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Arbuscular Mycorrhizal Status of the Plants Growing in the Alkaline/Sodic Soils of Pratapgarh, Allahabad, Uttar Pradesh

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Abstract: In India approximately 7 mha land is salt affected out of which 2.5 mha occurs in Indo-Gangetic plains. In Uttar Pradesh alone about 1.29 mha is salt affected. In the present study a survey was made to investigate arbuscular mycorrhizal status of the wild plants growing in alkaline/sodic soils of Pratapgarh, Allahabad, Uttar Pradesh, so that the adoptive and effective AMF isolates may be used in further studies to give the solution for the reclamation and utilization of such soils. A total of 18 plant species belonging to 18 genera and 9 families were identified from different zones of the selected site. *Cyperus rotundus* L., *Desmostachya* sp., *Saccharum munja* Roxb., and *Sporobolus diander* (Retz.), P. Beauv. were the most dominant grasses in I to III zones. However, zone IV, where cultivation was being practiced, was mainly dominated by the cultivated crops. All the plants showed mycorrhizal association in their roots, except the member of Brassicaceae and Cyperaceae. A total of 19 species belonging to three genera of AM fungi were isolated. *Glomus* was recorded as the most dominant genus with 13 species.

Key Words: AM fungi, Diversity, Alkaline/sodic soils, Indo-Gangetic plains.

Introduction

One of the important categories of wasteland is salt affected wasteland, which occupies extensive area in the world and in India as well, presenting a serious impediment to crop production. These soils are completely barren and practically produce nothing. Plant growth and development in such soils is adversely affected either due to excessive amounts of neutral soluble salts or high exchangeable sodium or both. In India approximately 7 mha land is salt affected out of which, 2.5 mha occurs in Indo-Gangetic plains covering the states of Uttar Pradesh, Haryana, Punjab, Delhi and parts of Bihar. In Uttar Pradesh alone about 1.29 mha is salt affected and commonly known as '*usar*' or '*reh*' in local language.

Salt affected soils can be classified into three different categories, *viz.* saline, alkaline/sodic and saline-alkaline/saline-sodic. Extensive occurrence of alkaline soils has been reported from the Indo-Gangetic plains of northern India. The agricultural history of the region suggests that these high alkaline and sodic lands have been left unproductive in this area for a long time.

In this respect, application of biological inputs in combination with some moderately salt tolerant plant species seems to be a better option for the reclamation and management of such soils. In the past few decades, it has been well established that the AM fungi enhance the ability of plants to cope with environmental stresses generally prevalent in the degraded ecosystems. Several workers have reported the presence of AM association in salt stress environments.

The objective of the present work was to investigate the distribution of AMF in the rhizosphere of different wild plants growing in the alkaline/sodic soils of Pratapgarh, Allahabad, Uttar Pradesh so that the adoptive and effective AMF isolates may be used in further studies to give the solutions for the reclamation and utilization of such soils.

Materials and Methods

A systematic survey of the alkaline/sodic site of Pratapgarh, Allahabad was undertaken during to assess the population and diversity of AM fungi in such soils and intensity of arbuscular mycorrhizal association in the roots of the plants growing in vicinity.

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Site Selection and Sample Collection

The site *i.e.* Pratapgarh (25^0 33' N, 82^0 6' E) Allahabad, Uttar Pradesh, were selected for the survey. At the site, large patches of unproductive fallow lands, commonly known as '*usar*' or '*reh*', were found intermixed with the agricultural field. The site was divided into four different zones consisting of barren unproductive land, grassy patches, scanty vegetation and cultivated fields.

Samples site were collected during summer (April - May), rainy (August – September) and winter (December – January) seasons. During each sampling, 200 g soil was collected randomly from each zone at different depths *i.e.* 0-15 cm, 15-30 cm and 30 cm below the surface. Root samples of the plants growing in different zones were also collected for the estimation of mycorrhizal infection.

Analysis of Soil Samples

Collected soil samples were air-dried, sieved and soil extract was prepared by the method of Adams *et al.* 1980. Soil pH and electrical conductivity (EC) were measured with a digital pH meter and a digital EC meter. Total organic carbon was estimated by the method given by Nelson and Sommers (1982), phosphorus by Watanabe and Olsen (1965), nitrogen by Bremner (1960) and potassium by flame photometer method (Richards, 1954). Soil characteristics of different sites are presented in table 1.

Table 1: Edaphic features of the alkaline/sodic soils of different zones collected from the selected site Pratapgarh

Sites/Zones	pH	EC (m.mhos.cm^{-1})	Organic carbon (%)	Nitrogen (%)	Phosphorus (kg/ha)	Potassium (kg/ha)
Zone I	9.9	4.12	0.31	2.35	10.0	162.0
Zone II	9.7	2.08	0.37	2.78	32.0	177.0
Zone III	9.3	1.12	0.42	2.72	36.0	207.0
Zone IV	8.7	1.00	0.59	2.88	44.0	275.0

Estimation of AM Association in the Roots

Intensity of AM association in the root samples was determined by the method given by Philips and Hayman (1970). Percent root bits colonized and the percent root length colonization were expressed as the mean of three replicates.

Determination of AM Spore Population

AM spore population was determined in 50g air-dried soil in triplicates for each sample by wet sieving and decanting method (Gerdemann and Nicolson, 1963). Number of spores was expressed as the mean of three replicates.

Identification of AM Fungi

AM spores were mounted in PVLG and PVLG + Melzer's reagent (1:1 v/v) and identified to the species level using the synoptic keys of Trappe (1982), Schenck and Perez (1990) and INVAM species guide (http://invam.caf.wvu.edu).

Results

Analysis of Soil Samples

The data of analysis of all the soil samples collected from different zones of the selected site, *viz.* Pratapgarh has been presented in Table 1. The elemental analysis of the soil samples indicates that the soils of this site are highly alkaline. The pH of the soils ranged from 8.7 (Zone IV) to 9.9 (Zone I). However, electrical conductivity (EC) was not more than 4.0 m.mhos.cm^{-1}. EC ranged from 1.0 m.mhos.cm^{-1} (Zone IV) to 4.12 m.mhos.cm^{-1} (Zone I).

The nutrient status of the soil was also very poor. The organic carbon and nitrogen contents were low in all the samples analyzed. Organic carbon ranged from 0.31% (Zone I) to 0.59% (Zone IV). Likewise, nitrogen content ranged from 2.35% (Zone I) to 2.88% (Zone IV).

The soils were also deficient in phosphorus and potassium content. Available phosphorus, estimated in terms of P_2O_5, ranged from 10 kg/ha (Zone I) to 44 kg/ha (Zone IV). Potassium content, estimated in terms of K_2O, ranged from 162 kg/ha (Zone I) to 275 kg/ha (Zone IV).

Vegetation Composition in the Different Zones of the Selected Site

The data of vegetation composition in the different zones of the selected site *i.e.* Pratapgarh, has been presented in Table 2. A total of 18 plant species belonging to 18 genera and 9 families were identified from the

different zones of the the selected site. Minimum vegetation was recorded in Zone I and the vegetation cover increased sequentially from Zone I to Zone III. Grasses such as *Cynodon dactylon* (L.) Pres, *Cyperus rotundus* L., *Desmostachya* sp. and *Sporobolus diander* (Retz.) P. Beauv. were the most dominant vegetation in these zones. However, Zone IV, where cultivation was being practiced, was mainly dominated by the cultivated crops.

Table 2: Vegetation composition of different zones of the alkaline/sodic soils at the selected site, Pratapgarh

Family	Plant/ Zones	Vegetation Composition											
		Winter				Rainy				Summer			
		I	II	III	IV	I	II	III	IV	I	II	III	IV
Amaranthaceae	*Amaranthus viridis* L	-	+	+	-	-	+	+	-	-	-	+	-
Brassicaceae	*Brassica campestris* L.	-	-	-	+	-	-	-	-	-	-	-	-
Chenopodiaceae	*Chenopodium album* L.	-	-	+	+	-	+	+	+	-	-	+	-
Cyperaceae	*Cyperus rotundus* L.	+	+	+	-	+	+	+	-	-	+	+	-
Euphorbiaceae	*Croton bonplandianum* Baill.	-	-	+	-	-	+	+	-	-	-	-	-
Euphorbiaceae	*Euphorbia hirta* L.	-	-	+	+	-	+	+	-	-	-	-	-
Fabaceae	*Cajanus cajan* (L.) Huth	-	-	-	+	-	-	-	+	-	-	-	-
Fabaceae	*Cicer arietinum* L.	-	-	-	+	-	-	-	-	-	-	-	-
Fabaceae	*Sesbania sesban* (L.)	-	-	-	-	-	-	+	-	-	-	-	-
Fabaceae	*Vigna mungo* (L.) Hepper	-	-	-	-	-	-	-	-	-	-	-	+
Fabaceae	*Vigna radiata* (L.) Wilczek	-	-	-	-	-	-	-	-	-	-	-	+
Mimosaceae	*Acacia nilotica* (L.) Willd.	-	-	+	-	-	-	+	-	-	-	+	-
Mimosaceae	*Prosopis juliflora* (Swartz.) DC	-	+	+	-	-	+	+	-	-	+	+	-
Poaceae	*Desmostachya sp*.	+	+	+		+	+	+	+	-	+	+	-
Poaceae	*Oryza sativa* L.	-	-	-	-	-	-	-	+	-	-	-	-
Poaceae	*Panicum repens* L.	-	-	-	-	-	-	-	-	-	-	-	+
Poaceae	*Sporobolus diander* (Retz.) P. Beauv.	+	+	-	-	+	+	+	-	-	-	-	-
Poaceae	*Triticum aestivum* L.	-	-	-	-	+	-	-	-	-	-	-	-

The largest families were Fabaceae and Poaceae each accounting for 27.7% of total flora recorded, followed by Euphorbiaceae and Mimosaceae each accounting for 11.1%. *Cyperus rotundus* L., *Desmostachya* sp. and *Sporobolus diander* (Retz.) P. Beauv. formed the pioneer vegetation in Zone I. Zone II and Zone III were dominated by the members of the family Amaranthaceae, Chenopodiaceae, Cyperaceae, Euphorbiaceae, Mimosaceae and Poaceae. *Brassica campestris* L., *Cajanus cajan* (L.) Huth., *Oryza sativa* L. and *Triticum aestivum* L. were the most dominant crops being cultivated in Zone IV.

AM association in the Roots of the Plants Growing in Different Zones of the Selected Site

Data of the arbuscular mycorrhizal intensity in the roots of the plants growing in different zones in different seasons has been presented in table 3. All the plants showed mycorrhizal association in their roots, except the members of Brassicaceae and Cyperaceae. However, the intensity of the mycorrhizal association varied with the plant species, magnitude of the stress and seasonal variations. *Chenopodium album* L., member of the family Chenopodiaceae, which is otherwise reported as non-mycorrhizal, showed mycorrhizal association under stress conditions, although the intensity of the mycorrhizal infection was quite low.

Hyphae, vesicles, arbuscules and intramatrical spores characteristic of AM fungi were observed in the roots of the plants. Maximum mycorrhizal infection was recorded in *Cajanus cajan* (L.) Huth. and *Vigna radiata* (L.) Wilczek, while minimum in *Desmostachya* sp.

In general, the intensity of AM infection showed a negative correlation with the magnitude of stress, as it increased from Zone I to Zone IV. Mycorrhizal intensity ranged from 14-18% in Zone I, 10-44% in Zone II, 15-64% in Zone III and 14-71% in Zone IV.

Maximum AM infection was recorded in winter season. However, no significant pattern of mycorrhization could be traced in relation to the seasonal variation. During winter season, mycorrhizal intensity ranged from 12-71%, during rainy season, mycorrhizal intensity ranged from 14-46% and during summer season, mycorrhizal intensity ranged from 10-71% at Pratapgarh.

Table 3: Seasonal variation in the mycorrhizal intensity in the roots of plants growing in different zones of alkaline/sodic soils of Pratapgarh

Plants/Zones	MYCORRHIZATION (% Root bits Infected)											
	Winter (Dec-Jan)				Rainy (Aug-Sep)				Summer (Apr-May)			
	I	II	III	IV	I	II	III	IV	I	II	III	IV
Acacia nilotica (L.) Willd.	-	-	48	-	-	-	38	-	-	-	42	-
Amaranthus viridis L.	-	22	15	-	-	18	25	-	-	-	15	-
Brassica campestris L.	-	-	-	0	-	-	-	-	-	-	-	-
Cajanus cajan (L.) Huth.	-	-	-	71	-	-	-	-	-	-	-	-
Chenopodium album L.	-	-	0	0	-	14	0	0	-	-	0	-
Cicer arietinum L.	-	-	-	65	-	-	-	-	-	-	-	-
Croton bonplandianum Baill.	-	44	64	-	-	-	38	-	-	-	-	-
Cyperus rotundus L.	0	0	-	-	0	0	0	-	-	0	0	-
Desmostachya sp.	18	28	37	-	0	0	18	15	-	10	18	-
Euphorbia hirta L.	-	-	20	44	-	-	33	-	-	-	-	-
Oryza sativa L.	-	-	-	-	-	-	-	46	-	-	-	-
Panicum repens L.	-	-	-	-	-	-	-	-	-	-	-	14
Prosopis juliflora (Swartz.) DC	-	30	52	-	-	25	44	-	-	40	44	-
Sesbania sesban (L.) Merr.	-	-	-	-	-	-	36	-	-	-	-	-
Sporobolus diander (Retz.) P. Beauv.	14	12	-	-	-	18	33	-	-	-	28	-
Triticum aestivum L.	-	-	-	55	-	-	-	-	-	-	-	-
Vigna mungo (L.) Hepper	-	-	-	-	-	-	-	-	-	-	-	62
Vigna radiata (L.) Wilczek	-	-	-	-	-	-	-	-	-	-	-	71

Spore Population and Diversity of AM Fungi in Different Zones of selected Site

Data on spore population and diversity of AM fungi in different zones of the selected site in different seasons has been presented in Table 4. Although the presence of the AM fungi was recorded in all the zones, their population and diversity varied along with the magnitude of stress and the seasonal variations.

Table 4: Seasonal variation in the population size and diversity of AM fungi in different zones of alkaline/sodic soils of Pratapgarh

AM Fungi/Zones	AM POPULATION SIZE/DIVERSITY (/50g soil)											
	Winter (Dec-Jan)				Rainy (Aug-Sep)				Summer (Apr-May)			
	I	II	III	IV	I	II	III	IV	I	II	III	IV
Acaulospora bireticulata Rothwell & Trappe	-	1	1	4	1	1	1	2	-	-	-	2
A. denticulata Sieverding & Toro	1	1	3	6	-	-	2	4	-	1	-	1
A. laevis Gerdemann & Trappe (*Archaeospora laevis*)	-	3	-	5	1	1	-	3	-	-	1	1
A. longula Spain & Schenck	-	-	3	11	-	1	3	8	2	1	2	2
Gigaspora sp. UVK1	-	1	1	2	-	-	-	3	-	-	-	-
Glomus aggregatum Schenck & Smith	1	1	3	8	-	1	4	6	-	1	2	4
G. claroideum Schenck & Smith	1	-	-	4	-	-	-	2	1	1	2	2
G. constrictum Trappe	-	-	3	8	1	1	2	4	-	-	-	-
G. fasciculatum (Thaxter) Gerdemann & Trappe	3	6	8	15	1	2	6	11	1	1	-	5
G. invermaium Hall	2	1	1	5	1	1	-	1	-	-	-	-
G. macrocarpum Tul. & Tul.	-	-	-	4	-	-	-	-	-	1	-	-
G. mosseae Grdemann & Trappe	1	4	8	10	-	2	7	9	-	2	4	4
G. occultum Walker (*Paraglomus occultum* Morton & Redecker)	-	1	-	3	-	-	-	1	-	-	-	-
G. tortuosum Schenck & Smith	-	-	3	5	1	1	-	-	-	-	-	-
Glomus sp. UVK1	-	-	2	8	-	-	3	5	-	-	-	-
Glomus sp. UVK3	-	-	4	10	-	-	4	7	1	1	-	3
Glomus sp. UVK4	1	-	2	2	-	-	-	-	-	-	3	3
Glomus sp. UVK8	-	1	3	5	1	-	1	2	-	-	-	-
Scutellospora sp.	-	-	2	-	-	-	-	-	-	-	-	1
	10	20	47	115	7	11	33	68	5	9	14	28

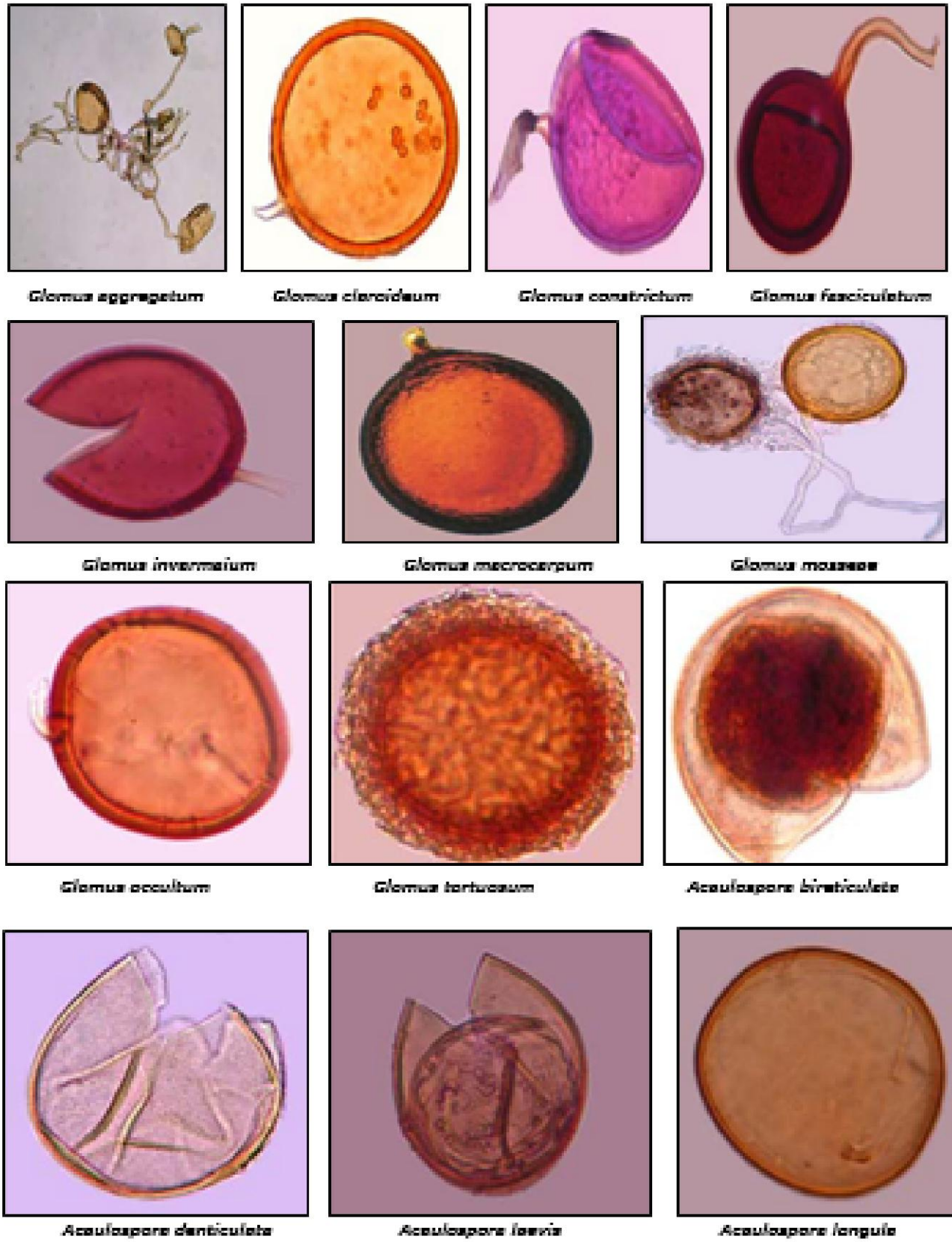
Glomus aggregatum
Glomus claroideum
Glomus constrictum
Glomus fasciculatum
Glomus invermaium
Glomus macrocarpum
Glomus mosseae
Glomus occultum
Glomus tortuosum
Acaulospora bireticulata
Acaulospora denticulata
Acaulospora laevis
Acaulospora longula

Plate-1

AM spore population showed a negative correlation with the magnitude of stress, as it increased from Zone I to Zone IV. Average AM spore population was 7.3, 13.3, 31.3 and 70.3 spores/50g air dried soil.

Significant variation was recorded in the AM spore population in relation to the seasonal changes. Maximum spore population was recorded in winter and minimum in summer. During winter season, spore population ranged from 10-115 spores/50g air dried soil during rainy season, spore population ranged from 7-68 spores/50g air dried soil and 7-68 and during summer season, AM spore population ranged from 5-28 spores/50g air dried soil.

A total of 19 species belonging to three genera of AM fungi (*Acaulospora, Glomus* and *Sclerocystis*) were isolated from the different zones of the selected alkaline/sodic site. *Glomus* was recorded as the most dominant genus with 12 species, *viz. G. aggregatum, G. oculltum, G. claroideum, G. constrictum, G. fasciculatum, G. macrocarpum, G. mosseae, G. tortuosum, G. invermaium* and 4 unidentified species named *Glomus* sp. UVK1, UVK3, UVK4, UVK8 followed by *Acaulospora* with four species, *viz. A. bireticulata, A. laevis, A. longula, A. denticulata* and *Sclerocystis* with single species, *viz. S. dussii*.

AM fungal diversity showed a negative correlation with the magnitude of stress, as it increased from Zone I to Zone IV. However, no significant seasonal variations were recorded in diversity.

Discussion and Conclusion

AM fungi can promote plant establishment by increasing resistance to environmental stresses, enhancing plant nutrient acquisition and improving soil quality (Schreiner *et al*., 1997; Jeffries and Barea, 2001). Because of the key ecological functions performed by AM associations (Jeffries and Barea, 2001), loss or diminution of mycorrhizal potential in degraded areas may limit the successful re-establishment of native plants (Van der Heijden *et al*., 1998; Requena *et al*., 2001). Van der Heijden *et al*. (1998) showed that below ground diversity of vesicular arbuscular mycorrhizal fungi (AMF) may be a major factor contributing to the maintenance of plant biodiversity and to ecosystem functioning.

AMF are reported to reduce the detrimental effects of soil associated plant stresses, such as lack of nutrients, organic matter, high salinity or high pH (Sylia and Williams, 1992; Entry *et al*. 2002). The mycorrhizal symbiosis, therefore, is an important potential strategy for phyto restoration schemes (Dodd *et al*; 2002; Renker *et al*. 2004). Clearly, multifunctional communities of AMF are important in the establishment and survival of plants in a wide range of habitats (Pfleger *et al*; 1994; Enkhtuya *et al*., 2003). With different levels of compatibility between host plant and AMF (Klironomos, 2003), appropriate isolates of AMF must be selected before setting an experiment, especially when native or non-native isolates are being considered (Dodd and Thomson,1994). Most of the studies show that especially in stressed conditions and degraded soils native AM fungi can grow and function better possibly as they are better adapted to their stressed microhabitat (Enkhtuya *et al.*, 2000; Caravaca *et al*., 2003; Calvente *et al*., 2004).

Distribution of AM fungi in different ecological regions and their relations to soil properties and native plants have been investigated by several researchers (Kim and Weber, 1985; Rozema *et al.*, 1986; Koske, 1987; Cook *et al.*, 1993; Janardhanan *et al.*, 1994; Aliasgharzadeh *et al.*, 2001; Wang *et al.*, 2004; Shi *et al.*, 2007). It is established that variation in AM distribution, spore density and colonization with different host plant species is generated by a variety of mechanisms, including variation in host species and their phenology, mycorrhizal dependency, soil properties, host plant-mediated alteration of the soil microenvironment, or other unknown host plant traits (Hayman, 1982; Eom *et al.*, 2000; Wang *et al.*, 2004). In addition, species and isolates of AM fungi differ in their tolerance to adverse physical and chemical conditions in soil (Sengupta and Chaudhuri, 1990; Juniper and Abbott, 1993; Joshi and Singh 1995; Aliasgharzadeh *et al.*, 2001).

All the plants, except Brassicaceae and Cyperaceae, surveyed in alkaline/sodic soils of the selected site were colonized by the AM fungi but the colonization percentages were significantly low. The results were in accordance to the earlier reports of Brown and Bledsoe (1996), Udaiyan *et al*. (1996) and Wang *et al.* (2004). Mycorrhizal intensity

showed a significant correlation with plant species and magnitude of stress. Lowest mycorrhizal intensity at Zone 1 may be attributed to the fact that high soil pH, low nutrients, poor plant diversity and vegetation cover severely restrict colonization of AM fungi (Janardhanan *et al.*, 1994; Aliasgharzadeh *et al.*, 2001; Wang *et al.*, 2004). However, AM fungi have the ability to form a zone of altered pH in the adjacent soil (Li *et al.*, 1971; Janardhanan *et al.*, 1994). Chenopod plants, which are otherwise regarded as non-mycorrhizal, have been reported to be mycorrhizal under alkaline stress in the present study. Mycorrhization in chenopods under different kind of stresses, such as drought, salinity *etc.*, have also been reported earlier by Hirrel *et al.* (1978), Rozema *et al.* (1986), Barrow *et al.* (1997) and Aliasgharzadeh *et al.* (2001). These results may also support the conclusion of Carvalho *et al*. (2001) that the colonization by vesicular arbuscular mycorrhizae under stress conditions depends more on host plant species than on environmental stresses.

It is also reported that the AM spore population and root colonization pattern changes with seasonal variation and host plant phenology (He *et al*., 2002; Bohrer *et al*., 2004). Patterns of seasonal changes in mycorrhizal intensity were more obvious in the annuals in comparison to perennials. Although no significant correlation could be established in the present study between the mycorrhizal intensity and seasonal variation, however in general mycorrhizal intensity was always higher in winter season. The highest degree of colonization of annuals in winter, i.e. during their flowering period, may have a possible explanation of increasing requirements of nutrients and water at this stage of growth (Shi *et al.*, 2007).

The species of *Glomus* were the most dominant in the alkaline/sodic soil of Indo-Gangatic plains of India and they were distributed widely in the rhizosphere of most of the plant species. *G. mosseae* and *G. fasciculatum* species were the most dominant and there is possibility that these species may improve tolerance to salt stress and may prove useful isolates for improving the overall performance of the crops under such conditions.

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