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Research Article

# Studies on mycorrhizal biodiversity in medicinal plant species of Pookode Lake area, Wayanad, India.

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**Abstract**: Forty different medicinal plant species were investigated for arbuscular mycorrhizal association from Pookode Lake area, Wayanad district, Kerala. The spore density and percentage of infection varied with plant species. The percentage of AM fungal colonization ranged from 30 to 80. The highest AM fungal infection was exhibited in *Centella asiatica* (80%) and maximum spore population in *Leucas aspera* (690/100g). Totally 36 arbuscular mycorrhizal fungal species were recovered from the rhizosphere of 40 medicinal plant species belongs to 20 plant families. The arbuscular mycorrhizal spore species *Glomus fasciculatum* was found dominant.

**Keywords**: *Glomus*, medicinal plants, mycorrhiza, wayanad

## Introduction

The plant in natural habitat forms association with large number of microorganisms; one among them is AM fungi. AM fungi are an important component in the soil microbial mass which regulates several essential biological processes at the plant soil interface. The majority of higher green plants and large number of AM fungi are involved in mycorrhiza formation (Harley, 1989). They regulate the abundance of plant community (Allen *et al.,* 1995). The symbiotic association between fungi and the roots of higher plants comes under the general name 'Mycorrhiza' which literally means fungus-roots. The mycorrhizal fungi as one of the important biological component of soil have been reported to play important role in the regeneration of the abandoned forests because of their symbiotic association with the plant roots.

Mycorrhizal fungi occur in nearly all soils throughout the world and form symbiotic associations, the fungal symbionts inhabiting the root forming a second absorptive organ for the host. As the most widespread symbiosis on earth (Brachmann and Parniske, 2006), AM fungi evolved concurrently with the first colonization of land by plants some 450 to 500 million years ago and persist in most extant plant taxa (Cairney, 2000). The AM associations can increase plant growth, in many cases by enhancing phosphorus uptake from soils with low to moderate phosphorus availability (Powell, 1984). AM fungi are major components of rhizosphere microflora in natural ecosystems and play significant role in the reestablishment of nutrient cycling (Peterson *et al.*, 1985). Due to their unique position at the root-soil interface, AMF have been described as "Keystone Mutualists" in ecosystems (O'Neill *et al.*, 1991). The great interest in AM in recent years has prompted numerous survey aimed at enumerating and assessing AM fungi in a particular region or in a natural environment. A study has been done to find the arbuscular mycorrhizal fungal spores in the rhizosphere soil samples of medicinal plant species and the rate of mycorrhizal infection in their roots.

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## Materials and Methods

### Study area- Pookode Lake

The Pookode Lake is a natural water body of about 7.5 ha surrounded by a chain of hills rising to the order of 800 m above mean sea level. This lake is located in Kunnathidavaka village between 11°34'O" and 11°32'24" N latitude and between 76°1'24" and 76°1'34" E longitude in the Vythiri taluk of the Wayanad district, 16 km towards the west of Kalpetta- the district headquarters.

### Soil sample collection:

Totally 40 medicinal plant species belonging to 20 families namely Apiaceae, Asteraceae, Acanthaceae, Apocynaceae, Chloranthaceae, Cyperaceae, Lilliaceae, Lamiaceae, Malvaceae, Melastomataceae, Mimosaceae, Onagraceae, Poaceae, Polygonaceae, Rubiaceae, Solanaceae, Scrophulariaceae, Tiliaceae, Urticaceae and Verbenaceae were collected from Pookode Lake in the period November 2009 to January 2010. Root samples and rhizosphere soil samples of medicinal plant species growing in and around the areas of Pookode Lake were collected. The root and soil samples were transported to the laboratory immediately after collection.

The plant species selected for the study are *Sida rhombifolia* L, *Urena lobata* L, *Triumfetta rhomboidea* Jacq, *Mimosa pudica* L, *Osbeckia octandra* DC, *Ludwigia octovalvis* (Jacq.) Raven, *Centella asiatica* (Linn) Urban, *Hydrocotyle javanica* Thunb, *Ophiorrhiza mungos* Linn, *Spermacoce ocymoides* Burm.f, *Ageratum conyzoides* Linn, *Blumea membranacea* (DC.) Hook.f, *Dichrocephala integrifolia* (L.f) O. Kuntz, *Elephantopus scaber* L, *Erechtites valerianifolia* (Wolf) DC, *Spilanthes ciliata* H.B.K, *Spilanthes radicans* Jacq, *Vernonia cinerea* (L.) Less, *Wedelia trilobata* (L.) A.S. Hitchc, *Rauwolfia serpentina* (L.) Benth.ex Kurz, *Rauwolfia verticillata* (Lour) Baill, *Solanum nigrum* L, *Solanum surattense* Burm.f, *Solanum torvum* Sw, *Lindernia antipoda* (L.)Alston, *Mecardonia procumbens* (Mill.) Small, *Scoparia dulcis* Linn, *Andrographis macrobotrys* Nees, *Strobilanthes heyneanus* Nees, *Lantana camara* L, *Stachytarpheta jamaicensis* (L.) Vahl, *Leucas aspera* (Willd.) Spreng, *Pogostemon paniculatus* (Willd.) Benth, *Polygonum chinense* Linn, *Sarcandra chloranthoides* Gard, *Elatostema lineolatum* Wight, *Asparagus racemosus* Willd, *Kyllinga monocephala* Rottb, *Paspalum conjugatum* Berg and *Panicum brevifolium* L.

**Estimation of Arbuscular Mycorrhizal colonization in roots**

**Root samples:**

Root samples, 5-15 cm long, were collected from the medicinal plant species during November 2009 - Jan 2010. During collection, care was taken to ascertain individual plants for which roots could be positively identified as belonging to a particular plant species. For identification and nomenclature of plant species the following manual was used (Mathew, 1983).

**Soil samples:**

The rhizosphere soil, dug up to a depth of 10cm, was collected from each plant species after removing the surface soil and litter covering. These samples were kept in sterilized bags and were transported to the laboratory immediately after collection for the examination of vesicular arbuscular mycorrhizal fungal spores association.

**Sample preservation:**

In the laboratory, the roots were separated from soil, by wet sieving. The roots were washed with water and processed afresh whenever possible. Otherwise the washed roots were fixed in formaldehyde-acetic acid-ethanol (FAA) solution (90:5:5 V/N) [modified method of Phillips and Hayman (1970) as mentioned in Cade-Menun*et al.* (1991). The soil sample was air dried and stored at 4˚C until processed. Each soil sample was used for chemical analysis, spore counts and classification into various types and multiplication, concentration and separation of AM fungal spore for identification.

**Preparation of soil samples for analysis:**

Each soil sample was spread on a flat wooden or plastic tray and was allowed to dry in air under shade. Stones and pieces of macro organic matter were removed. Large lumps were broken by hand and the soil was ground by rolling gently with a wooden roller. After grinding, the soil was screened through a 2 mm sieve and the fine soil was used for further analysis.

**Evaluation of AM infection**

The root samples were cleared and stained in tryphan blue with a modified version of the Philips and Hayman's (1970) method. Roots were cut into 1-2 cm pieces, heated at 90˚C in 10% KOH for about 1 hour. For thicker and older roots, the duration was increased. The root segments were rinsed in water and acidified with dilute HCL. The root pieces were stained with 0.05% tryphan blue in lacto phenol for 5 minutes and the excess stain was removed with clear lacto phenol.

The pigmented roots were heated at 90˚C in 10% KOH for 2 hours, washed with fresh 10% KOH and immersed in an alkaline solution of $H_2O_2$ for 30 minutes at 25˚C until bleached. They were rinsed thoroughly with water to remove the $H_2O_2$, acidified in dilute HCL and stained as described earlier. In some cases the modified method of Merryweather and Fitter (1991) was followed where autoclaving and bleaching with $H_2O_2$ were omitted. In a few cases, direct observation of unstained, fresh and intact roots (Arias *et al.*, 1987) was made.

Arbuscular mycorrhizal infection in the roots was assessed following the grid line-intersect method of Giovannetti and Mosse (1980). The stained root pieces were spread out evenly on a square plastic Petri dish (10.2 x 10.0 cm). A grid of lines was marked on the bottom of the dish to form 1 cm inch squares. Vertical and horizontal gridlines were scanned under a dissecting microscope and the presence of infection was recorded at each point where the roots intersected a line. Four sets of observations were made, recording 100, 200, 300 and all the root gridline intersects. Each of the three replicate records was made on a fresh re-arrangement of the same root sample.

The percentage of AM infection was calculated using the formula:

$$\%\ of\ infection\ = \frac{No.\ of\ root\ segments\ infected}{Total\ no.\ of\ root\ segments} \times 100$$

When sufficient root pieces are not available, the slide method Giovannetti and Mosse (1980) was followed. Root pieces, 1 cm long, were selected at random from a stained sample and mounted on microscope slide groups of 10. Presence of infection was recorded in each of the 10 pieces and percent infection was calculated. To observe hyphae, vesicles and arbuscules under light microscope, the root

pieces were mounted on glass slides either temporarily in lacto phenol or permanently in polyvinyl alcohol resin-lacto phenol. The cover slip was pressed gently to make the roots flattened and sealed with DPX medium.

**Isolation of Arbuscular mycorrhizal spores from soil samples:**

Spores were recovered from soil samples by the wet-sieving and decanting method (Gerdemann and Nicolson, 1963). From each soil sample, 100g soil was taken and mixed with 1:1 of lukewarm water in a large beaker until all the aggregates dispersed to leave a uniform suspension. Heavier particles were allowed to settle down. To remove organic matter and roots, the suspension was decanted through a 710µm sieve. The suspension that passed through 710µm was decanted through 425µm, 250µm, 150µm, 75µm and 45µm sieves consecutively. The residues in the respective sieve were collected in Petri dishes with about 10-20 ml water observed under a dissecting microscope for AM fungal spores.

Total spore count was calculated by counting the spores. Then the spores were separated using a glass pipette and segregated. The spores were mounted on clear glass slides using lacto phenol or polyvinyl alcohol lacto phenol (PVL), covered with cover slips and sealed with DPX medium.

**Identification of AM fungi:**

Based up on microscopic characters, the AM spores were identified. For identification and nomenclature, synoptic keys of the following authors were used: Raman and Mohankumar (1988); Schenck and Perez (1990). Classification was based on colour, size, shape, surface, structure, general nature of the spore contents and hyphal attachment. Photomicrographs were taken with the help of a Magnus Olympic Microscope.

## Results

Arbuscular mycorrhizal infection and spore population of 40 medicinal plant species belongs to 20 families present in the year 2009-2010 (Table: 1). Totally 40 plant species including shrubs, herbs belong to 20 families were examined for arbuscular mycorrhizal fungal association. Of these, maximum spore population displayed in the plant species of *Leucas aspera* (690/100g soil) belongs to the family Lamiaceae and minimum spore population was recorded in the plant species of *Spilanthes radicans* (170/100g soil) belongs to the family Asteraceae.

The highest AM fungal infection found in the roots of *Centella asiatica* (80%) belongs to the family of Apiaceae. The lowest fungal infection was found to the *Triumfetta rhomboidea* (30%) of Tiliaceae member.

The plant species *Urena lobata* (38%), *Triumfetta rhomboidea* (30%), *Osbeckia octandra* (32%), *Spermacoce ocymoides* (39%), *Elephantopus scaber* (40%), *Spilanthes radicans* (36%), *Spilanthes ciliata* (38%), *Scoparia dulcis* (38%), and *Elatostema lineolatum* (33%) showed 40% and less than 40% of infection.

The plant species *Sida rhombifolia* (46%), *Mimosa pudica* (43%), *Ludwigia octovalvis* (43%), *Hydrocotyle javanica* (48%), *Vernonia cinerea* (42%), *Pogostemon paniculatus* (50%) and *Asparagus racemosus* (45%) showed 50% and less than 50% of infection.

The plant species *Blumea membranaceae* (59%), *Solanum nigrum* (57%), *Lindernia antipoda* (56%), *Strobilanthes heyneanus* (58%) and *Panicum brevifolium* (60%) showed 60% and less than 60% infection.

The plant species *Erechtites valerianifolia* (62%), *Rauvolfia serpentine* (63%), *Solanum surattenses* (65%), *Ageratum conyzoides* (70%), *Mecardonia procumbens* (70%), *Lantana camara* (70%) and *Sarcandra chloranthoides* (70%) showed 70% and less than 70% of infection.

The medicinal plant species *Centella asiatica* (80%), *Dichrocephala integrifolia* (75%), *Wedelia trilobata* (73%), *Rauvolfia verticillata* (73%), *Solanum torvum* (74%), *Andrographis macrobotrys* (71%), *Stachytarpheta jamaicensis* (76%), *Leucas aspera* (78%), *Polygonum chinense* (78%), *Kyllinga monocephala* (73%), *Ophiorrhiza mungos* (76%) and *Paspalum conjugatum* (75%) showed 80% and less than 80% of infection.

Totally 36 arbuscular mycorrhizal fungal species were recovered from the rhizosphere of 40 medicinal plant species belongs to 20 plant families (Fig.1). Except for the genus *Entrophosphora* rest of the five genera of AM fungi were encountered during the present study. Of these one species from *Acaulospora* (*Aca. rehmii*), two species from *Gigaspora* (*Gig. rosea* and *Gig. albida*), 27 species from *Glomus, Gl. albidum, Gl. ambisporum, Gl. arborense, Gl. australe, Gl. boreale, Gl. botryoides, Gl. canadense, Gl. citricola, Gl. claroides, Gl. constrictum, Gl. delhiense, Gl. fecundisporum, Gl. fistulosum, Gl. flavisporum, Gl. fulvum, Gl. hoi, Gl. intraradices, Gl. invermayanum, Gl. macrocarpum, Gl. maculosum, Gl. magnicaule, Gl. multicaule, Gl. multisubtensum, Gl. radiatum, Gl. tenebrosum, Gl. tortuosum, Gl. vesiculiferum,* and one species from *Sclerocystis* (*Sclerocystis rubiformis*) and 6 species from *Scutellospora, Scu. auriglobosa, Scu. heterogama, Scu. nigra, Scu. reticulata* and *Scu. scutata.* The genus *Glomus* was recorded more frequently. The arbuscular mycorrhizal spores *Gl. fasciculatum* was found dominant followed by *Gl. intraradices.*

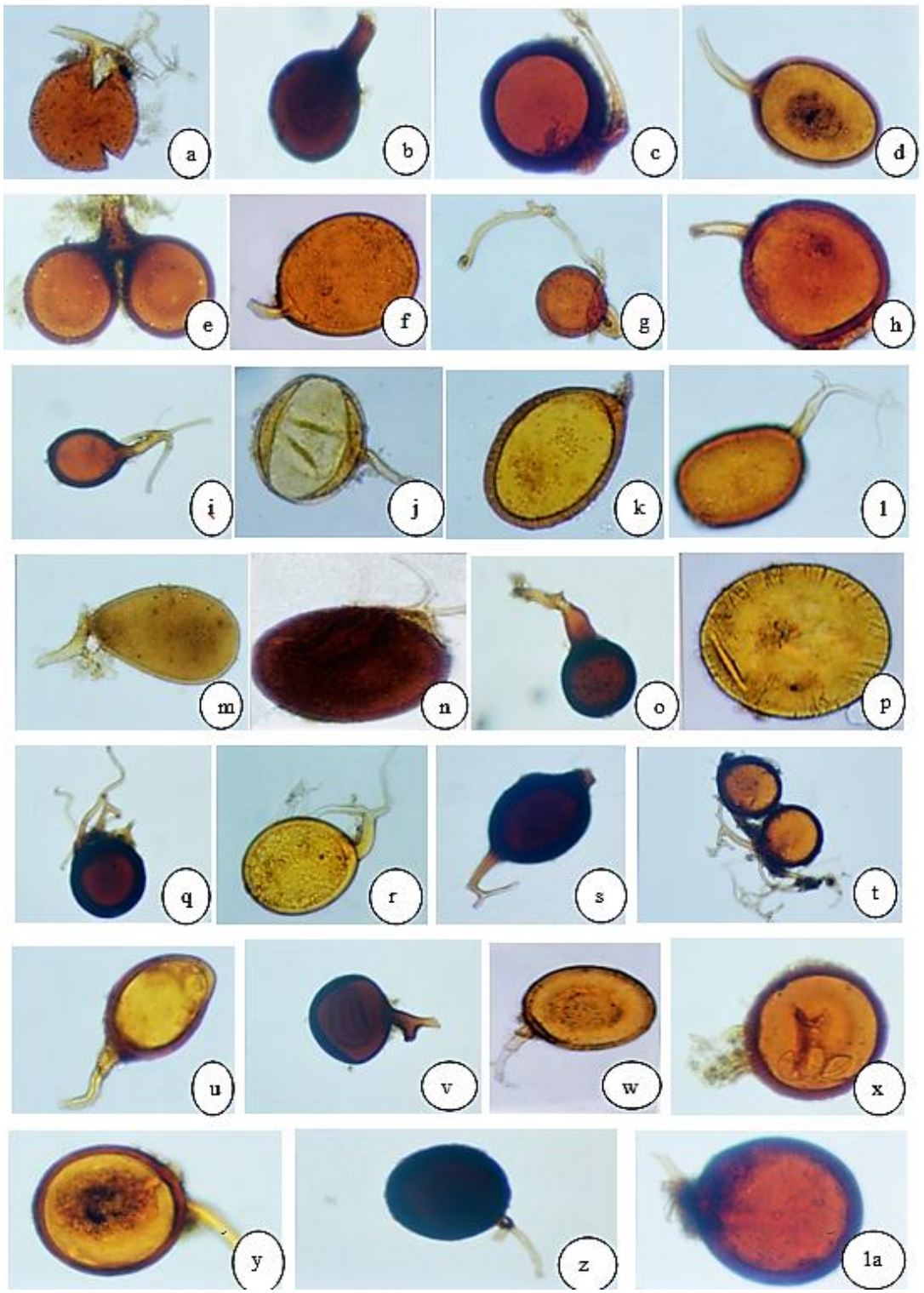


**Fig.1** Arbuscular mycorrhizal fungal species a. *Glomus ambisporum* (40X) b. *Gl. arborense* (40X) c. *Gl. australe* (40X) d. *Gl. boreale* (40X) e. *Gl. botriyoides* (40X) f. *Gl. canadense* (40X) g. *Gl. citricola* (20X) h. *Gl. claroides* (120X) i. *Gl. fasciculatum* (40X) j. *Gl. fecundisporum* (120X) k. *Gl. fistulosum* (120X) l. *Gl. flavisporum* (120X) m. *Gl. fulvum* (120X) n. *Gl. intraradices* (120X) o. *Gl. invermayanum* (20X) p. *Gl. macrocarpum* (120X) q. *Gl. maculosum* (40X) r. *Gl. magnicaule* (120X) s. *Gl. multicaule* (40X) t. *Gl. multisubtensum* (20X) u. *Gl. radiatum* (40X) v. *Gl. tenebrosum* (120X) w. *Gl. vesiculiferum* (180X) x. *Scutellospora auriglobosa* (180X) y. *Scu. heterogama* (180X) z. *Scu. nigra* (180X) 1a. *Scu. scutata* (180X).

**Table 1.** AM fungal infection and spore population of medicinal plant species collected in and around the areas of Pookode Lake

| S. No. | Name of the plant species | Habit | Type of Infection | | | % of Infection | Spore Population per 100 gm soil | AM Fungal spores |
|---|---|---|---|---|---|---|---|---|
| | | | Hyphae | Vesicles | Arbuscules | | | |
| **MALVACEAE** | | | | | | | | |
| 1. | *Sida rhombifolia* Linn. | Under shrub | + | - | + | 46% | 487 | *Gigaspora rosea*<br>*Glomus boreale*<br>*Gl. citricola*<br>*Gl. fistulosum*<br>*Gl. fasciculatum* |
| 2 | *Urena lobata* Linn. | Shrub | + | - | + | 38% | 285 | *Acaulospora rehmii*<br>*Glomus canadense*<br>*Gl. hoi*<br>*Gl. intraradices*<br>*Scutellospora nigra* |
| **TILIACEAE** | | | | | | | | |
| 3 | *Triumfetta rhomboidea,* Jacq. | Herb | + | - | + | 30% | 300 | *Acaulospora rehmii*<br>*Gigaspora albida*<br>*Glomus ambisporum*<br>*Gl. invermayanum*<br>*Scutellospora scutata* |
| **MIMOSACEAE** | | | | | | | | |
| 4 | *Mimosa pudica* L. | Herb | + | + | + | 43% | 525 | *Glomus delhiense*<br>*Gl. flavisporum*<br>*Gl. radiatum*<br>*Gl. fasciculatum*<br>*Scutellospora heterogama*<br>*Scu. nigra* |
| **MELASTOMACEAE** | | | | | | | | |
| 5 | *Osbeckia octandra* DC. | Shrub | + | + | - | 32% | 225 | *Acaulospora rehmii*<br>*Gigaspora rosea*<br>*Glomus constrictum*<br>*Gl. intraradices* |
| **ONAGRACEAE** | | | | | | | | |
| 6 | *Ludwigia octovalvis* (Jacq.) Raven. | Herb | + | + | - | 43% | 375 | *Glomus arborense*<br>*Gl. fulvum*<br>*Gl. fasciculatum*<br>*Gl. tortuosum*<br>*Sclerocystis rubiformis*<br>*Scutellospora nigra* |
| **APIACEAE** | | | | | | | | |
| 7 | *Centella asiatica* (L.) Urban. | Herb | + | - | + | 80% | 560 | *Acaulospora rehmii*<br>*Gigaspora albida*<br>*Glomus australe*<br>*Gl. fasciculatum*<br>*Gl. fecundisporum*<br>*Scutellospora nigra* |
| 8 | *Hydrocotyle javanica* Thunb. | Herb | + | + | - | 48% | 300 | *Gigaspora rosea*<br>*Glomus fasciculatum*<br>*Gl. magnicaule*<br>*Gl. tenebrosum* |
| **RUBIACEAE** | | | | | | | | |
| 9 | *Ophiorrhiza mungos* Linn | Under shrub | + | + | - | 76% | 645 | *Glomus claroides*<br>*Gl. fasciculatum*<br>*Gl. intraradices*<br>*Gl. macrocarpum*<br>*Gl. vesiculiferum* |
| 10 | *Spermacoce ocymoides* Burm. f. | Herb | + | + | - | 39% | 337 | *Gigaspora albida*<br>*Glomus multicaule*<br>*Scutellospora auriglobosum* |
| **ASTERACEAE** | | | | | | | | |
| 11 | *Ageratum conyzoides* Linn. | Herb | + | + | - | 70% | 525 | *Glomus albidum*<br>*Gl. hoi*<br>*Gl. tortuosum*<br>*Sclerocystis rubiformis* |
| 12 | *Blumea membranacea* (DC.) Hook. f. | Herb | + | + | - | 59% | 562 | *Glomus arborense*<br>*Gl. canadense*<br>*Gl. fulvum*<br>*Gl. intraradices* |
| 13 | *Dichrocephala integrifolia* (L.F.) O. Kuntze. | Herb | + | + | - | 75% | 568 | *Acaulospora rehmii*<br>*Glomus citricola*<br>*Gl. radiatum* |

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|---|---|---|---|---|---|---|---|---|
| | | | | | | | | *Sclerocystis rubiformis* |
| 14 | *Elephantopus scaber* Linn. | Herb | + | - | + | 40% | 382 | *Glomus constrictum*<br>*Gl. fistulosum*<br>*Gl. vesiculiferum* |
| 15 | *Erechtites valerianifolia* (Wolf) DC. | Herb | + | - | + | 62% | 600 | *Gigaspora rosea*<br>*Glomus botroyoides*<br>*Gl. multisubstensum*<br>*Gl. fecundisporum*<br>*Gl. tenebrosum* |
| 16 | *Spilanthes ciliata* H.B.K | Herb | + | - | + | 38% | 200 | *Glomus australe*<br>*Gl. fasciculatum*<br>*Gl. magnicaule*<br>*Scutellospora scutata* |
| 17 | *Spilanthes radicans* Jacq. | Herb | + | + | - | 36% | 170 | *Glomus canadense*<br>*Gl. flavisporum*<br>*Gl. hoi*<br>*Gl. intraradices* |
| 18 | *Vernonia cinerea* (Linn.) Less. | Herb | + | - | + | 42% | 487 | *Gl. albidum*<br>*Gl. fasciculatum*<br>*Gl. intraradices*<br>*Scutellospora sp.* |
| 19 | *Wedelia trilobata* (L.) A.S. Hitchc. | Herb | + | - | + | 73% | 600 | *Glomus canadense*<br>*Gl. maculosum*<br>*Gl. fecundisporum*<br>*Gl. boreale* |
| **APOCYNACEAE** | | | | | | | | |
| 20 | *Rauvolfia serpentina* (L.) benth. ex. Kurz. | Under shrub | + | + | + | 63% | 605 | *Gigaspora albida*<br>*Glomus multicaule*<br>*Gl. tenebrosum*<br>*Scutellospora auriglobosum* |
| 21 | *Rauvolfia verticillata* (Lour.) Baill. | Shrub | + | - | + | 73% | 668 | *Glomus albidum*<br>*Gl. ambisporum*<br>*Gl. constrictum*<br>*Gl. fasciculatum*<br>*Gl. invermayanum* |
| **SOLANACEAE** | | | | | | | | |
| 22 | *Solanum nigrum* (Linn.) | Herb | + | - | + | 57% | 517 | *Glomus macrocarpum*<br>*Gl. australe*<br>*Gl. radiatum*<br>*Gl. fasciculatum* |
| 23 | *Solanum surattense* Burm.f. | Under shrub | + | + | + | 65% | 490 | *Glomus ambisporum*<br>*Gl. citricola*<br>*Gl. fistulosum*<br>*Gl. intraradices*<br>*Scutellospora sp.* |
| 24 | *Solanum torvum* Swartz. | Herb | + | + | + | 74% | 645 | *Glomus hoi*<br>*Gl. claroides*<br>*Gl. radiatum*<br>*Gl. vesiculiferum*<br>*Gl. delhiense*<br>*Scutellospora scutata* |
| **SCROPHULARIACEAE** | | | | | | | | |
| 25 | *Lindernia antipoda* L. Alston. | Herb | + | + | - | 56% | 489 | *Acaulospora rehmii*<br>*Gigaspora albida*<br>*Glomus botryoides*<br>*Gl. intraradices* |
| 26 | *Mecardonia procumbens* (Mill.) Small. | Herb | + | + | - | 70% | 520 | *Glomus flavisporum*<br>*Gl. canadense*<br>*Gl. albidum*<br>*Gl. citricola*<br>*Gl. fasciculatum* |
| 27 | *Scoparia dulcis* Linn. | Herb | + | - | + | 38% | 240 | *Glomus australe*<br>*Gl. constrictum*<br>*Gl. fulvum*<br>*Gl. intraradices*<br>*Gl. invermayanum* |
| **ACANTHACEAE** | | | | | | | | |
| 28 | *Andrographis macrobotrys* Nees. | Herb | + | + | - | 71% | 600 | *Glomus ambisporum*<br>*Gl. boreale*<br>*Gl. claroides*<br>*Gl. fasciculatum*<br>*Gl. fecundisporum* |
| 29 | *Strobilanthes heyneanus* Nees. | Under shrub | + | + | - | 58% | 465 | *Glomus delhiense*<br>*Gl. maculosum*<br>*Gl. multisubstensum*<br>*Sclerocystis rubiformis* |

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|---|---|---|---|---|---|---|---|---|
| **VERBENACEAE** | | | | | | | | |
| 30 | *Lantana camara* Linn. Var. aculeate | Shrub | + | - | + | 70% | 600 | *Gigaspora rosea*<br>*Glomus citricola*<br>*Gl. fasciculatum*<br>*Gl. macrocarpum*<br>*Gl. tortuosum*<br>*Gl. radiatum* |
| 31 | *Stachytarpheta jamaicensis* (L.) Vahl. | Shrub | + | + | - | 76% | 660 | *Glomus arborense*<br>*Gl. canadense*<br>*Gl. fasciculatum*<br>*Gl. radiatum*<br>*Gl. tenebrosum* |
| **LAMIACEAE** | | | | | | | | |
| 32 | *Leucas aspera* (Willd.) Spreng. | Herb | + | + | - | 78% | 690 | *Glomus ambisporum*<br>*Gl. botroyoides*<br>*Gl. claroides*<br>*Gl. fasciculatum*<br>*Gl. fulvum*<br>*Gl. vesiculiferum* |
| 33 | *Pogostemon paniculatus* (Willd.) Benth. | Herb | + | - | + | 50% | 450 | *Acualosporarehmii*<br>*Glomus albidum*<br>*Gl. fulvum*<br>*Gl. intraradices*<br>*Gl. multisubstansum* |
| **POLYGONACEAE** | | | | | | | | |
| 34 | *Polygonum chinense* Linn. | Under shrub | + | + | - | 78% | 675 | *Glomus botroyoides*<br>*Gl. constrictum*<br>*Gl. fasciculatum*<br>*Gl. invermayanum*<br>*Gl. multicaule*<br>*Sclerocystis rubiformis*<br>*Scutellospora nigra* |
| **CHLORANTHACEAE** | | | | | | | | |
| 35 | *Sarcandra chloranthoides* Gardner. | Shrub | + | - | + | 70% | 600 | *Glomus magnicaule*<br>*Gl. arborense*<br>*Gl. claroides*<br>*Gl. delhiense*<br>*Gl. fasciculatum*<br>*Gl. intraradices* |
| **URTICACEAE** | | | | | | | | |
| 36 | *Elatostema lineolatum* Wight. | Under shrub | + | - | + | 33% | 300 | *Acaulospora rehmii*<br>*Glomus constrictum*<br>*Gl. fecundisporum*<br>*Gl. intraradices*<br>*Gl. maculosum* |
| **LILIACEAE** | | | | | | | | |
| 37 | *Asparagus racemosus* Willd. | Under shrub | + | + | + | 45% | 450 | *Gl. delhiense*<br>*Gl. fasciculatum*<br>*Gl. fistulosum*<br>*Gl. intraradices*<br>*Gl. invermayanum*<br>*Gl. vesiculiferum* |
| **CYPERACEAE** | | | | | | | | |
| 38 | *Kyllinga monocephala* Rottb. | Grass | + | + | - | 73% | 600 | *Gigaspora rosea*<br>*Glomus albida*<br>*Gl. boreale*<br>*Gl. constictum*<br>*Gl. flavisporum*<br>*Gl. macrocarpum* |
| **POACEAE** | | | | | | | | |
| 39 | *Paspalum conjugatum* Bergius. | Grass | + | + | - | 75% | 675 | *Glomus ambisporum*<br>*Gl. botroyoides*<br>*Gl. magnicaule*<br>*Gl. tenebrosum*<br>*Scutellospora heterogama* |
| 40 | *Panicum brevifolium* L. | Grass | + | + | - | 60% | 486 | *Acaulospora rehmii*<br>*Glomus citricola*<br>*Gl. fasciculatum*<br>*Gl. fulvum*<br>*Gl. intraradices*<br>*Gl. multisubstensum* |

## Discussion

In Kerala the Pookode lake area plant species were analyzed to determine for mycorrhizal infection and spore population for a period of one year. All the plant species showed mycorrhizal association. Totally 40 plant species belongs to 20 families were surveyed for AM fungal association. The root colonization ranged from 30% to 80% and the AM fungal spore density ranged from 170 to 690 per 100 gm of soil.

The populations of spores as well as the percentage of infection of all the samples were varied in all the plant species of Pookode area. Edaphic factors also play an important role in spore germination and colonization, along with plant factors like age, lifespan and root density (Abbott and Robson, 1991). Formations of AM fungal structures were inconsistent and fluctuating from site to site. Colonization and sporulation by AM fungi are influenced by a wide range of environmental, plants and fungal factors (Khalil *et al*., 1992).

Mohankumar and Mahadeven (1986) find out no mycorrhizal association in the families of Acanthaceae and Verbenaceae in mangrove vegetation. Muthukumar *et al*., (1994) observed the AM fungal colonization in *Mimosa pudica* (74.45%) belongs to the family Mimosaceae. Similarly, the same result was obtained in the present investigation and the plant species *Mimosa pudica* showed only 43% of AM fungal infection.

The present study reveals that there was no consistent relationship between organic matter, pH of the soil that of the number of spores in soils. Similar findings were reported by early workers (Warner & Mosse, 1980, Mohankumar and Mahadevan, 1988). Prasad (2005) displayed the AM infection and find out the maximum in *Cyperus rotundus* (40%), Cyperaceae followed by *Cynodon dactylon* (36%) (Graminae) and *Solanum sp* (Solanaceae) from Bettiah, Bihar. The AM fungal infection was observed 73%, 75% and 74% in the other species of families Cyperaceae, (*Kyllinga monocephala*), Graminae (*Paspalum conjugatum*), and Solanaceae (*Solanum torvum*) respectively.

The present study brought out that most of tree species were colonized by AM. Almost all forest trees recorded AM colonization in the roots besides the presence of their spores in the rhizosphere. Ragupathy *et al*., 1988 reported the absence of mycorrhizal infection in *Colocasia esculenta* (Araceae) and *Borreria hispida* belongs to the family Rubiaceae. But in contrast the present study reveals that the Rubiaceaea members *Ophiorrhiza mungos* and *Spermacoce ocymoides* infected 76% and 39% respectively.

Among the members of Astreraceae the species richness and diversity of AM fungi and infection has highest in *Dichrocephala integrifolia* (75%) followed by *Ageratum conyzoides* (70%). The same result was obtained in the species *Ageratum conyzoides* (70%) belongs to the family (Asteraceae) from coastal sand dunes of West coast of India (Beena *et al*., 2001). Their studies also indicates that the Lamiaceae member *Leucas aspera* infected (75%) by AM fungal infection (Beena *et al*., 2001). Kumar *et al.* (2003) also reported the AM fungal colonization (50%) in roots of the plant species *Ageratum conyzoides.*

The present study confirms the presence of AM colonization in medicinal plants. Similar observations were recorded earlier (Taber and Trappe, 1982; Muthukumar *et al*., 2001; Sadiq Gorsi, 2002). The mycorrhizal colonization differed among medicinal plant species and there was a considerable variation in percent root colonization and number of different arbuscular mycorrhizal fungal spores associated with rhizosphere soil samples of plant species.

Thirty-six AM fungal species belonging to five genera were recovered from the rhizosphere soil in the present study. This is in agreement with the findings of Francis and Read (1994) and Allen *et al*, (1995) who reported high AM fungal species diversity in medicinal plants. The medicinal plant species *Asparagus racemosus* and *Solanum nigrum* (Solanaceaea) was infected by AM fungal infection 40 & 30% respectively from Azad Jammu and Kashmir (Sadiq Gorsi, 2002). The present study reveals the similar results that the *Asparagus racemosus* (Liliaceaea) 45% and *Solanum nigrum* (Solanaceae) 57% infected by AM fungi. Kumar (2008) reported AM fungal association in *Sida cordifolia* (Meliaceae) from different parts of Karnataka which is in consistence with this present study.

Many common wetland plant families, most notably the cyperaceae had been previously as non-mycorrhizal because in few species roots observed to harbor the fungus (Powell, 1975; Newman and Reddell, 1987; Brundrett, 1991; Smith and Read, 1997) but there have been numerous reports of mycorrhizal colonization in certain Cyperaceae species (Wetzel and Van der Valk, 1996). The present study also confirmed that one of the Cyperaceae member *Kyllinga monocephala* (73%) was infected by AM fungi.

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