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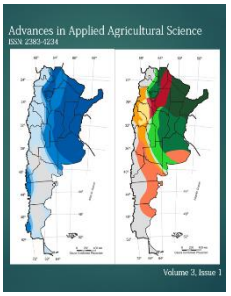

Research Article

Evaluation of two species of water ferns, *Azolla caroliniana* and *A. Pinnata* as soil amendments against *Meloidogyne javanica* infecting tomato in Egypt

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ABSTRACT

Under greenhouse conditions 25±5°C, the effect of two Azolla species, *Azolla caroliniana* and *A. pinnata* on controlling *Meloidogyne javanica* was determined as dried soil amendments. The treatments were applied at the rates 25 and 50 gm of dry materials of each species / pot. Application of *A. caroliniana* and *A. pinnata* significantly (P = 0.05 and or 0.01 levels) succeeded in reducing the development and reproduction of *M. javanica* and also significantly (P = 0.05 and or 0.01 levels) improved tomato growth when compared with those of the check. Application of *A. pinnata* species was significantly (P = 0.05 and or 0.01 levels) reducing number of nematode stages based on galls, egg-masses, females, developmental stages in roots, as well as, number of juveniles in soil per plant at both rates as compared with *A. caroliniana* did. Also, the growth of tomato plants was affected due to the application of azolla. Addition of azolla to the plant soil caused remarkable increase in the tomato growth. The higher dose of each azolla species was significantly (P = 0.05 and or 0.01 levels) more effective than the lower one. However, impact of dry materials of *A. pinnata* resulted in increasing the plant growth much more than *A. caroliniana* did.

In Egypt, root-knot nematodes, *Meloidogyne* spp. attack tomato (*Solanum lycopercicum* L.) roots and severely reduce the plant growth. Therefore, control of this nematode species has received attention to minimize damage. Synthetic nematicides have efficiently been used for a long time; however, hazards resulting from such chemicals have encouraged scientists to search for alternatives. Several studies showed significant reduction in the nematode infestation and avoidance of environmental pollution by using organic nitrogen fertilizers as one of the most effective control measure against phytonematode. (Nakhla et al., 1998, Ismail et al., 2007, 2009 and Usman and Siddiqui, 2013). Azolla, a free floating water fern commonly known as mosquito fern or duckweed fern or fairy moss (Yanni et al., 1994) heavily grows on water surface and fixes atmospheric nitrogen. There are three Azolla species namely: *Azolla pinnata* R. Br., *A. caroliniana*

Willd. and *A. filiculoides* Lam. (Azollaceae) which were introduced by soil microbiologists of the Agriculture Research Center (ARC), Egypt for green manuring in rice cultivation (Yanni et al., 1994). In addition, attempts have been devoted to utilize Azolla plants as a bio-fertilizers in management different plant-parasitic nematodes i.e. *Pratylenchus penetrans*, *Heterodera glycines*, *Meloidogyne incognita*, *Tylenchorhynchus vulgaris* and *M. incognita* in soil (Walker, 1969, Barker et al., 1971, Thaker et al., 1988, Patel et al., 1989 and 1994 and Abadir and El-Hamawi, 1995; respectively). The aim of the present work was to compare the role of two dry species of water ferns, *A. caroliniana* and *A. pinnata* as soil amendments in suppressing root knot nematode, *M. javanica* infecting tomato under greenhouse conditions in Egypt.

Materials and Methods

Two species of Azolla plants namely, *A.carolinaria* and *A. pinnata* were dried and used as soil amendments under greenhouse conditions. Pure culture of Azolla plants were originally obtained from Plant Nutrition Department, Institute of Soil and water Research, Agriculture Research Center (ARC), Giza, Egypt. Twenty three-day old of tomato, *Solanum lycopercicum* L. seedlings cv. Super Strain-B were transplanted in 15-cm diameter clay pots filled with 1 kg solarized sandy loam soil (1:1 w/w) and incorporated with dry Azolla plants at the rates of 25 and 50 gm / pot of each species. The dry Azolla rates of each species were added two weeks before transplanting to allow their decomposing in the soil. All the treatments were replicated five times, besides a set of five pots were left without adding Azolla to serve as the check. All pots were arranged in a greenhouse bench in a randomized block design (25± 5 ºC). Seven days later, all the pots were inoculated with 1000 freshly hatched juveniles of *M. javanica* / pot. The experiment was terminated seventy days after nematode inoculation. The nematodes in the soil were extracted by sieving and decanting methods (Barker, 1985). Also, the number of galls, egg-masses, females and the developmental stages in tomato roots were counted. All shoots and roots parameters e.g. lengths, fresh weights and dry weights of both root and shoot were recorded. Percentage of plant growth increase based on shoot and root fresh weights and percentages of nematode reduction were determined.

Statistical analysis

All nematode and plant growth data were subjected to analysis of variance by the least significant differences (LSD at P= 0.05 and 0.01 levels) according to Gomez and Gomez (1984).

Results and Discussion

The data in Table 1 revealed that, in general , that when dry azolla plants of both species were incorporated into soil of tomato plants cv. Super Strain B fourteen days before transplanting, it significantly (P = 0.05 and or 0.01 levels) affected on development and reproduction of *M. javanica* as compared to the check plants. It is clearly to notice that *Azolla pinnata* was significantly (P = 0.05 and or 0.01 levels) reducing all the nematode stages as compared with *A. caroliniana* did (Table 1). However, both Azolla species significantly (P = 0.05 and or 0.01 levels) decreasing gall formation, egg-masses number, females number, developmental stages number and number of juveniles per plant when compared with those of the control. Also, significantly (P = 0.05 and or 0.01

Table 1. *Effect of Azolla caroliniana and A. pinnata as dry soil additive on development and reproduction of M. javanica infecting tomato.*

Treatments & rates	No. of galls / root	R. %	No. of egg-masses / root	R. %	No. of females / root	R. %	No. of D.S./ root	R. %	No. of juveniles in soil	R. %
Azolla caroliniana										
25 g / pot	208	18.8	109	19.9	189	18.5	173	14.8	1320	29.8
50 g / pot	164	35.9	67	50.7	107	53.9	145	28.6	980	47.9
Azolla pinnata										
25 g / pot										
50 g / pot	187	27.0	56	58.8	87	62.5	92	54.7	1134	39.7
	78	69.5	45	66.9	51	78.0	52	74.4	675	64.1
Control	256		136		232		203		1880	
LSD (P= 0.05)	16		18		32		55		203	
LSD (P= 0.01)	23		32		40		63		245	

R. = Reduction %. D.S. = Developmental stages.

Table 2. *Effect of Azolla caroliniana and A. pinnata as dry soil additives on growth parameters of tomato plants infected with Meloidogyne javanica.*

Treatments & rates	Lengths (cm)				Fresh weights (g)				Dry weights (g)			
	root	Inc.%	shoot	Inc.%	root	Inc.%	shoot	Inc.%	root	Inc.%	shoot	Inc.%
Azolla caroliniana												
25 g / pot	24.9	4.6	73.2	4.6	6.3	28.6	27.3	10.1	1.4	16.7	4.3	16.2
50 g / pot	30.1	26.5	75.6	8.0	6.9	40.8	30.5	23.0	1.9	58.3	5.6	51.4
Azolla pinnata												
25 g / pot												
50 g / pot	31.3	31.5	77.3	10.4	8.7	77.6	36.2	46.0	2.1	75.0	5.8	56.8
	35.7	50.0	80.1	14.4	9.3	89.8	38.7	56.1	2.6	116.7	6.4	73.0
Control	23.8		70.0		4.9		24.8		1.2		3.7	
LSD (P= 0.05)	3.2		2.8		2.1		2.6		0.6		1.7	
LSD(P= 0.01)	5.7		3.4		3.7		4.2		0.9		2.4	

Inc. = Increase %.

levels) increase was attained with increasing the rates of Azolla. So, galls number was highly affected by using the higher rate (50 gm / pot) of each species than the lower one (25 gm /pot), as was significantly (p = 0.05 or 0.01) reduced by 35.9 and 18.8%, or by 69.5 and 27.0% when the soil was amended with *A. carolinia*na or *A. pinnata*; respectively (Table 1). Also, the reproductive potential of the nematode was significantly reduced affected when soil was amended with either *A. carolinia*na or *A. pinnata*. Noticeable decrease in number of egg-masses, females, developmental stages / root as well as number of juveniles / soil was obtained by using either higher or lower rate of Azolla plants as compared to the control. Using of dry materials of *A. pinnata* treatment was significantly (P = 0.05 and or 0.01 levels) more effective than *A. caroliniana*. Moreover, pronounced difference was observed between the rates of both Azolla species. Comparatively, percentages of reduction in number of egg-masses / root of *A. caroliniana* treatments were 19.9 and 50.7 for the lower

and higher rates; respectively; while they were 58.8 and 66.9% for the lower and higher rates of *A. pinnata*; respectively (Table 1).

Growth of tomato plants was also promoted due to soil amending with dry Azolla plants. Generally, all Azolla treatments significantly (P = 0.05 and or 0.01 levels) increased all plant growth parameters (Table 2). A positive correlation was detected between the plant growth increase and rates of both Azolla species. However, application of dry materials of *A. pinnata* was significantly (P = 0.05 and or 0.01 levels) more effective than *A. caroliniana*, and the higher rates were, also, more effective than the lower ones (Table 1). The obtained results proved the activity of azolla plants in reducing population of *M. javanica* when used as soil amendments. Some of the previous reports mentioned that the highly nutritive components of such plants from minerals (Calcium, Phosphorus, Potassium, Sodium, Magnesium, Manganese, Zinc, Copper and Iron) as well as amino acids e.g. Alanine, Arginine, Aspartic acid, Cystine, Glutamic acid, Glycine, Histidine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Proline, Serine, Threonine, Tryptophan, Tyrosine and Valine could significantly adversely interfere on the development and build-up of the nematode (Thomas et al., 1980 and Francisco et al., 2000). In addition, the decomposing dry azolla plants could help in improving the plant growth. Similar results has been obtained by Patel et al. (1989 & 1994) as they found that dry *A. pinnata* reduced the stunt nematode, *Tylenchorhynchus vulgaris* population and consequently improving the plant growth, as well as, Abadir and El-Hamawi (1995) who found that fresh materials of both *A. filiculoides* and *A. pinnata* decreased the root-knot nematode, *M. incognita* and improved the plant growth. In addition, Thaker et al. (1988) reported that extracts of both fresh and dry *A. pinnata* plants inhibited hatching of *M. javanica* and *M. incognita* eggs. However, further studies are needed to clearly elucidate proper effect against plant-parasitic nematodes.

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