to six times at one week interval, nematode criteria significantly decreased. These findings are in agreement with those reported by Shawky *et al.,* (2010) in respect to *Myrothecium verrucaria* (DiTera) especially at the highest concentration (1.5g/pot) that performed the highest decrease of *Meloidogyne incognita* parameters in both soil and root of tested cassava cultivars as well as, greatly improved the fresh weight of the whole plant. Moreover, oxamyl as a systemic nematicide acted successfully against *M. javanica* on tomato and grape trial as well; however, DiTera compound as a contact nematicide reduces *M. javanica* population to levels more or less comparable to that of oxamyl in this work.

In conclusion, DiTera compound is considered to be a safe contact nematicide as a promising biological agent against *M. javanica* on tomato and grape; and could be recommended to control such plant parasitic nematodes where organic crop production is needed.

REFERENCES

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

Duncan, D.B. (1955). Multiple rang and multiple, F-test Biometrics,11: 1-42.

FAOSTAT,ProdSTAT (Crops, 2005): The FAOSTAT ProdSTAT module on crops contains detailed agricultural Production data. Cited from: http://faostat.fao.org/site/PageID=567.

Gomez, K.A. and A.A. Gomez (1984). Statistical Procedures for Agricultural Research. 2nd Ed .,John Wiley&Sons. Inc. New York.

Goodey, J.B. (1957). Laboratory methods for work with plant and soil nematodes. Tech. Bull.No.2 Min. Agric. Fish Ed. London pp.47.

Hagag, Eman S. F. (2009). Biological control of root-knot nematode (*Meloidogyne* spp.) on Tomato by using different formulations from fungal antagonistics. Ph. D. Thesis, Fac. Agric. Kafr ElSheikh University, 112pp.

Hussey, R.S. and K. R. Baker (1973), A comparison of methods of controlling inoclua of *Meloidogyne* spp. Including a new technique. Plant Dis. Reptr, 57:1925-1928.

Luc, M.; R. A. Sikora and J. Bridge (2005). Plant parasitic nematodes in subtropical and tropical agriculture. CAB International, Wallingford, UK. 871 pp.

Netscher, C. and R. A. Sikora (1990). Nematode parasites of vegetables. 237-284 in M. Luc, R. A. Sikora and J. Bridge, eds. Plant parasitic nematodes in subtropical and tropical agriculture. CAB International.

Noling, J. W. and J. O. Becker (1994). The challenge of research and extension to define and implement alternatives to methyl bromide. J. Nematol., 26: 573-586.

Shawky, Samaa, M. ; A. E. Khalil and Manal, M. Soliman (2010). Seasonal fluctuation, inoculum levels and control of the root-knot nematode, *Meloidogyne incognita* on cassava. Minufiya J. Agric. Res., 35(3): 885-902.

Taylor, A.L.; and J. N. Sasser (1978). Biology , identification and control of root-knot nematodes (*Meloidogyne* species).Coop.Publ, Dep. Plant Pathol., North Carolina State Univ., and U.S. Agency Int.Dev., Raleigh, NC., 111pp.

Warrier, P.; A. Linda; M. Beach; P. A. Grau; G. W. Kirfman and J. M. Coleny. (1999). Commercial development and introduction of DiTera ™ , a new nematicide. Pestic. Sci. 55: 343-389.

اتجاه جديد لمكافحة نيماتودا *Meloidogyne javanica* بواسطة *Myrothecium verrucaria* (DiTera) كعامل حيوي واعد.

أشرف السعيد خليل *، أحمد جمال الشريف **، محمد علي بخيت * و عطف محروس كيله*

*** قسم النيماتودا ، معهد بحوث أمراض النباتات – مركز البحوث الزراعية – الجيزة – مصر.**

**** وحدة بحوث النيماتودا – قسم الحيوان الزراعي – كلية الزراعة – جامعة المنصورة – مصر.**

تم تقييم المركب الفطري DiTera ضد نيماتودا *Meloidogyne javanica* معمليا لتقدير نسبة الموت وأجريت تجارب في الصوبة والحقل علي الطماطم والعنب باستعمال معدلات وطرق إضافة عديدة. وأسفرت النتائج علي مايلي:

أولا: الاختبارات المعملية: لوحظ علاقة موجبة بين معدلات موت يرقات *M. javanica* ومعدلات المركب الفطري المختبرة (0.1، 0.2 ،0.3جم) وتركيزاتها (10% ، 20% ، 30%) عند فترات التعرض المختلفة المختبرة (24 ، 48 ،72 ساعة) حيث زادت معدلات الموت معنويا بزيادة معدلات المركب الحيوي وتركيزاتها من 10 الي 30% وكانت هناك زيادة طفيفة غير معنوية في موت اليرقات طبقا لدرجتي الحرارة المختبرة (20 ، 30 مº).

ثانيا: تجارب الصوبة علي الطماطم:

١. أدت كل المعاملات من معدلات DiTera وطرق اضافتها المختبرة الي زيادة معنوية واضحة في مقاييس نباتات الطماطم صنف كاستل روك وخفض واضح في مقاييس النيماتودا المختبرة.
٢. احتلت المعاملة 0.3 جم لكل نبات ضمن المعاملات المختبرة للمركب DiTera المركز الاول في معدل الزيادة وتحسين الوزن الرطب الكلي للنبات بقيم 56.73 ، 41.7 ، 77.32% عند إضافتها وقت الزراعة ، وبعد الزراعة وقبل الزراعة بأسبوع علي التوالي.
٣. كلما زادت معدلات المركب الفطري المختبرة من 0.1 الي 0.3 جم لكل نبات زادت نسب الوزن الرطب الكلي للنباتات من 21.26% الي 41.7% ومن 36.6 الي 56.73% و57.5 الي 77.32% عند إضافتها بعد الزراعة وعند الزراعة وقبل الزراعة بأسبوع علي التوالي.
٤. لوحظت نفس النتيجة بالنسبة لخفض مقاييس النيماتودا المختبرة حيث احتلت طريقة اضافة المركب قبل الزراعة باسبوع المركز الاول بقيم 85.2% ، 89.2% و 94.1% لمعدلات الاضافة 0.1 ، 0.2 ، 0.3 جم / نبات علي التوالي.
٥. لوحظ علاقة موجبة بين معدل تكاثر النيماتودا ومعدلات ووقت الاضافة للمركب الحيوي DiTera خاصة المعاملة 0.3 جم / نبات قبل الزراعة باسبوع حيث اعطت اقل القيم في المعاملات المختبرة وقدرها 1.22 بينما اعطت معاملة الاوكساميل قيمة أقل منها وهي 0.26.

و في تجربة الطماطم في الحقل:

٦. أدت المعاملة 0.3 جم / نبات الي نقص معنوي واضح في أعداد العقد النيماتودية علي جذور الطماطم بقيم 85.8% ، 71.0 % و 59.7% عند اضافتها قبل الرزاعة باسبوع وعند الزراعة وبعد الزراعة بأسبوع علي التوالي.
٧. أدت جميع معدلات اضافة المركب الفطري DiTera 0.1 ، 0.2 و 0.3 جم / نبات بكل طرق الاضافة المختبرة الي نقص واضح في نسب مقاييس النيماتودا.
٨. حيث أعطت طريقة الاضافة قبل الزراعة باسبوع أعلي معدل نقص في اعداد النيماتودا بقيم 74.3 ، 82.2، 85.2% بنسب اضافة المركب الفطري DiTera 0.1 ، 0.2 ، 0.3 جم / نبات علي التوالي.

ثالثا : تجارب العنب في الصوبة والحقل:

٩. أدت كل المعدلات المختبرة للمركب الفطري 2جم /نبات 4، 5، 6 مرات بين المرة والاخري أسبوع سواء في الصوبة او الحقل الي حماية نبات العنب صنف طومسون ضد عدوي النيماتودا الطبيعية في نقص معدلات مقاييس النيماتودا علي جذور العنب .
١٠. وكلما زادت عدد المرات لإضافة 2 جم / نبات من أربعة الي ستة زادت معدل نقص مقاييس النيماتودا.
١١. أعطت النباتات المعاملة 6 مرات بالمركب الفطري DiTera خلال التجربة أعلي معدلات نقص في مقاييس النيماتودا يليها المعاملة بخمس مرات ثم الاربع مرات.
١٢. أن اضافة المبيد الاوكساميل مرتين بين المرة والاخري شهر أدت الي نقص معنوي واضح في أعداد النيماتودا والعقد وكتل البيض في تجربتي العنب في الصوبة والحقل متفوق بذلك علي جميع المعاملات للمركب الفطري DiTera المختبرة.

ملحوظة هامة : تم اجراء التجارب لهذا البحث في معمل و صوبة ابحاث قسم النيماتودا معهد بحوث أمراض النبات مركز البحوث الزراعية – الجيزة أما تجارب الحقل أجريت في حقل مصاب طبيعيا بنيماتودا تعقد الجذور *M. javanica* كان مزروعا بالخيار وآخر كان مزوع بالعنب صنف طومسون بقرية عمر مكرم – مركز بدر – جنوب التحرير – محافظة البحيرة، مصر.

قام بتحكيم البحث

أ.د / فاطمة عبد المحسن مصطفي — كلية الزراعة – جامعة المنصورة
أ.د / عبد المنعم ياسين الجندي — كلية الزراعة – جامعة القاهرة

Table (3): Effect of three DiTera (*Myrothecium verrucaria*) rates as a new biological agent at three types of application infesting tomato cv. Castle Rock under greenhouse conditions at 25± 2C°.

Treatments	Time of application	DiTera rate		*Nematode population in				Final nematode population	Rate of build-up Pf/Pi	Reduction %	No of Galls	RGI	No. of Egg-masses	EI
					Ten grams root									
		pot (eg)	Feddan (kg)	Soil /pot	Develop. Stages	Females	Eggs/ root							
DiTera	one week before planting	0.1	2	290.0 ef	30.3 e	40.7 cde	5821.2	6182.1 e	3.1	85.2	40.7 cde	4	36.0 d	4
		0.2	4	250.0 f	24.3 f	31.7 def	4220.1	4526.1 e	2.3	89.2	31.7 def	4	27.0 e	3
		0.3	6	243.3 f	11.0 g	18.0 ef	2175.0	2447.3 f	1.22	94.1	18.0 ef	3	15.0 f	3
	at the same time of planting	0.1	2	350.0 cd	45.7 c	59.7 bc	9340.7	9796.1 b	4.9	76.6	59.7 bc	4	50.3 c	4
		0.2	4	320.0 de	40.7 cd	53.3 bcd	6749.0	7163.0 d	3.6	82.9	53.3 bcd	4	39.7 d	4
		0.3	6	267.7 ef	24.3 f	36.7 cde	4717.3	5064.0 e	2.5	87.9	36.7 cde	4	29.3 e	4
	one week after planting	0.1	2	416.0 b	56.7 b	74.3 b	14586.0	15133.0 b	7.6	63.8	74.3 b	4	66.3 b	3
		0.2	4	380.0 bc	40.3 cd	60.7 bc	9592.2	10073.2	5.04	75.9	60.7 bc	4	50.3 c	4
		0.3	6	323.3 de	37.7 d	51.0 bcd	7000.4	7412.4 b	3.7	82.3	51.0 bcd	4	40.7 d	4
Oxamyl 24% L		0.1 ml	3 lt+3 lt	243.3 f	3.7 h	14.7 f	256.2	617.6 f	0.26	95.4	14.7 f	3	14.0 f	3
N alone		-	-	1550.7 a	112.7 a	162.6 a	399446.1	41808.4 a	2.09	--	126.6 a	5	131.7 a	5

N=2000 J2 of *M. javanica* /Kg

***Each value is a mean of three replicates.**

***Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.**

*** Root gall index (RGI) or eggmass index (EGI): 0= no galling or eggmasses, 1=1-2 galls or eggmasses; 2=3-10 galls or eggmasses; 3= 11-30 galls or eggmasses; 4= 31-100 galls or eggmasses and 5= more than 100 galls or eggmasses. (Talyor and sasser, 1978).**

Table (4): Population density, rate of build-up and egg-masses number of *Meloidogyne javanica*. Infecting tomato cv. Castle Rock as affected by three rates of Di-Tera compound (*Myrothecium verrucaria*) as a new biological agent under field conditions.

Treatments	Time of application	DiTera Rate		*Nematode population in				Final nematode population	Rate of build-up Pf/Pi	Reduction %	No of Galls	RGI	No. of Egg-masses	EI
				Soil /1Kg	Ten grams root									
		Plant (eg)	**Feddan/ Kg		Develop. Stages	Females	Eggs/ root							
Di-tera	one week before planting	0.1	2	400.0 dcd	57.0 e	87.7 e	17568.39	18113.1 f	9.1	74.3	87.7 e	4	80.7 cd	4
		0.2	4	333.3 cde	46.3 f	77.7 fg	12122.89	12580.2 g	6.3	82.2	77.7 fg	4	59.3 gh	4
		0.3	6	293.3 de	34.7 g	55.0 h	10081.8	10464.8 h	5.2	85.2	55.0 h	4	54.0 h	4
	at the same time of planting	0.1	2	393.3 bcde	77.3 cd	112.7 d	23038.01	23621.4 c	11.8	66.5	112.7 d	5	97.3 b	4
		0.2	4	380.0 bcde	60.3 e	92.3 e	21105.56	21638.2 d	10.8	69.3	92.3 e	4	81.7 de	4
		0.3	6	346.7 bcde	55.7 ef	73.7 g	15454.39	15930.5 g	8.0	77.4	73.7 g	4	66.7 fg	4
	one week after planting	0.1	2	446.7 b	91.7 b	134.0 b	24576.58	25248.9 e	12.6	64.2	134.0 b	5	93.3 bc	4
		0.2	4	413.3 bc	84.7 be	121.0 c	20545.9	21164.9 b	10.6	70.02	121.0 c	5	89.3 bcd	4
		0.3	6	366.7 bcde	74.3 d	79.7 f	16792.6	17313.3 f	8.7	75.5	79.7 f	4	76.3 ef	4
Oxamyl 24% L.		0.1 ml	3 lt+3 lt	286.7 e	33.0 g	46.3 i	7516.53	7882.5i	3.9	88.8	46.3 i	4	41.0 i	4
Control		-	-	2000 a	212.0 a	276.3 a	68100.0	70588.3 a	35.3	0	276.3 a	5	227.0 a	5

N=2000 J2 of *M. javanica* /Kg

***Each value is a mean of four replicates.**

***Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.**

*** Root gall index (RGI) or eggmass index (EGI): 0= no galling or eggmasses, 1=1-2 galls or eggmasses; 2=3-10 galls or eggmasses; 3= 11-30 galls or eggmasses; 4= 31-100 galls or eggmasses and 5= more than 100 galls or eggmasses. (Talyor and sasser, 1978).**

Table (5): Effect of of Di-Tera compound (*Myrothecium verrucaria*) as a new biological nematicides with different times on *Meloidogyne javanica*. Infesting grape cv. Thompson under greenhouse conditions.

Treatments	Dose / Tree (g)	N. of applications	Dose /Feddan (Kg)	*Nematode population in				Final nematode population	Rate of build-up Pf/Pi	Reduction %	No of Galls	GI	No. of Egg-masses	EI
				Soil /1Kg	Ten grams root									
					Develop. Stages	Females	Eggs/ root							
Di-tera	2	Four time	8	340.0 b	32.0 b	42.0 b	9806.79	10219.6 b	5.1	68.7	42.0 b	4	35.7 b	4
	2	Five time	10	303.3 b	27.3 bc	36.7 b	6456.51	6831.8 bc	3.4	79.0	36.7 b	4	24.3 c	3
	2	Sex time	12	276.7 b	18.7 cd	30.7 c	5146.0	5472.7 c	2.7	83.23	30.7 c	4	20.0 d	3
Oxamyl 24% L	5 ml	2	5 lt	283.3 b	15.0 d	26.3 c	2950.5	3275.2 d	1.6	90.0	26.3 c	3	15.0 e	3
Control				2100.0 a	74.7 a	98.0 a	30367.89	32640.6 a	16.3	0	98.0 a	4	81.7 a	4

N=2000 J2 of *M. javanica* /Kg

***Each value is a mean of three replicates.**

***Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.**

*** Root gall index (RGI) or eggmass index (EGI): 0= no galling or eggmasses, 1=1-2 galls or eggmasses; 2=3-10 galls or eggmasses; 3= 11-30 galls or eggmasses; 4= 31-100 galls or eggmasses and 5= more than 100 galls or eggmasses. (Talyor and sasser, 1978).**

Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.

Table (6): Effect of of Di-Tera compound (*Myrothecium verrucaria*) as a new biological nematicides with different times on *Meloidogyne javanica*. Infesting grape cv. Thompson under field conditions.

Treatments	Dose / Tree (g)	No of Applications	Dose /Feddan (Kg)	*Nematode population in				Final nematode population	Rate of build-up Pf/Pi	Reduction %	No of Galls	RGI	No. of Egg-masses	EI
				Soil /1Kg	Ten grams root									
					Develop. Stages	Females	Eggs /root							
Di-tera	2	Four times	8	893.3 b	22.3 b	40.3 b	9189.99	10156.0 b	5.1	78.5	40.3 b	4	30.3 b	4
	2	Five times	10	720.0 c	15.3 c	31.3 c	5263.11	6038.0 bc	3.0	87.2	31.3 c	4	19.3 c	3
	2	Six times	12	413.3 d	11.3 cd	21.7 d	2902.8	3349.1	1.7	92.9	21.7 d	3	12.3 d	3
Oxamyl 24% L	5 ml	2	5 lt	400.0 d	9.0 d	18 .0d	1829.7	2265.7	1.1	95.2	18.0 d	3	9.0 d	2
Control				1826.7 a	124.7 a	171.3 a	45047.1	47169.8	23.6	0	171.3 a	5	133.0 a	5

N=2000 J2 of *M. javanica* /Kg
***Each value is a mean of four replicates.**
***Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.**
*** Root gall index (RGI) or eggmass index (EGI): 0= no galling or eggmasses, 1=1-2 galls or eggmasses; 2=3-10 galls or eggmasses; 3= 11-30 galls or eggmasses; 4= 31-100 galls or eggmasses and 5= more than 100 galls or eggmasses. (Talyor and sasser, 1978).**
Means in each column followed by the same letter(s) did not differ at p<0.05 according to Duncan multiple-range test.