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M. S. Chaudhry · Z. Batool · A. G. Khan

Preliminary assessment of plant community structure and arbuscular mycorrhizas in rangeland habitats of Cholistan desert, Pakistan

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Abstract Plant species dominance and arbuscular mycorrhizas (AMs) were evaluated in four different habitats of Cholistan desert, Pakistan. The sites were selected on the basis of variations in topography, floristic composition and biotic interference. The phytosociological data revealed obvious differences in the floristic composition, plant species diversity, AM colonization and arbuscular mycorrhizal fungi (AMF) spore densities in the rhizospheres. The vegetation pattern and composition was governed by topography and edaphic characteristics of a particular site. Grasses were dominant and main components of vegetation at all the study sites and were invariably mycorrhizal. Some plant species were mycorrhizal at one site but nonmycorrhizal at the other. The AMF survey reported here offers an important starting point from which to analyse AMF community structure in different phytosociological habitats and land uses of Cholistan desert.

Keywords Arbuscular mycorrhiza · Xerophytes · Desert

Introduction

In view of the increasing degradation of nutrient-deficient Pakistani soils due to salinity, water-logging, industrial and agroforestry practices, information about the potential significance of mycorrhizas for restoration of derelict land in Pakistan is lacking. There is an increasing body of evidence suggesting that the distribution and abundance of natural vegetation is influenced by the composition of indigenous

M. S. Chaudhry \cdot Z. Batool Cholistan Institute of Desert Studies, Islamia University, Bahawalpur, Pakistan

A. G. Khan (⊠) Department of Environmental Sciences, Allama Iqbal Open University, Islamabad, Pakistan e-mail: lasara37@hotmail.com microbial communities. Studies over the past few years have demonstrated that arbuscular mycorrhizas (AM) influence several aspects of plant community structure and function by influencing various parameters such as plant diversity, species composition, and successional dynamics (Hartnett and Wilson 2002). Plant species diversity may be positively (van der Heijden et al. 1998) or negatively (O'Connor et al. 2001) correlated with arbuscular mycorrhizal fungi (AMF) populations. AMF may also alter plant species composition with no net effect on species richness (Smilauer and Smilauerova 2000), or may alter rates of succession (Smith et al. 1998). AM status of plants and their succession can vary depending on the moisture and nutrient conditions of the soil (Allen and Allen 1990).

In deserts, osmotic stress, caused by drought, is considered the most important abiotic factor limiting plant growth and yield (Kramer and Boyer 1997). Various morphological, anatomical and physiological responses by plants allow them to alleviate such stresses or to increase their tolerance (Bray 1997). In addition to these intrinsic protective features of plants against osmotic stress, AMF are also known to enhance plant tolerance to drought (Ruiz-Lozano et al. 1995, 2001; Auge 2001).

The potential of AMF in arid zones to enhance plant establishment and productivity has now been recognized. However, little is known about the effects of urbanization of deserts on the indigenous AMF flora, as human activity may entail disturbance and/or removal of AMF species richness and infectivity compared with non-urbanized surroundings. The role of AMF in the xerophytic plant community structure and succession has not been widely examined due to lack of knowledge of most of the complex AMF-plant associations and lack of practical field studies. In view of these facts, a preliminary field survey was conducted to ascertain the community dynamics of plant species and their mycorrhizal status in various rangeland habitats of Cholistan desert in Pakistan. Information on the mycorrhization of Cholistan desert plant species is important for re-establishment programmes.

Materials and methods

Sample sites

The plant communities in Cholistan deserts are open, with distinct phytogeographical distribution. The available land is principally used for grazing. Apart from natural stresses such as drought, ethnic destructive activities and overgrazing are the major deterrents of ecosystems. The climate of Cholistan is the subtropical continental desert type characterized by low and sporadic rainfall, high temperatures, low relative humidity, high rate of evaporation and strong summer winds. The highest rainfall months are July and August, and the maximum temperature is recorded during the month of June (Fig. 1). Four sites were selected on the bases of variations in topography, floristic composition and biotic interference. The sites S1 and S2 were near Fort Moujgarh and Fort Bajnot, respectively, whereas site S3 was inside the fenced area of the Pakistan Council of Research in Water Resources Regional Office in Bahawalpur near Fort Dinegargh. Because grazing and cutting of vegetation inside the fenced area has been strictly prohibited in the last ten years this site provides the best opportunity for the study of mycorrhizas in undisturbed vegetation. Site S4 was located in the desert area of the Baghdad-ul-Jadeed Campus on the north side of the Cholistan Institute building. The description of these sites is given in Table 1. An area of 1,000 m² was sampled on each site.

Soil collection and AM spore extraction

The sampling of roots and soil for mycorrhization took place between 9 and 23 September 2002 (Table 1). Three soil samples were collected to the depth of approximately 30 cm from the root zones of each plant. The soil samples collected from different study sites were pooled so that each plant species had a single pooled soil sample, and stored at 4°C until analysed. Three aliquots of 100 g soil were obtained for every plant species. Spores were extracted from the soil samples by wet sieving and decanting (Gerdemann and Nicolson 1963). Healthy spores of each distinctive morphotype were directly counted under a dissecting microscope and recorded as mean spores per 100 g. No attempts were made to identify the spores to the species level, nor were any trap cultures established due to lack of appropriate resources.

Root collection and AMF colonization assessment

Three root samples representative of each plant species were collected when found growing at a particular site. The root samples for every plant species were thoroughly mixed separately to give a single composite sample. Five subsamples of 100 g for every plant species were washed and fixed in formaldehyde–acetic acid–alcohol (FAA, 5:5:5, v/v) and stored at 4°C for detailed mycorrhizal studies.

The fine feeder roots were washed to remove the fixative and stained as per Phillips and Hayman (1970). The roots

Fig. 1 Climatic data showing monthly average mean temperature and monthly average precipitation recorded at Dingrah in Cholistan desert (from 1989 to 2002). *m* Relative period of drought



Processed from the raw data provided by Council of Research in water Resources, Regional Office, Bahawalpur

Site	Location	Dominant plant species	Date of sample collection	Habitat	Disturbance type and level
Fort Moujgarh (S1)	28°58'N, 72°06'E	Sporobolus iocladus, Cenchrus ciliaris, Cyprus rotundus	9 Sept 2002	Large interdunal sandy flat	Livestock grazing and cutting of vegetation by humans (3)
Fort Bajnot (S2)	28°04′N, 71°40′E	L. sciendicus, Cymbopogon jwarancusa, Calligonum polygonoides	10 Sept 2002	Large dunes of about 6–9 m height with small interdunal areas	Livestock grazing with cutting of vegetation. High human intervention due to nearby semi- settlement of nomads (3)
Fort Dinegargh (S3)	28°57′N, 71°50′E	L. sciendicus, Cymbopogon jwarancusa, Cenchrus ciliaris	14 Sept 2002	Sandy slope of about 30% inclination	Not disturbed during the last ten years. For details, see text (1)
Desert area of Baghdad- ul-Jaded campus, Islamia University, Bahawalpur Pakistan (S4)	29°22'N, 71°44'E	Cenchrus ciliaris, Calligonum polygonoides, Suaeda fruticosa	23 Sept 2002	Very small dunes with flat sandy areas	Being near the building of Cholistan Institute; less livestock and human intervention (2)

Table 1 Description of sites for plant species sampling, date of sample collection and level of disturbance in the Cholistan desert

Disturbance was rated as 1=no disturbance, 2=less disturbance, and 3=disturbed

were cut into 1-cm pieces. After mixing, 100 pieces of fine roots were taken randomly, mounted in lectophenol on a microscopic slide (10 on each slide) and analysed for the presence or absence of coils and vesicles. Percent frequency was calculated as $100 \times [(number of segments$ observed with coils or vesicles)/(total number of segmentstaken)]. Percentage root AM and dark septate endophyte(DSE) colonization values were obtained by the gridlineintersect method of Giovannetti and Mosse (1980) byexamining 100–150 intersects. These procedures werereplicated five times with five subsamples and averaged togive a single value. Heavily pigmented roots were cleared $with <math>H_2O_2$ before staining.

Soil analysis

The physicochemical parameters such as soluble cations and anions, total N and P contents, percentages of organic matter, water-holding capacity and moisture, pH and electrical conductivity (EC) values were measured and are given in Table 2. Statistical analysis

The average AMF spore densities and AM root colonization values were statistically analysed using SPSS software on Pentium III.

Results and discussion

The data presented in Table 1 reveal obvious differences in the floristic composition and species diversity. The vegetation was a typical xeric type and well adapted to the harsh conditions of low precipitation and high temperatures. The level of biotic intervention largely affected the species composition; e.g. the relative disturbed sites S1 and S2 had less species diversity as compared to less intruded S3 and S4 sites. Grasses were dominant and the main components of vegetation at all the study sites.

The vegetation at the S1 site was scanty due to overgrazing and was dominated by the perennial grasses, *Sporobolus iocladus* and *Cenchrus ciliaris*, growing mostly on the interdunal sandy flats in the Cholistan desert.

Table 2 Physicochemical analysis of soils from the four study sites

Sites	tes Soluble cations (mEq/100 g)			Soluble anions (mEq/100 g)		Total P (ppm) N (%)		Organic matter (%)	Water-holding capacity (%)	Moisture (%)	рН (1:2 Н ₂ О)	EC (mEq/ 100 g)	Textural classes
	Ca^{2+} and Mg^{2+}	Na ⁺	K ⁺	$\mathrm{CO}_3^- + \mathrm{HCO}_3^-$	Cl								
S1	1.98	0.31	0.54	2.62	1.04	0.20	0.81	0.34	25.2	2.28	8.66	109.6	Sandy
S2	1.49	0.71	0.54	2.48	1.21	0.23	0.81	0.40	20.1	2.38	8.36	86.1	Sandy loam
S3	1.78	0.26	0.11	1.79	3.27	0.25	2.83	0.75	25.9	2.964	8.89	138.4	Sandy loam
S4	5.39	0.89	1.01	2.84	1.89	0.34	3.52	0.94	29.5	3.023	8.21	160.7	Loamy sand

Lasiurus sciendicus was the single dominant plant species growing in association with large bushes of Cymbopogon jwarancusa and Calligonum polygonoides at site S2 near Fort Moujgarh (Table 1). This site comprised small dunes with flat areas covered with eolian sand up to 2–4 ft depth and was also disturbed due to over grazing. Site S3 was dominated by L. sciendicus, with Cymbopogon jwarancusa as the codominant. This site was inside an enclosed area and consisted of large shifting sand dunes of 4-5 ft high on the West and large sandy flats on the E and EN sides. The shifting dunes were mostly without any vegetation, but bushes of Calligonum polygonoides, Dipterygium glacum and Caparis deciduas were found to be growing at the base of the dunes. Since the area was fenced with barbed wire and inaccessible to the livestock and nomads, the vegetation was not disturbed and had relatively more diversity in floristic composition compared to the other sites (Table 1). Although site S4 was also not accessible by nomads and animals it exhibited a different floristic composition from S3. Here the dominant plant species was *Cenchrus ciliaris* with *Calligonum polygonoides* as the codominant (Table 1). This study also showed a considerable variation in the AMF colonization and spore densities in the various plant communities (Table 3). Heavy AMF colonization occurred in all the perennial grasses, especially the dominant ones such as *Sporobolus iocladus*, *L. sciendicus* and *Cymbopogon jwarancusa* in their respective communities. Most of the xerophytic plants that were examined displayed AMF colonization and AMF spores in their rhizospheres. These observations are consistent with those of Khan (1974), who first reported AMF in the plants growing in the deserts of Pakistan. Many studies by other researchers suggest that AM protect plants against stress caused by drought and increase their tolerance to drought (e.g. Ruiz-Lozano 2003), although the mechanism(s) requires further research by modern molecular techniques.

AMF colonization was of general occurrence in most of the plants in the S3 and S4 community types, whereas the dynamics of AM in the other rangeland communities, i.e. S1 and S2, were sporadic (Table 3). It is also evident that *Crotolaria burhia* (Papilionaceae) was non-mycorrhizal in S2 but mycorrhizal in S4. This may be attributed to the

Table 3 Mycorrhizal and DSE status of plant species collected from different rangeland habitats of Cholistan desert

Plant species	Family	Frequ	ency of coiling	Colonized root		Vesicles (%)	Spore no.
		in root system (%)		length (%)			
		AM	DSE	AM	DSE		
Acacia jacquemontii Benth.	Mimosaceae	15	0	23.2±0.5	0	6.6±1.0	106.4±0.5
Prosopis cineraria (Linn.) Druce.	Mimosaceae	0	5	0	$7.0{\pm}0.8$	0	103.6±0.4
A. persica (Burm.f.) Merrill.	Amaranthaceae	0	16	0	$10.0{\pm}0.8$	0	104.4 ± 0.8
Aristida mutabilis Trin. and Rupr.	Poaceae	20	54	56.6±0.9	41.4±1.0	27.0±1.0	22.2±1.0
Cenchrus biflorus Roxb.	Poaceae	8	10	52.0±0.7	41.2±1.2	45.2±0.8	97.2±0.5
Cenchrus ciliaris Linn.	Poaceae	68	28	85.2±1.4	26.8±1.0	63.4±1.0	285.8±1.6
Cymbopogon jwarancusa (Jones) Schult.	Poaceae	58	7	79.2±0.9	35.2±1.2	74.6±1.1	124.6±0.6
L. sciendicus Henrard.	Poaceae	0	32	93.4±1.5	25.2±1.2	90.4±0.5	97.2±0.7
Ochthochloa compressa (Forsskal.) Hilu.	Poaceae	2	6	32.6±1.2	44.4±1.5	22.2±1.4	33.2±0.5
Panicum antidotale Retz.	Poaceae	13	18	67.6±0.9	34.2±1.2	49.0±1.0	$343.0{\pm}0.4$
Panicum turgidum Forsskal	Poaceae	0	12	46.2±1.3	55.0±1.2	16.6 ± 1.0	63.0±1.3
Sacharum bengalense Retz.	Poaceae	32	58	94.8±1.2	27.2±1.0	66.6±0.9	144.4±0.9
Sporobolus iocladus (Nees. Ex. Trin.) Nees.	Poaceae	2	24	74.0±1.3	40.4 ± 0.5	19.2±0.9	73.4±0.5
Aristida hystricula Edgew.	Poaceae	24	0	54.4±1.3	6.2±1.0	26.4±0.7	6.8±0.3
Calligonum polygonoides Linn	Polygonaceae	6	10	65.2±1.3	23.8±1.3	34.4±0.6	34.4 ± 0.8
Caparis deciduas (Forsskal.) Edgew	Capparidaceae	0	14	18.0±0.7	26.6±1.2	4.6±0.5	56.2±0.6
Crotolaria burhia Ham. Ex. Bth.	Papilionaceae	0	3	4.6±0.5	20.8 ± 0.5	16.2±0.8	12.2±0.7
Cyprus rotundus Linn.	Cyperaceae	24	18	45.6±1.4	$6.0{\pm}1.0$	16.4±0.5	156.0±0.7
D. glacum Decne.	Brassicaceae	0	0	0	9.6±0.5	0	6.4±0.5
<i>E. prostrate</i> Ait.	Euphorbiaceae	0	0	0	0	0	20.2±0.7
Fagonia cretica Linn.	Zygophyllaceae	0	0	0	0	0	23.8±0.5
Haloxylon recurvum Bunge. Ex. Boiss.	Chenopodiaceae	0	20	34.8±1.3	24.8 ± 1.4	$14.0{\pm}1.7$	19.0±0.3
Haloxylon salicornium (Moq.) Bunge.	Chenopodiaceae	7	0	10.6±0.8	0	4.8±0.3	23.4±1.2
Salsola baryosma (Roem. et. Schult.) Dany	Chenopodiaceae	0	7	36.4±1.0	$13.0{\pm}0.7$	34.8 ± 0.8	38.6±0.6
Suaeda fruticosa (Linn.) Forsskal	Chenopodiaceae	0	0	15.8±1.2	0	11.6 ± 1.0	44.0±1.4
Heliotropium crispum Desf.	Boraginaceae	0	0	0	0	0	8.6±0.5
Leptadenia pyrotechnica (Forsskal) Decne.	Asclepidaceae	18	36	45.6±2.0	57.6 ± 0.9	10.2±0.5	86.8±0.7
P. rajputanae Blatt. & Hall	Compositae	0	0	0	5.0 ± 0.9	0	44.4±1.6
T. aphylla (Linn.) Karst.	Tamaricaceae	0	0	0	0	0	14.4 ± 0.9

Values for AM and DSE are means \pm SEM (N=5)

inoculum potential available and the genetic compatibility between the host and AMF at a particular site. Although root cortices of Cyprus rotundus growing in the S2 and S4 communities did not show any AMF colonization, those growing in the S1 community were heavily mycorrhizal (46%) and their rhizosphere contained 156 spores 100 g^{-1} soil. These results are in contrast with the commonly held view that sedges are generally considered to be nonmycorrhizal (Khan and Belik 1995; Powell 1975). Lovera and Cuenca (1996) reported AM colonization and functional arbuscules in the Cyperaceae in the tropical savannah of Venezuela. Many researchers have reported seasonality in the morphology and occurrence of AM colonization in members of Cyperaceae (Muthukumar and Udaiyan 2002; Meney et al. 1993). Contrary to the generally held view that the members of the families Amaranthaceae, Chenopodiaceae and Brassicaceae are non-mycorrhizal (e.g. Gerdemann 1968), we found them mycorrhizal to a lesser or greater degree in all the communities studied, except D. glacum (Brassicaceae) in S2 and S4 and Haloxylon saliconicum (Chenopodiaceae) in S3 (Table 3). AMF vesicles and intercellular hyphae were observed in their root cortices, in agreement with observations by Hirrel et al. (1978).

The present study reveals that Glomus was the dominant AMF genus in the arid deserts of Cholistan. These findings are in agreement with the results of other researchers in other arid or semi-arid areas (e.g. Lamont 1982; Pande and Tarafdar 2004). The mean AMF spore density per site in different plant community members was variable, with the highest in Cenchrus ciliaris occurring at S1 and S4, i.e. 508 and 286 spores 100^{-1} soil, respectively. In contrast, although AMF spores were recovered from the rhizospheres of Pulicaria rajputanae (Compositae) in the S2 and S4 communities, no AM colonization was detected in their cortices (Table 3). Similarly, roots of Aerva persica (Amaranthaceae) in the S3 community, Heliotropium crispum (Boraginaceae) in the S4 community, Tamarix aphylla (Tamariceae) in the S1 community, Haloxylon salicornium (Chenopodiaceae) and A. persica (Amaranthaceae) in the S3 and *Euphorbia prostrate* (Euphorbiaceae) in the S3 and S4 were non-mycorrhizal although their rhizospheres harboured AMF spores (Table 3). In a previous survey, Khan (1971) also reported a few AMF spores in the rhizospheres of Euphorbia spp. from the semideserts of Pakistan but no AMF colonization. In contrast, Nicolson (1960) reported high AMF colonization but no AMF spores in sand dunes and Kowalchuk et al. (2002) reported AMF present at a site even though their spores are absent from soils.

In addition to AMF colonization, roots were also commonly associated with DSE (Table 3). The real nature of DSE is still obscure so far; however, the DSE complex has been found to be highly diverse in structure including several taxa that probably belong to the ascomycetes (Carlsen 2002; Vrålstad et al. 2002). Many DSE fungal strains have been shown capable of utilizing organic molecules in their nutrition (Currah and Tsuneda 1993; Caldwell et al. 2000). If DSE colonization is thought to be mutualistic in nature then their importance for the host plant may be as equally important as AMF colonization for improving nutrition (Jumpponen and Trappe 1998; Caldwell et al. 2000).

Sporulation of different AMF species has been shown to be seasonal (McGee 1989) and to be influenced by host plant phenology (Pringle and Bever 2002). The collection of soil samples in the beginning of the hottest period of the year, when many desert plants are actively growing, may have influenced sporulation patterns. Carvalho et al. (2003) attributed the spatial distribution of AMF spores to plant composition and plant distribution and soil properties. There is a need to collect samples during different seasons and to use trap cultures grown on more than one host plant to determine AMF diversity (Bever et al. 2001). Further research is required to obtain a better understanding of whether and how plant community structure affects the AMF spore distribution and vice versa. Despite these deficiencies, our data give some preliminary assessment of the plant community structure and AM in rangeland habitats of Cholistan desert in Pakistan. The AMF survey reported here offers an important starting point from which to analyse AMF community structure in different phytosociological habitats and land uses of Cholistan desert. Any future attempts to manage desert plant ecosystems would benefit from knowledge about mycorrhizas in the indigenous major grasses.

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