

Nematicidal Effect of Some Botanical Powders and Poultry Manure Against the Root-Knot Nematode *Meloidogyne incognita* (Kofoid and White) Chitwood infecting Tomato Plants

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Abstract

A greenhouse experiment was conducted to evaluate the efficacy of soil amendment with six botanical leaves powders and poultry manure in controlling *Meloidogyne incognita* infecting tomato and their potency in growth improvement. The result of the study showed that all the treatments significantly ($p \leq 0.05$) reduced root galling, second-stage juveniles in soil, developmental stages and egg masses and reductions differed according to the amended materials. Although all treatments were effective in reducing root galls and nematode population, soil treated with leaves powder of *Peganum harmala* gave the highest reduction in nematode criteria followed by *Datura stramonium* and *Nicotiana glauca*, However, *Azadirachta indica* had the least reduction compared to the untreated check. The efficacy increased with the increase in dosage rates except with poultry manure at the highest dosage. Most soil amendments at different dosage rates significantly improved the shoot and root weights. *Peganum harmala* achieved the highest increase, while *Azadirachta indica* gave the lowest one.

Keywords: *Meloidogyne incognita*, botanical amendments, tomato, poultry manure.

Introduction

Tomato (*Lycopersicon esculentum* Mill.) is the second important vegetable crop in Yemen and is cultivated in over 18274 hectares with a production of about 258654 meter tons in 2012 (Anonymous, 2013). Root knot nematodes (*Meloidogyne spp.*) are one of the major pests of tomato worldwide and limit its production (Sikora and Fernandez, 2005). Tomato infected by nematodes record yield losses up to 80 % in heavily infested soils (Kaskavalci, 2007). In Yemen, *M. incognita* (Kofoid & White, 1919) Chitwood, 1949 has been regarded as a serious pest of tomato causing severe losses in tomato yield especially in plastic houses and tunnels. Because of the extensive use of chemical nematicides such as methyl bromide in Yemen, especially in greenhouses, they create hazardous problems to human, animals and environment; and are relatively unaffordable to the average small scale farmer (Tsay et al., 2004 and Washira et al., 2009). Therefore, soil amendment with dry ground leaves of plant products and poultry manures have been successfully used for the control of plant-parasitic nematodes and improve plant growth when added to soil (Muller and Gooch, 1982; Alam, 1990; Akhtar

and Mahmood, 1996; Akhtar and Malik, 2000; Kheir, et al., 2000; Wani, 2006; Ahmad, et al., 2007; Ibrahim, et al., 2007; Radwan, et al., 2007; Wiratno et al., 2009; Moosavi, 2012; Ishaku and Ahaji, 2013; Ojo and Umar, 2013 and Youssef and Lashein, 2013).

Soil amendments of either plant or animal origin and poultry manure have beneficial advantageous effects on soil physical condition, nutrients, fertility and biological activities (Hungalle et al., 1986; Akhtar and Malik, 2000). Moreover, they release chemical compounds during their decomposition in soil those act as nematicides (Mian and Rodriguez-Kabana 1982; Sukul, 1992; Akhtar and Alam, 1993; Miano, 1999 and Chitwood, 2002).

The objective of this study was to:

1. Evaluate the nematicidal activity of dried-ground leaves of six plant species (*Argemone mexicana*, *Azadirachta indica*, *Datura innoxia*, *Datura stramonium*, *Nicotiana glauca* and *Peganum harmala*);
2. Poultry manure when used as soil amendments against the root-knot nematode, *M. incognita* infecting tomato under glasshouse conditions.

Materials and Methods

Collection and Preparation of plant Leaves and poultry manure:

Leaves of *Argemone mexicana*, *Azadirachta indica*, *Datura innoxia*, *Datura stramonium*, *Nicotiana glauca* and *Peganum harmala* were separately collected during the summer season of 2013 from different plants at different locations of Sana'a University, Yemen. The leaves were air-dried on laboratory benches for two weeks at 20-30°C. Then, they were ground separately into fine powders with Thomas Wiley laboratory mill and electric Waring blinder and stored in sealed containers until using. Dried chicken manure was obtained from a poultry farm ground and sieved over a 0.5-cm sieve.

Preparation of Inocula:

The root-knot nematode, *M. incognita* culture was initiated from single egg masses of previously identified females (Taylor et al., 1955) and isolated from galled roots of highly infected tomatoes collected from Sahar, Sadah governorate and propagated on eggplant cv. Long purple in a glasshouse at Sana'a University. Adequate second stage juveniles (J2) were obtained from the infected host plant to prepare the needed nematode inocula.

Greenhouse Experiment:

Rio Grande tomato seeds were germinated in foam boxes filled with sterilized sand and peatmoss (1: 2, w:w). Six weeks-old seedlings were singly transplanted in 15-cm. diameter plastic pots filled with steam sterilized loamy sand soil (1:2, w:w). Ten days after transplanting 5, 10 and 15 g (equivalent to 5.7, 11.4 and 17.1 t/ha,

respectively) of leaves powders were incorporated on top of soil around the base of each plant except those of the check. Poultry manure was applied at the rate of 2, 4 and 6 g/pot. Each treatment including the check was replicated 5 times.

Ten days later each plant was inoculated with 800 freshly hatched juveniles of *M. incognita* by pipetting the juveniles suspension in 5 holes around the root system. Five plants were kept without inoculation to serve as a check treatment. All pots were arranged in a randomized block design on a greenhouse bench at temperature degrees of $25\text{ C}^{\circ} \pm 5$. All plants received similar treatments of fertilizing and irrigation. Sixty days after inoculation, the experiment was ended and all plants were rooted-off and nematode counts in both soil and roots were estimated. Plant growth criteria on the basis of shoot and root length and fresh weight were measured. Data were subjected to analysis of variance and means were compared at $P = 0.05$ using SPSS software version 19.

Results

1. Antagonistic effect of the tested materials on the nematode development and reproduction:

As shown in Table (1) all applied treatments caused significant reduction in numbers of root galls, nematode population in soil, development of stages and egg masses in all treatments in comparison to that of the check. Efficacy of the materials differed according to the plant species and dosage rate. Generally, by increasing the amendment dosage rate, the efficacy was also increased (except in treatment of poultry manure). Leaves powder of *Peganum harmala* was the most effective in reducing gall formation, counts in soil, developmental stages, egg masses and final population at the different rates, followed by *Datura stramonium* and *Nicotiana glauca*; while the lowest % reduction in such nematode parameters was recorded in case of the soil amended with *Azadirachta indica* followed by *Argemone mexicana*. Insignificant differences were observed between treatments of leaves powder of *Peganum harmala* and *Datura stramonium* at all dosage rates in values of numbers of galls, counts in soil, and developmental stages. The modest dosage rate (4gm) of poultry manure had considerable more effect on number of galls and nematode criteria with 2.9 build up than other dosage rates. There were no significant differences in number of eggs per egg mass between the check and those treated with *Argemone mexicana*, *Azadirachta indica* and poultry manure at different rates; and with *Datura innoxia* and *Datura stramonium* at rate 5gm. Accordingly, the nematode final population and its rate of build-up were also affected. Treatments of all soil amendments at different dosage rates had lower rates of build-up (0.4 – 15.5) than that of the check (39.3). *Peganum harmala* at all dosage rates achieved the lowest rate of build-up (0.4, 0.6 and 1.7), followed by *Datura stramonium* (0.5, 2.2 and 1.8). The highest rates of build up. (15, 14 and 12) were obtained by *Azadirachta indica* at lowest and modest dosage rate and *Nicotiana glauca* at lowest dosage rate, respectively.

2. Effect of the tested materials on growth of tomato plants:

With regard to the effects of botanical and poultry soil amendments on tomato growth, data in table (2) reveal that most soil amendment, at different dosage rates, significantly improved growth parameters of shoot and root fresh weights except for *Datura stramonium* and poultry manure at lowest dosage rates in case of shoot weight and for *Azadirachta indica* at different dosage rates and *Argemone mexicana* at the lowest and modest dosage rates and lowest dosage rate for *Peganum harmala* in case of root weight. On the other hand, most tested soil amendments at different dosage rates caused insignificant% increase in shoot and root lengths except for *Argemone mexicana* and poultry manure at modest dosage rates and *Azadirachta indica* at different dosage rates in case shoot length and for *Azadirachta indica* and *Datura stramonium* at highest dosage rate in case of root length. Generally, shoot fresh weight increased with increasing in the dosage rates of all leaves powders and poultry manure. The highest shoot fresh weight was recorded in plants amended with leaves powder of *Peganum harmala* and *Nicotiana glauca* at highest and modest dosage rates where they caused 192, 187, 154 and 139 % increase, respectively. Contrary, the lowest percentages of increase were obtained by lowest dosage rate of poultry manure, *Datura stramonium* and *Azadirachta indica* with 26, 39 and 54 % increase, respectively. On the other hand, the highest root fresh weight was recorded in the plants treated with the leaves powder of *Peganum harmala* at highest dosage rate with 213 % increase followed by the lowest dosage rate of poultry manure and the highest dosage rate of *Datura stramonium* with 191% and 184 % increase, respectively.

Discussion

Amending soil with leaves powder of the tested plants and poultry manure suppressed gall formation and population of *M. incognita* both in soil and roots of tomato plants with a concomitant increase in plant growth compared to the untreated check.

When the dosage rate of leave powder was increased, nematode infection and reproduction rate decreased. These results are parallel with those obtained by **Kheir et al., 2000; Hosseininejad, 2004; Radwan, et al., 2007; Ntalli et al., 2010b; Onyeke and Akueshi, 2012 and Cavoski et al., 2012.** *Peganum harmala* performed the best effect in reducing numbers of root galls, juveniles in soil, developmental stages and egg masses of *M. incognita* followed by *Datura stramonium* at the different dosage rates in comparison to those of the check. This could be attributed to production of strong nematicidal substances like harmal's alkaloids including β -carboline as harmine, harmaline, harmalol, harmol and harman and quinazolines as vaccine and vasicinone (**EI-Hassan et al., 2013**). Active ingredients of *D. stramonium* are atropine, hyoscyamine and scopolamine which

Table (1): Effect of some botanical powders and Poultry manure on development and reproduction of *Meloidogyne incognita* infecting tomato.

Treatments	Dose gm.	Galls				Nematode counts						Final population	Build up (PI/PI)
		In soil		In root		D. stages		Egg masses		Eggs/EM.			
		No.	%Red.	No.	%Red.	No.	%Red.	No.	%Red.	No.	%Red.		
<i>Argemone mexicana</i>	5	556 efgn	59	6913 g	76	574 efgnl	61	359 lj	43	995 hl	0	7846	9.8 f
	10	542 efgn	60	6673 fg	77	561 efgn	62	305 hl	52	982 ghl	2	7539	9.4 f
	15	481 def	64	5015 ef	83	499 def	66	273 gn	57	899 efgnl	10	5787	7.2 ef
<i>Azadirachia indica</i>	5	756 h	44	11137 l	62	772 l	48	488 l	23	1102 l	-10	12397	15.5 h
	10	739 hl	45	10611.2 hl	64	758 hl	49	489.8 l	22	938 fghl	6	11859	14.8 h
	15	729 ghl	46	4323 de	85	747 ghl	50	424 k	33	1031 hl	-3	5494	6.9 e
<i>Datura innoxia</i>	5	625 fghl	54	3981 de	86	652 fghl	56	376 jk	40	837 defgn	16	5009	6.3 de
	10	428 ode	68	3671 ode	87	449 ode	70	258 gn	59	765 cdef	23	4378	5.5 de
	15	360 ode	73	1045 ab	96	376 ode	75	173 ef	72	648 bod	35	1595	2 abc
<i>Datura stramonium</i>	5	252 abc	81	1060 ab	96	267 abc	82	151 de	76	895 efgnl	10	1478	1.8 abc
	10	235 abc	83	1315 ab	96	249 abc	83	175 ef	72	767 cdef	23	1739	2.2 abc
	15	130 ab	90	152 a	99	146 ab	90	82 abc	87	640 bod	36	380	0.5 a
<i>Nicotiana glauca</i>	5	493 def	63	9282 h	68	492 def	67	308 hl	51	783 odefg	22	10083	12.6 g
	10	320 bod	76	2053 abc	93	286 abc	81	126 ode	80	713 ode	29	2466	3.1 bc
	15	222 abc	83	855 ab	97	260 abc	82	126 ode	80	597 abc	40	1241	1.6 ab
<i>Peganum harmala</i>	5	274 abc	80	989 ab	97	291 abc	80	97 abod	86	508 ab	49	1377	1.7 ab
	10	123 ab	91	313 a	99	134 ab	91	44 ab	93	485 ab	52	491	0.6 a
	15	80 a	94	200 a	99	91 a	94	39 a	94	435 a	57	330	0.4 a
Poultry manure	2	1062 j	21	4636 e	84	1090 j	26	257 gn	59	1046 hl	-6	5963	7.5 ef
	4	309 bod	77	1901 abc	94	334 bod	77	105 bod	83	1021 hl	-2	2340	2.9 abc
	6	533 efg	60	2590 bod	91	554 efg	63	233 fg	63	843 defgn	16	3376	4.2 cd
Check		1346 k	0	29336 k	0	1482 k	0	630 m	0	999 hl	0	31448	39.3 l

Means followed by the same letter(s) within a column are not significantly different ($p=0.05$) according to Duncan's multiple range test.

Treatments	Dose gm.	Shoot				Root			
		Weight(gm)	% Increase	Length(cm)	% Increase	Weight(gm)	% Increase	Length(cm)	% Increase
<i>Argemone mexicana</i>	5	82.2 bdef	72	56.4 abc	13	26.5 abc	48	27.3 ab	6
	10	99.8 defgh	109	67.4 c	35	25.1 ab	40	25.2 a	-2
<i>Azadirachta indica</i>	5	107.8 fgh	126	62 abc	24	37.4 def	109	27.7 abc	7
	10	73.7bcd	54	67.7 c	35	19.6 a	9	26.8 a	4
<i>Datura innoxia</i>	5	79 bode	65	66.8 c	34	25.9 abc	45	27.6 abc	7
	15	86.9 cdef	82	67.7 c	35	25.9 abc	45	31.7 bc	23
<i>Datura stramonium</i>	5	81.3 bodef	70	57.3 abc	15	35 bode	96	29.1 abc	13
	10	85.2 bodef	78	60.6 abc	21	37.3 def	108	26.6 a	3
<i>Nicotiana glauca</i>	5	96.2 defgh	101	62.7 abc	25	33.7 bcd	88	24.6 a	-6
	10	66.4 abc	39	57.4 abc	15	44.1 efg	146	27.3 ab	6
<i>Peganum harmala</i>	5	95.7 defgh	100	54.8 abc	10	45.2 efg	153	28.8 abc	12
	15	105.6 efg	121	60.5 abc	21	50.8 gh	184	32.2 c	25
<i>Poultry manure</i>	5	108.2 fgh	126	59.8 abc	20	30.6 bcd	71	25.9 a	0
	10	114.4 ghi	139	57.7 abc	15	38.5 def	115	28.3 abc	10
<i>Check</i>	5	137.2 ij	187	57.8 abc	16	44.2 efg	147	25.9 a	0
	10	91.3 cdefg	91	51.5 ab	3	27.1 abc	51	26.4 a	2
<i>Check</i>	5	121.3 hij	154	57.3 abc	15	36.2 cdef	102	28.4 abc	10
	15	139.6 i	192	51.3 a	3	56 h	213	28 abc	9
<i>Check</i>	2	60.1 ab	26	52.6 ab	5	52 gh	191	29.3 abc	14
	4	94.6 defgh	98	64.6 bc	29	49 gh	174	27.2 ab	5
<i>Check</i>	6	94.4 defgh	97	50.6 a	1	46.1 fgh	158	27 ab	5
	Check	47.8 a	0	50 a	0	18 a	0	25.8 a	0

are classified as deliriant, or anticholinergics (**Oduor-Owino, 1993**); and amending soil with leaves were effective against the root-knot nematodes *M. incognita* on tomato (**Chattopadhyay, 1991 and Radwan, et al., 2007**). The *N. glauca* activity might be due to the presence of nicotine, the piperidine alkaloid from tobacco plant which can be toxic in high doses to both animals and human. Its high solubility in water could explain the high effect of its water extracts (**Webb and Dalzell, 1997**). Also, anabasine could be regarded as a highly toxic piperidine like alkaloid constituting about 70% of the plant as a whole (**Mizrachi et al., 2000 and Mhinana et al., 2010**). The nematicidal activity of *A. indica* can be attributed to its contents of tannins, azadirachtine, alkaloids and lipids associates such as nimbidol, nimbine, nimbidine, nimbinin, kemferol, pyronimbin, salannin, thionemone, etc., which possess nematicidal properties (**Rossner and Zebitz, 1987; Akhtar, 2000; Fatoki, 2001 and Abbasi et al., 2004**). Triglyceride was reported as a nematicidal compound in seeds of *Argemone mexicana*. Sanguinarine is a quaternary benzophenanthridine alkaloid with a strong bactericidal effect on Gram-positive bacteria (**Rao and Dave, 2001**) which may confer protection against diverse pathogens (**Facchini, 2001**). Sanguinarine and other identified alkaloids in *Argemone* include berberine, protopine and coptisine may cause nematicidal effects (**Facchini, 2001**).

Amending the soil with poultry manure significantly reduced counts of root galls, juveniles in soil, developmental stages, egg masses in root and final population of *M. incognita*, in addition, the usage of such materials did improve the plant growth. These result are in agreement with the findings of **Kablan and Noe (1993)**, **El-Zawahry, (2000)**, **Ibrahim and Ibrahim, (2000)** **Maareg et al., (2000)**, **Devi and Hassan (2002)**, **Farahat et al., 2008 (Karmani et al., (2011) and Farahat et al. (2012)**). The nematicidal activity of poultry manure is mainly attributed to its contents of volatile fatty acids, phenols, amino acids, ammonia, nitrites and gasses which released during the decomposition of the manure; and to enhance soil populations of micro-organisms antagonistic to nematodes (**Badra, et al., 1979; Mian, Rodrequez-Kabana, 1982; Lazarovits et al., 2001; Oka and Pivonia, 2002; and Farahat et al., 2008**). The observed increase in growth in tomato plants grown in treated soils compared to those in the untreated soils may be attributed to the effective nematode control and to improvement in soil nutrients, soil physical conditions and soil biological activity (**Akhtar and Malik, 2000; Ramesh et al., 2009; Oka, 2010 and Moosavi, 2012**). Therefore, the using of dried leaves of the tested plants and poultry manure could offer a promising tool for nematode management far away from the traditional nematicides -Moreover, such materials are available, cheap, and safe environmentally.

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الملخص العربي

التأثير الإباضي لمطحون بعض النباتات وسيلة الدجاج ضد نيماتودا تعقد الجذور ميلويد وجين

انكوجيتا على نباتات الطماطم

محمد راوح محمد سعيد و جمان نشأت شوكت

قسم وقاية النبات - كلية الزراعة - جامعة صنعاء

قيمت كفاءة المطحون الجاف لأوراق ٦ أنواع نباتية وسيلة الدجاج في مكافحة نيماتودا تعقد الجذور على نباتات الطماطم تحت ظروف الصوبة ولقد أظهرت نتائج الدراسة أن كل المعاملات خفضت معنوياً أعداد العقد النيماتودية وبقرات الطور الثاني في التربة والأطوار المختلفة في الجذور وعدد كتل البيض واختلفت نسبة الخفض باختلاف نوع النبات والجرعة المضافة، وعلى الرغم من أن كل المعاملات أعطت كفاءة عالية في خفض النيماتودا إلا أن مطحون أوراق الحرمل أعطى أعلى كفاءة خفض يليه مطحون أوراق الداتورا نوع استرامونيوم *Datura stramonium* ثم التبغ البري، بينما أعطى مطحون أوراق نبات النيم أقل نسبة خفض، وبزيادة الجرعة زادت معها كفاءة الخفض باستثناء سبة الدجاج عند الجرعة العالية. ولقد حسنت معظم المعاملات بمختلف الجرعات معنوياً من الأوزان الخضرية والجذرية للنباتات وسجل الحرمل أعلى نسبة زيادة بينما أعطى النيم أقل نسبة زيادة في الأوزان الخضرية والجذرية. مما سبق يتضح أن تلك النباتات ذات فعالية عالية في مكافحة النيماتودا وتحسن من خواص التربة وكذا النمو النباتي إضافة إلى توفرها وسهولة استخدامها وعدم وجود أضرار خطيرة منها فيمكن إضافة مطحون تلك النباتات وسيلة الدجاج للتربة في برامج مكافحة المتكاملة للنيماتودا.