Arbuscular mycorrhizal fungi, *Glomus* spp. (*Glomeromycetes*), associated with drought tolerant plants of the Indian Thar desert

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Abstract: Arbuscular mycorrhizal fungi are well known for their benefit to the plants surviving under harsh environmental conditions. To understand better their interaction with arid region plants various arbuscular mycorrhizal species are to be studied in greater detail. Here, the aim was to investigate the species diversity of the mycorrhizal genus *Glomus* associated with some important plants of the Indian Thar Desert. Identification and characterization were made on spore morphology. *Glomus aggregatum* turned out to be the most frequent and abundant *Glomus* species in the arid Indian Thar Desert.

Zusammenfassung: Arbuskuläre Mykorrhizapilze sind bekannt für ihren Nutzen für das Überleben von Pflanzen unter rauen Umgebungsbedingungen. Um ihre Wechselwirkungen mit Pflanzen arider Regionen besser zu verstehen, sind verschiedene arbuskuläre Mykorrhiza-Arten näher zu untersuchen. Ziel der vorliegenden Arbeit war es, die Artenvielfalt der mykorrhizabildenden Gattung *Glomus*, die mit einigen wichtigen Pflanzen der indischen Wüste Thar assoziiert ist, zu untersuchen. Identifizierung und Charakterisierung basieren auf der Sporenmorphologie. *Glomus aggregatum* stellte sich als die häufigste und am reichlichsten auftretende *Glomus*-Art der ariden indischen Thar-Wüste heraus.

In many arid and semiarid regions of the world, drought limits plant growth. The incorporation of the factors enabling plants to withstand drought stress would be helpful to them under drought conditions (AL-KARAKI & al. 2004). Arbuscular-mycorrhizal fungi (AMF) are essential for vegetation in arid regions, as they improve plant water relations and thus increase the drought resistance of host plants (QUEREJETA & al. 2006), they improve disease control (GRAHAM 2001), and they increase mineral uptake (SIEH & al. 2013). Improved water uptake by the plant is also related to increase some mineral ions essentially required for the metabolism in the plants growing in these adverse conditions (BELTRANO & al. 2013). The abilities of specific fungus-plant associations to tolerate drought are of great interest.

Glomus TUL. & TUL. is a large genus of AMF with 90 species (MUKERJI 1996). The name *Glomus* (=a ball of yarn) possibly refers to the sometimes rounded, globose or subglobose spores of these fungi. *Glomus* reproduces asexually and spores are formed at the tip of growing hyphae either in the host or in the soil. The chlamydo-

spores germinate in the soil until they come in contact with the roots of the host plants. Then the hyphae penetrate the epidermal cells and grow between the cortical cells of the root. Inside the cortical cells, the fungus forms arbuscules, which are highly branched, and also swollen structures, called vesicles.

The widespread interest in AM fungi makes it desirable to describe precisely the characters of the individual fungal species or isolate. There is very little or only scattered information available in the literature as far as identification, characters and microphotography of different species of the genus *Glomus* are concerned. Hence, the purpose of present study is to enlist and characterize *Glomus* species associated with rhizosphere soils of some important Indian Thar Desert plants.

Materials and methods

Site description

The present study was conducted in arid regions of the Thar Desert of India, mainly covering the districts of Jaisalmer, Barmer, Bikaner and Jodhpur (26.9844° N to 71.0017° E) of Rajasthan state. Soils of the region are generally sandy to sandy loamy in texture with poor nutrient status and low water holding capacity. A bioclimatic and environmental limitation makes the region deprived of vegetation. Small trees and shrubs occur mainly in small clumps scattered more or less openly.

Collection of soil samples

Composite soil samples from rhizosphere soil of different herbs, shrubs and trees of different age growing in desert areas were collected. The samples were taken after removing the top layer and by digging out a small amount of soil close to the plant roots up to a depth of 15–30 cm. The samples were brought to the laboratory and stored at 4 $^{\circ}$ C to assure the viability of collected mycorrhizal spores.

Extraction of mycorrhizal spores from the rhizosphere soil

Isolation of *Glomus* spores was performed by the wet sieving and decanting technique of GERDEMANN & NICOLSON (1963). 50 g of soil was soaked in 500 ml of water for 2 h. The supernatant was then decanted through a gradient of sieves with pore sizes ranging from $45-350 \mu m$ arranged one above another in an ascending order. Each sieve was then washed in water and filtered through Whatman No.1 filter paper. Filter paper was further observed under stereo binocular microscope for the presence of various kinds of spores. Spores were subsequently picked with a hypodermal needle and mounted on polyvinyl lactic acid alcohol (PVLA) and examined under an optiphot-2 (Nikon) microscope. Spores and other spore structures were microphotographed.

Identification of collected mycorrhizal spores

For identification of *Glomus* spores, the conventional morphological characters, i.e., shape, size and colour of spore, wall structure, surface ornamentation of spore, suspensor, subtending hyphae and arrangement of spores in sporocarp were considered. They were identified up to the species level with the help of the synoptic keys of TRAPPE (1982), WALKER (1983), SCHENCK & PEREZ (1990), MORTAN & BENNY (1991), MUKERJI (1996) and MORTAN & REDECKER (2002).

Results

Sixteen species of *Glomus* were isolated and identified by their specific shape, size, spore wall, ornamentation of the spore wall and subtending hyphae. Descriptions of individual species are given below with their microphotographs.



Fig. 1. *Glomus* species isolatd from rhizosphere soil of plants growing in Indian Thar Desert. A *Glomus aggregatum*, B *Glomus fasciculatum*, C *Glomus albidum*, D *Glomus fulvum*, E *Glomus ambisporum*, F *Glomus constrictum* (×100), G *Glomus deserticola*, H *Glomus geosporum*. Bars: A, B, E = 250 μ m, C, D, G, H = 150 μ m.

Glomus aggregatum SCHENCK & SMITH

Chlamydospores formed in loose cluster or in sporocarp without a peridium. Sporocarp of variable size ranging from 650–1400 μ m. Sporocarp light brown to brown. Chlamydospore globose, subglobose, obovate or cylindrical, 50–100 μ m, brown. Spore walls brown, consisting of an outer wall slightly thicker and lighter in colour than the inner wall. Spore contents confluent with hyphal contents in young spores, but separated from hyphae in older spores by an inner spore wall, spore not occluded by hyphal wall thickening (Fig. 1 A).

Glomus fasciculatum (THAXTER Sensu GERD) GERD & TRAPPE

Chlamydospores borne free in soil or dead rootlets, in loose aggregation or in small compact clusters, and in sporocarp. Sporocarp irregularly globose or flattened, tuber-culate, brown, peridium absent. Chlamydospore globose, subglobose to obovate 75–150 μ m. Spore walls highly variable in thickness, yellow or yellow brown. Hyphal attachment occluded at maturity, hyphae branched and producing new spores at the tip (Fig. 1 B).

Glomus albidum WALKER & RHODES

Sporocarp unknown. Chlamydospore yellowish to brownish-yellow. Spore with one subtending hypha borne singly in the soil on coenocytic hyphae. Mature spore 150–200 μ m, globose to subglobose, occasionally ovoid or irregular. Spore walls continuous with hyphal walls consisting of an outer hyaline wall and light yellow inner wall. Subtending hyphae 2–walled, usually straight and simple, but sometimes constricted at the spore base or expanded by thickening of the outer wall (Fig. 1 C).

Glomus fulvum (BERK. & BROOME) TRAPPE & GERD.

Chlamydospore pale, yellowish. Spore oblong elliptical to oval or even subpyriform, rarely nearly spherical, $125-150 \mu m$. Attachment of the spore often sublateral. Sporogenous hypha often, though by no means invariably, somewhat narrow just below the point of attachment (Fig. 1 D).

Glomus ambisporum SMITH & SCHENCK

Sporocarp dark brown to black, subglobose to highly variable in shape consisting of a single layer of chlamydospores originating from a central core of thick interwoven hyphae, peridium absent. Chlamydospores produced singly in sporocarp in soil or aggregated around roots, dark brown to black. Sporocarp originating from thick-walled hyphae. Spores with three walls. Inner wall membranous, middle wall laminate, dark brown to black and outer wall reticulate and light brown. Spore globose to subglobose. Spore size 190–270 μ m. Hyphal attachment of spore simple (Fig. 1 E).

Glomus constrictum TRAPPE

Chlamydospore naked, formed singly or in loose cluster in soil, subglobose to globose, $150-330 \mu m$, dark brown to black, shiny-smooth. Spore wall thick, dark brown, two layered, attachment occluded by wall thickening. Attached hypha straight to recurved and with the following features appearing in sequence away from the spore; point of



Fig. 1 cont. I Glomus macrocarpum, J Glomus radiatum, K Glomus halonatum, L Glomus reticulatum, M Glomus invermaium, N Glomus monosporum, O Glomus warcupii, P Glomus multisubstensum. Bars: I, $J = 250 \mu m$, $K-P = 150 \mu m$.

attachment with dark brown walls; just beyond the point of attachment the hypha constricted; just beyond the constriction the hypha inflated with yellow to yellow brown walls (Fig. 1 F).

Glomus deserticola TRAPPE, BLOSS & MENGE

Spores borne singly or in loose fascicles in soil or within the roots, globose to subglobose, $115-185 \mu m$, shiny smooth, reddish brown with thick wall. Attached hypha 6–12 μm in diam., cylindrical to occasionally somewhat funnel shaped. The interior of the spore wall at the hyphal attachment thickened and plugged by dense materials. Outer wall of spore continuous with subtending hyphae. Spore wall divided into outer yellow, middle brownish and inner thick blackish wall (Fig. 1 G).

Glomus geosporum (NICOL & GERD.) WALKER

Chlamydospore formed singly in soil, globose to subglobose or broadly ellipsoid, 110–290 μ m, and of dull appearance, yellow-brown to dark yellow brown. Spore wall thick, 3-layered, with an outer wall, laminated middle wall and yellow to yellow brown inner wall. Spores with one straight to recurved, simple to slightly funnel shaped hyaline subtending hypha. Spore contents with few oil droplets when young (Fig. 1 H).

Glomus macrocarpum TUL. & TUL.

Sporocarp dark brown, globose to subglobose, sometimes irregular, consisting of a single or double layer of chlamydospores originating from a central core of thick interwoven hyphae. Peridium absent. Spores usually slightly longer than wide, subglobose, 150–250 μ m. Spore wall composed of two distinct layers, spores tapering to the point of attachment of the single persistent hypha. The inner wall at maturity thickened to occlude the pore of the attached hypha. Spores characteristically bearing a straight, long subtending hypha which may extend up to 100 μ m before branching or breaking (Fig. 1 I).

Glomus radiatum (THAXTER) TRAPPE & GERD.

Sporocarp generally flattened and lobed, firmly attached to roots or organic matter. Peridium absent or present. Sporocarp developed acrogenously. Chlamydospores at or near the surface thin walled and vesicular, becoming progressively thicker walled in direction of the sporocarp base, ellipsoid to oblong, ovoid or rarely globose, arranged in a distinct radial pattern, light brown, 150–250 μ m, wall thick laminate (Fig. 1 J).

Glomus halonatum (ROSE & TRAPPE)

Chlamydospore borne singly in soil or in small loose clusters of 3-7 spores, globose to subglobose, $200-280 \ \mu m$ in diam., light brown to brown. Spore wall thick, outer wall brown, inner wall hyaline and laminated. Attached hypha straight, hyaline. Spore contents of oil globules of varying size (Fig. 1 K).

Glomus reticulatum BHATTACHARJEE & MUKERJI

Chlamydospore borne freely and singly in the soil, not known to occur in sporocarp. Spore light brownish to black, globose, 130–220 μ m in diam. Clearly differentiated into an outer and inner wall. Outer wall thick, two layered and fissured, inner wall with regular geometric reticulate marking on its outer surface. Subtending hypha funnel shaped, wide at the point of attachment (Fig. 1 L).

Glomus invermaium HALL

Spores globose, light brown to brown, formed in loose sporocarp. Peridium lacking, spore wall double, outer wall hyaline to subhyaline, thick, inner wall light brown to brown, thick. Outer wall extending down the subtending hypha. Subtending hyphae broad, nonseptate and slightly curved (Fig. 1 M).

Glomus monosporum GERD. & TRAPPE

Sporocarp 350–650 μ m, globose to ellipsoid, containing mostly 1, occasionally 2, or rarely 3 chlamydospores. Chlamydospores 140–320 μ m, generally subglobose or rarely ellipsoid. Spore walls thick, dull brown to pale brown, outer wall and inner wall extending into subtending hypha. Spores partially enclosed and with visible pore. Subtending hyphae recurved and oppressed to spore wall (Fig. 1 N).

Glomus warcupii MC. GEE

Chlamydospore globose to subglobose. Spore size $150-200 \ \mu m$ in diam. Spore wall thick, outer wall brownish, irregular, unequally thick, more thickened at spore base, inner wall laminated. Subtending hyphae cylindrical slightly curved with a typical lateral projection (Fig. 1 O).

Glomus multisubstensum MUKERJI, BHATACHARJEE & TEWARI

Chlamydospore singly in the soil or in compact clusters. Spore golden brown, 150–210 μ m. Spore wall thick, outer wall regular, inner wall laminated. Subtending hyphae 2 in number, attached at one end of the spore, hyaline to yellow, wall thick (Fig. 1 P).

Discussion

In this study we determined different *Glomus* species associated with diverse plants of an arid region, the Indian Thar desert. AMF association is the essential requirement for the survival and establishment in these adverse conditions. Among the AMF, *Glomus* species are the predominant organisms in neutral to alkaline soils of agricultural interest (TEIXEIRA-RIOS & al. 2013). *Glomus* species are an important component of the soil in all the types of environments and are beneficial for plant growth and development by increasing the nutrient uptake. These species are also important to contribute substantially to the establishment, productivity and longevity of natural and man made ecosystem. SCHENCK & KINLOCK (1980) found that the soil or the plant types were more or less important factors for such an existence of *Glomus*. NAROLIA & al. (2008) recorded the dominance of *Glomus* species from rhizosphere soil of agricultural crops. In the present study authors found that *Glomus* species are also dominant in Desert soil.

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