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MANAGEMENT OF THE ROOT-KNOT NEMATODE OF TOMATO BY INOCULATION WITH ARBUSCULAR MYCORRHIZAL FUNGI

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ABSTRACT

Momotaz R, Alam MM, Islam MN, Alam KM, Rahman MZ (2015) Management of the root-knot nematode of tomato by inoculation with arbuscular mycorrhizal fungi. *Int. J. Sustain. Crop Prod.* 10(2), 48-54.

Arbuscular mycorrhizal fungi (AMF) have prospective role in biocontrol of soil-borne diseases. In winter season the experiments were conducted to quantify the interactions between AM fungi [*Glomus versiforme* (Karsten) Berch] and *Meloidogyne incognita* and rhizosphere of tomato plants (var. BARI Tomato 2) and to determine their combined effects on the root-knot nematode, *Meloidogyne incognita* and on tomato growth. AMF spore inoculation includes 60 g, 70 g, 80 g, 90 g, and 100 g spore along with Furadan 5G and without inoculation or control agent was as control. The highest shoot length was recorded in plants inoculated with 70 g AMF spore. The maximum shoot weight was recorded in 100 g AMF spore inoculation (44.33 g). The root length was ranged from 13.40-24.20 g. Significantly higher root length was computed in 100 g AMF spore and it differed significantly with all other inoculation. All the treatments amended with AMF proved their efficiency to establish the root infection. It was found that root infection increased with the increasing of AMF inocula concentration. The minimum gall index of 2.0 at 0-10 scale was assessed in T_3, T_5 and T_6. The maximum yield was harvested from T_5 (2740 kg/ha). T_5 (100 g AMF spore) gave satisfactory results to reduce root-knot disease and to improve plant growth and yield of Tomato. The results indicate that specific AM fungi can interact to suppress *M. incognita*, disease development and increases tomato yield.

Key words: *arbuscular mycorrhizal fungi, Glomus versiforme, disease control, meloidogyne incognita, root knot nematode*

INTRODUCTION

Bangladesh benefited from the green revolution in cereal production in the past but was not able to substantially reduce poverty and malnutrition. Vegetable production can help farmers to generate income which eventually alleviate poverty. Among the vegetables tomato (*Lycopersicon esculentum* L.) is one of the most important vegetables in terms of acreage production, yield, commercial use and consumption. At present 6.10% area is under tomato cultivation both in winter and summer (BBS 2005). It is the most consumable vegetable crop after potato and sweet potato occupying the top of the list of canned vegetable (Chowdhury 1979). It is cultivated all over the country due to its adaptability to wide range of soil and climate (Ahmed 1976). It is also a good source of vitamin C (31 mg per 100 g), vitamin A, calcium, iron etc (Matin *et al*. 1996). Its demand for both domestic and foreign markets has increased manifold due to its excellent nutritional and processing qualities (Hossain *et al*. 1999). The average yield of tomato is too low in our country as compared to that of other tomato growing countries and several yield limiting factors of tomato are enumerated. Among them, diseases caused by fungi, bacteria, nematodes and viruses play major role. The root-knot disease caused by *Meloidogyne incognita* is highly damaging and yield reducing factor of tomato throughout the country (Mian 1986). In some cases, especially in the intensive tomato growing areas, root-knot nematode is one of the major limiting factors affecting tomato production. The average losses due to root-knot nematode infestation are 20.6% in tomato (Sasser 1989). It causes about 40% yield loss of tomato in Bangladesh and about 46.2% yield reduction in India (Mohsin 1987). A number of approaches aimed for controlling root-knot nematodes through application of nematicides (Hossain *et al*. 1989), organic soil amendments (Faruk *et al*. 2001), cultural management, physical methods like soil solarization and biological measures like *Trichoderma* spp., *Pacecilornyces lilacinus*, *Pasturia penetrans* and *Pseudomonas aeruginosa* (Rao *et al*. 1997; Reddy *et al*. 1998; Siddiqui *et al*. 1999). Tomato cultivar resistant to root-knot nematode is not available in Bangladesh. Chemical control of this nematode is very costly and also undesirable because chemical nematicides may affect the agro-eco system and also has harmful effect on numerous beneficial parasites, predators and other microbes prevailing in the soil. Therefore, alternate management options against the disease are to be sought.

New biological methods of control including AMF are now considered as important alternatives to pesticides. The AMF contributed to the control of plant disease and the mechanisms by which they do so have been well documented (Ahmed *et al*. 2009; Vierheilig *et al*. 2008; Whipps 2004; Elsheikh and Mirghani, 1997). The presence of AMF in roots can reduce development of some soil-borne pathogenic bacteria, fungi, and nematodes and can also induce increased tolerance to plant diseases (Elsen *et al*. 2008). Li *et al*. (2004) observed that the AMF *Glomus versiforme* (Karsten) Berch, *Glomus mosseae* (Nicol. & Gerd.) and *Gigaspora rosea* decreased the propagule density of the pathogen, *Fusarium oxysporum* f. sp. *niveum* (E.F. Sm.). The mycorrhizal roots decreased disease incidence and severity on watermelon plants with *G. versiforme*. AMF have the ability to induce systemic resistance against plant-parasitic nematodes in a root system (Elsen *et al*. 2008). The establishment of an AMF before nematode infection reduced reproduction of the root-knot nematode *Meloidogyne incognita* and reduced disease severity in infested soil (Dos Anjos *et al*. 2010). A consortium of AMF suppressed *Fusarium* wilt of cucumber and showed potential for biocontrol in greenhouse agroecosystems (Hu *et al*. 2010).

Therefore, the present study was undertaken to find out the efficacy of mycorrhiza to control root-knot nematode, increase plant growth and yield of tomato. In addition, effect of AMF on plant growth and disease suppression was also determined.

MATERIALS AND METHODS

A pot experiment was conducted to test efficacy of AMF and a nematicide Furadan 5G (Carbofuran) to control root-knot nematode of tomato at the Plant Pathology Division pot house, Bangladesh Agricultural Research Institute (BARI), Gazipur during winter season. Standard cultivation procedures recommended by BARI were followed to grow tomato with little modification (Anon. 2007). The experiment was laid out in a complete block design with a total of seven treatments including a control with five replications each were included in the study *viz.* T_1 = 60 g AMF spore, T_2 = 70 g AMF spore, T_3 = 80 g AMF spore, T_4 = 90 g AMF spore, T_5 = 100 g AMF spore, T_6 = Furadan 5G and T_7 = control were maintained in this experiment. The 35 pots were used and each pot contains 5 kg of soil. Furadan 5G was applied to the experimental pot just before transplanting of seedlings. To ensure inocula of the nematode, chopped severely galled tomato roots infected with *M. incognita* were mixed with soil around the tomato seedlings @ 2 g/plant. AMF inoculums were collected from Soil Science Division of BARI. Firstly, AMF inoculums was placed in each pot then 3 seedling planted in each pot. Twenty five days old and apparently healthy tomato seedlings of variety BARI Tomato 2 were transplanted. During crop season, necessary weeding, irrigation and other intercultural operations were done as per recommendation of the crop (Anon. 2007). The root-knot disease severity was recorded at 60 days after transplanting. Data of length and weight of shoot and root were recorded. The fruit yield was expressed in g/pot. At the end of the growing period, plants were uprooted from the pot carefully to minimize the damage of roots. The root systems were washed under running tap water and data on shoot height and fresh weight, weight of shoots and roots were recorded. The number of galls per gram of roots, and yield contributes characters was recorded. The degree of root galling was indexed on the basis of 0-10 scale (Zeck 1971), where, 0 = (Zero) represented free from gall, 1 = very few small galls can only be detected upon close examination, 2 = small galls but easily detected, 3 = numerous small galls, some grown together, but function of roots not seriously affected, 4 = numerous small galls, some big galls, majority of roots still functioning, 5 = 25% of root system severely galled and not functioning, 6 = 75% of root system severely galled and not functioning, 7 = 75% of root system severely galled and not functioning, 8 = no healthy roots, nourishment of plants interrupted, plants still green, 9 = completely galled root system is rooting, plant is dying and 10 severely galled root system. Soil sample from the pot were collected during harvesting of the crop for counting AMF spore population. Assessment of spore population was done following the wet Sieving and Decanting method (Gerdermann and Nicolson, 1963). Spores were observed under Inverted stereomicroscope, the number of spores was counted and the result was expressed as numbers per 50 g of dry soil basis. Percent root colonization was calculated as follows:

$$\% \text{ Root colonization} = \frac{\text{Number of AMF positive segments}}{\text{Total number of segments observed}} \times 100$$

RESULTS AND DISCUSSION

Efficacy of doses of AMF on the shoot character of tomato infected with *M. incognita*

The dry weight of the apical shoot of single inoculated plant was greater than that of the non-inoculated control ones (Fig. 1). Plant growth was lower in the control treatment with *M. incognita* inoculated plants but was greater in the treatment with *M. incognita* added with AM fungi. There were significant variations among the treatments on growth and yield parameters of tomato due to *M. incognita.* Shoot length ranged 21.20-50.60 cm, significantly higher shoot length was recorded by T_2 (70 g AMF spore) while statistically insignificant with all the rest treatment except T_1 (60 g AMF spore) and T_7 (Control). The maximum shoot weight was recorded in T_6 (44.33 g) followed by T_2, T_5 (100 g AMF spore), T_4 (90 g AMF spore), T_1, T_3 (80 g AMF spore) and T_7. The effect of T_6 and T_2 were statistically similar on shoot weight (Fig. 1).

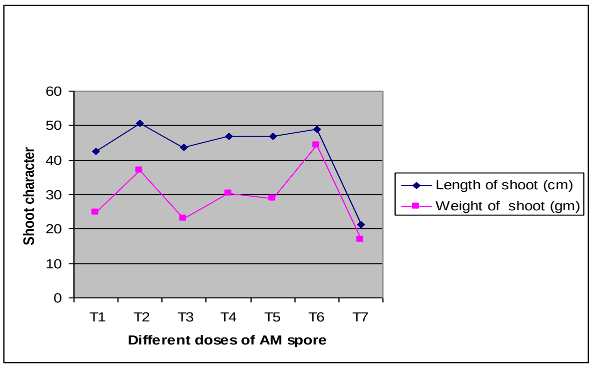

Fig. 1. Effect of doses of AMF on shoot character of tomato infected with *M. incognita.* T_2 (70 g AMF spore) and T_6 (Furadan 3G) showed higher shoot length and shoot weight. Blue line indicates shoot length (cm) and pink line indicates shoot weight (g)

Efficacy of doses of AMF on Root character of tomato infected with *M. incognita*

Plant roots were also significantly higher with AMF treatments inoculated plants than the control. In case of root length, it ranged 13.40-24.20 g. Significantly higher root length was computed in T_5 and it differed significantly with all the rest treatments. The effect of T_1, T_2, T_3, T_5 and T_6 were statistically similar on root length. Weight of root by the treatments varied 4.80-29.18 g. Although T_6 produced maximum root length but showed statistically similar to T_3, T_4 and T_5 (Fig. 2).

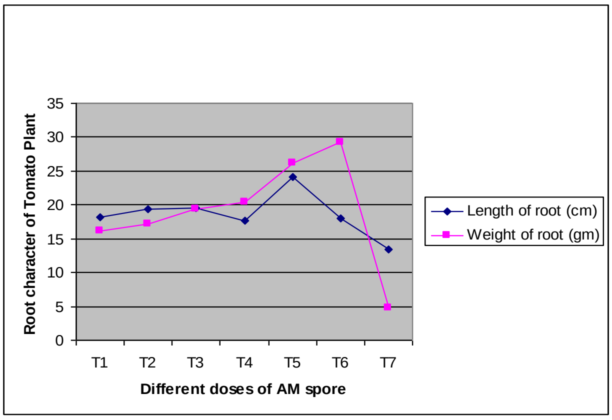

Fig. 2. Effect of doses of AMF on Root character of tomato infected with *M. incognita*. T_5 (70 g AMF spore) and T_6 (Furadan 3G) showed higher root length and T_5 also increased root weight. Blue line indicates root length (cm) and pink line indicates root weight (g)

Efficacy of doses of AMF on root infection of tomato infected with *M. incognita*

All the treatments amended with AMF proved their efficiency to establish the root infection. It was found that root infection increased with the increasing of AMF inocula concentration (Fig. 3 & 4). *M. incognita* in roots increased rapidly the control treatments. In other words, penetration of roots by *M. incognita* was lowest with the AMF and *M. incognita* added at transplanting.

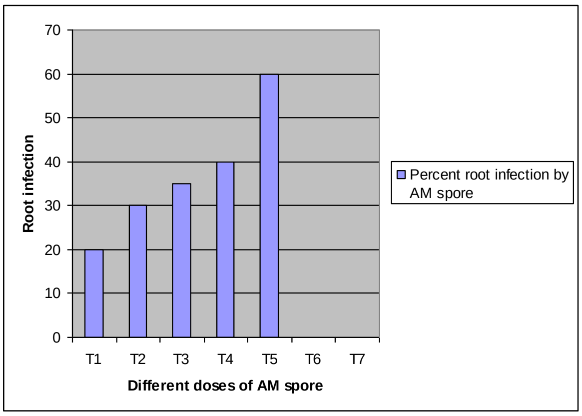

Fig. 3. Effect of doses of AMF on root infection of tomato infected with *M. incognita*. Among the different doses of AM T_1 showed lowest AMF root infection, where as T_5 gave highest infection

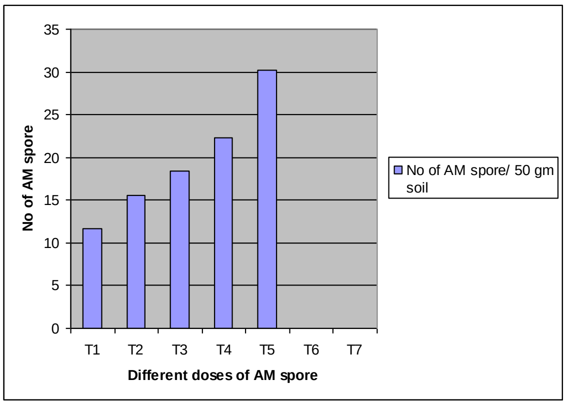

Fig. 4. Effect of doses of AMF on root colonization of tomato infected with *M. incognita*. Among the different doses of AM T_1 showed lowest AMF root infection, where as T_5 gave highest infection

Effect of doses of AMF on the Gall severity of *M. incognita* and yield of Tomato infected plant

The disease incidences and disease indexes were all significantly lower in the treatment with *M. incognita* added with AM fungi than in the control treatment with *M. incognita* (Table 1). The severity of root gall of tomato was drastically reduced over control due to treatment of soil with different doses of AMF and application of Furadan 5G. Number of gall per gram root among the treatments varied widely, ranged 39.40-162.0, respectively, with the T_5 and T_7. Significantly lower gall number per gram root was counted in T_5 and it showed statistically insignificant with T_3, T_4 and T_6. Significantly lower gall number per gram root was recorded in T_7 and it differed significantly with all the rest treatments. The minimum gall index of 2.0 at 0-10 scale was assessed in T_3, T_5 and T_6. The maximum yield was harvested in T_5 (2740 g) followed by T_6 (2520 g), T_4 (2230 g), T_3 (2180 g), T_2, (1920 g) T_1, (1760 g) and T_7 (1200 g). Statistical analysis revealed that the effect of the treatments T_4, T_5 and T_6 significantly higher yield compared to T_1, T_2 and T_7. The treatment T_7 gave lower yield which differed with all the rest treatments (Table 1).

Table 1. Effect of doses of AMF on the Gall severity of *M. incognita* and yield of Tomato infected plant

AMF inoculation	Gall number/gram roots	Gall index (0-10) scale	Yield (g)
T_1 = 60 g AMF spore	86.80 b	3.0	1760 b
T_2 = 70 g AMF spore	81.00 b	3.0	1920 b
T_3 = 80 g AMF spore	50.00 bc	2.0	2180 ab
T_4 = 90 g AMF spore	41.60 c	3.0	2230 ab
T_5 = 100 g AMF spore	39.40 c	2.0	2740 a
T_6 = Furadan 5G	55.80 bc	2.0	2520 a
T_7 = Control	162.0 a	5.0	1200 c

Values within the same column with a common letter do not differ significantly (P = 0.05)

Relationship of fruit yield with gall number and gall index

Correlation and regression analysis was performed to find out the relationship of fruit yield with gall number and gall index values of tomato grown in soil inoculated with *M. incognita* and treated with AMF and Furadan 5G (Fig. 5 & 6). The relationship was linear and negative for fruit yield with gall number and gall index values with coefficient of correlations (r) 0.915 and 0.903, respectively. The relationship was significant in case of fruit yield with gall number and influence of gall index on those two parameters may be attributed to 83.90% (R^2 = 0.839) and 81.70% (R^2 = 0.817), respectively. The results indicated that AMF improved tomato yield. It may be due to addition of plant nutrients to the soil. Their higher doses caused phytotoxicity resulting lower root weight compared to lower dose. Results of the present study reveal that AMF 100 g gave the highest yield (2740 g), and are effective to reduce root-knot severity and to increase plant growth and fruit yield of tomato grown in *M. incognita* inoculated soil. Among the treatments tested in the present study, the most effective treatment was T_5, which was followed by T_3, T_4 and T_6. Based on findings of the present investigation this treatment may be recommended for controlling root-knot of tomato.

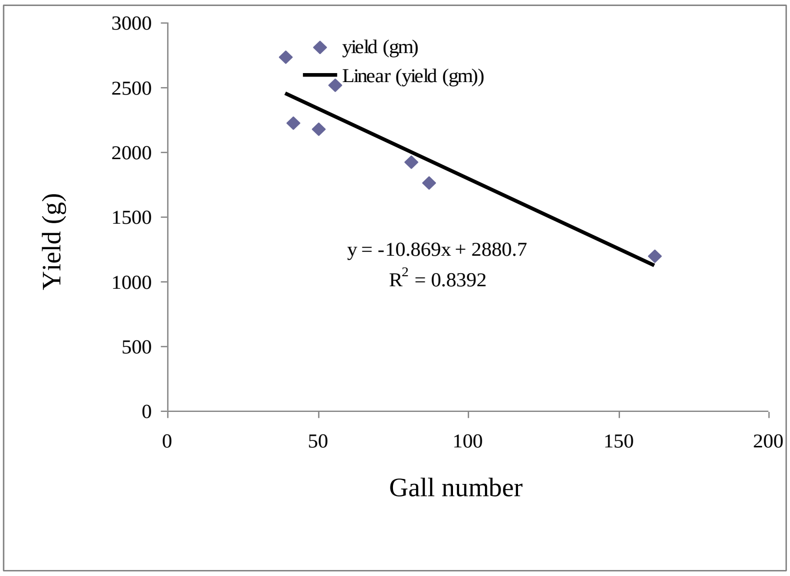

Fig. 5. Relationship of shoot fruit yield with gall number of tomato grown in soil inoculated with *M. incognita* and treated with AMF and Furadan 5G

The yield increase over control was significant under all treatments with AMF and Furadan 5G. The highest yield was obtained with T_5, which was statistically similar to T_3, T_4 and T_6. The lowest yield increase was found under T_7 followed by T_1 and T_2.

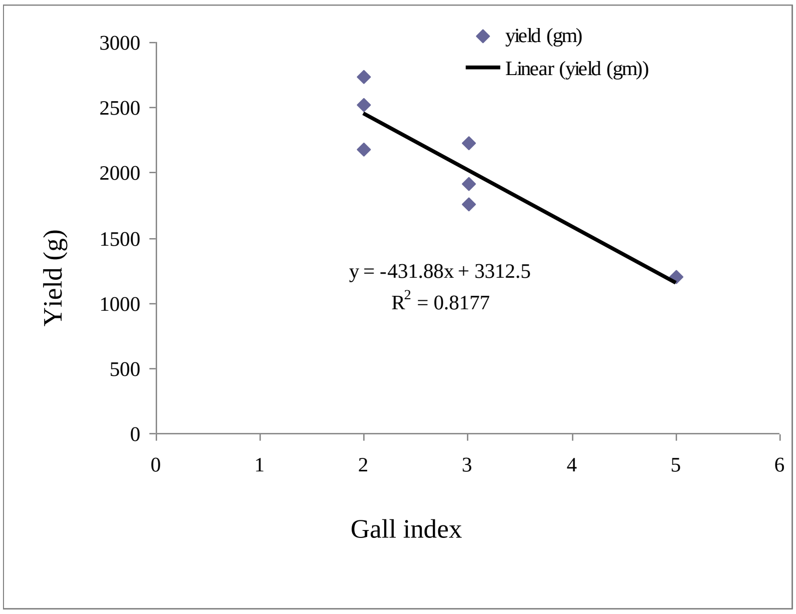

Fig. 6. Relationship of shoot fruit yield with gall index of tomato grown, in soil inoculated with *M. incognita* and treated with AMF and Furadan 5G

The yield increase over control was significant under all treatments with AMF and Furadan 5G. The highest yield was obtained with T_5, which was statistically similar to T_3, T_4 and T_6. The lowest yield increase was found under T_7 followed by T_1 and T_2.

It has been exposed that microbial inoculation as an approach of protecting plants against environmental stress and increasing the sustainability of plant production was anticipated. AM fungi are well recognized as a microorganism that can improve plant nutrition and growth and also reduce plant disease (Miroslav and Milan, 2000; Attia and Awad, 2003; Akhtar and Siddiqui, 2008; Siddiqui and Akhtar, 2009). For instance, inoculation with the AM fungus *Glomus fasciculatum* enhanced nitrogen acquisition and growth of *Medicago sativa* L. (Biró *et al*. 2000). In the present study, tomato plant growth was improved more by inoculation with AM fungi inoculations. Meyer and Linderman (1986) found that plant growth and nodulation of subterranean clover were enhanced by indigenous AM fungi.

However, root colonization by AM fungi, had a negative effect on the development of *M. incognita*. Nematode population build up decreased with time as indicated by the differences in nematode development in the roots for the different treatments. So, it may be concluded that nematode penetration of roots, nematode reproduction, and nematode-incited disease was decreased more by dual inoculations with AMF with *G. mosseae* gave the best results. This may be due to AMF strong induction of systemic resistance in plants towards nematodes (Elsen *et al*. 2008), directly affect the plant metabolism or affect the plant by "helping" another beneficial microorganism to function better (Shreenivasa *et al*. 2007; Bashan and Holguin, 1998; Linderman 1988). However, the mechanisms of nematode suppression in the roots are unknown but would seem to be related to physiological changes in roots affecting nematode food source or feeding rather than a direct competition for space. Peña *et al*. (2006) even considered that nematode suppression by AM fungi did not occur through a systemic plant response but through local mechanisms. Moreover, this beneficial fungus can be used without problems in combination with AMF in some instances even with increasing positive effects on the host plant.

CONCLUSION

In conclusion, the trial showed that the efficacy of AMF in controlling root-knot disease and growth enhancement in tomato. Thus, application of AMF could be used to manage the pest in a more sustainable and eco-friendly way.

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