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Impact of *Azolla caroliniana* and *A. pinnata* as soil amendments on *Rotylenchulus reniformis* and plant growth of cowpea in Egypt

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ABSTRACT

Two Azolla species, *Azolla caroliniana* and *A. pinnata* were evaluated throughout two successive seasons (2015 & 2016) as green manures on *Rotylenchulus reniformis* and plant growth of cowpea cv. Baladi under greenhouse conditions (25±5°C). The substrates were applied at the rates of 25 and 50 gm of dry material of each species / pot. The use of *A. caroliniana* and *A. pinnata* significantly (P = 0.05, 0.01 levels) succeeded in reducing the number of swollen females and number of egg-laying females and also significantly (P = 0.05, 0.01 levels) improved cowpea growth when compared with those of the check. The use of *A. pinnata* significantly (P = 0.05, 0.01 levels) reduced the number of swollen females and number of egg-laying females per plant, at both rates, when compared to *A. caroliniana*. Also, the growth of cowpea plants was affected due to the application of *Azolla*. Addition of *Azolla* to the plant caused a remarkable increase in the cowpea growth. The higher dose of each *Azolla* species was significantly (P = 0.05, 0.01 levels) more effective than the lower one. In general, the use of dry materials of *A. pinnata* resulted in increase in the plant growth, more than *A. caroliniana*.

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Introduction

The reniform nematode, *Rotylenchulus reniformis* attacks a wide range of crops including cowpea, *Vigna sinensis* in Egypt as well as in many parts of the world (Oteifa, 1987 and Johnson & Fassuliutis, 1984). Therefore, management of this nematode has received attention to reduce damage. However, hazards resulting from synthetic nematicides have encouraged scientists to search for alternatives. A free floating water fern commonly known as azolla or mosquito fern or duckweed fern or fairy moss heavily grows on water surface and fixes atmospheric nitrogen. Some studies 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*, *M. incognita and M. javanica* in soil (Walker, 1969, Barker *et al.*, 1971, Thaker *et al.*, 1988, Patel and Thaker, 1989 and Patel *et al.*, 1994 , Abadir and El-Hamawi, 1995 and Ismail, 2015; respectively).Therefore, the aim of the present work wasto compare the effective role of

two species of *Azolla* plants namely *A. caroliniana* and *A. pinnata* as soil additives in suppressing the reniform nematode, *Rotylenchulus reniformis* infecting cowpea throughout two successive seasons (2015 & 2016) under greenhouse conditions in Egypt.

Materials and Methods

Seeds of cowpea, *Vigna sinensis* cv. Baladi were sown in 15 cm diam. plastic pots filled with 1 kg autoclaved sandy loam soil (1:1 w/w) with pH 7.3 and E.C. 0.56 throughout two successive seasons (2015 & 2016). After germination, the seedlings in each pot were thinned to one seedling / pot. Two species of *Azolla* plants namely, *A. caroliniana* and *A. pinnata* were air dried under shade for 2 weeks and finally powdered by an electric grinder and thoroughly incorporated with the soil as soil amendments at two_rates i.e. 25 and 50 gm / pot of each species. The pure culture of *Azolla* plants were obtained from Plant Nutrition Department, Institute of Soil and Water Research, Agricultural Research Center (ARC), Giza, Egypt. The dry *Azolla* rates of each species were added two weeks before seeding to allow their decomposing in the soil. The chemical analyses of the tested *Azolla* species are shown in table (1). The treatments were replicated five times. Untreated pots served as control. The pots were arranged in a greenhouse bench in a completely randomized block design (25± 5 ºC) and irrigated with tap water as needed. One thousand of infective stage (J4) of *R. renoformis* (equivalent one J4 per one gm soil) were poured into three holes at the base of the standing plant 15 days after germination.The nematode inoculum was obtained from tomato (*Lycopersicon esculentum* L. cv. Super Strain B) roots .After dissolving the gelatinous matrix of the nematode egg masses using sodium hypochlorite solution, centrifuging and washing with tap water, the nematode suspension was left in aerated water for three days at 27 ± 2ºC and then examined under stereomicroscope to confirm that all viable eggs had hatched just before application. Seventy days after the nematode inoculation time, cowpea plants were carefully uprooted and nematodes in pot soil and on cowpea roots were determined. The nematode juveniles in the soil were extracted by sieving and decanting method (Barker, 1985). Also,the number of swollen females / root and egg- laying females / root were determined. All plant growth parameters e.g. lengths, fresh weights and dry weights of both root and shootwere recorded. Percentages of plant growth increase based on shoot and root fresh weights and percentages of nematode reduction as compared to untreated pots were calculated .

Statistical analysis

In both experiments, the obtained data on cowpea growth components (lengths and fresh weights of both shoot and root growth) were collected. Data were also collected on number of larvae in soil, both females and egg-laying females on roots from all the treatments. All the data were pooled together and means were analyzed statistically using the Fisher's Least Significant Difference (L.S.D.) according to Gomez and Gomez (1984).Correlation analyses were also used to determine the relationships between doses of tested substances, each of decreases in the different nematode stages and increases in the cowpea growth components.

Results and Discussion

When dry *Azolla* additives of both species were incorporatedinto soil of cowpea plants cv. Baladi fourteen days before seeding, it significantly (P = 0.05 and or 0.01 levels) affected on development and reproduction of *R. reniformis* as compared to the check plants (Table 2). Clearly, *Azolla pinnata* was significantly (P = 0.05 and or 0.01 levels) reducing all the nematode stages as compared with *A. caroliniana* did. Both *Azolla* species significantly (P = 0.05 and or 0.01 levels) decreasing swollen females / root, egg- laying females / root and the

Table 1. Chemical analyses of the tested *Azolla* species.

Azolla species	Total nitrogen%	Organic carbon%	Organic matter%	C:N	E.C.	pH*
Azolla caroliniana	0.44	48.00	80.38	108:1	1.09	6.0
Azolla pinnata	0.67	45.80	81.64	79:1	1.05	5.8

*pH= 1: 2.5 *Azolla*: water ratio, E.C. = Electrical conductivity (m mhos / cm 2)

Table 2. Effect of *Azolla caroliniana* and *A. pinnata*as dry soil additives on the final population of *Rotylenchulus reniformis* infecting cowpea. (Mean of two successive seasons, 2015 & 2016)

Treatments & rates	No. of swollen females / root	Reduction %	No. of egg-laying females / root	Reduction %	No. of juveniles in soil / pot	Reduction %
Azolla caroliniana						
25 g / pot	180	26.8	89	36.0	1220	28.2
50 g / pot	134	45.5	51	63.3	880	48.2
Azolla pinnata						
25 g / pot	157	36.2	43	69.1	1100	35.3
50 g / pot	58	76.4	39	71.9	600	64.7
Control	246		139		1700	
L.S.D. 0.05	13		16		198	
L.S.D. 0.01	21		29		233	

Table 3. Cowpea growth as influenced with dry soil additives of *Azolla caroliniana* and *A. pinnata* and infected with *Rotylenchulus reniformis.* (Mean of two successive seasons, 2015 & 2016)

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	26.1	3.5	66.1	2.3	5.9	18.0	26.3	9.6	1.3	8.3	4.1	2.5
50 g / pot	32.0	27.0	70.2	8.7	8.4	68.0	28.7	19.6	2.2	83.3	5.5	37.5
Azolla pinnata												
25 g / pot	34.6	37.3	79.7	23.4	9.4	88.0	37.4	55.8	2.6	116.7	6.3	57.5
50 g / pot	37.0	46.8	83.7	29.6	11.8	136.0	39.4	64.2	3.4	183.3	7.1	77.5
Control	25.2		64.6		5.0		24.0		1.2		4.0	
L.S.D. 0.05	3.1		2.7		2.0		2.5		0.5		1.6	
L.S.D. 0.01	5.6		3.3		3.6		4.1		0.8		2.3	

Inc. = Increase %.

juveniles in soil when compared with those of the control. Significantly (P = 0.05 and or 0.01 levels) increasewas attainedwith increasing the evaluated rates of *Azolla*. So, swollen females was highly affected by using the higher rate (50 gm / pot) of each azolla species than the lower one (25 gm /pot), as was significantly (p = 0.05 or 0.01)reduced by 45.5% and 26.8%, or by 76.4% and 36.2% when the soil was amended with *A. caroliniana* or *A. pinnata*; respectively (Table 2). The reproductive potential of the nematode was significantly decreased when soil was treated with either *A. carolinia*na or *A. pinnata*. Evidently, decrease in number of swollen females/ root, egg- laying females / root as well as number of juveniles in soil was attained by using the different rates of *Azolla* plants as compared to the control. The application of dry substrates of *A. pinnata* treatment was significantly (P = 0.05 and or 0.01 levels) more effective than *A. caroliniana*. Also, a pronounced difference was observed between the rates of both *Azolla* species. As shown in Table 2, percentages of reduction in number of egg- laying females / root of*A. caroliniana* treatments were 36.0 % and 63.3% for the lower and higher rates; respectively; while they were 69.1 and 71.9% for the lower and higher rates of *A. pinnata*; respectively.

Cowpea plants showed luxuriant growth due to soil amending with dry *Azolla* plants. The two rates significantly (P = 0.05 and or 0.01 levels) increased plant growth criteria's (Table 3). A positive trend was detected between the plant growth increase and the used rates of both *Azolla* species. 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 as shown in Table 2.The present results proved the activity of *Azolla* plants in eliminating the reniform nematode population and improved the cowpea growth when used as soil additives. Some studies stated that the highly nutritive components of such plants from minerals such as 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 reproduction of the nematode (Thomas *et al.,* 1980 and Francisco *et al.,* 2000). Consequently, the decomposing dry *Azolla* plants as green manures could help in improving the plant growth. Similar results has been attained by Patel et al. (1989 & 1994) as they found that dry *A. pinnata*as soil additives reduced the stunt nematode, *Tylenchorhynchus vulgaris* population and consequently improving the plant growth, as well as, Ismail (2015) found that both of *A. caroliniana* and *A. pinnata* as green manures reduced the root-knot nematode, *M. javanica* and improving the tomato growth. Fresh materials of both *A. filiculoides* and *A. pinnata* decreased *M. incognita* and improved the plant growth (Abadir and El-Hamawi, 1995). Moreover, the extracts of both fresh and dry*A. Pinnata* plants inhibited egg hatching of *M. javanica* and *M. incognita* (Thaker *et al.,* 1988). Further studies are needed to clearly elucidate proper effect against plant-parasitic nematodes.

References

Abadir S K, El-Hamawi M H. (1995). Activity of azolla plants against *Meloidogyne incognita* infecting tomato. Egyptian J. Applied Sciences 10(11): 316-321.

Barker K R, Lehman P S and Huisingh D. (1971). Influence of nitrogen and *Rhizobium jabonicum* on the activity of *Heterodera glycines*. Nematologica 17: 377-400.

Barker T R. (1985). Nematode extraction and bioassays.19-35 pp. In: An Advanced Treatise on *Meloidogyne* Vol. II. (Eds.)Barker,T.R.,Carter,C.C. and Sasser,J.N., North Carolina State University.

Francisco C, Generosa T and Diniz M A. (2000). *Azolla* as a Biofertiliser in Africa.A Challenge for the future. Revista de Ciencias Agrarias 23(3-4): 120-138.

Johnson A Y, Fassuliutis G (1984).Nematodes parasites of vegetable crops. In: Nickle WR, editor. Plant and insect nematodes. New York: Marcel Dekker; p.323-372.

Gomez K A & Gomez A. A. (1984). Statistical procedures for agriculture research, 2nded. New York (NY): John Wiley 780 p.

Ismail A E (2015). Evaluation of two species of water ferns, *Azolla caroliniana* and *A. pinnata* as soil amendments against *Meloidogyne javanica* infecting tomato in Egypt. Advances in Applied Agricultural Science 3(1):18-22.

Oteifa BA. (1987).Nematode problems of winter season crops in the Mediterranean region. In: Saxens MC, Sikora JP, editors. Proceedings of a workshop on parasitic nematodes in temperate semi-arid regions, held at Larnaca, Cyprus, 1987 Mar 105; Aleppo; ICARDA. P.199-207.

Patel H R, Patel D J, Patel C C and Thaker N A.(1994).Effectivity of *Clerodendron inerme* L., *Catharanthus roseus* L. and *Azolla pinnata* for management of root-knot nematode in okra. Pak. J. Nematol., 12: 95-98.

Patel P N and Thaker N A. (1989). Organic amendments in control of the stunt nematode, *Tylenchorhynchus vulgaris* on wheat. Indian J. of Nematology 19:81-82.

Thaker N A, Patel C C& Patel H R.(1988).Effect of extracts of *Azolla pinnata* on egg hatching of root-knot nematode *Meloidogyne incognita* and *M. javanica*. Madras Agricultural Journal 75: 297-299.

Thomas A, Lumpkin A& Donald L P. (1980). *Azolla*: Botany, Physiology, and Use as a Green Manure. Economic Botany 34(2): 111-153.

Walker J T. (1969). Depression of *Pratylenchus penetrans* populations by nitrogenous amendments. Phytopathology 59: 403-404.

Yanni Y G, Shalaan S Nand El- Haddad M. (1994).Potential role of *Azolla* as green manure for rice in Nile delta under different levels of inorganic fertilization.In:Nitrogen Fixation with non-legumes. N. A. Hegazi, M. Fayez and M. Monib (Eds.). The American University in Cairo Press: 127-132.