

RESEARCH ARTICLE

STUDIES ON THE ARBUSCULAR MYCORRHIZAL FUNGAL BIODIVERSITY IN THE PLANT SPECIES OF YELLANAHALLI HILLS, VALLEY VIEW OF NILGIRIS, UDHAGAMANDALAM, TAMIL NADU, INDIA

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ABSTRACT

The present study to investigated that the arbuscular mycorrhizal fungal root colonization and spore population in some medicinal at Yellanahalli hills, valley view of Nilgiris, Udhagamandalam, Tamilnadu, India. Root and rhizosphere soil samples were collected during the month of August, 2017 - March, 2018 Soil pH was to be recorded. From the study results revealed that totally 25 plant species belonging to 13 families were recorded root colonization and rhizosphere spore population. A totally 12 Arbuscular mycorrhizal fungal species belonging to 7 genera and 2 different Orders were isolated and identified. The maximum spore population was found in the rhizosphere soil samples of *Justicia procumbens* (380 /100 g of soil) which belongs to the family Acanthaceae and the lowest spore population was observed in the *Crotalariaeae juncea* (102 / 100 g of soil) belongs to Fabaceae. Among these plant species the highest 81% AM fungal infection was found in roots of *Solanum nigrum* belongs to the family Solanaceae While the lowest 23 % AM fungal association was found in the root of *Verbascum thapsus* belongs to Scrophulariaceae.

Keywords: AMF Spore population, medicinal plants, Yellanahalli hills.

1. INTRODUCTION

As the world population continues to increase, the demands placed on agriculture to supply future food and fiber needs will be one of the greatest challenges facing the agricultural community. In particularly soil is one of the most important along with various microorganisms colonizing the rhizosphere soil surface, mycorrhizae, the mutualistic symbiotic, play an important role in mobilizing phosphorus from the deeper layers of the soil and supplying it to the host plants. Among the mycorrhizae, Arbuscular mycorrhizae (AM) is the most prevalent type (1).

In recently, considerable importance is being given to AM fungi, because of awareness of environmental pollution and health hazards by the use of chemicals. The responsibility of AM fungi and PGPR's, in improving crop plants growth is well documented (2, 3). Arbuscular Mycorrhizal fungi are also known to several benefits of the hosts by improving the uptake of other nutrients such as nitrogen (4), copper (5), sulphur, potassium and calcium (6) and by limiting uptake of toxic heavy metals such as Zn and Cd from soil (5) and they also increase drought tolerance (7), disease resistance (8). Hence in this present research work, the arbuscular mycorrhizal fungal root colonization and spore population in the rhizosphere soil samples were investigated in Yellanahalli hills, valley view of Nilgiris, Udhagamandalam, Tamilnadu.

2. MATERIALS AND METHODS

2.1. Study area

The present study area of Yellanahalli valley Coonoor (taluk) located in the Nilgiris District of Tamil Nadu State, India. The hill is located 11.404457°N 76. 712843°E (Fig. 1). The elevation of valley view ranges 2,400 msl (7,900ft). Near Yellanahalli are another two villages called Ketti and Aruvankadu. The Ketti is located to the south-west of Yellanahalli and is also sometimes referred to as the Switzerland of Southern India due to the year-round climatic conditions. The maximum annual rainfall 991mm and maximum temperature 24.3°C and minimum were 4.8°C.

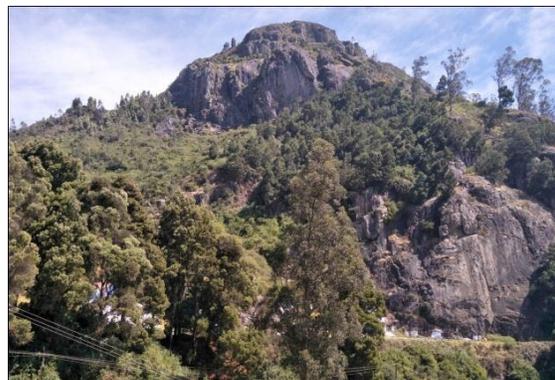


Fig. 1. View of the study area of Yellanahalli hills.

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2.2. Sample collection

The present study root and rhizosphere soils samples were collected from 25 plant species during the year August, 2017 to March, 2018. All the samples were placed in the polyethylene bags, labeled and then transported to the laboratory. The root samples were freshly processed, whereas rhizosphere soil samples were analyzed for mycorrhizal spore population and AM fungal root colonization.

2.3. Estimation of AM fungal root colonization

The fresh root samples were cleared and stained in trypan blue following method of (9). Root samples of each plant species were washed gently under tap water and cleared in 2.5% KOH, acidified in 5 N HCL and stained in lacto glycerol with 0.05% Trypan blue. The stained roots were examined under a compound microscope (40x-100x). Hundred root segments for each sample were randomly selected for microscopic observation and the degree of colonization was estimated using the slide method (10).

The percentage of AM fungal infection was calculated using the formula:

$$\text{Percentage of colonization} = \frac{\text{No. of root segments colonized}}{\text{Total no of root segments of observed}} \times 100$$

2.4. AMF spore identification

AM fungal spores were extracted from 100 g rhizosphere soil by wet-sieving and decanting method (11) through a series of 710 to 37 μ m size sieve filter. For the identification and nomenclature of these AM fungal spore synoptic keys developed by (12, 13, 14) were used. The classification was based upon the color, shape, hyphae, structure, size, and cell wall thickness and spore diameter.

2.5. Soil pH

The pH of the rhizosphere soil samples was determined (soil-water suspensions 1:5) with the help of pH meter (Elico) and values were recorded.

3. RESULTS AND DISCUSSION

In the present research, revealed that AM fungal colonization and spore population totally 25 plant species belongs to 13 families and pH of rhizosphere soil samples ranges between 4.8 to 6.6 were recorded from the study region. The detailed information about the plant species and their family habit, parts used and medicinal uses presented in (Table-1, 2; Fig.2).

In this study, analysis of life forms indicates that 72% of them are herbs and 28% of them are

shrubs (Fig. 3). As far as the plant part used is concerned, it was noted that the local people especially in Badagas employed almost all part of plant used as ethnomedicine. The leaf is most predominantly used 44 % followed by whole plant 32%, seed 8 %, flower 8%, fruit 4% and root 4% (Fig.4) respectively. Based on the present study, it has been found that the Badagas tribal community of Yellanahalli hills is rich in ethnobiological knowledge and this knowledge is being transmitted from one generation to another generation. These traditional medicines are the primary health care resources for the Badagas tribes to protect their health.

Our present study findings that AM fungal colonization, the highest percent root colonization 81% was observed in the root samples from the plant species *Solanum nigrum*. A least number of 23% AM fungal infection was observed in *Verbascum thapsus*. The maximum spore population was noted in *Justicia procumbens* (380/100 g of soil) belongs to the family Acanthaceae and minimum spore population was recorded in *Crotalariaeae juncea* (102/100 g of soil) belongs to Fabaceae (Fig. 5 and 6).

The Plant species like *Agapanthus africanus* 27% (Amaryllidaceae), *Helichrysum arenarium* 28% (Asteraceae), *Verbascum thapsus* 23% (Scrophulariaceae), *Rumex nepalensis* 29% (Polygonaceae), *Cestrum aurantiacum* 30% (Solanaceae), showed 20 and less than 30% of infection. The Plant species *Agertina adenophora* 33% (Asteraceae), *Dahlia imperialis* 35% (Asteraceae), *Plectranthus rugosus* 31% (Lamiaceae), showed 30 and less than 40% of AM fungal infection. The Plant species like *Anaphalis aristata* 45% (Asteraceae), *Crotalariaeae juncea* 49% (Fabaceae), *Erigeron karvinkianus* 48% and *Helichrysum bracteatum* 45% both belongs to the family Asteraceae 48 %. *Euphorbia rothiana* 47% (Euphorbiaceae), *Leucas suffruticosa* 42% (Lamiaceae), *Phytolacca octandra* 44% (Phytolaccaceae), showed 40 and less than 50% of infection. The Asteraceae member *Parthinium hysterophorus* 55%, *Tricholepis amplexicaulis* 59% and Fabaceae member *Ulex europaeus* 60% showed 50 and less than 60% infection. The Plant species like *Diplazium esculentum* 61% (Athyriaceae), *Hypochaeris radicata* 66% (Asteraceae), *Ipomoea carnea* 69% (Convolvulaceae), showed 60 and less than 70% of infection. The Plant species like *Bidens trichosperma* 71% (Asteraceae), *Solanum nigrum* 81% (Solanaceae), *Vinca major* 77% (Apocynaceae) showed 70 and less than (90%) of infection.

Table 1. List plant species and their medicinal uses.

S. No	Plant Species	Family	Habit	Parts Used	Medicinal uses
1.	<i>Agapanthus africanus</i> (L.) Hoffmanns.	Amaryllidaceae	Herb	Whole plant	Allergy, fever, impotence, skin diseases
2.	<i>Ageratina adenophora</i> (Spreng.) King & H. Rob	Asteraceae	Shrub	Leaves	Itching, menses scanty,
3.	<i>Anaphalis aristata</i> (D. C.)	Asteraceae	Herb	Whole Plant	Stomach Problems
4.	<i>Bidens trichosperma</i> (Michx.) Britton	Asteraceae	Herb	Flowers	Skin diseases and Itching
5.	<i>Cestrum aurantiacum</i> Lindl.	Solanaceae	Shrub	Leaves	Epilepsy
6.	<i>Crotalaria juncea</i> L.	Fabaceae	Herb	Whole plant	Swelling and Ulcers
7.	<i>Dahlia imperialis</i> Roetzl ex Ortgies	Asteraceae	Herb	Flower	Skin treatments,
8.	<i>Diplazium esculentum</i> (Retz.) Sw.	Athyriaceae	Herb	Leaves	Fever cold, cough
9.	<i>Erigeron karvinskianus</i> D. C.	Asteraceae	Herb	Leaves	Bee attractive Plant and Skin diseases
10.	<i>Euphorbia rothiana</i> Spreng.	Euphorbiaceae	Shrub	Leaves	cough, Abscesses, ulcer
11.	<i>Helichrysum aurantiacum</i> Boiss. & A. Huet	Asteraceae	Herb	Fruits	Gall bladder disorders,
12.	<i>Helichrysum bracteatum</i> (Vent.) Haw	Asteraceae	Herb	Seeds	Chest complaints
13.	<i>Hypochaeris radicata</i> L.	Asteraceae	Herb	Whole plant	Cough and cold
14.	<i>Ipomoea carnea</i> Jacq.	Convolvulaceae	Shrub	Leaves	Diabetic, Cancer,
15.	<i>Justicia procumbens</i> L.	Acanthaceae	Herb	Leaves	Diuretic, Asthma, Cough
16.	<i>Leucas suffruticosa</i> Benth.	Lamiaceae	Herb	Whole plant	Scorpion bites, Reduce fever
17.	<i>Parthenium hysterophorus</i> L.	Asteraceae	Herb	Roots	Rheumatic pain, Diarrhea
18.	<i>Phytolacca octandra</i> L.	Phytolaccaceae	Shrub	Whole plant	Impotency and also in down fever.
19.	<i>Plectranthus rugosus</i> Wall. ex Benth	Lamiaceae	Herb	Leaves	Cough and Cold,
20.	<i>Rumex nepalensis</i> Spreng.	Polygonaceae	Herb	Leaves	Skin sores
21.	<i>Solanum nigrum</i> L.	Solanaceae	Herb	Whole plant	Nonetheless and Locales
22.	<i>Tricholepis amplexicaulis</i> C. B. Clark	Asteraceae	Herb	Whole plant	Skin disease, Cough and Urinary troubles
23.	<i>Ulex europaeus</i> L.	Fabaceae	Shrub	Seeds	Blood problems
24.	<i>Verbascum thapsus</i> L.	Scrophulariaceae	Herb	Leaves	Respiratory, problems and ear pain, eczema
25.	<i>Vinca major</i> L.	Apocynaceae	Shrub	Leaves	Stomach problems, Cerebral stimulant

Table 2. AM fungal Colonization and spore Population of some Plant species in Yellanahalli, Valley view during, 2017-2018.

S. No	Plant Species	Family	pH	Types of infection			Spore Population (100g/soil)	(% of root colonization)
				Hyphal	Arbuscule	Vesicle		
1.	<i>Agapanthus africanus</i> (L.) Hoffmanns.	Amaryllidaceae	5.1	+	-	+	220	27
2.	<i>Ageratina adenophora</i> (Spreng.) King & H. Rob	Asteraceae	4.8	+	+		108	33
3.	<i>Anaphalis aristata</i> (D C.)	Asteraceae	6.1	+	-	+	110	45
4.	<i>Bidens trichosperma</i> (Michx.) Britton	Asteraceae	5.3	+	+	-	226	71
5.	<i>Cestrum aurantiacum</i> Lindl.	Solanaceae	6.4	+	-	+	177	30
6.	<i>Crotalaria juncea</i> L.	Fabaceae	5.5	+	+		102	49
7.	<i>Dahlia imperialis</i> Roehl ex Ortgies	Asteraceae	4.8	+	-	+	310	35
8.	<i>Diplazium esculentum</i> (Retz.) Sw.	Athyriaceae	5.2	+	+		265	61
9.	<i>Erigeron karvinskianus</i> D C.	Asteraceae	5.9	+	-	+	238	48
10.	<i>Euphorbia rothiana</i> Spreng.	Euphorbiaceae	6.0	+	+		224	47
11.	<i>Helichrysum aurantiacum</i> Boiss. & A. Huet	Asteraceae	5.3	+	-	+	188	28
12.	<i>Helichrysum bracteatum</i> (Vent.) Haw	Asteraceae	5.6	+	+		134	45
13.	<i>Hypochaeris radicata</i> L.	Asteraceae	5.8	+	-	+	199	66
14.	<i>Ipomoea carnea</i> Jacq.	Convolvulaceae	5.1	+	+		256	69
15.	<i>Justicia procumbens</i> L.	Acanthaceae	6.3	+	-	+	380	67
16.	<i>Leucas suffruticosa</i> Benth.	Lamiaceae	6.6	+	+		320	42
17.	<i>Parthenium hysterophorus</i> L.	Asteraceae	5.6	+	-	+	277	55
18.	<i>Phytolacca octandra</i> L.	Phytolaccaceae	5.8	+	+		219	44
19.	<i>Plectranthus rugosus</i> Wall. ex Benth	Lamiaceae	5.4	+	-	+	173	31
20.	<i>Rumex nepalensis</i> Spreng.	Polygonaceae	5.7	+	+	-	342	29
21.	<i>Solanum nigrum</i> L.	Solanaceae	5.9	+	-	+	287	81
22.	<i>Tricholepis amplexicaulis</i> C.B. Clark	Asteraceae	5.1	+	-	+	202	59
23.	<i>Ulex europaeus</i> L.	Fabaceae	5.0	+	-	+	258	60
24.	<i>Verbascum thapsus</i> L.	Scrophulariaceae	6.4	+	+	-	293	23
25.	<i>Vinca major</i> L.	Apocynaceae	6.2	+	-	+	328	77

Table 3. AM fungal genera and species were isolated from the rhizosphere soil samples in Yellannahalli hills Valley view of Nilgiri's.

S. No	AM fungal genera	Order	Family	Species
1	<i>Acaulospora</i>	Diversisporales	Acaulosporaceae	<i>levies</i> and <i>thomii</i>
2	<i>Claroideoglomus</i>	Glomerales	Claroideoglomeraceae	<i>etunicatum</i>
3	<i>Gigaspora</i>	Diversisporales	Gigasporaceae	<i>candida</i>
4	<i>Glomus</i>	Glomerales	Glomeraceae	<i>Glomus hoi</i> , <i>G. invermeyanum</i> , <i>G. macrocarpum</i> , <i>G. magnicaule</i> , <i>G. multicaulis</i> ,
5	<i>Racocetra</i>	Diversisporales	Gigasporaceae	<i>verrucosa</i>
6	<i>Rhizophagus</i>	Glomerales	Glomeraceae	<i>fasciculatus</i>
7	<i>Sclerocystis</i>	Glomerales	Glomeraceae	<i>pachycaulis</i>



Fig. 2. Identification of collected plant species at Yellannahalli hills, Nilgiris.

a) *Agapanthus africanus* (L.) Hoffmanns. b) *Bidens trichosperma* (Michx.) Britton c) *Dahlia imperialis* Roetzl ex Ortgies d) *Hypochaeris radicata* L.

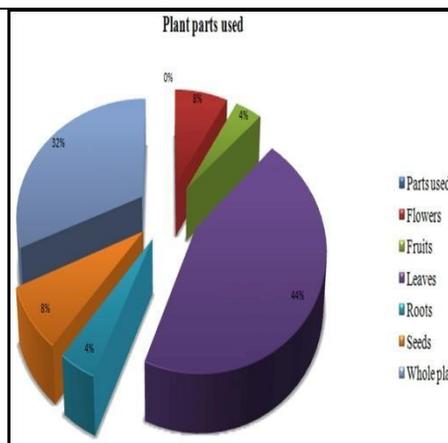
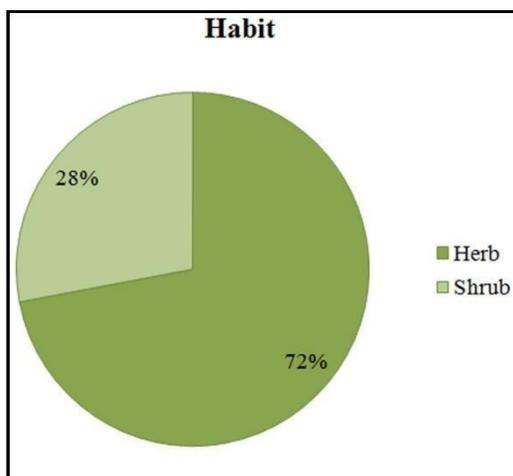


Fig. 4. Plant Parts Used.

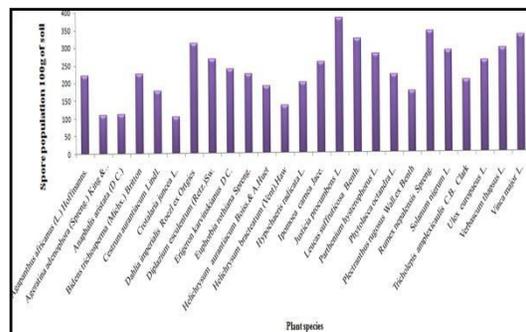


Fig. 5. AM fungal spore population in the plant species of Yellannahalli hills.

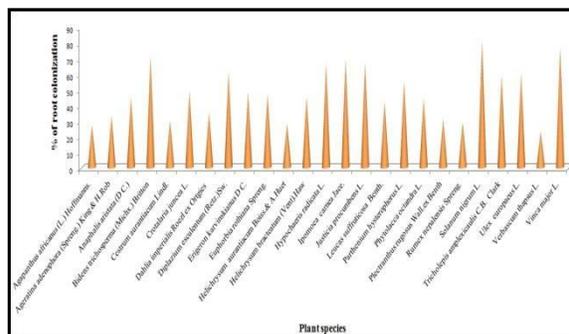


Fig. 6. Percentage of root colonization in the plant species of Collected plant families.

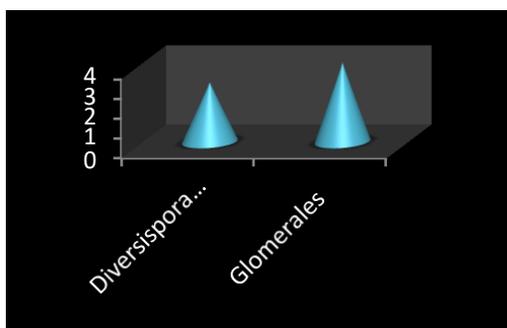


Fig. 7. Different orders of AM fungal species.

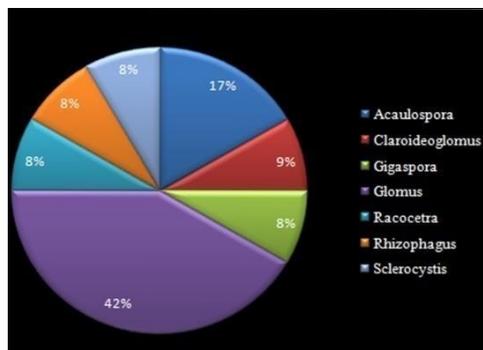


Fig. 8. Dominant species of the AM fungal genera.

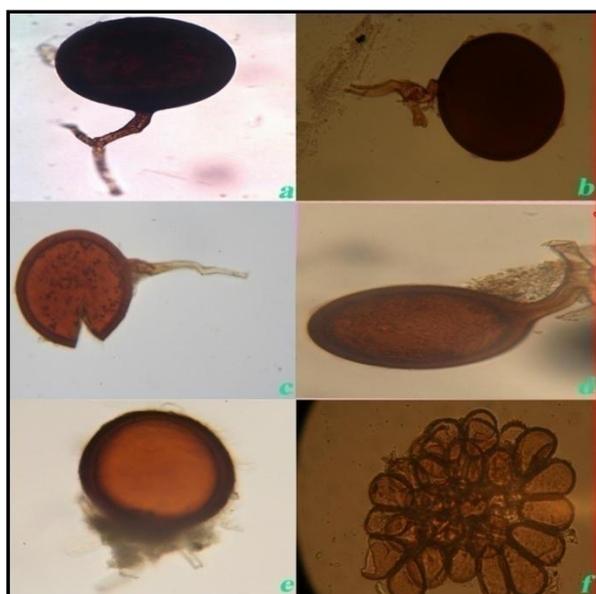


Fig. 9. Isolation and Identification of Arbuscular mycorrhizal fungal spores in Yellanahalli hills, Valley view, Nilgiris.

- a) *Acaulospora levies* b) *Gigaspora candida* c) *Glomus invermayanum*, d) *Rhizophagus fasciculatus* e) *Glomus multicaulis* f) *Sclerocystis pachycaulis*

From rhizosphere soil samples of Yellanahalli hills, totally 12 AM fungal species belongs to 7 genera and 2 different Orders were isolated and identified (Fig. 7). Of these 2 species of *Acaulospora*, *Aca. levies*, *Aca. thomii*, 1 species of

Claroideoglomus, *Cl. etunicatum*, 1 species of *Gigaspora*, *Gig. candida*, 5 species of *Glomus*, *Gl. hoi*, *Gl. invermayanum*, *Gl. macrocarpum*, *Gl. magnicaule*, *Gl. multicaulis*, 1 species of *Racocetra*, *Rac. verrucosa*, 1 species of *Rhizophagus*, *Rhiz. fasciculatus*, 1 species of *Sclerocystis*, *Scl. pachycaulis*. The genus *Glomus* was dominant and the name of the species were present in (Table. 3, Fig. 8, 9). Santhoshkumar and Nagarajan (15) reported that arbuscular mycorrhizal fungal association in the rhizosphere soil and root colonization of some medicinal plant Species in Sirumalai Hills Eastern Ghats of Dindugul District, Tamilnadu and they were reported totally 39 AM fungal species belonging to six genera were isolated and identified. The genus *Glomus* were found dominate followed by *Acaulospora*, *Sclerocystis*, *Entrophospora* and *Gigaspora*. Priyadarshini *et al.* (16) also reported that occurrence of VAM fungi in Kalasalingam University campus. They were isolated totally 26 species of vesicular arbuscular mycorrhizal fungal spores from the rhizosphere soil samples of the plant species belonging to 14 families was reported.

4. CONCLUSION

Based on this result, concluded that arbuscular mycorrhizal fungal root colonization and spore population were observed in the plant species of Yellanahalli hills. The symbiotic association of these arbuscular mycorrhizal fungal species *Glomus* was more abundant in all rhizosphere soil of the plant species. Further studies need for the tissue culture technique using mycorrhizal inoculation for ensuring enhanced the plant growth especially in agricultural crops..

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