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Original Article

Assessment of Arbuscular Mycorrhizal Association in Some Fruit Plants in Bangladesh

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Rhizosphere soils of 19 fruit plants from horticultural farm of Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur were assessed for arbuscular mycorrhiza (AM) spore population and determining colonization in their roots. The spore numbers recorded ranged from 48.0 (*Citrus limon*) to 1,050 (*Annona reticulata*) per 100 g soil in 2004, 41.0 (*Citrus grandis*) to 916.7 (*Phyllanthus emblica*) in 2005 and 44.3 (*Citrus grandis*) to 575.0 (*Syzygium samarangense*, white) in 2006. A considerable variation was observed in average spore numbers recorded in three consecutive years. Different fruit plants showed different percentages of root colonization by AM fungi. Among all the fruit plants, the highest colonization (86.7%) was found in *Syzygium jambos* and *Elaeocarpus floribundus* and the lowest colonization (20.0%) was recorded in *Syzygium samarangense* (red).

Keywords: Arbuscular mycorrhiza (AM), Colonization, Spore population, Fruit plants

Introduction

Mycorrhizae are symbiotic association between beneficial soil fungi and plant roots. They have an important role in increasing plant uptake of P and other poorly mobile micronutrients particularly Zn and Cu[1]. Out of the different types of mycorrhizae, the arbuscular mycorrhiza (AM) fungi are the most widely occurring mycorrhizae and are very important in relation to the improvement of agricultural and horticultural crops and forest trees in hilly areas[2]. They form three-way associations involving plants, fungi and soils. Wide diversity exists within the group of fungi responsible for the formation of AM by most plants in the majority of terrestrial ecosystems. The selection of the most specific appropriate plant-fungus association for each specific environmental and ecological situation is one of the main challenges in current research on AM. Therefore, knowledge of the different factors influencing the population biology of AM fungi is essential in any attempt to use them in environmental conservation[3], biotechnology[4] or in sustainable agriculture[5].

The identification of indigenous AM fungi is a fundamental requirement to understand biodiversity and essential for monitoring changes in natural, managed or disturbed ecosystems. Diversity in AM fungi can be explored at this level by studying spore characteristics, ultra-structural features and infection patterns in different agricultural crops including different varieties of the same crop. The AM fungi are abundant and ecologically very important in the tropics[6] and have been recognized as a promising alternative technology for reducing fertilizer requirement of the major crop species[6-7]. Soils in the tropics are either poor in phosphorus (P) and other essential nutrients or have an immobile form of P[8]. Hence AM fungal inoculum could be added to the soil for better uptake of P to enhance crop production in tropical countries. Bangladesh produces a variety of fruits.

It seems that there is an important role of arbuscular mycorrhizal fungi in nutrient availability for these fruit plants. But still no work has been done to assess the mycorrhizal association with different fruit plants. So, this present work was taken to know the percent root colonization of fruit plants and the number of AM spores in the rhizosphere soils for producing suitable inoculum for future use in different fruit crops.

Materials and Methods

Nineteen fruit plants from Horticultural farm of Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur were selected for the assessment of arbuscular mycorrhizal association. Rhizosphere soils were collected in three consecutive years (2004-2006) from the same plants and places. Assessment of spore population was done by following the wet sieving and decanting method[9]. Spores were observed under stereomicroscope and the number of spores was counted. Spore numbers from the three replicates per sample were averaged and the result was expressed as number per 100 g of dry soil basis.

The root pieces of each plant species were stained according to Koske and Gemma[10]. The root pieces were boiled in 2.5% KOH solution for 30 min at 90°C. Then, the root segments were washed in water several times and acidified with 1% HCl solution for 24 h.

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Heavily pigmented roots were bleached in 10% H_2O_2 for 20 to 60 min. Again these segments were boiled for 30 min in 0.05% aniline blue at a temperature of 90°C. Subsequently, the stained roots were destained at room temperature using acidic glycerol.

Root colonization (%) of arbuscular mycorrhiza (AM) was estimated by root slide technique[11]. One hundred root segments were examined for each sample. The stained root pieces were mounted in acidic glycerol on slides covered with cover slips that were slightly pressed. The stained roots were observed under a microscope. A root segment was considered as positively infected, if it showed mycelium, vesicles and arbuscules, or any other combination of these structural characteristics of AM colonization. The presence or absence of colonization in the root pieces was recorded and the percent of root colonization was calculated as follows:

$$\textit{Root colonization (\%)} = \frac{\textit{No. of AM positive segments}}{\textit{Total No. of segments scored}} \times 100$$

Physical and chemical properties were determined of a composite soil sample following ASI method[12].

Results and Discussion

Association between arbuscular mycorrhiza (AM) fungi and root of agricultural or horticultural plants is very important[2]. In this study, a horticultural farm of BARI, Gazipur was selected for the assessment of arbuscular mycorrhizal association with some fruit plants. The physicochemical properties of the farm are summarized in Table 1.

Table 1. *Physical and chemical properties of soil of horticultural farm*

Soil variable	Content	Critical level
pH	5.7	-
Organic matter (%)	2.34	-
Total N (%)	0.124	-
Available P (ppm)	73	14
Available S (ppm)	35	14
Exchangeable K (meq/100ml)	0.32	0.2
Exchangeable Ca (meq/100ml)	4.1	2
Exchangeable Mg (meq/100ml)	1.3	0.8
Available Zn (ppm)	2.4	2
Available Cu (ppm)	2.9	1
Available Fe (ppm)	427	10
Available Mn (ppm)	40	5

Spore population of arbuscular mycorrhiza (AM) in the rhizosphere of some fruit plants was assessed in three consecutive years. A considerable variation was observed in average spore number recorded in three consecutive years (Figure 1). The average spore numbers per 100 g soil of three years were 397.7 in the year 2004, 321.3 in the year 2005 and 139.5 in the year 2006. The spore density decreased abruptly in 2006 as compared to that in the year 2004 and 2005. This change might be due to climatic variation from year to year that also led to the changes of some edaphic factors critical for distribution and abundance of AM fungi[13-15].

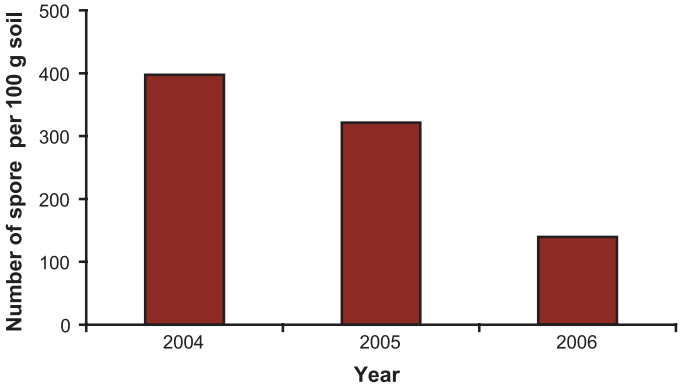

Figure 1. *Variation of number of arbuscular mycorrhiza (AM) spores in the rhizosphere soils of different fruit plants in three consecutive years.*

Table 2 shows the spore numbers associated with rhizosphere soils of different fruit plants. The spore counts of 8 plants were high (>100/g soil) in the three consecutive years. These included *Annona reticulata*, *Averrhoa carambola*, *Carissa congesta*, *Litchi chinensis*, *Manilkara zapota*, *Spondias mangifera*, *Syzygium jambos* and *Syzygium samarangense* (white). Very high (>500/g soil) count was observed in the rhizosphere soil of *Annona reticulata*, *Averrhoa carambola*, *Phyllanthus emblica*, *Psidium guajava* and *Zizyphus mauritiana* in the year 2004, *Mangifera indica*, *Phyllanthus emblica*, *Syzygium jambos* and *Syzygium samarangense* (white) in the year 2005, and *Syzygium samarangense* (white) in the year 2006. The counts were low (<100/g soil) in the three consecutive years in case of *Citrus grandis* and *Syzygium samarangense* (red).

In present study, the spore density in the rhizosphere soil varied in different fruit plants, which was supported by Howeler *et al*.[16] who reported that the intensity of spore density varied on different factors like plant species and genera and nature of rhizosphere soil. Moreover, higher spore population was observed in some fruit plants. The stimulating effects of organic matter, comparatively high level of N and P might have created a favourable condition for maximum sporulation of AM fungi in that particular field.

Table 3 represents the percent root colonization of different fruit plants. Different fruit plants showed different degrees of root colonization by AM fungi. The percent root colonization varied from 16.7 to 86.7%. More than 50% colonization was recorded with *Carissa congesta*, *Citrus grandis*, *Diospyros discolor*, *Elaeocarpus floribundus*, *Litchi chinensis*, *Psidium guajava*, *Spondias mangifera*, *Syzygium jambos*, *Tamarindus indica* and *Zizyphus mauritiana*. Among all the fruit plants, the highest colonization (86.7%) was found in *Elaeocarpus floribundus* and *Syzygium jambos*. The lowest colonization (16.7%) was recorded in white *Carissa congesta*. The variation observed in the colonization of AM fungi among different fruit plants might be due to the differences in the structure of root system, phosphorus uptake[17] and also might be genetical variations[18].

Table 2. *Spore population of arbuscular mycorrhizae in rhizosphere soil of different fruit plants during 2004-2006*

Fruit plant	Spore number per 100 g soil[a]		
(Local name / English name)	2004	2005	2006
Annona reticulata (Ata / Bullock's heart)	1,050.0 ± 50.0	270.0 ± 15.3	105.3 ± 7.5
Artocarpus heterophyllus (Kathal / Jackfruit)	448.0 ± 68.0	270.0 ± 15.3	89.3 ± 8.7
Averrhoa carambola (Kamranga / Carambola)	527. 0 ± 25.0	306.7 ± 23.3	127.7 ± 14.7
Carissa congesta (Karomcha / Bengal currant)	483.0 ± 7.3	225.0 ± 14.4	220.0 ± 16.1
Citrus grandis (Batabi lebu / Shaddock)	62.0 ± 13.0	41.7 ± 6.0	44.3 ± 3.5
Citrus limon (Lebu / Lemon)	48.0 ± 18.0	116.7 ± 20.3	53.3 ± 3.5
Diospyros discolor (Bilati gab / Velvet apple)	55.0 ± 5.0	270.0 ± 15.3	62.0 ± 7.2
Elaeocarpus floribundus (Jalpai / Indian olive)	163.0 ± 47.0	85.0 ± 10.4	194.0 ± 8.3
Litchi chinensis (Lichu / Litchi)	480.0 ± 72.0	433.0 ± 44.1	271.7 ± 14.8
Mangifera indica (Aam / Mango)	102.0 ± 18.0	697.3 ± 57.8	53.3 ± 3.5
Manilkara zapota (Shafata / Sapota)	177.0 ± 25.0	295.0 ± 13.2	96.7 ± 8.8
Phyllanthus emblica (Amloki / Aonla)	950.0 ± 50.0	916.7 ± 44.1	66.7 ± 6.0
Psidium guajava (Payara / Guava)	787.0 ± 13.0	83.3 ± 16.9	89.0 ± 6.7
Spondias mangifera (Amra / Hog plum)	250.0 ± 50.0	225.0 ± 14.4	105.3 ± 7.5
Syzygium jambos (Golapjam / Rose apple)	185.0 ± 7.6	856.7 ± 34.8	169.0 ± 9.7
Syzygium samarangense (Jamrul / Wax jambu, red)	74.0 ± 7.0	91.7 ± 6.0	54.0 ± 4.6
Syzygium samarangense (Jamrul / Wax jambu, white)	497.0 ± 25.0	650.0 ± 32.2	575.0 ± 13.2
Tamarindus indica (Tetul / Tamarind)	467.0 ± 35.0	191.7 ± 6.0	86.3 ± 8.8
Zizyphus mauritiana (Deshi kul / Jujube	752.0 ± 48.0	80.0 ± 2.9	188.3 ± 6.0

[a]Spore numbers are the means ± SE of three independent counts.

Table 3. *Root colonization (%) of arbuscular mycorrhiza (AM) fungi in different fruit plants*

Fruit plant	Root	AM structure			
(Local name / English name)	colonization (%)	Hypae	Arbuscle	Vesicle	Vesicle shape
Annona reticulata (Ata / Bullock's heart)	40.0 ± 5.8	+	+	-	-
Artocarpus heterophyllus (Kathal / Jackfruit)	56.7 ± 3.3	+	-	-	-
Averrhoa carambola (Kamranga / Carambola)	16.7 ± 3.3	+	-	-	-
Carissa congesta (Karomcha / Bengal currant)	66.7 ± 3.3	+	+	-	-
Citrus grandis (Batabi Lebu / Shaddock)	76.7 ± 3.3	+	+	+	Spherical
Citrus limon (Lebu / Lemon)	23.3 ± 3.3	+	-	-	-
Diospyros discolor (Bilati gab / Velvet apple)	60.0 ± 5.8	+	-	+	Oval
Elaeocarpus floribundus (Jalpai / Indian olive)	86.7 ± 3.3	-	+	+	Oval
Litchi chinensis (Lichu / Litchi)	60.0 ± 5.8	+	+	-	-
Mangifera indica (Aam / Mango)	30.0 ± 5.8	+	-	-	-
Manilkara zapota (Shafata / Sapota)	43.3 ± 3.3	+	-	-	-
Phyllanthus emblica (Amloki / Aonla)	40.0 ± 5.8	+	-	-	-
Psidium guajava (Payara / Guava)	26.7 ± 3.3	+	-	-	-
Spondias mangifera (Amra / Hog plum)	66.7 ± 3.3	+	+	+	Spherical
Syzygium jambos (Golapjam / Rose apple)	86.7 ± 3.3	+	-	-	Oval
Syzygium samarangense (Jamrul / Wax jambu, red)	30.0 ± 5.8	+	-	-	-
Syzygium samarangense (Jamrul / Wax jambu, white)	20.0 ± 5.8	+	-	-	-
Tamarindus indica (Tetul / Tamarind)	73.3 ± 3.3	-	+	+	Oval
Zizyphus mauritiana (Deshi kul / Jujube	60.0 ± 5.8	-	+	+	Spherical

[a]Percent root colonization are the means ± SE of three independent counts.

The AM fungal structure in the root system of the selected fruit plants varied irrespective of fruit species (Table 3). Out of 19 plants only 7 had vesicles. Hyphae were not present in *Elaeocarpus floribundus*, *Syzygium jambos*, *Tamarindus indica* and *Zizyphus mauritiana*. Arbuscules were found in 9 plants. Both oval and spherical vesicles were found in this study, which was supported, by Muthukumar *et al*.[13] and Khanam *et al.*[14-15].

Spore number and root colonization varied from plant to plant in the present study. But variations in spore numbers in different plants were not related to per cent root colonization which is in agreement with Khalil *et al*.[19]. As a wide range of host, fungal and environmental factors are known to influence AM formation and subsequent spore production; these two phenomena may not necessarily be related.

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