

# Identification and characterization of arbuscular mycorrhizal strains from the roots and rhizospheres of tomato, brinjal, chilli and onion

## ABSTRACT

Rhizosphere soils and roots of two vegetables (tomato and brinjal) and two spices (chilli and onion) crops were collected from each of four Agro Ecological Zones (AEZs) of Bangladesh viz. AEZ-9 (RARS, Jamalpur), AEZ-11 (RARS, Jashore), AEZ-25 (ARS, Bogura) and AEZ-28 (BARI, Joydebpur) in 2014-2015. Characterization and identification of arbuscular mycorrhizal strains was studied in the microbiology laboratory of Bangladesh Agricultural Research Institute (BARI). Different crops showed positive responses in per cent root colonization such as 74.11-83.78%, 22.11-24.33%, 7.33-8.89% and 6.67-9.33% in onion, tomato, chilli and brinjal, respectively. Rhizosphere soil of individual crops had also variation in number of spore population such as 120.11-200.33, 33.44-50.78, 36.11-39.22 and 28.56-41.89 per 100g soil in onion, tomato, chilli and brinjal, respectively. The formation of AM structures were found inconsistent and fluctuating from site to site. Both oval and spherical shaped vesicles were found among the AM structures. Eleven AM strains were identified in tomato: *Glomus geosporum*, *G. mosseae*, *G. fasciculatum*, *G. spp.*, *Acaulospora dilatata*, *A. bireticulata*, *A. mellea*, *A. morrowiae*, *Entrophospora infrequens*, *Sclerocystis coremioides* and *Gigaspora margarita*. Nine each were identified from other three crops. *G. spp.* and *Acaulospora bireticulata* were absent in brinjal and chilli. *Acaulospora dilatata* and *A. mellea* were absent in onion. The formation of AM structures found inconsistent and fluctuating from site to site in the present study. Both oval and spherical shaped vesicles were found among the AM structures.

**Key words:** Arbuscular mycorrhizal (AM) strains, root colonization, spore population, AM structures

## 1.Introduction

Arbuscular mycorrhizal fungi are obligatory biotrophic symbionts occurring in nearly all natural and agricultural soils and commonly colonize roots of many plant species [1]. Arbuscular mycorrhizae are most important type of fungi and have worldwide recognition for their role in plant survival and nutrient cycling in the ecosystem. Mycorrhizal fungi are well known to have a wide range of benefits to their host plants. They can enhance nutrient uptake especially P, N and Zn. They can also suppress soil pathogens, enhance tolerance to drought stress and reduce sensitivity to toxic substances contaminated to the soil [2]. Arbuscular mycorrhizal fungi have received considerable attention in the literature due to their potential benefits to host plants by enhancing plant nutrient uptake and increasing tolerance to adverse conditions [1].

[3] reported an average of 35.99% mycorrhiza colonization of tomato roots in soils. The distribution of different genera in various locations and crop species varied to a greater extent. *Glomus* species were reported to be most common throughout the world [4]. [5] worked with different agricultural crops and they identified *Acaulospora*, *Entrophospora*, *Gigaspora*, *Glomus* and *Scutellospora*. *Glomus* species were the most common followed by *Gigaspora* and *Scutellospora* in vegetables and rice. [6] studied the genus diversity of AM fungi in some vegetable crops in Bangladesh.

[7] studied the effects of arbuscular mycorrhizal fungi on the morphological and biochemical changes of four different vegetable seedlings such as tomato (*Lycopersicon esculentum* L.), brinjal (*Solanum melongena* L.), chilli (*Capsicum annum* L.) and bhendhi (*Abelmoschus esculentus* Moench.) grown under nursery conditions. They revealed the symbiotic association

between AM fungi and plant roots provides a significant contribution to plant nutrition and growth.

Arbuscular mycorrhizal fungi improved the absorption of several plant nutrients like N, P, K, Mg, Cu, Ca and Fe by the roots of plants [8]. The AM fungi can increase plant uptake of nutrients and consequently increase root and shoot biomass and improve plant growth and yield [9]. The mycorrhizal infection enhances nutrient uptake through increasing the absorbing area of root and by mobilizing sparingly available nutrient sources; or by excretion of chelating compounds or ecto enzymes [10].

Arbuscular mycorrhizal fungi inoculation produced significantly higher plant growth compared to the untreated control pepper [11], onions [12], and vegetable crops [13]. Bioaugmentation with native arbuscular mycorrhiza fungi was reported to improve the qualities of seedlings in nurseries [14]. Considering the above facts the present study was undertaken to characterize and identify the arbuscular mycorrhizal strains from the roots and rhizosphere soils of tomato, brinjal, chilli and onion.

## 2. Materials and methods

### 2.1 Sample collection sites

For conducting the study the soil and root samples were collected from four different sites situated in four Agro Ecological Zones (AEZs) of Bangladesh. The selected sites were: RARS, Jamalpur (AEZ 9 Old Brahmaputra Floodplain), RARS, Jashore (AEZ 11 High Ganges River Floodplain), ARS, Bogura (AEZ 25 Level Barind Tract), BARI, Joydebpur (AEZ 28 Madhupur Tract)

### 2.2 Selection of crops

Studies included a number of crops under vegetable and spices crops such as Tomato (*Solanum lycopersicum* L.), Brinjal (*Solanum melongena* L.), Chilli (*Capsicum annuum* L) of Solanaceae family and Onion (*Allium cepa* L.) of Liliaceae family.

### 2.3 Collection of samples

Roots and rhizosphere soil samples were collected from crop fields of four AEZs during December 2014 to February 2015. Four plants for each crop were sampled. Plant roots were dug out, washed thoroughly with water to remove the adhering soil particles and then cut into approximately 1 cm segments. The root samples were then preserved in screw cap test tubes with 50% ethanol for future use. Soil samples were collected up to a depth of 0-15 cm with roots (4 samples/crop) thereafter the collected samples were air-dried, packed in airtight polyethylene bags and stored at 4°C for assessing spore density.

### 2.4 Analysis of soil physico-chemical properties

The chemical and physical properties of soil were determined by following methods. such as pH [15], Texture [16], wet oxidation method [17], organic carbon with the van Bemmelen factor 1.73 [18], Total N [19], Available P [20], Exchangeable Ca, Mg & K [21], Available S [22]; [23], Available Zn, Cu, Mn & Fe [24] Available B [25].

#### Physical and chemical properties of soils under study

AEZ and Locations	Texture	pH	OM (%)	Ca	Mg	K	Total N (%)	P	S	Zn	B	Fe	Cu	Mn
				meq/100 g soil										

AEZ-09 (Jamalpur)	Sandy Clay loam	7.1	1.63	6.0	2.4	0.16	0.086	10	17	1.19	0.28	44	1.6	3.1
AEZ-11 (Jashore)	Clay loam	7.4	1.42	6.4	2.1	0.16	0.075	14	15	1.71	0.35	30	2.4	2.3
AEZ-25 (Bogura)	Silty clay loam	6.3	1.42	6.8	2.3	0.13	0.075	22	19	1.64	0.30	36	2.2	4.1
AEZ-28 (Joydebpur)	Clay loam	6.5	0.87	4.8	1.7	0.20	0.042	11	12	0.90	0.30	51	1.7	2.8

## 2.5 Determination of mycorrhizal colonization

### 2.5.1 Staining of roots

The root pieces of each crop were stained according to [26] with some modifications followed in the Soil Microbiology Laboratory of Bangladesh Agricultural Research Institute [27].

### 2.5.2 Assessment of mycorrhizal colonization

The percentage of AM colonization was estimated by root slide technique [28].

The presence or absence of colonization in the root pieces was recorded and the per cent colonization was calculated as follows:

$$\% \text{ root colonization} = \frac{\text{Number of AM positive segments}}{\text{Total number of segments scored}} \times 100$$

## 2.6 Identification of AM fungi

For identification of fungi Melzer's Reagents and Polyvinyl Alcohol-Lacto-Glycerol were needed. The preparation of these reagents is given below:

**2.6.1 Melzer's Reagents:** Potassium Iodide (1.5 g), Iodine (0.5 g), Chloral hydrate (100.0 g), Distilled water (22 mL).

**2.6.2 Polyvinyl Alcohol-Lacto-Glycerol (PVLG):** Polyvinyl alcohol (1.66g), Water (10mL), Lactic acid(10 mL), Glycerine(1 mL).

The slides with spores were observed under a compound microscope for identification. The spores were then identified into different genera according to the sporocarpic and spore characters such as spore size, colour, spore walls, hyphal attachments and other morphological characters [29]; [30].

## 3. Results and Discussion

### 3.1 Root colonization, spore population in rhizosphere soils and occurrence of AM structure in different crops of AEZ 9

Data were presented as per cent root colonization and number of spores/100g soil, of AM structure of different crops such as tomato, brinjal, chilli and onion in AEZ 9 (RARS, Jamalpur

(Table 1). Per cent root colonization of different crops varied from 7.3-83.8% where the highest root colonization (83.8%) was recorded in onion which was followed by tomato (24.1%) and brinjal (8.11%) and the lowest root colonization was recorded in chilli (7.3%).

The spore numbers in the rhizosphere soil varied in different crops. The number of AM spores varied from 36.7-180.0/100g soil. The highest number of AM spores per 100g soil was recorded from the rhizosphere soil of onion (180.0) and the lowest in chilli (36.7). The rhizosphere soil of tomato and brinjal contained 43.1 and 38.6 spores per 100 g soil, respectively.

The AM fungal structures in root systems of the selected crops varied irrespective of crops. Out of 4 crops, hyphae were found in all crops at AEZ 9 (RARS, Jamalpur). Arbuscules were found in chilli and onion, vesicles in tomato and onion which showed both oval and spherical shapes.

**Table 1. Occurrence of AM fungi in tomato, brinjal, chilli and onion crops grown under AEZ 9 (RARS, Jamalpur)**

Crops	Root colonization (%) <sup>a</sup>	Number of spore/100g soil <sup>b</sup>	AM structure			
			H	A	V	VS
Tomato	24.11 ± 0.61	43.11 ± 1.14	+	-	+	-
Brinjal	8.11 ± 0.59	38.56 ± 0.85	+	-	-	-
Chilli	7.33 ± 0.47	36.67 ± 0.88	+	+	-	-
Onion	83.78 ± 0.70	180.00 ± 1.28	+	+	+	O,S

<sup>a</sup>Per cent root colonization are the means ± S.E of eight independent observations

<sup>b</sup>Spore number are the means ± S.E of eight independent counts.

<sup>c</sup>H=Hyphae, A=Arbuscule, V=Vesicle, VS=Vesicle shape, O=Oval, S=Spherical  
+ = Present, - = Absent

### 3.2 Root colonization, spore population and M structure in different crops of AEZ 11

Per cent root colonization of different crops varied from 8.89-82.00% (Table 2). The highest root colonization by AM fungi was observed in onion (82.00%) which was followed by tomato (22.11%) and brinjal (9.33%). The lowest per cent of root colonization was observed in chilli (8.89%). The number of AM spores ranged from 33.44 to 174.00 per 100 g soil (Table 2). The highest number of spores per 100g soil was found in rhizosphere soil of onion (174.00) and the lowest in tomato (33.44). The rhizosphere soils of brinjal and chilli had 41.89 and 38.33 spores per 100 g soil, respectively. Out of 4 crops, hyphae were observed in all crops at AEZ 11 (RARS, Jashore). Both arbuscules and vesicles were found in tomato and onion. Again both oval and spherical shaped vesicles were found in onion (Table 2).

**Table 2. Occurrence of AM fungi in tomato, brinjal, chilli and onion crops grown under AEZ-11 (RARS, Jashore)**

Crops	Root colonization	Number of	AM structure <sup>c</sup>
-------	-------------------	-----------	---------------------------

	(%) <sup>a</sup>	spore/100g soil <sup>b</sup>	H	A	V	VS
Tomato	22.11 ± 1.21	33.44 ± 1.52	+	+	+	-
Brinjal	9.33 ± 0.83	41.89 ± 1.81	+	-	-	-
Chilli	8.89 ± 0.79	38.33 ± 1.41	+	-	-	-
Onion	82.00 ± 1.19	174.00 ± 1.17	+	+	+	O,S

<sup>a</sup>Per cent root colonization are the means ± S.E of eight independent observations

<sup>b</sup>Spore number are the means ± S.E of eight independent counts.

<sup>c</sup>H=Hyphae, A=Arbuscule, V=Vesicle, VS=Vesicle shape, O=Oval, S=Spherical

+ = Present, - = Absent

### 3.3 Root colonization, spore population in rhizosphere soil, AM structure in different crops of AEZ 25

The crops showed different percentage of root colonization by AM fungi. Per cent root colonization differed from 7.78 to 78.56%. The highest root colonization (78.56%) was recorded in onion and the lowest in brinjal (7.78%). Root colonization in tomato and chilli was recorded as 23.22 and 8.56%, respectively (Table 3).

The spore number in rhizosphere soils varied in different crops (Table 3). The number of AM spores varied from 38.56 to 200.33 per 100 g soil. The highest number of spores was recorded from rhizosphere soil of onion (200.33 per 100 g soil) and the lowest in brinjal (38.56 per 100 g soil). The rhizosphere soil of tomato and chilli had 42.11 and 39.22 spores per 100 g soil, respectively. Hyphae were observed in all crops at AEZ 25 (ARS, Bogura). Arbuscules were found in tomato, chilli and onion but vesicles were found in tomato and onion. Both oval and spherical shaped vesicles were recorded in onion (Table 3).

**Table 3. Occurrence of AM fungi in tomato, brinjal, chilli and onion crops grown under AEZ 25 (ARS, Bogura)**

Crops	Root colonization (%) <sup>a</sup>	Number of spore/100g soil <sup>b</sup>	AM structure <sup>c</sup>			
			H	A	V	VS
Tomato	23.22 ± 1.15	42.11 ± 1.84	+	+	+	-
Brinjal	7.78 ± 0.46	38.56 ± 1.68	+	-	-	-
Chilli	8.56 ± 0.71	39.22 ± 3.83	+	+	-	-
Onion	78.56 ± 1.12	200.33 ± 1.39	+	+	+	O,S

<sup>a</sup>Per cent root colonization are the means ± S.E of eight independent observations

<sup>b</sup>Spore number are the means ± S.E of eight independent counts.

<sup>c</sup>H=Hyphae, A=Arbuscule, V=Vesicle, VS=Vesicle shape, O=Oval, S=Spherical

+ = Present, - = Absent

### 3.4 Root colonization, spore population in rhizosphere and AM structure in different crops of AEZ 28

Different crops showed different percentage of root colonization by AM fungi varied from 6.67-74.11% (Table -4). The highest root colonization (74.11%) was recorded in onion and the lowest in brinjal (6.67%). Root colonization of tomato and chilli was recorded as 24.33% and 7.33%, respectively.

The number of AM spores varied from 36.11-120.11 per 100 g soil. The highest number of spores (120.11) per 100g soil was recorded from the rhizosphere soil of onion and the lowest

from brinjal (28.56). The rhizosphere soils of tomato and chilli had 50.78 and 36.11 spores per 100 g soil, respectively.

Out of 4 crops, hyphae were observed in all crops. Arbuscules were observed in tomato and onion whereas vesicles were recorded only in onion. Both spherical and oval shaped vesicles were found in onion (Table 4).

**Table 4. Occurrence of AM fungi in tomato, brinjal, chilli and onion crops grown under AEZ 28 (BARI, Joydebpur)**

Crops	Root colonization (%) <sup>a</sup>	Number of spore/100g soil <sup>b</sup>	AM structure <sup>c</sup>			
			H	A	V	VS
Tomato	24.33 ± 0.89	50.78 ± 1.35	+	+	-	-
Brinjal	6.67 ± 0.60	28.56 ± 1.25	+	-	-	-
Chilli	7.33 ± 0.78	36.11 ± 0.98	+	-	-	-
Onion	74.11 ± 1.09	120.11 ± 1.56	+	+	+	O,S

<sup>a</sup>Per cent root colonization are the means ± S.E of eight independent observations

<sup>b</sup>Spore number are the means ± S.E of eight independent counts.

<sup>c</sup>H=Hyphae, A=Arbuscule, V=Vesicle, VS=Vesicle shape, O=Oval, S=Spherical  
+ = Present, - = Absent

### 3.5 Identification of AM species

#### 3.5.1 Occurrence of AM species in AEZ-9 soils

All the eleven AM fungi species identified in the rhizosphere soils of Jamalpur (AEZ 9) (Table 5). Here *G. geosporum*, *G. mosseae*, *G. spp.* and *Gigaspora margarita* were present in tomato rhizosphere, *G. fasciculatum*, *A. dilatata*, and *A. morrowiae* in brinjal rhizosphere, *A. mellea*, *A. morrowiae* and *Sclerocystis coremioides* in chilli rhizosphere and *Acaulospora bireticulata*, *Entrophospora infrequens* and *Gigaspora margarita* in onion rhizosphere. It was observed that none of the AM fungi was common in all the four crops except *A. morrowiae* in brinjal and chilli rhizosphere and *Gigaspora margarita* in tomato and onion rhizosphere (Table 5).

**Table 5. Occurrence of AM fungal strains in the rhizospheres of tomato, brinjal, chilli and onion from AEZ 9 soils**

Sl. No.	Fungal strains	Crops															
		Tomato				Brinjal				Chilli				Onion			
		F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>
1	<i>Glomus geosporum</i>	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	<i>G. mosseae</i>	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3	<i>G. fasciculatum</i>	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
4	<i>G. Spp.</i>	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-
5	<i>Acaulospora dilatata</i>	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
6	<i>A. bireticulata</i>	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-

7	<i>A. mellea</i>	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
8	<i>A. morrowiae</i>	-	-	-	-	-	-	+	-	-	-	+	-	-	-	-
9	<i>Entrophospora infrequens</i>	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-
10	<i>Sclerocystis coremioides</i>	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
11	<i>Gigaspora margarita</i>	-	-	-	+	-	-	-	-	-	-	-	-	-	-	+

+ = Present, - = Absent, F<sub>1</sub> -- Field 1, F<sub>2</sub> -- Field 2, F<sub>3</sub> -- Field 3, F<sub>4</sub> -- Field -4

### 3.5.2 Occurrence of AM species in AEZ 11 soils

Six species were identified in the Jashore location (AEZ 11) (Table 6). It was observed that only one species *Entrophospora infrequens* was present in tomato rhizosphere but the rest three crops had two species each such as *A. dilatata* and *Entrophospora infrequens* in brinjal rhizosphere, *G. geosporum* and *G. mosseae* in chilli rhizosphere and *G. fasciculatum* and *G. spp.* in onion rhizosphere (Table 6).

**Table 6. Occurrence of AM fungal strains in the rhizospheres of tomato, brinjal, chilli and onion from AEZ 11 soils**

Sl. No.	Fungal strains	Crops															
		Tomato				Brinjal				Chilli				Onion			
		F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>
1	<i>Glomus geosporum</i>	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-
2	<i>G. mosseae</i>	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
3	<i>G. fasciculatum</i>	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-
4	<i>G. Spp.</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-
5	<i>Acaulospora dilatata</i>	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
6	<i>Entrophospora infrequens</i>	+	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-

+ = Present, - = Absent, F<sub>1</sub>- Field 1, F<sub>2</sub>- Field 2, F<sub>3</sub>- Field 3, F<sub>4</sub>- Field -4

### 3.5.3 Occurrence of AM species in AEZ 25 soils

Both tomato and brinjal rhizospheres had four species (Table 7). *Entrophospora infrequens* and *Gigaspora margarita* were found in chilli rhizosphere and only *G. spp.* was identified from onion rhizosphere. Here it was observed that none of the strains were common in all crops except *Gigaspora margarita* in brinjal and chilli rhizosphere.

**Table 7. Occurrence of AM fungal strains in the rhizospheres of tomato, brinjal, chilli and onion from AEZ 25 soils**

Sl. No.	Fungal strains	Crops															
		Tomato				Brinjal				Chilli				Onion			
		F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>
1	<i>Glomus geosporum</i>	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
2	<i>G. mosseae</i>	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
3	<i>G. fasciculatum</i>	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
4	<i>G. Spp.</i>	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-
5	<i>Acaulospora dilatata</i>	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	<i>A. mellea</i>	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-
7	<i>A. morrowiae</i>	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
8	<i>Entrophospora infrequens</i>	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-

9	<i>Sclerocystis coremioides</i>	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-
10	<i>Gigaspora margarita</i>	-	-	-	-	-	-	-	+	-	+	-	-	-	-	-	-	-

+ = Present, - = Absent, F<sub>1</sub>- Field 1, F<sub>2</sub>- Field 2, F<sub>3</sub>- Field 3, F<sub>4</sub>- Field -4

### 3.5.4 Occurrence of AM species in AEZ 28 soils

Nine AM species were identified in AEZ 28 soils (Table 8). Maximum numbers of AM species were identified in onion rhizosphere with four species which was followed by three in chilli, two in tomato and brinjal rhizosphere.

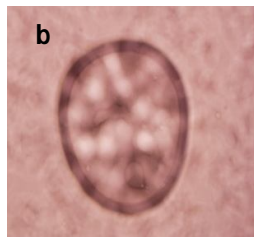
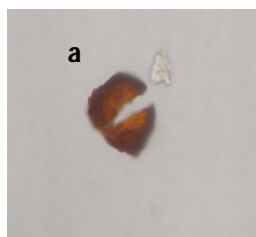
**Table 8. Occurrence of AM fungal strains in the rhizospheres of tomato, brinjal, chilli and onion from AEZ 28 soils**

Sl. No.	Fungal strains	Crops															
		Tomato				Brinjal				Chilli				Onion			
		F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>
1	<i>Glomus geosporum</i>	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-
2	<i>G. mosseae</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-
3	<i>G. fasciculatum</i>	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-
4	<i>Acaulospora dilatata</i>	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
5	<i>A. bireticulata</i>	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	<i>A. mellea</i>	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
7	<i>A. morrowiae</i>	-	+	-	-	-	-	-	-	-	-	-	-	-	-	+	-
8	<i>Sclerocystis coremioides</i>	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	+
9	<i>Gigaspora margarita</i>	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-

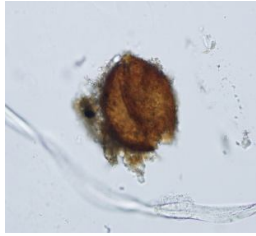
+ = Present, - = Absent, F<sub>1</sub>- Field 1, F<sub>2</sub>- Field 2, F<sub>3</sub>- Field 3, F<sub>4</sub>- Field -4

The Characteristics of AM species isolated from rhizosphere of tomato, brinjal, chilli and onion from different locations were in accordance with the following authors (Plate 1-9)-

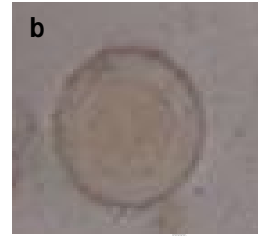
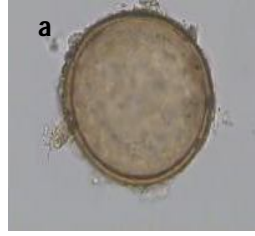
*Glomus geosporum* [31], *Glomus mosseae* [32], *Glomus fasciculatum* [31], *Glomus spp.* [31], *Acaulospora dilatata* [32], *Acaulospora bireticulata* [33], *Acaulospora mellea* [32], *Acaulospora morrowiae* [32], *Entrophospora infrequens* layers [34], [35], and [36], *Sclerocystis coremioides* [37] *Gigaspora margarita* [38].



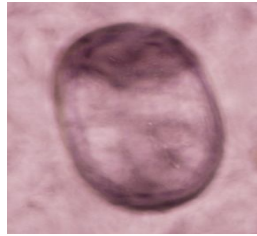
**Plate No.1. *Glomus fasciculatum* (a, b and c)**



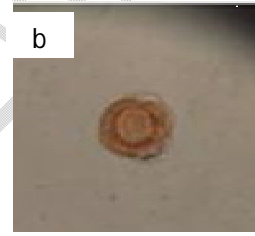
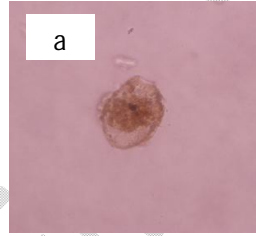
**Plate No. 2. *Glomus* spp.**



**Plate No. 3. *Acaulospora dilatata* (a, b)**



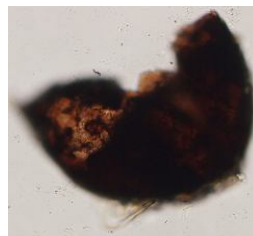
**Plate No. 4. *Acaulospora bireticulata***



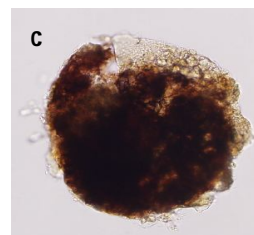
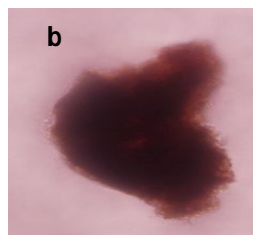
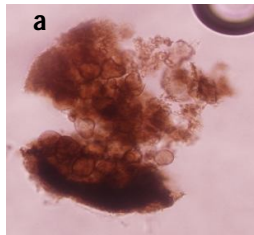
**Plate No. 5. *Acaulospora mellea* (a, b)**



**Plate No. 6. *Acaulospora morrowiae***



**Plate No. 7. *Entrophospora infrequens***



**Plate No. 8. *Sclerocystis coremioides* (a, b and c)**



**Plate No. 9. *Gigaspora margarita***

Eleven AM fungal species were identified representing five genera namely *Glomus*, *Acaulospora*, *Entrophospora*, *Sclerocystis* and *Gigaspora* over the locations (Table 5 to 8). Both *Glomus* and *Acaulospora* had four species each while three other genera namely *Entrophospora*, *Sclerocystis* and *Gigaspora* had single species each.

Eleven AM fungi species were recorded in tomato: that was followed by brinjal, chilli and onion having nine strains for each. In tomato fields *Glomus geosporum*, *G. mosseae*, *G. fasciculatum*, *G. spp.*, *Acaulospora dilatata*, *A. bireticulata*, *A. mellea*, *A. morrowiae*, *Entrophospora infrequens*, *Sclerocystis coremioides* and *Gigaspora margarita*. Nine species were recorded in other crops. *G. spp.*, and *Acaulospora bireticulata* were absent in both brinjal and chilli while *Acaulospora dilatata* and *A. mellea* were absent in onion.

#### **4. Discussion**

The variation in spore number might be due to structure of root system and in-built capacity of AM. Variation in spore density from site to site under the same crop species had also been reported by [39]. [40] stated that the higher spore population might be either due to conducive edaphic conditions like low nutrient status, high aeration and optimum moisture.

Variation between AEZs for the same crop was not that prominent, although not negligible. Almost similar trend was found in the spore population, the values were 120.11-200.33, 33.44-50.78, 36.11-39.22 and 28.56-41 per 100 g soil in onion, tomato, chilli and brinjal, respectively. The colonization rate of AM fungi differed with the plant species. It could be due to differences in the architecture of root system and efficacy of the genera and species of AM and soil moisture. Variation among different crops and sites observed could be due to fungal species, climatic and edaphic factors as stated by other researchers [41], [42] and [43]. The formation of arbuscules was influenced by the stages of plant development and was short-lived structure [44]. The formation of AM structures was inconsistent and fluctuating from site to site in the present study. The differences in morphological features of AM structures especially vesicles indicated the infection by different AM fungal species. Both oval and spherical vesicles were found in the study, which was also demonstrated by [45], [50].

The higher numbers of AM fungal species were recorded in the rhizosphere soil of AEZ 9 (11), followed by that of AEZ 25 (10), AEZ 28 (9) and AEZ 11 (6). The present study revealed that the AM fungi were common occurring in all the crops assessed from the four different locations.

This study revealed that AM fungi were common in all the crops assessed from the different locations. Among all the genera recorded in this study, *Glomus* and *Acaulospora* were the most common genera. These findings were in agreement with the numerous wide-spread occurrence of *Glomus spp.* throughout the world [46]; [47] and [4]. [48] reported that *Glomus* and *Acaulospora* were predominant in all rhizosphere soils in their study. [32] also found some AM fungi such as *G. fasciculatum*, *G. mosseae*, *A. dilatata*, *A. mellea*, *A. morrowiae* and *Gigaspora margarita* in the rhizosphere soils of tomato. [49] reported fifteen AM fungi in the rhizosphere soils of three solanaceous vegetables namely, tomato, chilli and brinjal collected from five different locations. The abundance of *Glomus*, *Acaulospora*, *Entrophospora*, *Sclerocystis* and

*Gigaspora* in tomato, brinjal, chilli and onion under four AEZ -9, 11, 23 and 28 had been stated by [27].

Edaphic factors play a critical role in the distribution and abundance of AM fungi [50]. The distribution of AM spores in rhizosphere soil is governed by edaphic and certain other climatic factors [48].

## 5. Conclusions

Rhizosphere soils and roots of tomato, brinjal, chilli and onion crops were collected from each of four AEZs, viz. AEZ-9 (RARS, Jamalpur), AEZ-11 (RARS, Jashore), AEZ-25 (ARS, Bogura) and AEZ-28 (BARI, Joydebpur) in 2013-2014 for characterization and identification of arbuscular mycorrhizal strains. Different crops showed positive responses in per cent root colonization. The AM fungus are ubiquitous in rhizospheres irrespective of crops and locations. *Glomus* and *Acaulospora* of AM fungi were the most common genera. The per cent root colonization showed positive responses in different crops such as 74.11-83.78%, 22.11-24.33%, 7.33-8.89% and 6.67-9.33% in onion, tomato, chilli and brinjal, respectively. Rhizosphere soil of individual crops had also variation in number of spore population such as 120.11-200.33, 33.44-50.78, 36.11-39.22 and 28.56-41.89 in onion, tomato, chilli and brinjal, respectively. The formation of AM structures found inconsistent and fluctuating from site to site in the present study. Both oval and spherical shaped vesicles were found among the AM structures. Eleven AM strains were identified in tomato. Nine each were identified from other three crops. *G. spp.* and *Acaulospora bireticulata* were absent in brinjal and chilli. *Acaulospora dilatata* and *A. mellea* were absent in onion.

## References

- [1] Smith, S.E. and Read, D.J. (1997) *Mycorrhizal Symbiosis*, 2<sup>nd</sup> edition. San Diego, CA Academic Press, London, U.K.
- [2] Solaiman, M.Z., and Mickan, B. (2014) "Use of mycorrhiza in sustainable agriculture and land restoration". In: MZ Solaiman, LK Abbott and A Varma (Editors), *Mycorrhizal Fungi: Use in sustainable agriculture and land restoration*. pp.10.
- [3] Akond, M.A., Mubassara, S., Rahman, M.M., Alam, S. and M. Khan, Z.U. (2008) "Status of Vesicular–arbuscular (VA) Mycorrhiza in vegetable crop plants of Bangladesh." *World Journal of Agricultural Science*, **4**, 704–708.
- [4] Blaszkowski, J. (1989) "The occurrence of the Endogonaceae in Poland". *Agricultural Ecosystem and Environment*, **29**, 45-50.
- [5] Mahmud, R., Mridha, M.A.U., Sultana, A., Sultana, N., Xu, H.L. and Umemura, H. (1999) "Biodiversity of VAM fungi in soils of crop field". *Nature Farming and Sustainable Environment*, **2**, 43-50.
- [6] Mridha, M.A.U., Sultana, A., Sultana, N., Xu, H.L. and H. Umemura. (1999) "Biodiversity of VA mycorrhizal fungi of some vegetable crops in Bangladesh". In: H. Xu (editor), *Proceedings International Symposium on World Food Security and Crop Production Technologies for Tomorrow*, October 8-9, 1998. Kyoto, Japan. pp. 330-331.

- [7] Lenin, M., Selvakumar, G., Thamizhiniyan, P. and Rajendiran. R. (2010) "Growth and biochemical changes of vegetable seedlings induced by arbuscular mycorrhizal fungus". *Journal of Experimental Science*, **1 (4)**, 27-31.
- [8] Liu, A., Hamal, C., Elmi, A., Costa, C.B. Ma. and Smith, D.L. (2002) "Concentrations of K, Ca and Mg in maize colonized by arbuscular mycorrhizal fungi under field conditions". *Canadian Journal of Soil Science*, **82 (3)**, 271-278.
- [9] Ryan, M.H., and Angus, J.F. (2003) "Arbuscular mycorrhizae in wheat and field pea crops on a low P soil: increased Zn-uptake but no increase in P uptake or yield". *Plant and Soil* **250**, 225–239.
- [10] Marschner, H., and Dell, B.(. (1994.) "Nutrient uptake in mycorrhizal symbiosis". *Plant and Soil*, **159**, 89-102.
- [11] Ikiz. O., Abak, K. H.Y., Dasgan, K. and Ortas. I. (2009) "Effects of mycorrhizal inoculation in soilless culture on pepper plant growth". *Acta Horticulturae*, **807**,533–540.
- [12] Goussous, S.J., Mohammad, M.J. (2009) "Effect of two arbuscular mycorrhizae and N and P fertilizers on growth and nutrient uptake of onions". *International Journal of Agriculture and Biology*, **11**, 463–467.
- [13] Matsubara, Y., Harada, T. and Yakuwa, T. (1994) "Effect of vesicular arbuscular mycorrhizal fungi inoculation on seedling growth in several species of vegetable crops". *Journal of Japanese Society for Horticultural Science*, **63**, 619–628.
- [14] Blaszkowski, J. (1989) The occurrence of the Endogonaceae in Poland. *Agricultural Ecosystem and Environment*, **29** 45-50.
- [15] Jackson, M.L. (1962) "Soil Chemical Analysis". Prentice Hall Inc. Englewood Cliffs, N.J.
- [16] Gee, G.W. and Bauder, J.W. (1986) Particle size Analysis. In: *Methods of Soil Analysis*, Part 1 (2<sup>nd</sup> edition). A. Klute (editor), American Society of Agronomy Inc. and Soil Science Society of America Inc., Madison, Wisconsin, USA. pp. 383-411.
- [17] Nelson, D.W. and Sommers, L.E. (1996) *Total Carbon, Organic Carbon, and Organic Matter*. In: DL Sparks, AL Page, PA Helmke, RH Loeppert, PN Soltanpour, MA Tabatabai, CT Johnston and ME Sumner (editors), *Methods of Soil Analysis, Part 3, Chemical Methods*, American Society of Agronomy Inc., Madison, Wisconsin, USA. pp. 961-1010.
- [18] Piper, C.S. (1950) *Soil and Plant Analysis*. Adelaide Univ. Hasel Press, Australia.
- [19] Bremner, J.M. and Mulvaney, C.S. (1982). *Total Nitrogen*. In: AL Page, RH Miller and DR Keeney (editors), *Methods of Soil Analysis, Part 2, Chemical and Microbiological Properties*, American Society of Agronomy and Soil Science Society of America, Inc., Madison, Wisconsin, USA. pp. 595-624.
- [20] Olsen, S.R., and Sommers, L.E. (1982) *Phosphorus*. In: AL Page, RH Miller and DR Keeney (editors), *Methods of Soil Analysis, Part 2, Chemical and Microbiological Properties*, American Society of Agronomy Inc., Madison, WI, USA. pp. 403-430.

- [21] Knudsen, D., Peterson, G.A. and Pratt, P.F. (1982) *Lithium, Sodium, and Potassium*. In: AL Page, RH Miller and DR Keeney (editors), *Methods of Soil Analysis, Part 2, Chemical and Microbiological Properties*, American Society of Agronomy Inc., Madison, Wisconsin, USA. pp. 225-246.
- [22] Fox, R.L., Olson, R. A. and Rhoades, H.F. (1964) "Evaluating the sulfur status of soils by plants and soil tests". *Soil Science Society of America Proceeding*, **28**, 243-246.
- [23] Jones, L.H.P., Cowling, D.W. and Lockyer, D.R. (1972) "Plant-available and extractable sulphur in some soils of England and Wales". *Soil Science*, **114**, 104-114.
- [24] Lindsay, W.L., and Norvell, W.A. (1978) "Development of a DTPA soil test for Zn, Fe, Mn and Cu". *Soil Science Society of America Journal*, **42**, 421-428.
- [25] Keren, R. (1996) *Boron*. In: DL Sparks, AL Page, PA Helmke, RH Loeppert, PN Soltanpour, MA Tabatabai, CT Johnston and ME Sumner (editors), *Methods of Soil Analysis, Part 3, Chemical methods*, American Society of Agronomy Inc., Madison, Wisconsin, USA. pp. 603-626.
- [26] Koske, R.E., and Gemma, J.N. (1989) "A modified procedure for staining roots to detect VA mycorrhizas". *Mycological Research* 92: 486-488.
- [27] Khanam, D. (2002) Biodiversity of arbuscular mycorrhizal fungi in agricultural crops and chickpea (*Cicer arietinum* L.). PhD thesis, Dept. of Soil Science. Bangabandhu Sheikh Mujibur Rahman Agricultural University (BSMRAU), Gazipur-1703, Bangladesh.
- [28] Read, D.J., Koucheiki, H.K. and Hodgaon, J. (1976) "Vesicular arbuscular mycorrhiza in natural vegetation systems". *New Phytologist*, **77**, 641-653.
- [29] Morton, J.B. and Benny. G.L. (1990) "Revised classification of arbuscular mycorrhizal fungi (Zygomycetes): a New order, Glomales, two New sub-orders, Glomineae and Gigasporaceae and two new families, Acaulosporaceae and Gigasporaceae with an amendment of Glomaceae". *Mycotaxonomy* **37**, 471-491.
- [30] Schenck, N.C., and Perez, Y. (1990). *Manual for the identification of VA mycorrhizal fungi*. Synergistic Publications, USA. p. 286.
- [31] Sharma, S., V. Parkash, and Aggrawal. A. (2008) "Glomales I: A monograph of *Glomus* spp. (Glomaceae) in the sunflower rhizosphere of Haryana, India". *HELIA* **31 (49)**, 13-18.
- [32] Sreevani, S., and Reddy, B.N. (2004) "Arbuscular mycorrhizal fungi associated with tomato (*Lycopersicon esculentum* Mill.) as influenced by soil physico-chemical properties". *Philippines Journal of Science*, **133 (2)**, 115-129.
- [33] Rothwell, .FM., and Trappe (2004) [file:///H:/ Acaulospora bireticulata.htm](file:///H:/Acaulospora%20bireticulata.htm), retrived on 1.1.2004.
- [34] Hall, I.R. (1977) "Species and mycorrhizal infections of New Zealand Endogonaceae". *Transactions of the British Mycological Society*, **68**, 341-356.
- [35] Ames, R.N., and Schneider, R.W. (1979) "*Entrophospora*, a new genus in the Endogonaceae". *Myeotaxon*, **8**, 347-352.

- [36] Oehl, F., and Sieverding, E. (2006) "Revision of *Entrophospora* and description of *Kuklospora* and *Intraspora*, two new genera in the arbuscular mycorrhizal Glomeromycetes". *Journal of applied botany and food quality*, 80, 69-81.
- [37] Almeida, R.T. and Schenck. (1990) "A revision of the genus *Sclerocystis* (Glomaceae, Glomales)". *Mycologia*, **82** (6), 703-714.
- [38] Khade, S.W. (2011) "Gigaspora (Gigasporaceae) from India, with morpho-taxonomic records. *Acta Biologica Paranaense*". *Curitiba*, **40**(1-2), 9-17.
- [39] Howeler, R.H., Seiverding, E. and Saif, S. (1987) "Practical aspects of mycorrhizal technology in some tropical crops and pastures". *Plant and Soil*, **100**, 249-283.
- [40] Azizah, H. and Omar, M. (1991) "Incidence of VAM spores in some Malaysian soils." *Pertanika*, **14**, 133-137.
- [41] Khalil, S., Loynachan, T.E. and McNabb, H.S. (1992) "Colonization of soybean by mycorrhizal fungi and spore population in Iowa soils". *Agronomy Journal*, **84**, 832-836.
- [42] Udaiyan, K., Karthikeyan, A. and Muthukumar, T. (1996) Influence of edaphic and climatic factors on dynamic of root colonization and spore density of vesicular arbuscular mycorrhizal fungi in *Acacia farnesiana* Willd, and *A. planifrons* W et A. *Trees*, **11**, 65-71.
- [43] Hetrick, B.A.D. and Bloom, J. (1986) "The influence of host plant on production and colonization ability of vesicular arbuscular mycorrhizal spores". *Mycologia*, **78**, 32-36.
- [44] Neeraj, A., Mathew, S. J. and Varma, A. (1991) "Occurrence of vesicular arbuscular mycorrhizae with Amaranthaceae in soils of the Indian semi arid region". *Biology and Fertility of Soils*, **11**, 140-144.
- [45] Muthukumar, T., Udaiyan, K. and Manian, S. (1994a) "Role of edaphic factors on VAM fungal colonization and spore populations in certain tropical wild legumes. *Pertanika*". *Journal of Tropical Agricultural Science*, **17**, 33-42.
- [46] Mosse, B. and Stribley DP Le Tacon F (1981) Ecology of mycorrhizae and mycorrhizal fungi. *Advances in Microbial Ecology*, **5**, 137.
- [47] Sylvia, D.M., Hammond, L.C., Bennett, J.M., Haas, J.H. and Linda, S. B. (1993) "Field response of maize to VAM fungus and water management". *Agronomy Journal*, **85**, 193-198.
- [48] Bhuvaneshwari, N. and Sadhana, B. (2014) "Arbuscular mycorrhizal status of non-leguminous plants". *Indian Journal of Advances in Plant Research (IJAPR)* 1 (2): 31-38.
- [49] Reddy, B.N., Sreevani, A., Raghavender, C.R. (2006) "Association of AM fungi in three solanaceous vegetable crop". *Journal of Mycology and Plant Pathology*, **36** (1), 52-56.

[50] Muthukumar, T., Udaiyan, K. and Manian, S. (1994b). “Vesicular arbuscular mycorrhizae in certain tropical wild legumes”. *Annals of Forestry*, **2**, 33-43.

UNDER PEER REVIEW