

OCCURRENCE AND SYSTEMATICS OF ARBUSCULAR MYCORRHIZAL FUNGI ASSOCIATED WITH SORGHUM

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The rhizosphere soils of sorghum (*Sorghum vulgare*) were collected from kharif (rainy) and rabi (post-rainy) seasons during 2004 and 2005 from the fields of National Research Centre for Sorghum, Rajendranagar, Hyderabad, India. The samples were screened to study the AM (Arbuscular Mycorrhizal) fungi associated with the roots of host plants. Altogether, nineteen species of AM fungi representing three genera namely *Acaulospora*, *Glomus* and *Gigaspora* were isolated. The genus *Glomus* dominated among the entire AM fungal flora by representing twelve species, followed by five species of *Gigaspora* and two of *Acaulospora*. *Glomus mosseae*, *G. fasciculatum*, *G. constrictum* and *G. monosporium* were detected frequently from the rhizosphere samples of which *G. fasciculatum* was found to be predominant in almost all the soil samples, whereas *G. multisubtensum* was reported only in the months of October and November. The other species of AM fungi like *Glomus aggregatum*, *G. monosporium*, *G. caledonium*, *G. deserticola*, *Gigaspora gigantea*, *Acaulospora laevis* and *A. foveata* were observed in few numbers that too in certain periods of study.

Keywords: AM fungi; Sorghum; Systematics; Taxonomy.

Introduction

There are many reports on the studies of taxonomy and systematics of arbuscular mycorrhizal (AM) fungi from different parts of the world. AM fungi are geographically ubiquitous occurring over a broad ecological range in plants from arctic to desert-environments. The available information indicates that these obligate symbionts have been reported from cultivated and non-cultivated soils, moist forests, open wood-lands, scrub, savanna heaths, grasslands, sand-dunes¹, semi-deserts and coal wastes²⁻⁵. Gerdemann⁶ reported that mycorrhizal condition is the rule under natural situations and non-mycorrhizal is the exception. AM fungi are associated with bryophytes, pteridophytes, gymnosperms and angiosperms⁷. However, families of cruciferae, chaenopodiaceae, commelinaceae, cyperaceae, pumeriaceae, polygonaceae and utricaceae do not harbour mycorrhizal association. Few members of chaenopodiaceae^{8,9} and cruciferae were reported to have mycorrhizal association. Khan¹⁰ reported the absence of AM in halophytes and few xerophytes in Pakistan. Most of the plants which lack AM are xerophytes, for example: capparidaceae, nyctaginaceae, portulacaceae and phytolocaceae^{7,10}. Black¹¹ reported that AM fungi were absent in a number of cosmopolitan hydrophytic families. The occurrence of AM fungal association was confirmed from India in the tropics¹². Mishra and Sharma¹³ and

Kharbuli and Mishra¹⁴ reported the occurrence of AM fungi from north eastern regions of Himalayas and Trappe¹⁵ from forage plants of arid and semi-arid range lands. Hayman¹⁶ reported from plants growing in arctic, temperate and tropical regions, however, their abundance being greatest in tropical regions. Howeler *et al.*¹⁷ have discussed the practical aspects of mycorrhizal technology in some tropical crops and pastures. The association of AM fungi has been recorded in Guayule¹⁸, orchards and plantation crops like citrus, tea, coffee and rubber¹⁹, tomato²⁰, maize²¹, raspberry²², coconut²³, apples²⁴ and cocoa²⁵. AM fungal association has been reported in field crops^{3, 26}, cereal crops²⁷⁻²⁹, legumes³⁰⁻³², vegetable crops³³⁻³⁵, in some oil seeds^{32, 36, 37}, herbaceous plants³⁸, mangrove plants³⁹ and xerophytic plants⁴⁰. Mukerji *et al.*⁴¹ discussed in detail the concepts and advances related to AM fungi from Indian sub-continent.

In spite of having good taxonomic account about AM fungi, the problems seem to be many, as the fungi appear either in the form of chlamydo spores, azygospores, or very rarely as zygo spores. In view of the importance of the problem the present study was undertaken to know the occurrence and systematics of AM fungi associated with sorghum.

Materials and Methods

The rhizosphere soils of different cultivars of sorghum

Table 1. Occurrence of AM fungal species in the rhizosphere soils of sorghum during kharif and rabi seasons.

AM fungal species	June	July	Aug	Sept	Oct	Nov	Dec	Jan	Feb
<i>Acaulospora laevis</i>	+	+	+	+	+	-	-	-	+
<i>A. foveata</i>	+	+	+	+	-	+	-	-	+
<i>Glomus mosseae</i>	+	+	+	+	-	-	+	-	+
<i>G. fasciculatum</i>	+	+	+	+	+	+	+	+	+
<i>G. aggregatum</i>	+	+	+	-	+	+	-	-	+
<i>G. constrictum</i>	+	+	-	+	+	-	-	+	-
<i>G. monosporum</i>	+	+	+	+	-	+	+	-	+
<i>G. geosporum</i>	-	+	-	-	+	+	-	+	+
<i>G. heterogenum</i>	-	-	-	-	-	-	+	+	-
<i>G. diphanum</i>	-	+	-	-	+	+	+	-	-
<i>G. caledonium</i>	+	-	+	-	-	+	+	+	-
<i>G. deserticola</i>	+	-	+	+	+	+	-	-	+
<i>G. multisubstensum</i>	-	-	-	-	+	+	-	-	-
<i>G. rubiforme</i>	+	-	-	-	+	+	-	-	+
<i>Gigaspora gigantea</i>	+	+	-	+	+	-	+	+	-
<i>Gigaspora</i> spp.	+	+	-	+	+	-	-	+	+
<i>Gigaspora</i> spp.	+	-	+	-	+	+	-	+	+
<i>Gigaspora</i> spp.	-	-	+	-	-	+	-	-	-
<i>Gigaspora</i> spp.	+	-	-	+	-	-	+	-	-

were surveyed in both kharif (June/July-October/November, 2004) and rabi (October/November-January/February, 2005) seasons at regular intervals by collecting the samples from the fields of National Research Centre for Sorghum, Hyderabad, Andhra Pradesh. The rhizosphere soil samples were taken with the help of a widger by lifting up gently a block of soil with the plant roots intact. The soils were collected in fresh polythene bags and stored in refrigerator at 5°C for further study. The soils were thoroughly investigated for the qualitative and quantitative estimation of AM fungi using the modified wet sieving and decanting technique^{42, 43}.

100 g of soil was taken and mixed in 400 ml of luke-warm water in a large beaker until all soil aggregates disperse to leave a uniform suspension. A pinch of sodium hexametaphosphate was added for easy dispersal of soil aggregates: 710 µm, 420 µm, 250 µm, 105 µm and 53 µm sieves were arranged in the descending order with 710 µm at the top and 53 µm at the bottom. 710 µm sieve was used for the removal of large organic matter and roots. The

contents of the beaker were decanted through the sieves. The process was repeated 4 to 5 times till only the sand and stones were left in the beaker. The debris retained on the sieves was carefully collected into the beaker with the help of a level pipe separately through single synthetic fibered imported white cloth. The imported mesh was kept in a petridish with some water. The AM fungi associated with each filter cloth after sieving at various levels were scanned, picked up with micro-needle and mounted on slides in polyvinyl lactic acid as mounting medium. Later, all such slides were observed carefully under high power research microscope for segregation into general and followed by species identification. Permanent slides with PVL (Polyvinyl alcohol-lactophenol) as mountant were prepared which allow slides to remain useable for years. The AM fungi were identified using the monographs of Hall⁴⁴⁻⁴⁶, Mehrotra and Bajjal⁴⁷, Morton⁴⁸, Raman and Mohan Kumar⁴⁹, Schenck and Perez^{50, 51}, Trappe⁵² and Walker⁵³⁻⁵⁵. The important criteria for identifying and classifying the AM fungi are:

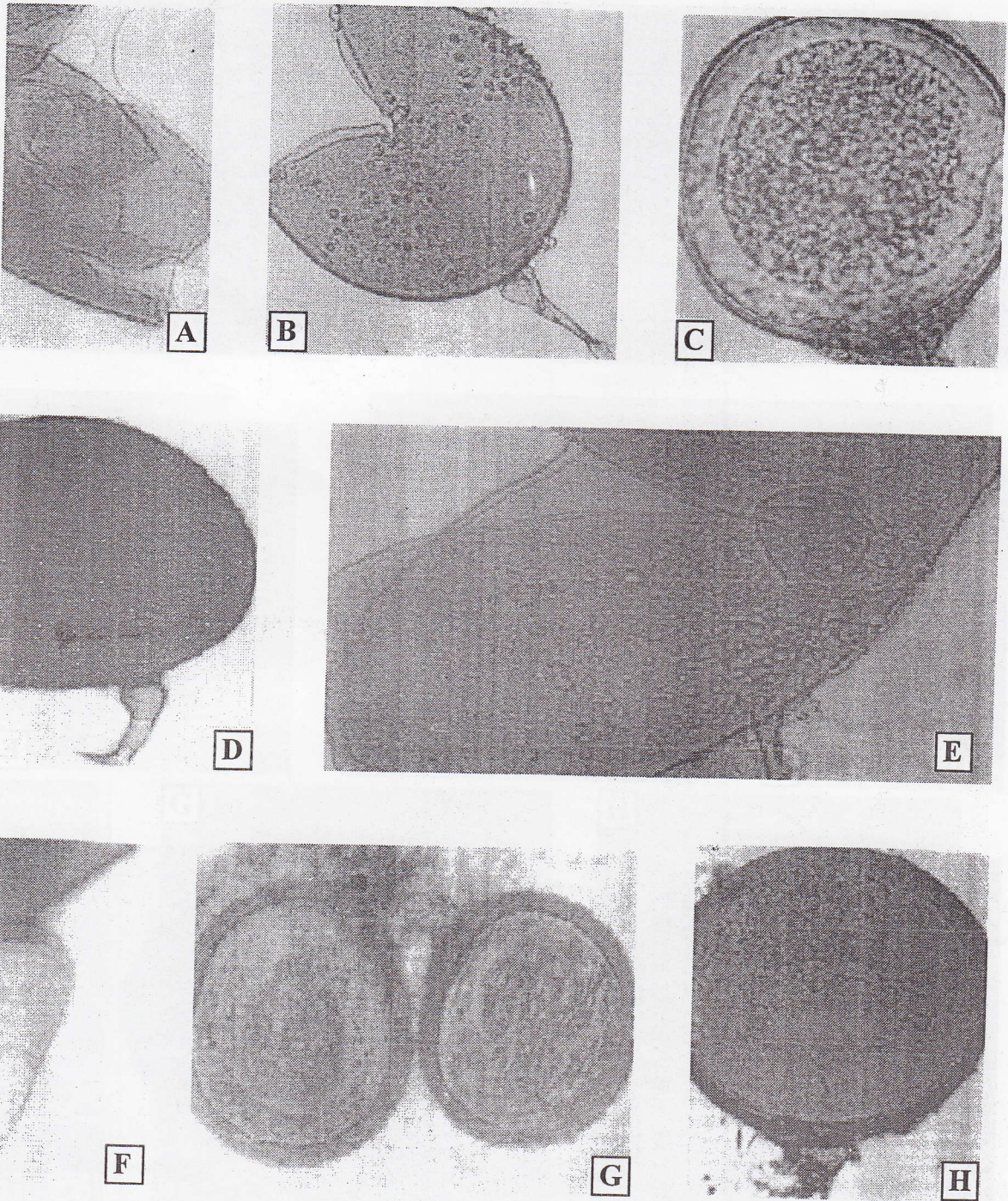


Fig. 1 (A-H). (A) Azygospore of *Gigaspora gigantea* (X100); (B) Azygospore of *Gigaspora* sp. (X200); (C) Single spore – *Acaulospora laevis* (X200); (D) Azygospore of *Gigaspora* sp. (X200); (E) Azygospore of *Gigaspora* sp. (X450); (F) Bulbous portion of subtending hypha (X450); (G) Spores of *Acaulospora foveata* (X200); (H) Azygospore of *Gigaspora* sp. (X200)

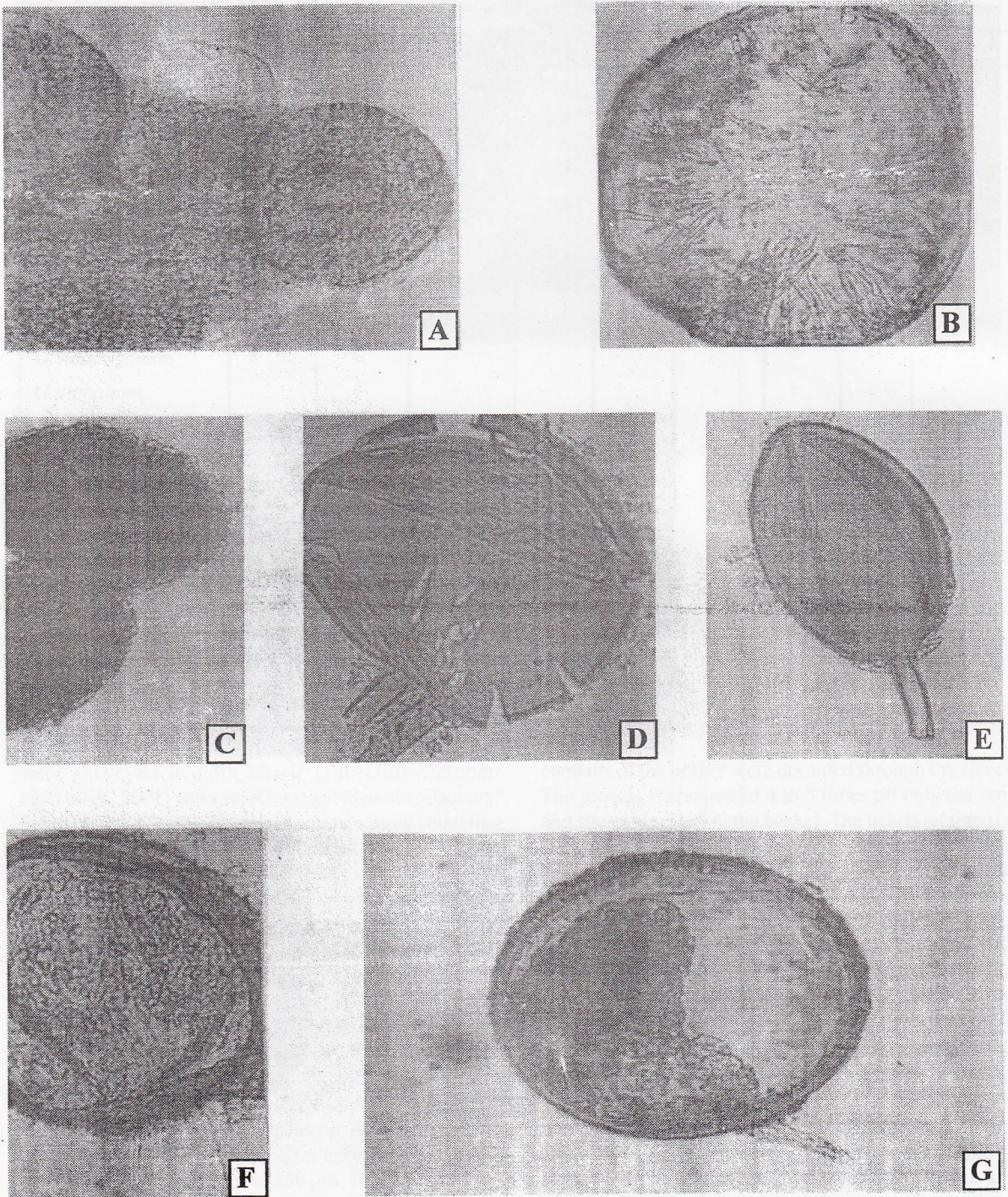


Fig. 2 (A-G). (A) Spore cluster of *Glomus heterogenum* (X200); (B) Single spore of *Glomus fasciculatum* (X450); (C) *Glomus multisporum* (X200); (D) Chlamydospore of *Glomus multisubtensum* (X200); (E) Single spore of *Glomus geosporum* (X200); (F) Single spore of *Glomus mosseae* (X200); (G) Single spore of *Glomus monosporum* (X200)

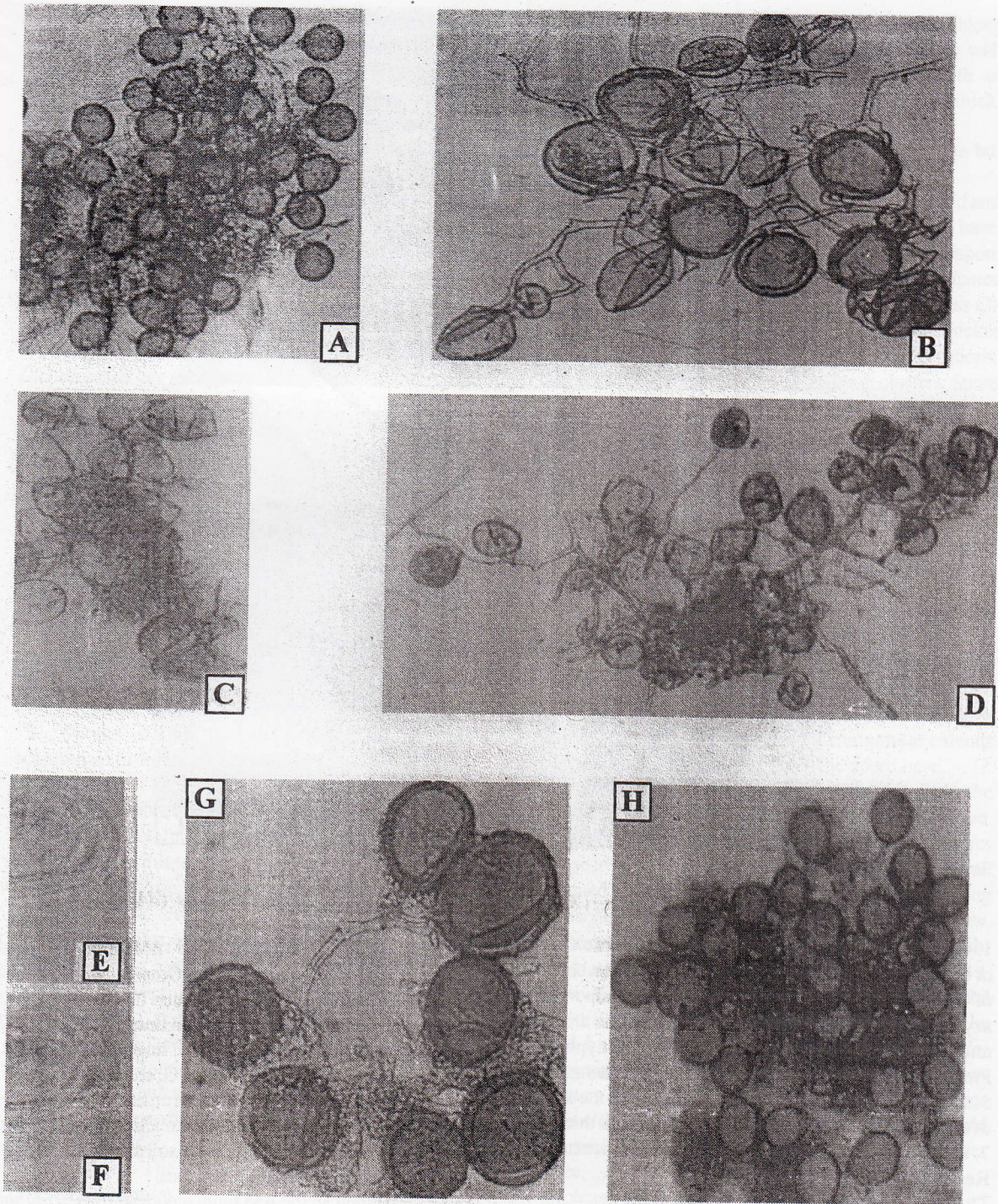


Fig. 3 (A-H). (A) Sporocarp of *Glomus caledonium* (X100); (B) Sporocarp of *Glomus fasciculatum* (X100); (C) Spores of *Glomus diphanum* (X100); (D) Sporocarp of *Glomus deserticola* (X100); (E, F) Single spores of *Glomus diphanum* (X200); (G) Sporocarp of *Glomus constrictum* (X100); (H) Sporocarp of *Glomus aggregatum* (X100)

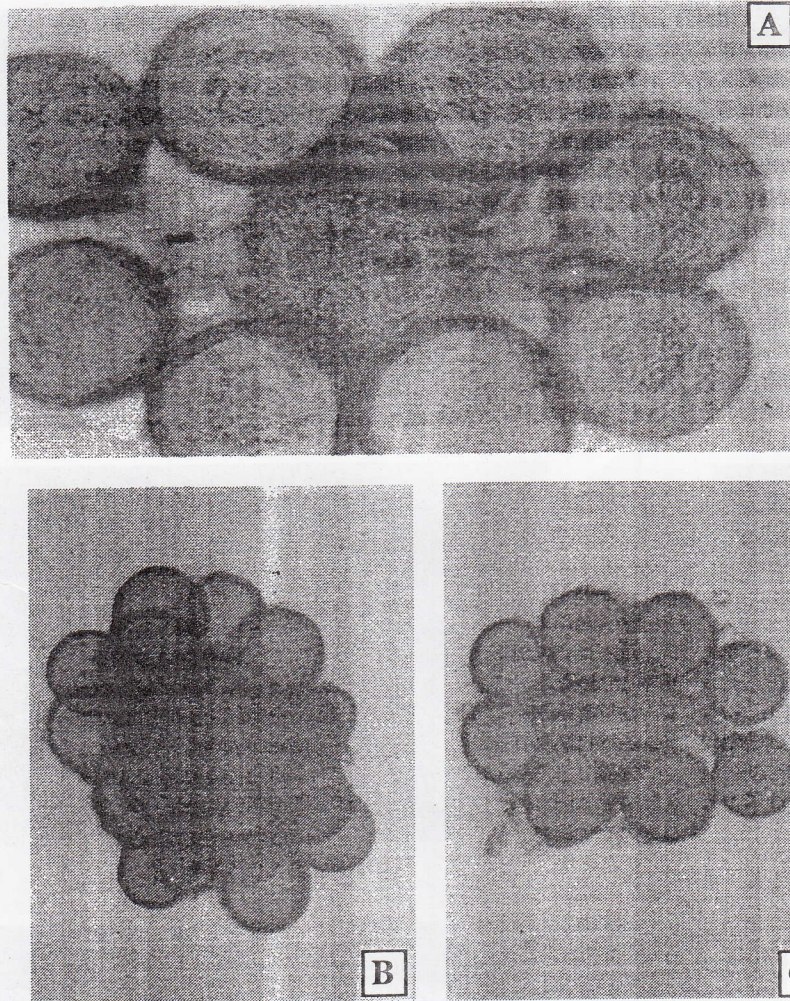


Fig. 4. (A-C), (A) Sporocarp of *Glomus rubiforme* (X200); (B & C) Sporocarp of *Glomus rubiforme* (X100)

1. Occurrence of spores either singly or in loose clusters or in tightly packed sporocarps with or without a covering of hyphae enclosing them; 2. Nature of zygospores and azygospores; 3. The manner in which the spores are borne and arise on the hyphae or branching of hyphae; 4. Presence or absence of basal swellings, germinating shield; 5. Size, shape, colour and ornamentation on the surface of the spores; 6. Number of wall layers and their thickness; 7. Extra-matrical vesicles and other minor characters.

Results and Discussion

The rhizosphere soils of sorghum were collected during kharif and rabi seasons from the fields of National Research Centre for Sorghum, Rajendranagar, Hyderabad and screened to study the AM fungi associated with the

roots of host plants. Altogether, 19 species of AM fungi representing 3 genera namely *Acaulospora*, *Glomus* and *Gigaspora* (Table 1) were isolated. The genus *Glomus* dominated among all the AM fungal flora representing 12 species, followed by *Gigaspora* 5, and *Acaulospora* 2. *Glomus mosseae*, *G. fasciculatum*, *G. constrictum* and *G. monosporium* were detected frequently from the rhizosphere soils samples but *G. fasciculatum* was found to be predominant in almost all the soil samples collected from both the kharif and rabi seasons.

Among all the AM fungi reported *G. fasciculatum* was reported throughout the study i.e., both in kharif and rabi. The other species of *G. aggregatum*, *G. monosporium*, *G. caledonium*, *Gigaspora gigantea* and *Acaulospora*

laevis and *A.foveata*, whereas *Glomus multisubtensum* was reported only in the months of October and November through this study. *Glomus rubiforme*, *Glomus heterogenum* and *G.deserticola* were observed rarely. It is clear from the study that in general, more AM fungal propagules were recorded with rhizosphere soils of sorghum during kharif than rabi season. It is also evident from the study that there was a gradual increase in the AM propagules during early stages (up to 90 days) of plant growth. There are also fluctuations in the number of AM fungi in different months and such variations are common for all the fields and seasons.

Dichotomous key has been given below followed by the segregation of species collected under each genus.

Dichotomous key for separation of genera:

- 1a. Spores produced as chlamydo-spores - 2
- 1b. Spores not produced as chlamydo-spores - 3
- 2a. Spores formed singly in soil or in sporocarps, spores not radiating from a central core of hyphae- *Glomus*.
- 3a. Azygospores formed near or below a swollen hyphal terminus - 4
- 3b. Azygospores formed on swollen hyphal terminus-5
- 4a. Spores formed laterally on hyphae, below a swollen hyphal terminus - *Acaulospora*
- 4b. Spores formed within the hypha, below a swollen hyphal terminus - *Entrophospora*
- 5a. Spores with 2 or more wall groups, the inner containing a coriaceous or membranous wall - *Scutellospora*
- 5b. Spores of only 1 wall group, auxilliary cells echinulate or finely papillate - *Gigaspora*

Acaulospora -

1. Spores smooth with three walls; outer wall 2-4 μm thick and inner two hyaline membranous walls - *A.laevis*
2. Spores with pitted surface. Pits with round to oblong depression with rounded bottoms separated by ridges, 1-12 μm broad- *A.foveata*.

1. *Acaulospora laevis* Gerdemann and Trappe: Sporocarps unknown. Spores formed singly in the soil, sessile, spores smooth, 181.60 μm , globose, deep yellow brown to reddish-brown at maturity. Spore wall continuous, consisting of 3 layers, a rigid yellow brown to reddish brown outer wall and two hyaline inner membranes. Spores contain oil globules and the contents of the spores are globose to somewhat polygonal (Reticulate in optical section) (Fig. 1C).

Critical Notes - The present species closely resembles the original description.

2. *Acaulospora foveata* Trappe & Janos- Sporocarps are unknown. Azygospores are formed singly in the soil, sessile, spores globose, 256.7 μm in diameter. Yellowish

brown, spore surface uniformly pitted with round to oblong or occasionally irregular depressions. Outer spore wall yellowish to reddish brown 13.63 μm thick with an adherent but separable, hyaline inner layer 3.4 μm thick. Spore contents of small hyaline guttles (Fig. 1G).

Critical Notes- It resembles the original description but differs in having comparatively smaller spores.

Glomus - 1. Chlamydo-spores are globose to ellipsoid and yellow brown. Chlamydo-spores have funnel shaped base, which merges into the subtending hyphae. Chlamydo-spore walls are 3-6 μm thick with very thin outer wall.... *Glomus mosseae*; 2. Spore walls highly variable in thickness (3-17 μm), hyaline to yellow or yellow to brown, the thicker walls often minutely perforate with thickened projections.*G.fasciculatum*; 3. Spores arranged in clusters. Spore contents confluent with hyphal contents on young spores but separated from hyphae on older spores by inner spore wall; pore not occluded by hyphal wall thickening. Spores produced inside the spores by internal proliferation.*G. aggregatum*; 4. Spores naked, singly or in loose clusters, sub globose to globose. One layered seeming two layered. Yellow brown when young to dark brown when mature. Spore walls 7-12 μm thick.*G. constrictum*.

5. Inner wall laminate with minute, abundant to scattered echinulations that protrude into the outer wall....*G. monosporum*; 6. Spore contents cut off by a septum that protrudes slightly into the subtending hypha. Spores with one straight funnel shaped subtending hypha with yellow to dark yellow to brown. Wall thickening that extends 30-100 μm along the hypha from the spore base.*G. geosporum*; 7. Spore walls and spore contents lacks the pigment formation, due to which the spores are hyaline.

....*G. diphanum*; 8. Spores single or in sporocarps. Spores globose to sub-globose, ellipsoid and irregular. Outer wall thickened at the hyphal attachment and extending along the attached hyphae for some distance. *G. caledonium*.

9. Spores single or in loose fascicles. Globose to sub-globose, shiny, smooth, yellow to brown or reddish brown. Interior of the spore wall at the hypal attachment thickened at maturity to form an inner rounded collar, appear to be closed by a membranous septum. *G. deserticola*;

10. Compact chlamydo-spores formed singly in the soil or in clusters of 5-8 spores. Subtending hyphae 2-4 in number.*G. multisubtensum*; 11. Spore wall often perforated and often with thick, perforated projections on the inner surface. *Glomus rubiforme*.

1. *Glomus mossea* (Nicolson and Gerdemann) Gerdemann and Trappe - Spores are globose to ellipsoid to obovoid or irregular and yellow to brown. Spore walls are 2-7 μm thick with a very thin outer wall. Peridium loosely

interwoven, irregularly branched, septate hyphae 2-12 μm , walls 0.5 μm , frequently anastomosing to form a thin network, enclosing chlamydospores. Spores with one or occasionally two funnel shaped bases 2-30 (-50) μm in diameter, derived from subtending hypha by a curved septum. Walls 2-7 μm thick, often barely perceptible hyaline outer membrane and a thick brownish yellow, inner wall (Fig. 2F). *Critical Notes*- This spore agrees with the original description in all the characters.

2. *Glomus fasciculatum* (Thaxter Sensus Gerd) Gerdemann and Trappe- Chlamydospores are formed in loose aggregations in the soil, in small compact clusters and in sporocarps. Sporocarp size 438.15 X 353.32 μm in diameter, irregularly globose, yellowish brown; peridium absent. Chlamydospores 48.35-138.63 μm when globose, 72-135.13 X 53-129.41 μm in diameter when sub-globose. Smooth or seemingly roughened from adherent debris. Spore walls 3.6-13 μm thick. Spores are hyaline to yellow or yellow brown, the thicker walls often minutely perforate with thickened inward projections. Subtending hyphae 15-16 μm in diameter, occluded at maturity. Walls of the hypha often thickened to 3-5 μm near the spore (Fig. 2B, 3B).

3. *Glomus aggregatum* Schenck and Smith- Chlamydospores are found in loose clusters or in sporocarps without a peridium. Sporocarps are hyaline to yellow with a greenish tinge, becoming brown with age. Chlamydospores globose to sub-globose. Spore diameter (32-) 79.41 (-103) μm when globose and 86.13 X 73.14 μm when subglobose. Spores hyaline to yellow, spore walls 1-2 layered, yellow to yellow brown varying from 1.4-2.8 μm thick consisting of an outerwall slightly thicker and lighter in colour than the inner wall, hyphae at the point of spore attachment 9.31 μm wide. Spore contents confluent with hyphal contents on young spores but separated from hyphae in older spores by inner spore wall. Spore wall in older spores not occluded by hyphal wall thickening. Hyphal attachment straight or recurved sharply at the spore base. Most hyphae 6-8 μm in diameter (Fig. 3H).

Critical Notes- This spore agrees with the original description with slight difference in the spore size.

4. *Glomus constrictum* Trappe- Spores naked, found singly or in loose clusters. Spores sub-globose to globose, spore size 148.33-258.56 μm . Spores are yellow brown when young to dark brown or black when mature. One layered, dark brown, seemingly two layered, 8.3-13.12 (-17) μm thick. Base straight or occasionally with a short funnel shaped projection, attachment occluded by wall thickenings. Hypha straight or recurved, point of attachment with dark brown walls. Spore walls 3.5-7.8 μm . Just beyond point of attachment, hypha constricted

to-11.2-19.4 μm beyond it inflated to 18-34.8 μm . Yellow to yellow brown walls, 2.6-3.4 μm thick. Pegs arise, thin dichotomy. Spores have shiny, smooth contents of oil globules of widely varying sizes (Fig. 3G).

Critical Notes- This species agrees with the original description in all the characters. However, the size of the spores and the spore wall thickenings are slightly bigger than the original description.

5. *Glomus monosporum* Gerdemann and Trappe- Spores globose to ellipsoid. Spores frequently are enclosed in sporocarps, containing mostly 1, occasionally 2 or rarely 3 chlamydospores. Chlamydospores 239.8-293.3 μm in diameter when globose. Spores 2 layered, spore walls 8-32.5 μm thick, dull brown, composed of a thin outer wall which often flakes off and a thick inner wall. Inner wall laminated with minute scattered echinulations that protrude into the outer wall. Thickening of inner wall extend into hypha. Hypha 8.4-13.8 μm in diameter strongly recurved and addressed to spore walls occasionally with 2 subtending hyphae. Spore contains oil globules and with hyphae thin walled, 4-6.8 μm in diameter (Fig. 2G).

Critical Notes- It resembles the original description in all the characters with slight differences in the spore size and wall thickness.

6. *Glomus geosporum* (Nicolson and Gerdemann) Walker- Sporocarps are unknown, chlamydospores formed singly in soil, globose, 263.53 μm in diameter. Smooth, light brown (transparent to translucent) when young and dark reddish-brown, when mature. Spore walls, 18.35 μm thick. Spore wall 3 layered, outer hyaline thin wall, tightly adherent, a reddish-brown laminated middle wall, 11.34 μm and a yellow brown inner wall 1.3 μm that appears membranous and forms a septum separating the spore contents from the lumen of the subtending hypha. Spores with one straight to recurved or 2 adjacent attachments simple to slightly funnel shaped subtending hypha measuring 148 μm long, 16.37 μm in diameter. With yellow to dark yellow brown, wall thickening that extends 30-100 μm along the hyphae from the spore base (Fig. 2E).

Critical Notes-It agrees with the original description in all the characters.

7. *Glomus diphanum* Mortar and Walker- Sporocarps are unknown, spores found singly or in loose clusters in the soil. Spores mostly globose or sub-globose. 62.34 μm in diameter, ellipsoid, spores which are more frequently in roots, measure 68 X 73.62 μm in diameter. Spores hyaline throughout life. Spore wall structure consists of 2 walls (walls 1 and 2) in a single group. Wall 1 is 5.3 μm thick, brittle and finely laminate, breaking into segments, when a spore is broken. Wall 2 is closely addressed, not adherent

to wall 1, consisting of thin membranous wall, 1.3 μm thick, which separates from the outer wall when the spore is crushed subtending hypha is single, straight or slightly funnel shaped 8.4 μm in diameter at the spore base. Hyphal wall at the point of attachment is 3.2 μm thick. It occasionally breaks. Contents of the spore are hyaline and consist of one to many oil globules (Fig. 3C, E, and F).

Critical Notes-The description of the present isolate agrees with the original description in all the characters.

8. *Glomus caledonium* (Nicolson & Gerdemann) Trappe and Gerdemann-Chlamydo spores are found singly or in sporocarps. Sub-globose, 5.3 μm in diameter, yellow to light brown. Spores globose to sub-globose, ellipsoid or irregular. Spore walls 2 layered, 5.1-7.6 μm . Outer wall hyaline, 1-3.8 μm , easily separable, thickened at the hyphal attachment and extending along the attached hyphae for some distance. Inner wall yellow to brown, 3.1-6.9 μm thick, thickening extending into the hyphae a short distance. Spore contents separated from the subtending hypha by a thin yellow, curved wall formed at the attachment (Fig. 3A).

Critical Notes-The present isolate resembles the original description in all the characters.

9. *Glomus deserticola* Trappe, Bloss and Menge-Spores found in single or in loose fascicles. Spores globose to sub-globose, measuring 5.2-103.6 X 46 X 83.4 μm . Spores shiny, smooth, yellow to brown or reddish to brown. Spores are single layered 2-2.8 μm . Spore walls simple or sometimes laminated. Attached hypha 5-11.3 μm in diameter, cylindrical to occasionally somewhat funnel shaped. The walls thickened and reddish brown, especially thick, adjacent to spores, but not occluding the hypha. Interior of the spore wall at the hyphal attachment thickened at maturity to form an inner rounded collar, appear to be closed by a membranous septum (Fig. 3D).

Critical Notes-This isolate resembles the original description in all the characters except the minimum spore size a little bigger than the original spore size.

10. *Glomus multisubtensum* Mukerji, Bhattacharjee and Tewari:-Chlamydo spores formed singly in the soil or in compact clusters of 5-8 spores, globose, light brown, 100-150 μm in diameter. Spore wall 10-15 μm thick, with two inseparable layers. Outer layer 10-12 μm thick, brown, inner layer 1-4 μm thick and pale yellow brown. Subtending hyphae 2-4 in number, attached at one end of the spore. Hyaline, pale yellow, thin walled, 10-15 μm wide at the point of attachment, tapering to 5-7 μm width, septum present in some cases, 20-25 μm along the subtending hyphae which may be branched. Spores of *G. multisubtensum* sometimes have 3 or more subtending

hyphae. Spores globose, smooth walled, with multiple hyphal attachment being always present on one end of the spore (Fig. 2D).

11. *Glomus rubiforme* Gerdemann and Trappe-Sporocarps yellowish brown, sub-globose or ellipsoid, 206 X 268 μm consisting of a single layer of chlamydo spores surrounding a central plexus of hyphae. Peridium absent, individual spores at times partially enclosed in a thin network of a tightly adpressed hyphae. Chlamydo spores dark brown, obovoid to ellipsoid or sub-globose 50-60.36 X 35.23-51.37 μm with a small pore opening into a thick-walled subtending hypha. Spore wall laminate, 3.86 μm thick, up to 7.56 μm thick at spore base, often perforated and often with thick, perforated projections on the inner surface (Fig. 4).

Critical Notes-This isolate agrees with the original description in all the characters.

Gigaspora- 1. Azygo spores bud from the bulbous, suspensor like tip of a hypha. Azygo spores found singly in the soils. Spore walls 2 layered - *G. gigantea*; 2. Subtending hyphae are bulbous, thin walled and hyaline. A small hyphal branch projecting from the bulbous portion of the subtending hyphae - *Gigaspora* spp.

1. *Gigaspora gigantea* (Nicolson and Gerdemann) Gerdemann and Trappe- Azygo spores formed singly in soil, 178-434 X 256-786 μm , globose to ellipsoid. Spores yellow to green or greenish yellow. Spore wall 2 layered, 2.8-6.8 μm , outer wall tightly covering the inner wall. The inner wall is 6.2 μm thick and continuous except for an occluded pore at the attachment, 38-45.3 μm , suspensor bulbous, giving rise to slender hyphae that projects to spore. Mature spores bright yellow with greenish tinge. Germ tubes produced directly through the spore wall in the basal region. Spore wall 2 layered with walls 7.6 μm thicknesses (Fig. 1A).

Critical Notes-The above isolate agrees with the original description in all the characters.

2. *Gigaspora* spp. (Fig. 1B, D, E, F & H).

Vesicular-arbuscular mycorrhiza is characterized by the presence of two specialized structures: arbuscules and vesicles. Arbuscules are produced by the internal mycelium intra-cellularly in the form of highly ramified minute arborescence within a few days of infection. Arbuscules are short-lived with a life of approximately 4 days^{56, 57} and digested by host, leaving the cortical cells intact and available for further colonization. Vesicles are also produced by the internal mycelium but mostly intercellular. They are regarded as structures of storage and are absent in certain forms. In addition to internal growth, the fungus also develops a network of hyphae that

extend out of the root into the soil. These hyphae are capable of re-initiating colonization and are also responsible for the acquisition of mineral nutrients from the environment. This most common type of vesicular arbuscular mycorrhizal (VAM) association is also referred as the arbuscular mycorrhiza (AM) but because the arbuscule is the unifying feature of these associations and vesicles are formed by few mycorrhizal fungi, it has been proposed recently that the name be simplified to arbuscular mycorrhiza^{58,59}. Currently both the terms are found in the literature.

Dangeard⁶⁰ was the first to name vesicular arbuscular fungus. Peyronel^{61,62} was the first to recognize the AM fungi as the members of *Endogonales*, rather than Chytrids, *Pythium* species or other fungi suggested by earlier workers. Thaxter⁶³ in his monograph of the family Endogonaceae described all the species known at that time. He recognized four genera in *Endogonaceae* – *Endogone*, *Sphaeroceas*, *Sclerocystis* and *Glaziella*.

Gerdemann and Trappe⁶⁴ and Nicolson and Gerdemann⁶⁵ divided *Endogone* into *Glomus*, *Gigaspora* and *Modicella* along with *Sclerocystis* and *Glaziella*. Gerdemann and Trappe⁶⁴ added another genus, *Acaulospora*. Thus the family endogonaceae of zygomycotina includes the genera, *Acaulospora*, *Entrophosphora*, *Gigaspora*, *Glomus*, *Sclerocystis* and *Scutellospora*⁶⁵. In the last 25 years, a number of workers have added new information to the taxonomy of AM fungi⁶⁶⁻⁷⁶. Berch⁷⁷, Trappe and Schenck⁷⁸ have compiled additional information to the taxonomic knowledge of AM fungi.

Identification of taxa of AM fungi has relied extensively on the morphology of the spores. The disadvantage of the morphological characters is that they hide all of the diversity that occurs with in strains of the species. It has been stressed that new genetic and molecular characters must be searched for fungal systematics and they must offer answer at different resolution levels from species to genus and family⁷⁹.

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