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CONTROL OF ROOT KNOT NEMATODE, *Meloidogyne incognita* BY CERTAIN BOTANICAL SOIL AMENDMENTS ON EGGPLANT

Amin, A.W.; A. Y. El-Gendi ; A. I. Afia and Manar M. Sanad
Agricultural Zoology and Nematology Department, Fac. of Agric., Cairo Univ., Egypt.

ABSTRACT

Nine medicinal and aromatic plants viz., santonia, *Artemisia cinae*; neem, *Azadirachta indica*; calotropis, *Calotropis procera*; stramonium, *Datura stramonium* ;eucalyptus, *Eucalyptus globulus*; melia, *Melia azedarach*; hamd, *Mesembryanthemum forsskalie*; geranium, *Pelargonium graveolens;* schinus, *Schinus terebinthifolius* were evaluated as botanical organic soil amendments against root-knot nematode, *Meloidogyne incognita* activity in natural and sterilized soils on eggplant under greenhouse conditions. All of botanical organic amendments satisfactory reduced ($p \leq 0.01$) the total number of galls, females and egg-laying females as well as the total number of nematodes in eggplant roots compared with non treated check. Pots receiving hamd, schinus and stramonium performed the best minimizing females number with percentage of redaction 97.9, 95.5 and 94.6% respectively in sterilized soil compaired with Mocap (99.1%). While, powder of geranium, neem, calotopis and eucalyptus had comparatively lower effects. In non sterilized soil the best reduction in galls number induced by nematode infection performed with santonia (9), hamd (13), and neem (30.5) compared with Mocap (7) and untreated check (700.0 galls).therefore, the best minimizing female number was recorded when pots received these materials. The best of the percentage of female reduction were 98.8, 98.6 and 97.8 for hamd, santonia and schinus respectively compared with Mocap (99.7). While, powder of calotopis, eucalyptus, neem and geranium had comparatively lower effects. In sterilized soil, nematodes multiplied greater on eggplant than in natural one. The plant growth of eggplant as measured by length of shoot and root and weight of fresh and dry shoot and number of leaves showed an improvement over healthy plant in some treatment. As results, there was a positive reaction, in most cases, between botanical soil amendments treatment and plant growth.

Keywords: eggplant, medicinal and aromatic plants, *Meloidogyne incognita*, natural soil, sterilized soil, soil amendments.

INTRODUCTION

Root-knot nematode, *Meloidogyne* spp. are major pests of economic plants. Throughout the world, root-knot nematode causes an average annual yield losses. Several control measures are employed to suppress this pest in infested areas. Chemicals include fumigants and non-fumigants are the most common methods used to suppress root-knot nematodes (Walker and Wachtel, 1988; Lamberti *et al.*, 2000). Since, the ferequently use of nematicides were expensive and highly toxic and of harmful effects on health, efforts are needed to develop alternative nematode management strategies,

effective, safe and low coast methods of control. Many plants are known to have nematicidal effect against plant parasitic nematodes when they are used as botanical soil amendments or plant extracts. There are many reports for using of plant parts as organic amendments for the control of nematodes (Amin and Youssif, 1997 and 1998; Rao *et al.*,1996; Youssif and Amin 1997 and Ismail 1998). Other investigators are used the plant as extracts or oils (Khurma *et al.*, 1997; Nagesh *et al.*, 1997 and Abbott *et al.*, 1998). In the present study, the evaluation of nine medicinal and aromatic plants compared with Mocap (10 G) on reproduction and development of *Meloidogyne incognita* (Kofoid&White)Chitwood were studied in non sterelized and sterelized soils.

MATERIALS AND METHODS

One-month eggplant seedlings, *Solanum melongena* cv. Long Purble were implanted in 15 cm diam., clay pots. Pots were arranged in the greenhouse into two groups according to the soil type condition; sterilized soil (autoclaved at 121° atmospheric pressure for 30 min.), and 2nd group was natural soil (free of any parasitic nematodes) and non-sterilized.

Nine aromatic and medicinal plants were air-dried and ground. These plant species were Santonia, *Artemisia cinae*, neem, *Azadirachta indica*, caotropis, *Calotropis procera*, stramonium, *Datura stramonium*, eucalyptus, *Eucalyptus globulus*, melia, *Melia azedarach*, hamd, *Mesembryanthemum forsskalie*, geranium, *Pelargonium graveolens*, schinus, *Schinus terebinthifolius*. Five gram per pot ground plant were throughly mixed with the soil seven days before nematode inoculation. Each pot was inoculated with 1000 infective stages of *Meloidogyne incognita*. Un-inoculated - untreated four pots were served as a healthy plant and other four pots inoculated-untreated served as a nematode control. There were four replicates of each treatment. The experiment was repeated twice for two soil conditions. The pots were arranged in a completely randomized design in a greenhouse at 30±5C°. The pots were watered daily. After six weeks of nematodes inoculation, eggplant plants were carefully uprooted and nematodes in roots were counted by staining the roots in a lactophenol-acid Fuchsin solution for 2-3 minutes and cleared in pure lacto-phenol (Franklin, 1949). The number of galls, females and egg-masses were examined and calculated under stereomicroscope. The total number of non-parasitic and parasitic nematodes in soil were extracted by sieving method followed by Baermann tray technique and counted. Length and weight of shoots and roots, shoot dry weight and number of leaves were recorded. Data was statisticaly analyzed using New Least Significant Difference (New LSD).

RESULTS

Data represented in Tables 1 and 2 showed that all tested botanical soil amendements , hamd, stramonium (leaves and seeds), schinus, neem, santonia, calotropis, melia, eucalyptus and geranium had highly segnificant reduction in root-knot nematode population on eggplant in both soil conditions.

Table 1: Effect of certain botanical soil amendments in controlling *Meloidogyne incognita* on eggplant implanted in sterilized soil.

Treatments	No. Nematodes in soil	No.Galls	No. Nematodes in root					Total No. Nematodes	RF*	No free living nematodes
			immature stages	Females	Egg-masses	Eggs/egg-mass	Total eggs			
Artemisia cinae	28.8	93.5	84.0	83.5	34.5	65.5	2259.8	196.3	0.20	562.5
Azadirachta indica	30.0	144.5	86.5	133.5	94.5	71.0	6709.5	250.0	0.25	417.5
Calotropis procera	21.3	144.5	89.0	119.5	83.5	43.3	3615.6	229.8	0.23	647.5
Datura stramonium	0.0	43.3	31.0	35.8	22.5	26.0	585.0	66.8	0.07	241.3
Datura stramonium♣	146.3	201.0	146.0	198.0	145.0	25.8	3741.0	490.3	0.49	551.3
Eucalyptus globulus	86.3	167.0	148.0	114.0	79.0	43.3	3420.7	348.3	0.35	953.8
Melia azedarach	168.8	126.0	103.0	75.5	49.5	41.0	2029.5	347.3	0.35	1895.0
Mesembryanthemum forsskalei	35.0	27.0	30.0	14.0	11.0	13.0	143.0	79.0	0.08	1360.0
Pelargonium graveolens	147.5	172.0	130.0	142.0	57.0	50.5	2878.5	419.0	0.42	902.5
Schinus terebinthifolius	115.0	53.0	51.0	30.0	24.0	45.8	1099.2	196.0	0.20	197.5
Mocap (10G)	32.5	14.0	9.0	6.0	3.0	10.5	31.5	47.5	0.05	268.8
Inoculated plants (Check)	6089.0	829.0	289.0	668.0	357.0	196.8	70257.6	7046.0	7.05	2041.3
New LSD ($p \leq 0.05$)	**342.8**	**64.5**	**50.1**	**55.4**	**66.1**	**22.2**				
New LSD ($p \leq 0.01$)	**460.5**	**460.5**	**67.4**	**74.5**	**88.8**	**29.8**				

* RF(Rate of build up) = $\frac{\text{Pf (Nematode final population)}}{\text{Pi (Nematode initial population)}}$

♣ = seeds

Table 2: Effect of certain botanical soil amendments in controlling *Meloidogyne incognita* on eggplant implanted in natural soil (non sterilized).

Treatments	No. Nematodes in soil	No.Galls	No. Nematodes in root					Total No. Nematodes	RF**	No free living nematodes
			Immature stages	Females	Egg-masses	Eggs/egg-mass	Total eggs			
Artemisia cinae	20.0	9.0	5.5	8.0	2.5	19.3	48.3	33.5	0.03	353.8
Azadirachta indica	8.8	30.5	19.0	19.5	10.5	33.5	351.8	47.3	0.05	293.8
Calotropis procera	30.0	100.5	63.5	100.5	60.8	46.0	2796.8	194.5	0.19	283.8
Datura stramonium	13.8	41.0	31.5	17.0	11.0	16.3	179.3	62.3	0.06	322.5
Datura stramonium♣	25.0	128.5	107.5	93.5	60.0	27.5	1650.0	226.0	0.23	231.3
Eucalyptus globulus	58.8	87.5	80.0	72.5	34.0	26.0	884.0	211.3	0.21	318.8
Melia azedarach	43.8	82.0	53.5	67.0	33.0	25.8	851.4	164.3	0.16	526.3
Mesembryanthemum forsskalei	0.0	13.0	16.0	7.0	1.0	4.5	4.5	23.0	0.02	757.5
Pelargonium graveolens	93.8	92.0	95.0	63.5	8.5	27.0	229.5	252.3	0.25	176.3
Schinus terebinthifolius	26.3	47.0	52.0	12.5	7.0	13.8	96.6	90.8	0.09	666.3
Mocap (10G)	0.0	7.0	6.0	2.0	1.0	3.0	3.0	8.0	0.01	200.0
Inoculated plants (Check)	4762.0	700.0	254.0	577.0	341.0	119.5	40749.5	5593.0	5.59	3827.5
New LSD ($p \leq 0.05$)	338.9	52.7	41.6	45.1	16.0	8.5				
New LSD ($p \leq 0.01$)	455.2	70.8	55.8	60.6	21.5	11.5				

♣ Seeds

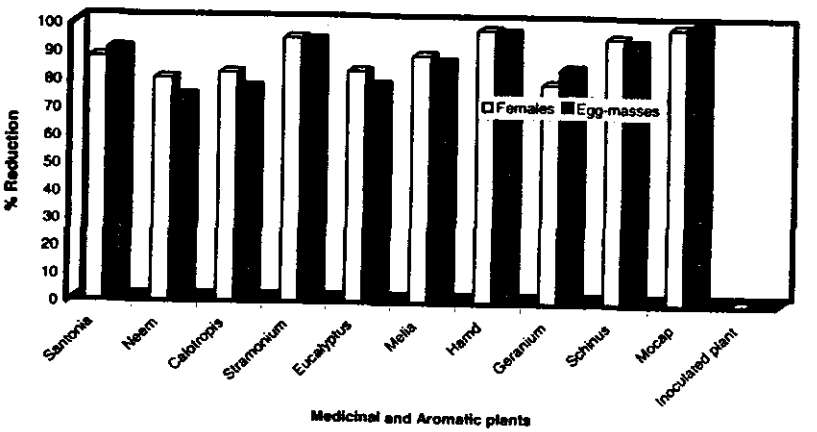
** RF (Rate of build up) = $\frac{\text{Pf (Nematode final population)}}{\text{Pi (Nematode initial population)}}$

On the other hand, in sterilized soil, pots received hamd, stramonium or schinus were highly significant reduced the number of galls, females, egg-masses, total number of nematodes and rate of build up followed by neem and santonia. Population of nematodes multiplied greater in sterilized soil than non sterilized one (Fig.3).

Regarding data in Table 1 , obviously that all treatments caused highly significant reduction in galls number induced by nematode infection. Best reduction was performed with hamd (27.0) , stramonium leaves (43.3) and schinus (53.0) compard with Mocap (14.0) and untreated check (829.0). Therefore , pots receiving hamd , stramonium or schinus performed the best minimizing females number with percentage of redaction 97.9 , 94.6 and 95.5 respectively in sterilized soil. Similar trend was noticed with egg-masses, total nematodes and rate of build up (Fig. 1). While, powder of geranium, neem, calotopis and eucalyptus had comparatively lower effects.

In natural soil (non-sterilized), hamd, santonia, and neem caused highly significant reduction in gall, females, egg-masses numbers and rate of build up of root-knot nematode population on eggplant. Best reduction in galls number induced by nematode infection performed with santonia (9), hamd (13), and neem (30.5) compared with Mocap (7) and untreated check (700 galls). Therefor, the best minimizing female number was recorded when pots received these materials with percentage of female reduction (98.8), (98.6) and (96.6) respectively compared with Mocap (99.7) Fig. 2.

Fig 1: Effect of botanical soil amendments on the rerduction of females and egg-masses of *Meloidogyne incognita* on eggplant implanted in sterilized soil.



Similar trend was noticed with egg-masses, total number of nematodes and rate of build up. While, powder of calotopis, eucalyptus, neem and geranium had comparatively lower effects. In both soil condition, it's clearly noticed that, hamd had not effected in decreasing free living nematodes in soil while, other materials did. However, pots received santonia showed better results over healthy plants in fresh weight, number of leaves and root length folowed by neem, stramonium and calotopis in both soil conditions (Tables 3 and 4).

Fig. 2: Effect of cerain botanical soil amendments on reduction of femal and egg-masses of *Meloidogyne incognita* on eggplant implanted in non-sterilized (natural soil).

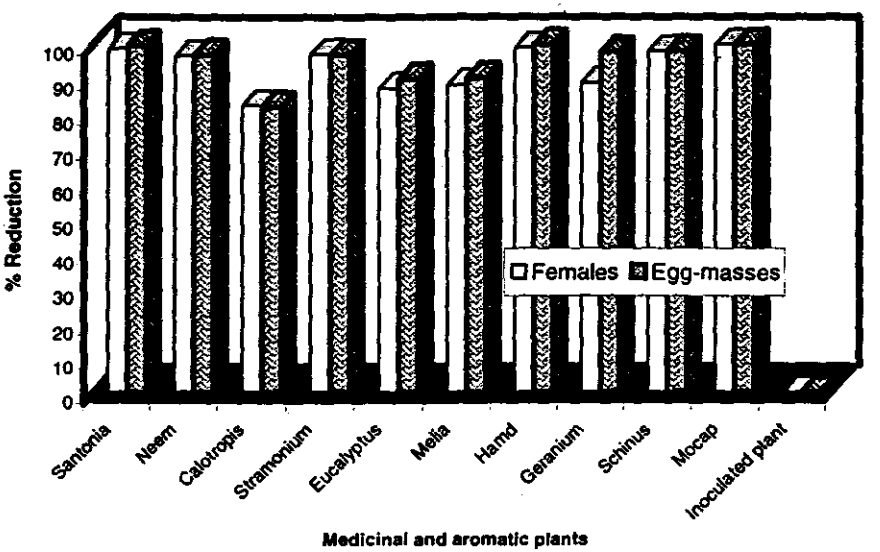


Fig.3 : Effect of botanical soil amendments on *Meloidogyne incognita* rate of reproduction (RF) on eggplant implanted in two soil conditions.

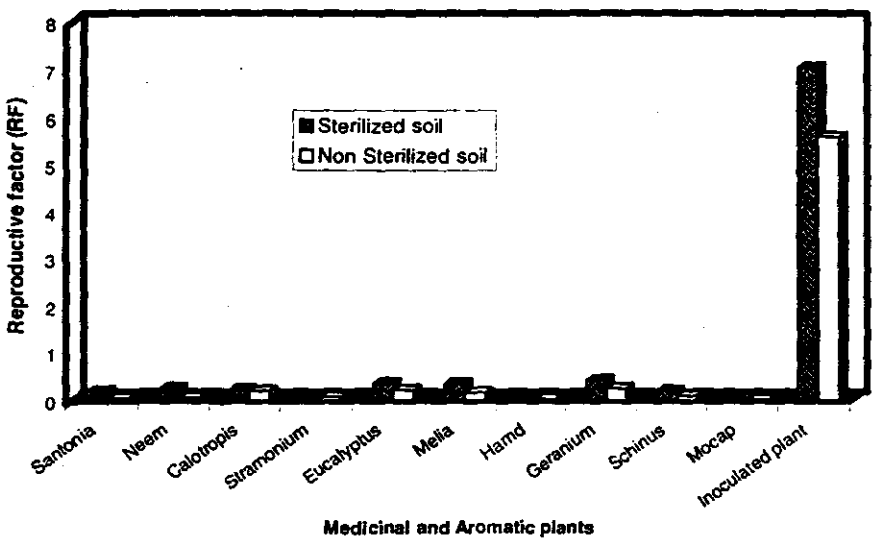


Table 3: Effect of certain botanical soil amendments on plant growth of eggplant seedling infecting with *Meloidogyne incognita* implanted in sterilized soil.

Treatments	Shoot				Root	
	length	Fresh weight	Dry weight	No. Leaves	Length	Fresh weight
Artemisia cinae	44.3	31.1	4.8	8.5	30.5	7.4
Azadirachta indica	40.5	27.0	3.8	7.0	25.3	5.8
Calotropis procera	41.8	26.8	3.4	8.3	27.8	6.9
Datura stramonium	38.8	22.9	3.4	7.5	22.0	3.9
Datura stramonium	39.3	20.3	2.3	7.3	23.3	3.1
Eucalyptus globulus	37.5	18.9	2.3	7.5	18.0	2.0
Melia azedarach	39.5	20.9	1.9	7.3	21.8	3.4
Mesembryanthemum forsskalei	39.8	16.0	2.0	6.0	18.5	2.5
Pelargonium graveolens	36.8	25.1	2.9	7.5	22.0	4.5
Schinus terebinthifolius	40.8	24.3	3.1	7.3	24.5	3.8
Mocap (10G)	36.0	4.5	3.1	8.0	21.3	4.1
Inoculated plants (Check)	39.0	15.1	3.5	7.3	16.8	1.9
Healthy plants	45.3	28.1	5.1	7.8	23.8	10.0
New LSD ($p \leq 0.05$)	5.3	7.7	1.6	N.S	4.3	3.6
New LSD ($p \leq 0.01$)	N.S	10.3	2.1	N.S	5.7	4.8

Table 4: Effect of certain botanical soil amendments on plant growth of eggplant seedling infecting with *Meloidogyne incognita* implanted in natural soil (non sterilizedsoi)l.

Treatments	Shoot				Root	
	length	Fresh weight	Dry weight	No. Leaves	Length	Fresh weight
Artemisia cinae	47.8	35.1	4.9	9.5	35.8	10.3
Azadirachta indica	43.8	27.8	3.9	8.5	26.8	12.3
Calotropis procera	44.5	27.4	3.8	9.8	30.3	9.5
Datura stramonium	39.5	26.3	3.5	7.5	26.5	8.3
Datura stramonium	42.3	21.6	2.6	8.0	24.3	4.0
Eucalyptus globulus	40.0	17.0	2.3	7.8	19.5	2.0
Melia azedarach	40.8	23.1	3.0	7.5	22.8	5.4
Mesembryanthemum forsskalei	40.5	21.0	2.0	6.5	22.0	2.5
Pelargonium graveolens	42.5	26.5	3.6	8.8	22.5	6.1
Schinus terebinthifolius	44.3	24.3	3.4	7.8	24.8	7.8
Mocap (10G)	36.0	06.0	3.9	11.5	21.8	4.4
Inoculated plants (Check)	42.3	17.5	3.6	8.0	20.0	2.5
Healthy plants	46.3	31.5	5.4	9.3	30.3	10.0
New LSD ($p \leq 0.05$)	5.1	09.0	1.3	2.4	4.5	6.6
New LSD ($p \leq 0.01$)	6.8	12.1	1.7	N.S	6.0	N.S

DISCUSSION

Non-chemical control measures were assessed under greenhouse conditions to find out a practical solution for controlling the root-knot nematode, *Meloidogyne incognita* infecting eggplant. Soil amended with decomposable organic matter is recognized as most efficient method of changing soil and rhizosphere environments thereby diversely affecting the life cycle of nematodes and enabling the plant to resist attack of nematode juveniles through better vigor and / or altered root physiology.

We are focusing in this discussion on certain components of the nematode integrated strategy including soil amended with organic manures and some residues or fragments of certain medicinal and aromatic plants, in relation to their efficancy in suppressing nematode activities and reproductive potentiality under pot conditions .

In this study soil nematode populations generally, showed highly significant downward trends at the end of trials in treatments containing *Meloidogyne incognita* infecting eggplant. There were differences in gall indices, numbers of egg-masses resulted from the tested materials in both soil condition. Either there were significant differences in fresh and dry shoot or fresh root weights due to the treatment effects.

Soils that suppress nematode multiplication usually contain range of natural enemies, which attack their host at different stages in its life cycle. Similar trends could be noticed after the decomposition of organic materials in soil. Each may kill relatively few nematodes, but the combined effects of several aspects of biological control agents may prevent nematode population from increasing.

The decomposition of organic matter at the site of nematode population is more effective as a control than partly or fully decomposed material, thus our organic substances were incorporated into soil under experimentation to decompose in nematode site. Our results indicate significant increase in plant growth criteria and remarkable nematode reduction in soil treatments amended with plant residues and organic manures. Plant growth increase due to soil amendments is attributable not only to control root-knot but also to amount and speed of availability of plant nutrients.

In some treatments the populations of free living nematode larvae were always lower in certain amended than in non-amended soil, and were correlated with decrease in the numbers of root-knot larvae in most treatment.

The root-knot nematode, *Meloidogyne incognita* was controlled more effectively and host plants were greater when hamd, stramonium, schinus and santonia were applied as soil amendments than in the un-treated check. Treatments containing *Meloidogyne incognita* and the previous materials exhibited the lowest numbers of galls, egg-masses, females, and number of eggs per egg-mass in the greenhouse tests. Soil amended with santonia, schinus, hamd, and melia had resulted in a significant increase in shoot fresh weight, root fresh weight and shoot dry weight compared with check.

The application of the previous medicinal and aromatic plant parts had resulted also in the best control of *Meloidogyne incognita* over all the screened materials. The addition of certain plant fragments of well documented and potent medicinal plants were found to be successful in reducing nematode biological activities and improved plant vigorness i. e. *Azadirachta indica, Melia azedarach, Asparagas sp., Ricinus communis, Lantana indica , and Eucalyptus restrate* (Akhtar and Alam, 1989 ; Montasser, 1991 (a,b), and Farahat *et al.*, 1994). They proved the great inhibitory effect of neem components on plant parasitic nematodes infecting wide range of host plants. Azadirachtin, nimbidine and thiomemone, are the major nematotoxic compounds that have been found in neem trees, (*Azadirachta indica* and *Melia azaderach*).

Al-Mihanna *et al.* (1999) fund that the addition of dry leaves of eucalyptus caused obvious reduction in wheat cyst nematode, *Heterodera avenae* under field conditions. Many kinds of medicinal plants have been reported in the literature possessing nematicidal properties against pathogenic nematodes. These plants attracted the attention of numerous research workers to explore this area of interest. Ramakrishnan *et al. (*1999) found that the highest nematode mortality (77.2%) was recorded by *Azadirachta indica* followed by *Melia azedarach* (73.3%). Mortality increased with increasing concentrations of plant extracts and exposure time. Our findings are in agreement with these results.

The majority of the tested plants has been found to possess nematicidal substances that may be released in soil during degradation. Sharma *et al,* (1985) stated that dry leaf powders of tagetes and neem reduced population of *Meloidogyne incognita* and improved the growth parameters of sunflower. The antagonistic action of the powder materials on the nematode development and reproduction, revealed the beneficial impact against the nematode. Powders of lantana, tagetes, garlic, neem, majoranum, eucalyptus, sea ambrosia, or asparagus achieved the best results, while powders of artemisia, periwinkle, bitter jasmine, castor or, rosemary had comparatively, lower effects. The great effect of these materials were experimentally proved by many assumptions of numerous investigators, El-Zawahry , 1994 ; Vats *et al.* 1995 ; El-Gonimy, 1996 ; Mostafa , 1997 and El-Rab, 2000.

Our results clearly proved that addition of botanical amendments to the soil reduced infestation level of *Meloidogyne incognita* in eggplant roots. The application of farmyard manures appeared to hamper the development of nematodes in the plant when they developed more slowly than in plants grown in unamended pots.

The addition of organic materials to soil stimulates activity of actinomycetes, algae, bacteria, fungi, microbivorous nematodes, and others (Rodriguez–Kabana and Morgan–Jones, 1987). The deleterious effect on plant–parasitic nematodes can be associated with the increased populations of parasitic or antagonistic organisms as well as the accumulations of decomposition products and microbial metabolites (Goodey *et al* .,1983).

Additionally, organic residues have been associated with build up of free–living nematodes in soil where nematophagous fungi were present, and

high infection of free–living nematodes contributed to the control of plant–parasitic nematodes (Esnard *et al* . , 1998).

As the organic matter decomposes in soil, saprophytic nematodes increase rapidly subsisting on decomposition products and on associated microorganisms. This rise in the saprophytic forms leads to subsequent increase in the numbers of predators. When decomposition neared completion, the availability of saprophytic nematodes decreases and the predators may consume a large portion of parasitic forms and minimize their inoculum in soil. Predacious fungi have been reported to decrease after organic addition (Ragab *et al* . , 1973).

It was realized by (Badra, 1977) in his study that the incorporation of soil organic amendments into soil undergoes a multitude of decomposition processes initiated and continued by inhabitant soil micro–organisms. This decomposition avail plants of abroad variety of macro – and micro–elements released slowly at a rate that matches plant uptake .

Fatty acids and phenol are the main substances included in or formed by the decomposition of these materials. It was verified that fatty acids and phenol had high nematicidal potency.

No doubt other nematicidal compounds could by partially involved in nematotoxicity of organic amendments. Microbial activity on organic substances liberates several gasses like ammonia. Ammonia is recognized as a decomposition product and toxic tool of considerable magnitude in this respect. Some studies gave few clues to the relationship between metabolism of aromatic amino acids (phenyl-alanine and tyrosine) and rapid synthesis of phenolics as induction of resistance in plants (Pegg and Sequeira , 1968).

Finally, it could be summerized that one of the important points in nematode control, the point refers to the understanding that botanical soil amendments should be seen as replacements, effective, safe and lower coast methods for control plant parasitc nematodes and decrease nematicides treatments and maintain higher efficient nematode management to integrate with other method.

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مكافحة نيماتودا تعقد الجذور *Meloidogyne incognita* بالتسميد النباتى العضوى لبعض النباتات الطبية والعطرية على نباتات الباذنجان

أمين وفدى أمين – عبد المنعم ياسين الجندى – أشرف عافية – منار مصطفى سند
قسم الحيوان الزراعي والنيماتولوجيا – كليه الزراعه – جامعه القاهرة – مصر

تم دراسة تسع أنواع من المسحوق الجاف لأوراق النباتات الطبية و العطرية وهى الحمد، الفلفل العريض ،العشار ، الشيح البلدى ، العتر، الزنزلخت، النيم، الكافور البلدى ، والداتورا الخضراء (أوراق و بذور) و إستخدامها كمواد عضوية نباتية فى مكافحة نيماتودا تعقد الجذور *Meloidogyne incognita* على نباتات الباذنجان فى تربة طبيعية غير معقمة وتربة معقمة بالبخار تحت ضغط. دلت النتائج أن كل المعاملات خفضت معنويا من أعداد العقد الجذرية للنيماتودا وأعداد الإناث وأكياس البيض وكذلك متوسط عدد البيض فى كيس البيض و بالتبعية إنخفض معدل تكاثر النيماتودا مقارنة بمبيد الموكاب المحبب (١٠%) والنباتات غير المعاملة. أعطت النباتات التى عوملت بأوراق الحمد ، الفلفل العريض و الداتورا الخضراء أحسن النتائج فى خفض نسبة أعداد الإناث فى التربة المعقمة والتى سجلت ٩٧,٩ و٩٥,٥ و٩٤,٦% لكل من المواد السابقة على التوالى. وكانت أحسن النتائج فى خفض أعداد العقد النيماتودية عند إستخدام مسحوق نباتات الشيح البلدى(٩ عقد) ، الحمد (عقدة١٣) والنيم الهندى (٣٠,٥ عقدة) فى التربة غير المعقمة مقارنة بالموكاب (٧ عقد) والكنترول غير المعامل (٧٠٠ عقدة). وعلى هذا إنخفضت أعداد الإناث بنسبة ٩٨,٨ و ٩٨,٦ و ٩٧,٨% لكل من المواد الحمد ، الشيح و الفلفل العريض على التوالى. وسجلت القياسات على النمو الخضرى متمثلة فى أطوال الجذر والمجموع الخضرى و أوزان المجموع الخضرى (جاف و طازج) والجذرى وعدد الاوراق لنباتات الباذنجان معدل نمو أعلى فى بعض المعاملات فاقت التى سجلت على النباتات السليمة غير المعاملة وغير المعداة بالنيماتودا.و لذلك لوحظ علاقة إيجابية فى أغلب الحالات بين إضافة تلك المواد الطيبية والعطرية كمواد عضوية نباتية فى التربة ومعدل النمو النباتى.

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