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Comparison Efficiency of Certain Mesofaunatic Predators against *Meloidogyne javanica* Infected Tomato Plants

Abo-Korah, M. S.
Economic Entomology and Agricultural Zoology Dept., Fac. of Agric., Menoufia Univ., Shibin El-Kom, Egypt.

ABSTRACT

A comparison between predation efficiency of certain soil mesofaunatic predators such as: *Macrocheles matrius* (mite); *Onychiurus armatus* (collembolan) and *Diplogaster lheritieri* (nematode) solely or combined in addition to a nematicide (Tervigo) as standard to these treatments on *Meloidogyne javanica* infected tomato plants in pot experiments under greenhouse conditions. The obtained results revealed that all tested soil mesofaunatic predators and Tervigo significant reduced *Meloidogyne javanica* population in soil and roots of tomato plants. Predation efficiency of the predatory nematode treatment, *D. lheritieri* recorded the first rank, followed by the predatory mite, *M. matrius* and collembolan, *O. armatus* with reduction percentages of 86.7; 79.2 and 73.1% on root-knot nematode , respectively. Combined treatment of (*M. matrius* + *O. armatus* + *D. lheritieri*) gave the highest reduction percentage (91.2%) on root-knot nematode compared with the application of the nematicide Tervigo (87.7%). In addition, effective of this combination gave a best effect of various growth parameters of tomato, as well as best results of gall index (0.5) decreased by (- 91.7%) and egg-masses production (3.0) decreased by (- 95.2%) compared with control value, respectively, while in Tervigo treatment revealed (1.0) and (5.0) gall index and egg-masses production which decreased by (- 83.3 and - 91.1%) comparing to control values, respectively. Application of the nematicide Tervigo as comparator with certain natural soil predators in nematode control sheds a light upon the implicitly importance of biological control agents as a successful methods in the control of plant-parasitic nematode, *M. javanica* instead of the application of dangerous nematicides on the environment.

Keywords: Biological control, *Macrocheles matrius*, *Onychiurus armatus*, *Diplogaster lheritieri.*

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is an important food and cash crop of the farmers (Lemma, 2004). It's one of the most wide cultivate vegetable crops in Africa and in the world as a whole (Opena and Kyomo, 1990). Tomato consider important source of human nutrition. It is an major source of phosphorus, iron and vitamin A, B and C; (Naika *et al.,* 2005). Tomato is heavily attacked by root-knot nematode, *Meloidogyne* spp. and the species *Meloidogyne javanica* is the dominant species (Lemma, 2002).

Root-knot nematode *Meloidogyne javanica* is a major constraint to successful vegetable production all over the world, causing severe damage that leads to dramatic yield losses (Sikora and Fernandez, 2005). Control of root knot nematodes has been primarily accomplished through chemical nematicides (Widmer and Abawi, 2000). However, due to the significant drawbacks of the chemical control including threats to human health and the environment, biological control has become one of the promising alternatives (Stirling, 1991).

Biological control of plant-parasitic menatode using predatory mites has been extensively practiced in many countries, especially in protected plants. Macrochelids have a potential role as biological control agents on vermiform nematodes and organisms which found associated include different species of flies, and other of soil fauna (Beaulieu and Weeks 2007).

Many common soil fauna feed on nematodes and may have potential in biological control. These include micro arthropods such as mites and Collembolan (springtails), as well as tardigrades (Yeates *et al.,* 1997). Most of these predators of *Meloidogyne javanica* nematode are widely distributed and common in soils, including mites, collembolan, predatory nematodes, and other organisms (Coleman and Crossley, 1996). Mites and collembola are abundant in soil and many species are known to prey on nematodes (Stirling, 2014).

Diplogaster sp. is a very important predatory nematode caused significantly reduction in nematode population in the roots of tomato. It has a voraciousness in its predation on *M. javanica* (Osman, 1988). Predatory and omnivorous nematodes are found in most soils and may feed on and suppress plant-parasitic nematodes. The diplogasterid predators are important and may prove to be promising biological control agents of plant parasitic nematode. Potential *of Diplogaster,* as a predator of some plant parasitic nematodes *in vitro* studies were carried out to assess the efficacy of 115 *Diplogaster* species as a predator of second stage juveniles *of Meloidogyne javanica* and *Tylenchulus semipenetrans* at 25°C and 25% relative humidity in an incubator (Khan and Kim, 2005; McSorley *et al.,* 2006; Sanchez-Moreno and Ferris, 2007).

The present study was carried out to compare the predation efficiency of certain soil mesofaunatic predators such as: *Macrocheles matrius* (mite); *Onychiurus armatus* (collembolan) and *Diplogaster lheritieri* (nematode) solely or combined compared with the nematicide (Tervigo) in biological control of *Meloidogyne javanica* infected tomato plants under greenhouse conditions.

MATERIALS AND METHODS

Nematode culture:

Juveniles of the root knot nematode, *M. javanica* were rearing of pure culture on black nightshade, planted with *Solanum nigrum* in the Nematode laboratory of the Entomology and Zoology Department, Faculty of Agriculture , Menoufia University.

Experimental preparation and design:

Potted experiment was carried out under greenhouse conditions at the Experimental Farm of the Faculty of Agriculture, Menoufia University, Shibin El-Kom, Egypt. Experiment layout was randomized complete block design. for treatments were represented with three replicates. Three seedlings of the tomato, *Solanum lycopersicum* L. (Variety super strain-b) five weeks old were planted in wood pot 25 cm in diameter filled with four kg sterilized clay-sand mixed soil (1:1, v/v). After seven days for seedlings adaptation, thousand (1000) J2 of *M. javanica* per 1kg soil were added by pipette into three holes around each seedling. One hundred individuals of soil mite *M. matrius*; collembolan, *O. armatus* and predator nematode, *D. lheritieri* were added solely or combined per pot at the same time.

Material preparation:

Mite individuals and collembolans were collected from chicken and farmyard manure, and extracted by using modified Tullgren funnels for 72 hours (Lindquest *et al*,.1979). The extracted predators were received in distilled water and then transferred into plastic rearing units then transferred onto plastic rearing units (Fouly, 1996). Manure samples were taken by an iron sampler vol, 1000cm3 from experimental Farm at Faculty of Agriculture, Menoufia University. Examination of soil mesofauna was carried out twice daily by a stereomicroscope to identify and count all the individuals. Pure culture of the predator nematode, *Diplogaster lheritieri* was obtained from the Nematology and Biotechnology Laboratory, Faculty of Agriculture, Fayoum University and reared under laboratory conditions.

TervigoTM is a suspension concentrate (SC) contain of 20g/L abamectin plus iron chelate Fe-EDDHA 400g/L produced by Syngenta East Africa Ltd. Abamectin as an active ingredient provides effective against nematodes, while the iron chelate is a micro fertilizer that provides crop enhancement effects especially in alkaline soils. After one week of seedlings adaptation, Tervigo was applied as 0.4 ml per plant as soil and pot around the roots (full recommended dose 2.5 liter/fedan).

Nematode Extraction and Counts:

Each composite soil sample was carefully mixed, and an aliquot of 100 cm3 was processed for nematode extraction according to methods described by (Southey, 1970) each treatment was replicated 3 times. An aliquant of 1 ml each of nematode suspensions were pipetted off, placed in a Hawksley counting slide and examined by using a stereomicroscope.

Nematode counts in soil and roots were done after 30, 60 and 90 days of application. At the finish of the experiment, we are recorded roots & shoots fresh weight and plant height. Roots were carefully washed by distill water, and the nematode galls were counted and rated as mentioned in Table (1), as well as (1 gram) per root was stained by acid fuchsin lactophenol to counted root knot nematode stages into the roots with the aid of a dissecting microscope. Egg masses were assessed by staining the roots with Phloxin-B solution (0.15 g/l tap water) for twenty minutes according to (Daykin and Hussey 1985).

Table 1. Rating scale levels of resistance or susceptible by gall numbers (Southey, 1970)

Number of galls/ root system	Gall index	Resistance rating
0	0	Immune
1-2	1	Highly resistant
3-10	2	Resistant
11-30	3	Moderately resistant
31-70	4	Moderately susceptible
71-100	5	Susceptible
>100	6	Highly susceptible

Statistical analysis:

The obtained results were subjected to analysis of variance (ANOVA) using CoStat Software, Version 6.4 (2008). The mean differences were compared by Least Significant Difference (L.S.D. 5%)

Reduction percentages were computed according to Abbott formula (1925).

Increase or decrease % = Control – treatment / Control x 100

RESULTS AND DISCUSSION

Data presented in Table (2) indicated that, the mean of *M. javanica* juveniles per 100 g soil 30, 60 and 90days after the applicated of three soil mesofaunatic predators solely or combined compared with a nematicide (Tervigo) for management *M. javanica* infected tomato plants under shield condition.

Statistical analysis indicated that all treatments significant different of suppressed the *M. javanica* population in the soil treated after thirteen, sixteen and nineteen days in comparison with untreated treatment.Combined treatment of (*M. matrius* + *O.armatus*+ *D. lheritieri*) recorded the lowest average of population density of *M. javanica* in the three months (96.8 ind's) compared with a nematicide (Tervigo) which recorded (136.0 ind's).

Table 2. Impact of soil mesofaunatic predators on the population density of *M. javanica* infected tomato plants and reduction percentage ,under greenhouse conditions.

Treatments	Aver. no. of *M. javanica* juveniles/ 100 g soil Days post-treatments				Reduction %			
	30 Days	60 Days	90 Days	Overall mean	30 Days	60 Days	90 Days	overall mean
M. matrius	317.0 c	292.0c	110.0c	239.6c	68.9	76.8	91.8	79.2
O. armatus	421.0b	331.0b	175.0b	309.0b	58.8	73.7	86.9	73.1
D. lheritieri	294.0d	101.3f	41.0g	145.4f	71.2	91.9	96.9	86.7
M. matrius + O.armatus	315.3c	241.0d	101.0d	185.8d	69.1	80.8	92.4	80.8
M. matrius + D. lheritieri	280.0e	83.0g	58.0f	140.3f	72.6	93.4	95.7	87.2
O.armatus + D. lheritieri	289.3d	123.0e	67.0e	169.7e	71.7	90.2	94.9	85.6
M. matrius+O.armatus+D. lheritieri	195.0g	72.3h	23.0h	96.8h	80.9	94.3	98.3	91.2
Tervigo	245.0f	127.0e	36.0g	136.0g	76.0	89.9	97.3	87.7
Control (*M. javanica*) only	1021.3a	1260.0a	1337.0a	1206.1a	-	-	-	-
LSD 5%	8.6	6.8	5.1	8.5	-	-	-	-

Average in each column followed by the same letter (s) are not significantly different at five% level.

Highest reduction percentages of the *M. javanica* population in the soil, were recorded by application of combined treatment (*M. matrius* + *O.armatus*+ *D. lheritieri*) followed by (Tervigo) with (91.2 and 87.7 %), respectively.

Results showed that, the nematode predator (*D. lheritieri*) considered the most powerful predator on *M. javanica*, recorded the highest reduction percentages of the *M. javanica* population in the soil, (86.7 %) followed by the two other predators mite and collembolan which recorded (79.2 and 73.1 %), respectively, descendingly.

The obtained results are in agreements with (McSorley and Koon, 2009) who reported that the relative suppression of *M. javanica* is greater in tomato soil, and that the occurrence of some invertebrate predators, especially mites; collembola and nematode predator are consistent with the nematode population declines observed.

Regarding to the influence of treatments solely or combined on some tomato plant characters, such as plant height; shoot weight; root weight; root gall index and egg-masses production. The statistical analysis of the obtained data in Table (3) recorded that there are significant different between all solely or combined treatment and untreated treatment.

These results indicated that, all tested led to considerable increment of plant height; shoot weight; root weight and consider decrement of root gall index and egg-masses production compared with control (pathogenic treatment). The highest increase of plant height, shoot weight and root weight were obtained by combined application with (*M. matrius* + *O. armatus*+ *D. lheritieri*) and recorded the least number of root gall index (0.5) and egg-masses production (3.0). While these parameters (gall index and egg-masses production) in Tervigo treatment are higher than that found in previously combinaed treatment which recorded (1.0 and 5.0) of gall index and egg-masses, respectively.

When the apply in soil number of mites and collembolan were over 300 ind's, the egg masses of *M. javanica* began to decrement significantly, which probability because mites and collembolan preferring to *M. javanica* on more active juveniles than fixed egg masses (Chen *et al.,* 2013).

Table 3. Influence of treatments solely or combined on some tomato plants characters; root gall index and egg-masses production.

Treatments	plant height cm	shoot weight g	root weight g	root gall index	Egg-masses production
M. matrius	45.7e	60.1f	10.0de	3.0c	27.0c
O. armatus	41.0f	59.0f	9.1ef	4.0b	29.0b
D. lheritieri	55.4c	70.3c	13.9bc	1.5def	13.0f
M. matrius + *O.armatus*	48.0de	64.3e	11.9cd	2.5cd	24.0d
M. matrius + *D. lheritieri*	57.6c	71.0c	15.1b	1.5def	9.0g
O.armatus + *D. lheritieri*	51.2d	67.5d	11.0de	2.0cde	19.0e
M. matrius + *O.armatus*+ *D. lheritieri*	66.3a	77.5a	18.0A	0.5f	3.0i
Tervigo	62.1b	74.0b	13.2bc	1.0ef	5.0h
Control (*M. javanica*) only	36.5g	49.0g	7.7f	6.0a	62.0a
LSD 5%	3.4	1.7	1.7	0.9	1.7

Average in each column followed by the same letter (s) are not significantly different at 5% level.

Table 4. Increase or decrease percentages of some growth characters, gall index and egg-masses production on tomato as influenced by treatments solely or combined application.

Treatments	Increase or decrease (%)				
	plant height	shoot weight	root weight	root gall index	Egg-mass
M. matrius	+25.2	+22.7	+29.9	-50.0	-56.5
O. armatus	+12.3	+20.4	+18.2	-33.3	-53.2
D. lheritieri	+51.8	+43.5	+80.5	-75.0	-79.0
M. matrius + *O.armatus*	+31.5	+31.2	+54.5	-58.3	-61.3
M. matrius + *D. lheritieri*	+57.8	+44.9	+96.1	-75.0	-85.5
O. armatus + *D. lheritieri*	+40.3	+37.8	+42.9	-66.7	-69.4
M. matrius + *O. armatus*+ *D. lheritieri*	+81.6	+58.2	+133.3	-91.7	-95.2
Tervigo	+70.1	+51.0	+71.4	-83.3	-91.9
Control (*M. javanica*) only	-	-	-	-	-

There for the increment or decrement of tested plants characters Table (4), the obtained results express that all plant treatments increased plant height; shoot weight and root weight, while it decrement root gall index and egg-masses production.

Our results are in agreements with (Bilgrami *et al.,* 2005) who decided that, the predator nematodes fed on many different spices of *Meloidogyne.* nematodes. Both mite and collembolan of micro arthropods are known to fed on plant parasites and other nematodes and decrease root-gall and egg masses (Manwaring *et al.,* 2015).

Machrochelidae mites are predaceous in nature and fed voraciously on plant and soil nematodes belonging to different trophic groups. Machrochelidae possess greater predatory potentials mainly because of their ability to kill a variety of nematodes in large numbers in culture and experimental dishes (Bilgrami, 1993).

Chemical control is the dominant and effective way against *M. javanica* nematode in the our world (Whitehead 1997). However, repeated and extensive use of chemical pesticides has brought a series of negative impacts on environment and human health. These limitations worthy of to introduce these predators in a successful strategy for controlling nematodes as a substituent long use of chemical pesticides which has brought contrariwise of preceding limitations (Chun *et al.,* 2014).

Finally it could be concluded that, predatory nematode *D. lheritieri* on *M. javanica* came in the first rank relative to its predation efficiency. *M. matrius* as a predatory mites play a very important role in its fed voraciously on *M. javanica* at the absence of mites as a natural prey for it, and occupied the 2nd position for its fed on *M. javanica*. *O.armatus* (collembolan) as a predatory insects fed on nematode relatively little as a substituent of its natural insect preys. Combined treatment (*M. matrius* + *O.armatus*+ *D. lheritieri*) gave a forceful and the highest reduction percentage on *M. javanica*, as a result of its accumulative effectiveness of predation rates which created consequently the higher predaciousness potentiality than solely treatments of these bio control agents, compared with the treatment of the nematicide Tervigo. Hence, the application of natural soil mesofaunatic predators for controlling *M. javanica* are more safe; effective; eco-friendly and without any costs at all, therefor these limitations should be introduce these creatures in successful strategies as management tools for controlling *M. javanica* by predators with high bio control potential. To take into that a nematicide Tervigo is contrariwise with previously limitation.

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مقارنة بين كفاءة بعض مفترسات حيوانات التربة لمقاومة نيماتودا تعقد الجذور التى تصيب نباتات الطماطم

محمد سعيد ابوقورة

قسم الحشرات الاقتصادية والحيوان الزراعى – كلية الزراعة – جامعة المنوفية – مصر

تمت المقارنة بين القدرة الأفتراسية لبعض مفترسات حيوانات التربة وهي أكاروس *Macrocheles matrius* وحشرة الكولمبولا *Onychiurus armatus* والنيماتودا المفترسة *Diplogaster lheritieri* وذلك في معاملات منفردة أو مركبة بالإضافة إلى المبيد النيماتودي Tervigo كمقارنة بين هذة المعاملات. وقد أجريت الدراسة على نيماتودا تعقد الجذور *Meloidogyne javanica* في شتلات الطماطم وذلك في أصص تحت ظروف الصوبة الزجاجية. أدت جميع المعاملات من المفترسات والمبيد النيماتودي إلى خفض معنوي في الكثافة العددية للنيماتودا في التربة والعقد الجذرية. وفي التجارب الفردية للمفترسات كان ترتيب المفترس النيماتودي *D. lheritieri* الأول حيث أدى إلى موت النيماتودا بنسبة عالية يتبعة المفترس الأكاروسي *M. matrius* وأخيراً *O. armatus* (الكولمبولا) كانت في المركز الثالث بنسب موت على النيماتودا (٨٦.٧ & ٧٩.٢ & ٧٣.١%) على التوالي. أما المعاملة المركبة (*M. matrius + O.armatus+ D. lheritieri*) فقد أدت إلى أعلى نسبة لموت نيماتودا تعقد الجذور (٩١.٢ %) ثم المبيد النيماتودي (٨٧.٧%). إضافة إلى ذلك أدت هذة المعاملة المركبة إلى تحسين صفات النمو المختلفة لنباتات للطماطم. كما أعطت لبمعاملة المركبة أفضل النتائج فيما يخص العقد الجذرية (٠.٥) بنسبة نقص بلغت (-٩١.٧%) وكذلك في إنتاج كتل البيض (٣.٠) بنسبة نقص بلغت (-٩٥.٢%) مقارنة بالكنترول. إن تطبيق المبيد النيماتودي كمقارنة لبيان كفاءة المفترسات الطبيعية في التربة وتأثيرها على النيماتودا ألقى الضوء على الأهمية القصوى لتطبيق عوامل المقاومة الحيوية كالمفترسات كاستراتيجية جديدة لمقاومة نيماتودا تعقد الجذور *M. javanica* بالمقارنة باستخدام المبيدات النيماتودية في هذا الغرض.