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Distribution of nematode genera and seasonal fluctuation of *Meloidogyne incognita* with reference to its control on date- palm trees in Egypt

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Abstract

Under field conditions, distribution (vertical and horizontal) of eleven nematode genera on three date-palm cvs. Barhi, Samani and Zaghlool. Seasonal fluctuation of *Meloidogyne incognita,* and its biological control on date-palm cv. Zaghlool were investigated during one year of 2012. Results indicated that all eleven nematode genera recovered were found in high densities at a depth of 30-50 cm for both types of distributions, whereas only three genera viz. *Criconemoides* , *Tylenchus* and *Trichodorus* were detected in low densities at depth over 50 up to 100 cm in the case of vertical. However, at depth of 50-100 cm only three genera e.g. *Aphelenchus, Criconemoides* and *Tylenchus* were recovered in low number in the case of horizontal. Meanwhile, seasonal fluctuation of *M. incognita* population (J2) showed generally high peaks on date - palm cvs. Zaghlool, Samani and Barhi soil in August 2012, where Zaghlool cultivar ranked first in this respect. *M. incognita* (J2) population increased gradually during July, August and September on tested cultivars, then decreased in November and December 2012. Regarding *M. incognita* integrated biological control on date-palm cv. Zaghlool, dual treatments gave better results than single one for both percent reduction of reproduction factor and percentage increase of crop yield (78.1%), especially, *Paecilomyces lilacinus*+ *Datura stramonium* treatment.

Results also indicated amount increase in concentration of total sugar, total phenol, reducing sugar and free phenols in date-palm cv. Zaghlool of dual treatment than the single one, where *P. lilacinus* plus *D. stramonium* ranked first, however, the opposite trend was recorded in the case of total amino acids.

Key words*:* Bioagents, date-palm, *Meloidogyne incognita*, seasonal fluctuation, reducing sugars, amino acids, total phenol.

Introduction

Date palm, *Phoenix dactylifera* L. has been successfully grown on a commerce scale. Date palm is attacked by pests and diseases wherever it grows. Plant parasitic nematodes, such as *Meloidogyne* and *Pratylenchus* cause severe damage to date-palm **(Eissa, *et al*., 2009).**

Interest in alternative nematode control practices have increased due to the environmental and healthy risks associated with nematicides use. The impact of drenching application with aqueous plant extracts on such plant parasitic nematodes have been reported by several authors **(Oduor-Owino, 1993 and Khalil, 2002).**

Trichoderma harzianum acts through different mechanisms including mycoparasitism, also through production of antibiotic substances **(Hayes, 1992).** *T. harzianum* also acts through production of destructive enzymes i.e., chitenase **(Bolar *et al*., 2000; Sharon *et al*., 2001; Faruk *et al*., 2002; Shawky and Abd El-Moneim, 2005; Sahebani & Hadavi, 2008 and Shawky *et al*., 2010).** *Trichoderma* spp. can produce various toxin metabolites and different enzymes that improve photolytic activity of the antagonist and control of nematodes. In addition *T. harzianum* has ability to conolization **(Devi *et al*., 2000; Sharon *et al*., 2001; Faruk *et al*., 2002 and Siddiqui and Shaukat, 2004)**.

On the other hand, the endoparasitic nematophagous fungus, *Hirsutella rhossiliensis* has been reported to suppress different species of phytoparasitic nematodes whether in soil or root **(Jaffee & Muldoon, 1995; Velvis. & Kamp, 1995; Mostafa *et al.,* 1998 and Khalil, 2000).** *H. rhossiliensis* parasitizes and kills nematodes in vitro, suppresses nematode numbers in greenhouse tests. Spores of *H. rhossiliensis* adheres to vermiform nematodes which are directly penetrated and subsequently killed by the fungus. Therefore, the purpose of this investi0gation is to study the distribution of nematode genera associated with three date – palm cvs. Barhi, Samani and Zaghlool, seasonal fluctuation of *Meloidogyne incognita* on the same cultivars for one year with reference to its biocontrol on date – palm c.v. Zaghlool under field conditions.

Materials and Methods

1. Distribution of nematode genera associated with three date – palm cultivars:

In order to study the distribution of nematode genera (horizontal & vertical) infested orchard of date – palm cvs. Barhi, Samani and Zaghlool (25- year old) located at Al-Busailiy village at Rasheed district, Behera Governorate was chosen. For horizontal distribution, soil samples were collected at a depth of 50 cm and at the distances of 0-30, 31-50 and 51-100 from tree trunks. For vertical distribution, soil samples were collected at 5 different depths of 0-30, 30-50 and 50-100 cm at 50 cm distance from tree trunks. Five samples were collected at each distance or depth. Nematode genera were extracted from soil by sieving and modified Baremann-pan-technique **(Goodey, 1957),** while root samples by carefully washed with tap water and cut into small pieces for nematode extraction by incubate in Baremann-pan **(Goodey, 1957).** Also, % nematode population potential was estimated according to formula **Oostenbrink, (1966):**

$$\% \text{ Nematode population potential} = \frac{\text{Population density of a given distance}}{\text{Population density at all distances}}$$

2. Seasonal fluctuation of *Meloidogyne incognita* on three date – palm cultivars:

In order to study the seasonal fluctuation of *M. incognita* on three date – palm cvs. Barhi, Samani and Zaghlool, an orchard (25- year old) cultivated with these date – palm cultivars in a sandy loam soil of Al-Busailiy village at Rasheed district, Behera Governorate was selected. The trees of each date – palm cultivar were also selected at random and labeled as permanent sampling sites. From each palm tree, 3 composite soil samples of 250 g each were collected at monthly intervals for one year (2012) by using manual auger. Samples positions were 50 cm around the tree trunks. In all cases, the soil collected was in close contact with the tree roots. Soil samples were processed for nematode extraction by sieves **(Goodey, 1957).**

3. Impact of three nematophagous fungi alone or combined with aqueous leaf extract of thorn apple on *Meloidogyne incognita* infecting date – palm cv. Zaghlool field conditions:

Preparation of fungal inocula:

An isolate of *Trichoderma harzianum* was obtained from Central Laboratory of Organic Agriculture, Agricultural Research Center, Giza, Egypt. The concentrations of *T. harzianum* 5×10^6 cfu.

An isolate of the nematophagous fungus, *Paecilomyces lilacinus* was obtained from Plant Pathology Department, Faculty of Agriculture, Cairo University.

The endoparasitic fungus was cultured on PDA (Potato Dextrose Agar) and then added aseptically as mycelial mat to a flask containing 50g of autoclaved wheat grains and incubated at 25 + 1°C for one week. The concentration of the fungus was determined to 1×10^6 cfu/g of colonized wheat. Three grams of wheat grains infected with or without the fungus were used.

An isolate of the nematophagous fungus, *Hirsutella rhossiliensis* was obtained from Nematology Department of U.C. Davis, Ca., U.S.A. by Dr. Fatma A. Mostafa in 1990, then recultured at the Nematological Research Unit of the Agricultural Zoology Department, Fac. of Agric., Mansoura University, and designated as strain IMI 265748.

The endoparasitic fungus was cultured on PDA (Potato Dextrose Agar) and then added aseptically as mycelial mat to a flask containing 50 g of autoclaved wheat grains and incubated at 25 ± 1 °C for one week. The concentration of the fungus was determined to be 3.6×10^6 cfu /gm of colonized wheat. One gram of wheat grains infected with or without the fungus was used.

Preparation of aqueous leaf extracts:

Fresh leaves of weed plant were collected from ornamental Research station, Faculty of Agriculture, Mansoura University and transferred to Nematology Department, Plant Pathology Research Institute, Agriculture Research Center, Giza for extraction. The tested plant was thorn apple, *Datura stramonium*.

Standard leaf extract was prepared by crushing and dissolving 20 g of leaves in 100 ml distilled water separately using mortar and pestle. The result solution was then centrifuged at 5000 rpm for five minutes. The supernatant was filtered through a layer of muslin cloth, and dilution of 5% was prepared from standard. Plants were drenched separately with 10 mls of tested previous plant, around plants root.

In order to study the effect of three nematophagous fungi i.e. *T. harzianum, P.lilacinus* and *H. rhossiliensis* singly or integrated with *Datura stramonium* on controlling *M. incognita* infecting date – palm cv. Zaghlool and crop yield in return under field conditions, forty trees were randomy chosen within the same date – palm orchard.The concentrations of each fungus tested, aqueous leaf extract and the systemic nematicide, vydate was separately added at the beginning the season of 2012.Each concentration of each tested materials for each tree was $5x10^6$ cfu, 1×10^6 cfu/g and $3.6x10^6$ cfu /g of colonized wheat for *T. harzianum, P.lilacinus* and *H. rhossiliensis*, while 10 mls for *D. stramonium* and 3g for vydate, and left four trees without any treatment, respectively.

Treatments were as follows:

1. N+ thorn apple (*D. stramonium*) 2. N+ fungus, *T. harzianum*

3. N+ fungus, *P. lilacinus* 4. N+ fungus*, H. rhossiliensis*

5. N+thorn apple (*D. stramonium*) + fungus, *T. harzianum*

6. N+thorn apple (*D. stramonium*) + fungus, *P. lilacinus*

7. N+thorn apple (D. stramonium) + fungus, H. rhossiliensis

8. N+ wheat. 9. N+Vydate, and 10. Nematode (N) alone (control).

Nematode determinations:

Nematode populations in soil (number of juveniles/250g. soil) were determined after one, two, three and four months from adding the tested materials according to **Goodey (1957).** Roots were stained by acid fuchsin in acetic acid according to **(Byrd *et. al*., 1983)** and examined for number of developmental stages and egg laying females/1g. root. Eggs /egg-mass of *M. incognita* were extracted by using sodium hypochlorite (NaOCl) method as described by (**Husssey and Baker, 1973).**

Chemical analyses:

Chemical analyses of sugars, phenolic compounds and amino acids were analyzed in Food Technology Research Institute. Total and reducing sugars were described by **(Thomas, 1924).** Phenolic compounds were determined by **(Snell and Snell, 1953).** The total free amino acids were determined by **(Rason, 1959).**

Statistical analysis procedure:

All obtained data were subjected to analysis of variance (ANOVA) **(Gomez and Gomez, 1984)** and means were compared by using L.S.D. at 5 % level of significance.

Results and Discussion

1. Distribution of nematode genera on three cultivars of date palm at different depths:

In vertical depth the highest density of *M. incognita* and their population potential % were also found at a depth of 50 cm from the soil surface, while the lowest density of *Tylenchus* and *Trichodorus Criconemoides*, *Helicotylenchus*, *Hoplolaimus*, *Longidorus, Pratylenchus*, *Rotylenchulus*, *Xiphinema* and *Tylenchorhynchus* found in the intermediates rank of population dynamics of nematodes in Egypt (Table 1).

In Horizontal depth the highest density of *M. incognita* and their population potential % were also found at a distance of 50 cm from the tree, while the lowest density of *Tylenchus* and *Aphelenchus. Criconemoides*, *Helicotylenchus*, *Hoplolaimus*, *Longidorus, Pratylenchus*, *Rotylenchulus*, and *Tylenchorhynchus* found in the intermediates rank of population dynamics of nematodes in Egypt (Table 2).

2. Seasonal fluctuation of root knot nematode, *M. incognita*:

M. incognita was found in the rhizosphere of date-palm trees cvs. Barhi, Samani and Zaghlool (Fig.1). Results showed that Zaghlool cultivar was more susceptible than Barhi and Samani cultivar. Population of *M. incognita* second stage juveniles (J2) in the soil showed a high peak in August. The lowest population density was found in Jaunary.

Table (1): Vertical distribution of populations densities of plant nematode genera associated with three date-palm cvs. Barhi ,Samani and Zaghlool at three depths from soil surface around tree trunk under field conditions.

Nematodes genera	Varieties								
	Barhi			Samani			Zaghlool		
	Vertical depth (cm.)								
	30 cm.	50 cm.	100 cm.	30 cm.	50 cm.	100 cm.	30 cm.	50 cm.	100 cm.
Criconemoides	36	64	27	58	92	54	105	123	89
Helicotylenchus	61	82	97	84	105	93	121	182	103
Hoplolaimus	84	105	56	107	144	123	162	224	142
Longidorus	0	43	68	0	66	84	0	82	121
Meloidogyne	177	216	157	327	413	238	627	856	536
Pratylenchus	97	136	89	164	210	171	230	342	202
Rotylenchulus	116	155	102	221	362	191	383	523	321
Trichodorus	8	27	33	27	45	62	36	55	76
Tylenchrohenchus	86	117	74	136	176	118	206	271	178
Tylenchus	26	56	17	46	77	37	78	96	58
Xiphinema	0	33	41	0	62	86	0	76	106
L.S.D (5%)	9.3	6.1	5.8	15.2	12.3	7.9	17.8	13.1	11.5

Values were the average of ten replicates of nematodes per 250 gm soil.

$$\% \text{ population potential} = \frac{\text{Population density of a given distance}}{\text{Population density at all distances}}$$

Rhizosphere of date-palm cv. Barhi , Samani and Zaghlool trees

Table (2): Horizontal distribution of populations densities of plant nematode genera associated with three date-palm cvs. Barhi ,Samani and Zaghlool at three distance from the trunk under field conditions.

Nematodes genera	Varieties								
	Barhi			Samani			Zaghlool		
	Horizontal distances (cm.)								
	30 cm.	50 cm.	100 cm.	30 cm.	50 cm.	100 cm.	30 cm.	50 cm.	100 cm.
Aphelenchus	22	43	24	41	64	33	62	85	52
Criconemoides	46	74	33	68	103	65	115	133	88
Helicotylenchus	72	94	103	95	116	112	133	195	117
Hoplolaimus	96	117	65	119	154	138	177	235	156
Meloidogyne	183	233	167	333	442	249	634	866	548
Pratylenchus	112	147	95	178	223	188	245	361	224
Rotylenchulus	137	167	115	233	337	201	393	534	339
Tylenchrohynchus	98	123	87	146	186	127	216	283	188
Tylenchus	33	68	47	52	87	43	89	112	69
L.S.D (5%)	8.9	5.7	7.1	12.6	12.8	9.7	15.2	18.4	17.6

Values were the average of ten replicates of nematodes per 250 gm soil.

$$\% \text{ population potential} = \frac{\text{Population density of a given distance}}{\text{Population density at all distances}}$$

Rhizosphere of date-palm cv. Barhi, Samani and Zaghlool trees

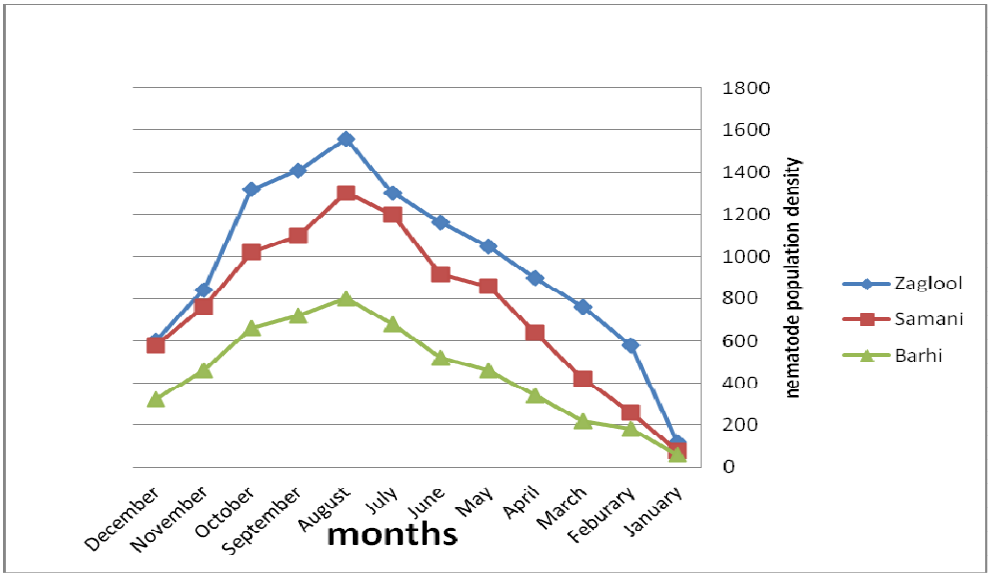

Fig. (1): Effect of seasonal fluctuation of *M. incognita* on three date palm cultivars under field conditions.

3. Effect of three nematophagous fungi, *T. harzianum, P. lilacinus* and *H. rhossiliensis* alone or combined with one plant leaf extract on crop yield of date-palm cv. Zaghlool infected with *M. incognita* under field conditions:

Data in Table (3) show that high reproduction of *M. incognita* was significantly suppressed in all tested treatments. Rate of nematode build-up ranged from 0.63 for *P. lilacinus* plus *D. stramonium* to 0.75 for wheat grains when compared with nematode alone (1.20) after one month Similar trend was obtained with the same treatments after two, three and four months.

However, significan variation in nematode population density were noticed between most treatments when applied either singly or combined with plant leaf extract. However, significant reduction in the final nematode population achieved by all tested treatments in relation to nematode or wheat grains alone (Table 3).

Results in Figure (2) reveal that different response in % crop yield of date palm cv. Zaghlool over control under field conditions.The treatments of *P. lilacinus* plus *D. stramonium* showed the highest % crop yield of date palm cv. Zaghlool over control after the treatment of vydate while treatment of *D. stramonium* alone showed the lowest increase over the control (Fig.2).

Table (3): Suppression of *M. incognita* reproduction infecting date palm cv. Zaghlool as affected by three nematophagous fungi alone or combined with one leaf extract under field conditions.

Treatments	Initial	*After one month		*After two months		*After three months		*After four months	
	Total population in soil/250g	Total population in soil/250g + in root/g	PF/PI	Total population in soil/250g + in root/g	PF/PI	Total population in soil/250g + in root/g	PF/PI	Total population in soil/250g+ in root/g	PF/PI
T. harzianum	1740	1240	0.71	1300	0.74	1160	0.66	1190	0.68
P. lilacinus	1700	1200	0.70	1040	0.61	860	0.50	1020	0.60
H. rhossiliensis	1840	1360	0.73	1200	0.65	1090	0.59	1180	0.64
D. stramonium	1780	1260	0.71	1380	0.77	1200	0.67	1300	0.75
T. harzianum + *D. stramonium*	1820	1250	0.68	1050	0.57	890	0.49	1060	0.58
P. lilacinus + *D. stramonium*	1900	1200	0.63	960	0.51	880	0.46	920	0.48
H. rhossiliensis +*D. stramonium*	1800	1160	0.64	980	0.55	920	0.51	980	0.54
Wheat	1840	1380	0.75	1470	0.79	1300	0.70	1400	0.76
Vydate	1640	840	0.51	720	0.43	680	0.41	520	0.31
Control (nematode)	1940	2340	1.20	3600	1.85	4200	2.10	4800	2.47
L.S.D (5%)	**17.3**	**15, 1**	**0.10**	**9.7**	**0.32**	**9.1**	**0.09**	**9.8**	**0.12**

* Each value is the mean of four replicates per treatment.

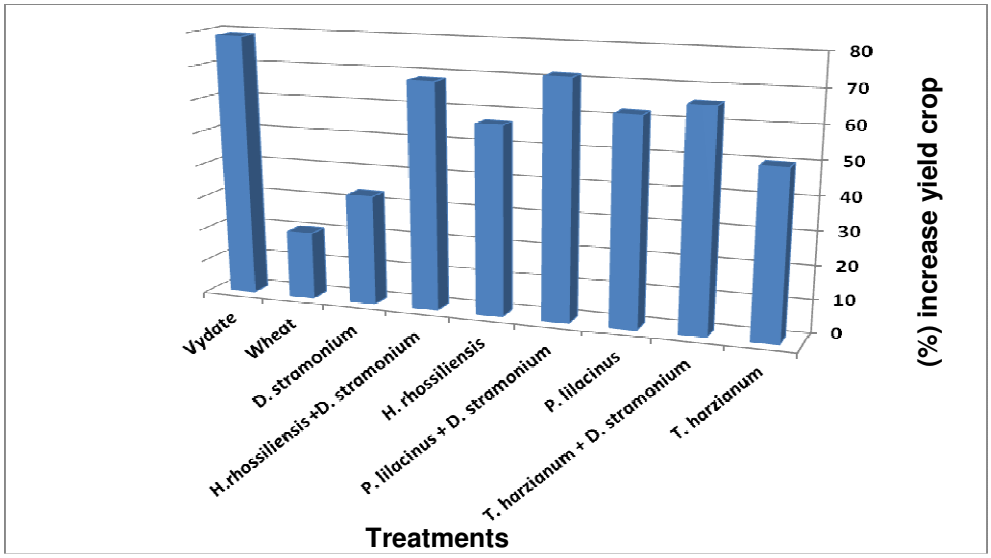

Fig. (2): Effect of *T. harzianum, P. lilacinus* and *H. rhossiliensis* alone or combined with *D. stramonium* on date palm cv. Zaghlool infected with *M. incognita* under field conditions.

Data in Table (4) showed a positive correlation between the amounts of total or reducing sugars and total or free phenols of the used treatments. Data also showed a negative correlation in total amino acids of the used treatments when compared with nematode alone. Amount of total sugars ranged from 1.789 to 3.145 mg/gm while amounts of reducing sugars were from 0.319 to 0.522 mg/gm. The highest amount of total phenols, (18.529 mg/gm) was found in trees treated with *P. lilacinus + D. stramonium*, while the lowest, (11.045 mg/gm) was for trees treated with wheat grains only. Amounts of the total amino acids ranged from 0.009 to 0.127 mg/gm from trees treated with *P. lilacinus + D. stramonium* or wheat grains, respectively.

Apparently, distribution of the eleven nematode genera recovered in the rhizosphere of the tested three date –palm tree roots showed high densities at a depth of 30-50 cm where date –palm trees root system percent either vertically or horizontally except for three nematode genera that were recorded at depth over 50cm up to 100cm.These results are not surprising since root distributions in both cases were in the same zones **(Eissa, *et al.*, 2009).**

In general, seasonal fluctuation of *M. incognita* in soil of the three date –palm cvs. Barhi, Samani and Zaghlool increased gradually during July, August, and September and then diminished in November and December 2012, where Zaghlool cultivar has the highest nematode population of *M. incognita* in August, followed by Samani and then Barhi. These findings are in agreement with these of **(Eissa, *et al.*, 2009)** in this respect.

Table (4): Efficacy of three nematophagous fungi and one plant leaf extract alone or combined with *D. stramonium* on some chemical components of date palm cv. Zaghlool) infecting with *M. incognita* under field conditions.

Treatments	Concentration (mg/g)				
	Total sugars	Reducing sugars	Total phenols	Free phenols	Total amino acids
T. harzianum	2.200	0.410	16.380	10.400	0.090
P. lilacinus	2.226	0.412	15.458	11.346	0.110
H. rhossiliensis	2.015	0.346	13.125	8.458	0.219
D. stramonium	2.135	0.369	16.254	9.456	0.085
T. harzianum + *D. stramonium*	2.621	0.459	18.325	12.789	0.068
P. lilacinus + *D. stramonium*	3.145	0.522	18.529	14.478	0.009
H.rhossiliensis +*D. stramonium*	2.568	0.419	16.547	12.312	0.098
Wheat	1.789	0.319	11.046	8.682	0.127
Vydate	4.155	0.654	18.789	15.714	0.012
Control(nematode)	1.756	0.255	9.358	4.254	0.458
L.S.D (5%)	0.020	0.022	0.413	0.251	0.003

Most treatments generally showed significant reduction in nematode population infected with *M. incognita* over that of the nematode alone. These results may be due to that of *Trichoderma* spp. has ability to conolization **(Devi *et al*., 2000; Sharon *et al*., 2001; Faruk *et al*., 2002 and Siddiqui and Shaukat, 2004 and Jegathambigai *et al.* 2008).** *T. harzianum* also acts through production of the destructive enzyme chitenase **(Paderes *et al*., 1992 and Bolar *et al*., 2000).** *Trichoderma* spp. can produce various toxin metabolites and different enzymes that improve photolytic activity of the antagonist and control of nematodes. In addition *T. harzianum* has the ability to conolization **(Devi *et al*., 2000; Sharon *et al*., 2001 and Faruk *et al*., 2002).**

These findings support the evaluation of reported by **Siddiqui and Mahmoud (1994)** and **Rao *et al.* (1997)** in respect to *Meloidogyne* spp. These results are in accordance with the work of **Nagesh *et al.* (1997)** who reported that combination of *P. lilacinus* with neem leaf extract resulted in significantly higher fresh plant weight and flower yield. Root gall index of *M. incognita* was least under *P. lilacinus* plus neem leaf extract combination followed by *P. lilacinus* plus castor leaf extract treatment. Although the percent egg and egg mass parasitization by *P. lilacinus* was higher when integrated with the leaf extracts, neem leaf extract improved the parasization by *P. lilacinus* more than that with castor leaf extract. Furthermore, results of **Saikia *et al.* (1999)** in respect to *P. lilacinus* with or without neem (*Azadirachta indica*) or mustard oil cake extract on *M. incognita* infecting tomato support also the present findings.

The active ingredients of *D. stramonium* are atropine, hyoscyamine and scopolamine which are classified as deliriants, or anticholinergics **(Oduor-Owino, 1993).**

The increase in sugar by variation of the used treatments could be due to enhancing the metabolism and accumulation of metabolites that contain sugars. Another possibility is that the variation of the used treatments gave higher nematode suppression and consequently less consumption of nutrient including sugars. The increased of free phenols by variation of the applied compounds could be attributed to increase of the plant defense against the invading nematode. The increase in total amino acids by variation of used compounds could be attributed to lower infection and consequently lower replenish of proteins from the plant cell that cell is adjacent to infected ones **(Abdel-Momen *et al.,* 2005 and Shawky *et al*., 2010).**

In conclusion, Zaghlool cultivar was the more susceptible cultivar than the others to the root knot nematode. It also can be said that drenching application with aqueous plant extracts; *D. stramonium* that applied singly or integrated with *T. harzianum* or *P. lilacinus* or *H. rhossiliensis* retained their nematicidal effects in soil. More research is needed to be done before using this trend for integrated control of such nematodes.

Referances

Abdel-Momen, S. M.; Hanaa S. Zawam and A.E.M. Khalil (2005). Effect of certain mineral salts, organic acids, amino acids and growth regulators on reproduction of *Meloidogyne javanica* infecting sunflower. J. Agric. Sci. Mansoura Univ., 30(5): 2951-2961.

Byrd, D.W., T. Kirkpatrick and K.R. Barker (1983). An improved technique for cleaning and staining plant tissues for detection of nematodes. J. Nematol., 15 (1): 142-143.

Bolar, J.P., J.L. Norelli, K.W. Wong, C.K. Hayes, Q.E. Harman and H.S. Aldwinckle. (2000). Expression of endochitinase from *Trichodermn harziianum* in transgenic apple increase resistance to apple scab and reduces vigour. Phytopathology, 90: 72-77.

Devi, L.S., S. Richa and R. Sharma. (2000). Effect of *Trichoderma* spp. against root-knot nematode *Meloidogyne incognita* on tomato. Indian Journal of Nematology, 32(2): 227-228.

Eissa, M. F. M.; El-Sherif, M. A.; Abd-El-Gawad, M. M.; Ismail, A. E. and Wafaa, M. A. E. (2009). Seasonal population dynamics, horizontal and vertical distributions of phytonematodes in date-palm cv. Samani in Egypt. J. Plant Pathology Bulletin, vol. 18 No. 1 pp. 45- 50.

Franklin, M. T. and J. B. Goodey (1957). A cotton-blue lactophenol technique for mounting plant parasitic nematodes. J. Helminthological abstracts, 23:175-178.

Faruk, M.I., M.L. Rahman and M.A. Bari. (2002). Mangement of root-knot nematodes of tomato using *Trichoderma harzianum* and organic soil amendment. Bangladesh Journal of Plant Pathology, 18: 33-37.

Gomez, K.W. and A.A. Gomez (1984). Statistical procedures for agricultural research 2nd Ed. John Wiley and Sons inc, New York 680p.

Hayes, C. K. (1992). Improvement of *Trichoderma* and *Gliocladium* by genetic manipulation. P 227- 286. In: Biological control of plant disease progress and challenges for the future (Eds: T. Jamos and R.J. Cook) Plenum Press, New York and London, Published in co operation with NATO affairs division, 462p.

Hussey, R.S. and K.R. Barker (1973). A comparison on methods of collecting inocula of *Meloidogyne* spp. including a new technique. Plant Dis. Reptr., 57:1925-1928.

Jaffee, B.A. and A.E. Muldoon (1995). Numerical responses of the nematophgous fungi *Hirsutella rhossiliensis, Monacrosporium cionopagum* and *M. ellippsosporum.* Mycologia, 87(5): 643-650.

Jegathambigai V, Karunaratne MD, Svinningen A, Mikunthan G. (2008). Biocontrol of root-knot nematode, *Meloidogyne incognita* damaging queen palm, Livistona rotundifolia using *Trichoderma* species. Commun Agric Appl Biol Sci.;73(4):681-7.

Khalil, A.E.M. (2000). Integrated control of the reniform nematode on certain plant crops. Ph. D. Thesis, Fac. of Agriculture, Mansoura Univ., pp. 112.

Khalil, A.E.M. (2002). Evaluation of nematicidal potential in some plant extracts either singly or integrated with *Hirsutella rhossiliensis* against *Rotylenchulus reniformis* on tomato. Zagazig J.Agric.Res., 29(4):1311-1322.

Mostafa, F.A.M. (1998). Impact of three nematophagus fungi separately and in combination with poultry manure on root-knot nematodes infecting sunflower. Egyptian J. Agronematol., 2(2): 245-256.

Nagesh, M.; P.P. Reddy and M.S. Rao (1997). Integrated management of *Meloidogyne incognita* on tuberose using *Paecilomyces lilacinus* in combination with plant extracts. Nematologia Mediterranea, 25(1): 3-7.

Oduor-Owino, P. (1993). Effects of aldicarb, *Datura stamonium*, *Datura metel* and *Tagetes minuta* on the pathogenicity of root-knot nematode in Kenya. **J.** Crop Protection**, 12 (4): 315-317.**

Oostenbrink, M. (1966). Major characteristics of the relation between nematodes and plants. Meded. Landbouwhogesch, Wageningen, 66-4, 46 pp.

Padares, D.F., J. Hockenbull, D.F. Jensen and S. B. Mathus (1992). *In vivo* screening of *Trichoderma* isolates for antagonism against *Sclerotium rolfsii* using rice seedlings. Bulletin oil B / srop 15 (1): 33-35. (C.F. Rev. Pl. Pathol., 1993.Vol. 72 No. 1, 208)

Rao, M.S.; P.P. Reddy and M. Nagesh (1997). Integration of *Paecilomyces lilacinus* with neem leaf suspension for the management of root-knot nematodes on egg plant. Nematologia Mediterranea, 25 (2): 249-252.

Rason, E.L. (1959). Modern methods of plant analysis, "non volatile Mon-, Di- and Tricarboxylic acids" Cd. Peak, K and M.C. Tracely, pp. 539-582.

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

Saikia, M.K.; K.C. Puzari and U.N. Saikia (1999). An integrated approach to control the root-knot nematode, *Meloidogyne incognita* on vegetables. Annals of Biology Ludhiana, 15(1): 81-84.

Sharon, E., E.M. Bar, I. Cheti, A.E. Herrera, O. Kleifeled and Y. Splegel. (2001). Biological control of the root-knot nematode *Meloidogyne javanica* by *Trichoderma harzianum*. Phytopathology, 91: 687-693.

Siddiqui, Z.A. and I. Mahmood (1994). Culture of *Paecilomyces lilacinus* on leaf extracts and leaf residues for nematode control. Bioresource Technol-ogy, 49(2): 187-189.

Siddiqui, I. A and S. S. Shaukat (2004). *Trichoderma harzianum* enhances the production of nematicidal compounds *in vitro* and improves biocontrol of *Meloidogyne javanica* by *Pseudomonas fluorescents* in tomato. Letters in Applied Microbiology, 38: 2, 169-175.

Shawky, Samaa M. and Abd El- Moneim, Maisa L. (2005). Non chemical control of root - knot nematode*, Meloidogyne incognita* on grape vines. Minufiya J. Agric. Res., 30(6): 1685- 1708.

Shawky, Samaa M., A.E. Khalil and Manal M. Soliman (2010). Non chemical control of root-knot nematode; *Meloidogyne javanica* on peanut in Egypt. Zagazig J. Agric. Res., 37 (1): 185-206.

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

Snell, F.D. and C.T. Snell. (1953). Colorimetric Methods of Analysis. Vol. 3 Van Nostrand Company, New York, 606pp.

Thomas, W. and R. D. Dutcher (1924). Picric method for carbohydrate. J. Am. Chem. Soc. 45: 1662-1669.

Velvis, H. and P. Kamp (1995). Infection of second stage juveniles of potato cyst nematodes by the nematophagus fungus *Hirsutella rhossiliensis* in Dutch potato field. Nematologica, 41(5): 617-627.

الملخص العربى

انتشار أجناس النيماتودا والتذبذب الموسمي لنيماتودا (ميلودوجين إنكوجنيتا) مع الإشارة إلى مكافحتها على نخيل البلح في مصر

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تم دراسة الانتشار الرأسى والأفقى لإحدى عشر جنسا من الأجناس النيماتودية المتطفلة على ثلاثة أصناف من نخيل البلح (برحي، سماني، زغلول) والتذبذب الموسمي لنيماتودا تعقد الجذور (ميلودوجين انكوجنيتا) وكذلك مكافحتها بيولوجيا على صنف زغلول لمدة عام واحد (٢٠١٢) تحت الظروف الحقلية. أوضحت النتائج ظهور أحد عشر من الأجناس النيماتودية بكثافة عالية عند المسافات من ٣٠–٥٠ سم فى كلا الاتجاهين الرأسى والأفقى، بينما وجدت الأجناس النيماتودية الثلاث (الحلقية، التيلنكس، التقصف) بكثافة قليله عند المسافات من ٥٠ –١٠٠ سم عند مستوى الانتشار الرأسى. كذلك تلاحظ ظهور الثلاثة أجناس (أفيلنكس، الحلقية، التيلنكس) بكثافة قليلة عند المسافات من ٥٠–١٠٠ سم فى الاتجاه الأفقى. أوضح التذبذب الموسمى لنيماتودا تعقد الجذور أن أعلى معدل للتكاثر كان على الأصناف (برحى، سمانى، زغلول) فى شهر أغسطس ٢٠١٢ حيث احتل الصنف زغلول المركز الأول. حدثت زيادة فى تعداد النيماتودا تدريجيًا خلال أشهر (يوليو، أغسطس، سبتمبر) على الأصناف المختبره ثم تناقصت فى شهري نوفمبر وديسمبر ٢٠١٢.

فيما يتعلق بالمكافحة البيولوجية المتكاملة لنيماتودا تعقد الجذور (ميلودوجين انكوجنيتا) على صنف زغلول لوحظ أن المعاملات المشتركة أعطت أفضل النتائج عن المعاملات المنفردة في خفض تعداد النيماتودا وزيادة فى المحصول وصلت إلى ٧٨,١% للمعاملة المشتركة (باسيلوميسيس ليلاسينس والمستخلص المائى للداتورة).

أوضحت النتائج زيادة السكريات الكلية والفينولات الكلية والسكريات المختزلة والفينولات الحرة فى أشجار النخيل (صنف زغلول) للمعاملة المركبة عن المعاملة الفردية حيث تلاحظ أن المعاملة بفطر باسيلوميسيس ليلاسينس والمستخلص المائي للداتورة معًا احتلت المركز الأول ولكن على العكس من ذلك حدث عند تقدير الأحماض الأمينية الكلية.