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Efficacy of garlic clove and oil aqueous extracts against *Meloidogyne incognita* infecting eggplant

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Abstract

A green house experiment was conducted to control root-knot nematode *Meloidogyne incognita* infecting eggplant *Solanum melongena* with aqueous extracts of garlic *Allium sativum* mashed clove and oil. The plant materials were diluted with distilled water at concentrations of 2.5, 1.25 and 0.625% and drenched in each plot. Results showed that the plant extracts showed nematicidal and nematode-hatching inhibitory activity reduced nematode criteria including number of galls, egg-masses and hatched juveniles on roots of eggplant and number of juveniles in soil at harvest stage compared to untreated plants significantly ($p \leq 0.05$). Lower concentration of the tested materials caused higher percentages reduction the mentioned nematode criteria. Vice versa, increase in length of shoots, dry and fresh weights of shoots and roots occurred by higher concentration of each material followed by those occurred by moderate and lower ones. The percentages of total soluble carbohydrates, proteins, phenolic and carotenoid contents increased at all tested concentrations as compared to untreated plants, but no any relation among them.

Keywords: Garlic clove, oil aqueous, *Meloidogyne incognita*, eggplant, green house

Eggplant *Solanum melongena* is an important vegetable crop globally. Root-knot nematodes, *Meloidogyne* spp., are obligate parasites, damaging plants and this pest limiting agricultural productivity. In Egypt, root-knot nematodes *Meloidogyne* spp., considered as limiting factors in crop production (Ibrahim, 2011). The use of botanical extracts for controlling the root-knot nematodes becoming appealing due to environmental pollution arising from the use of persistent pesticides. Efficacy of different plant extracts in nematode control has been reported by Khan *et al*., (2011) and Mousa *et al*., (2011).

Nematicidal effect of garlic has been reported (Ali, 1994; Ameen, 1996; Amin & Youssef, 1998; Osman *et al*., 2005; Agbenin *et al*., 2005; Bekhiet *et al*., 2010). Most of these studies focused on the use of the tested materials as smashed garlic treatment only. Cetintas & Yarba (2010) evaluated nematicidal effect of five plant essential oils on the southern root-knot nematode *Meloidogyne incognita* race 2, which was one of major pest in many vegetables plants including tomato which demonstrated that garlic and thyme essential oils were more effective in reducing the nematode infestation. Consequently, reducing number of egg-mass and root galling formation caused by *M. incognita* than the other plant essential oils tested. It was evident that garlic and thyme essential oils deserve serious consideration for inclusion into nematode management tactics. Not many studies have been conducted with effect of particular plant essential oils on *M. incognita*. Hence, the objective of this research is to compare the potentials of garlic clove and oil aqueous extracts as soil drench for the control of *M. incognita* infecting eggplant under greenhouse conditions.

Materials and Methods

Plant materials: Eggplant *Solanum melongena* was selected for this study. Aqueous extracts of garlic *Allium sativum* clove and oil were tested.

Preparation of aqueous extracts of garlic clove and oil: Aqueous extract of garlic clove was prepared by soaking 25 g of mashed garlic cloves in 1 lit distilled water and kept for 72 hrs before filtration through Whatman filter paper No.1 and other concentrations prepared by soaking 12.5 and 6.25 g in 1 lit distilled water (equivalent to 2.5, 1.25 and 0.625%). Garlic oil at the same concentrations was prepared by adding 25, 12.5 and 6.25 ml to 1 lit distilled water. These materials were applied in each pot at the rate of 200 ml at time of nematode inoculation. Four weeks-old eggplant cv. Baladi seedlings were transplanted on May 25, 2013 in pots 30 cm diameter containing 6 kg solarized sandy loam soil (1:1 w/w). Each pot was inoculated with 1,000 newly hatched juveniles of root-knot nematode *M. incognita* after establishment of eggplant seedlings in July 3, 2013. There were five pots (replicate) for each treatment. An equal number of pots inoculated with nematode only served as untreated check. All treatments were distributed in a randomized complete block design.

Root and soil samples were collected at harvest stage from each pot including the whole root system on September 9, 2013. Plants were uprooted, cleaned with tap water to avoid adhering soil and examined for the number of galls, egg-masses. Hatched juveniles from egg-masses on roots were extracted according to Young (1954). Juveniles in soil were extracted by sieving and decanting methods. Also, plant growth and yield parameters were recorded.

Chemical analysis: Total soluble carbohydrates (TSC) from dry leaves of eggplant were determined calorimetrically by the method of Yemm & Willis (1954) and total soluble proteins from dry leaves were determined as described by Bradford (1976). Carotenoid content was determined according to the method reported by AOAC (1990). Total phenolic compounds were extracted from dry leaves and determined colorimetrically according to the method defined by Snell & Snell (1953) using Folin Ciocalteu phenol reagent.

Results

Effect on nematode parameters: Data in Table 1 and 2 indicated that root-knot nematode *Meloidogyne incognita* infecting eggplant as affected by different concentrations of aqueous extracts of garlic clove and oil at the end of experiment. It was noticed that garlic clove extract as soil drench treatments at all concentrations reduced number of galls, egg-masses and hatched juveniles on roots and number of juveniles in soil of eggplant compared to untreated check and reduction (%) inversely proportional to the tested concentrations as lower concentration of extract caused higher nematode reduction (%) significantly ($p \leq 0.05$). The same trend was noticed in relation to oil extract as soil treatment. When the effects of the two materials were compared with each other, garlic clove extract was more efficient in reducing the number of juveniles in soil and roots of eggplant than extract of garlic oil, but they were equal in reducing number of galls. As for number of egg-masses, garlic oil was more efficient than garlic clove extract.

Effect on eggplant plant growth: As for eggplant plant growth, the tested materials differed in their efficacy on plant growth parameters according to the tested materials and concentrations at harvest. Higher concentrations of the tested material caused higher percentages increase of length of shoots, fresh and dry weights of shoots and roots than those occurred by lower ones. On the other hand, when the two materials were compared with each other, the results showed that garlic oil extract was more efficient than garlic clove extract in improving shoot length and fresh weight. In contrary, garlic clove extract was more efficient than garlic oil extract in improving dry weight of shoots and fresh and dry weights of roots (Table 2).

Effect on biochemical compounds: It was found that the percentages soluble carbohydrates, proteins and carotenoid contents increased at all tested concentrations compared to those of untreated plants, but without relation among

them. There were no gradual increases in the percentages mentioned contents with the respective concentrations. When the two tested materials were compared with each other, garlic clove extract proved to be more effective than garlic oil in increasing total soluble carbohydrates, proteins and phenolic contents. Garlic oil was better than garlic extract in increasing carotenoid contents without significant differences among them (Table 3).

Table 1. Effect of aqueous extracts of garlic clove and oil on *Meloidogyne incognita* infecting eggplant.

Treatments	Conce-ntration (%)	No. of J_2 in soil	Reduction (%)	No. of J_2 in roots	Reduction (%)	No. of galls	Reduction (%)	No. of egg-masses	Reduction (%)
	0.625	260 c	89.68	1162 d	84.32	33 b	69.72	25 b	69.10
Garlic clove	1.25	288 c	88.57	245 e	96.69	36 b	66.97	29 b	64.20
	2.5	490 bc	80.56	2100 c	71.66	40 b	63.30	31 b	61.70
Average	-	346 B	86.27	1169 B	84.22	36 A	66.97	38 A	53.09
	0.625	292 c	88.41	1320 cd	82.19	36 b	66.97	23 b	71.60
Garlic oil	1.25	713b	71.71	4378 b	40.92	35 b	67.89	25 b	69.14
	2.5	660 b	73.81	1820 c	75.44	33 b	69.72	25 b	69.14
Average	-	555 A	77.98	2506 A	66.11	35 A	67.89	24 B	69.96
Untreated (control)	-	2520 a	-	7410 a	-	109 a	-	81a	-

Figures followed by the same letter(s) are not significantly different at $p \geq 0.05$.

Table 2. Effect of garlic clove and oil aqueous extracts on plant growth of eggplant as affected by *Meloidogyne incognita*.

Treatments	Conce-ntration (%)	Shoot length (cm)	Increase (%)	Fresh weight of shoots	Increase (%)	Dry weight of shoots	Increase (%)	Fresh weight of roots	Increase (%)	Dry weight of roots	Increase (%)
	0.625	53.25 c	23.3	33.1 d	12.2	10.96 b	78.50	12.87 a	36.6	2.11 b	1.9
Garlic clove	1.25	54.30 c	25.7	39.50 c	33.9	11.38 a	85.30	13.10 a	39.1	2.18 b	5.3
	2.5	56.0 b	29.6	42.3 b	43.4	11.74 a	91.20	14.80 a	57.1	2.80 a	35.2
Average	-	54.52 B	26.2	38.30 B	29.8	11.36 A	85.02	13.59 A	44.3	2.36 A	14.0
	0.625	56.0 b	29.6	40.9 bc	38.7	9.22 c	50.2	12.3 a	30.6	2.29 b	10.6
Garlic oil	1.25	57.67 b	33.5	42.47 b	44.0	9.75 bc	58.80	12.57 a	33.4	2.12 b	2.4
	2.5	59.40 a	37.5	53.10 a	80.0	9.79 bc	59.50	14.23 a	51.1	2.30 b	11.1
Average	-	57.69 A	33.5	45.49 A	54.2	9.59 A	56.19	13.03 A	38.3	2.24 A	8.2
Untreated (control)	-	43.20 d	-	29.50 e	-	6.14 d	-	9.42 b	-	2.07 b	-

Figures followed by the same letter(s) are not significantly different according at $p \geq 0.05$.

Table 3. Effect of garlic aqueous extract and oil garlic on biochemical compounds of eggplant as affected by *Meloidogyne incognita*.

Treatments	Concentration (%)	Soluble carbohydrates (%)	Soluble proteins (%)	Phenolic content	Carotenoid (mg/l)
	0.625	10.024 a	11.27 a	0.984 b	0.507 g
Garlic clove	1.25	7.318 b	9.52 b	0.995 b	0.983 c
	2.5	10.162 a	9.42 b	1.177 a	1.068 b
Average	-	9.168 A	10.07 A	1.052 A	0.853 A
	0.625	7.052 b	9.90 b	0.906 c	0.847 e
Garlic oil	1.25	7.220 b	9.94 b	0.767 e	1.098 a
	2.5	5.072 d	8.46 c	0.972 b	0.922 d
Average	-	6.448 B	9.43 A	0.882 B	0.956 A
Untreated (control)	-	5.722 c	8.14 c	0.850 d	0.664 f

Figures followed by the same letter(s) are not significantly different at $p \geq 0.05$.

Discussion

In the present study, the nematicidal activity of the aqueous extracts of garlic clove and oil reduced number of galls, egg-masses and hatched juveniles on roots and number of juveniles in soil as compared to nematode infected plants. Abd-Elgawad *et al*., (2009) reported that soil treatment with a other commercial product aqueous garlic extract contained reduce the nematode root gall index and increased the activity of catalase, B-1,3 glucanase enzyme of tomato leaves in comparison to nematode infected plants. This trend continued in the roots as an indicator of inducing resistance against the root-knot nematode. In the present results, lower concentration caused higher percentages nematode reduction which emphasizes that garlic and oil extracts may serve as inducer for resistance against the root-knot nematode. Also, garlic extract showed no phytotoxicity in contrast to Sukul *et al*., (1974) as they used 250 ml of 50% garlic extract concentration per plant. These concentrations (2.5, 1.25 and 0.25%) were higher than tested concentrations in the present study. High concentrations showed osmotic loss of water from the root tissues as wilting. On the other hand, Tibugari *et al*., (2012) reported that castor aqueous extract > garlic aqueous extract > no treatment regarding gall inhibition, 60 days after nematode inoculum by *M. javanica* on tomato. The present results agreed with Ali (1994) and Osman *et al*., (2005) who attributed the nematode reduction to that garlic clove contain pyruvic acid and ammonia together with diallyl disulphide. Also, Nigh (1985) mentioned that garlic possess biochemical substances of which allelopathic substances were toxic to nematodes. Ameen (1996) reported that soil population of *M. incognita* and *Rotylenchulus reniformis* were significantly reduced in garlic intercropped with cowpea or tomato or garlic monoculture. Nematode population reduced garlic contents of allelopathic substances or garlic did not supported essential elements for nematode development. At the same trend, Youssef & El-Nagdi (2012) reported that garlic plants intercropped with sugar beet infected with *M. incognita* achieved the highest percentage gall reduction (72%) compared to untreated check. El-Saedy *et al*., (2014) studied the efficacy of seven essential oils of which garlic oil and cultivation of two garlic cultivars on *M. incognita* infecting tomato. Garlic oil recorded the best control among the tested oils. On the other hand, garlic has indirect effects on nematode populations and disrupted their mobility, food absorption and reproduction (Fadzirayi *et al*., 2010). Garlic oil

provided significant protection against free-living soil inhabiting nematodes (Block, 2010). Garlic essential oil possessed nematicidal activity on root-knot nematodes (Agbenin *et al*., 2005; Park *et al*., 2005; Ibrahim & Traboulsi, 2009). The volatile antimicrobial substance allicin produced in garlic is active against several plant pathogenic bacteria and fungi and suppressed different plant parasitic nematodes (Sukul, 1992; Zasada *et al*., 2002; Gupta & Sharma, 1993; Curtis *et al*., 2004; Agbenin *et al*., 2005). The immobility of J_2 after 24 hrs of exposure may reach 100% immobility of *M. incognita* J_2 induced by the active bulb extract of garlic which contain component, allicin (Gupta & Sharma, 1993; Korayem & Hasabo, 1994; Curtis *et al*., 2004). An environmentally benign garlic derived polysulfide product is approved for use in the European Union and the UK as a nematicide (Anwar *et al*., 2009).

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