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RESEARCH ARTICLE

STUDIES ON THE ARBUSCULAR MYCORRHIZAL FUNGAL ASSOCIATION IN THE PLANT SPECIES OF PONNUTHU HILLS, WESTERN GHATS COIMBATORE DISTRICT, TAMILNADU

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ABSTRACT

The present study analyzed the arbuscular mycorrhizal fungal spores in root colonization and spore population in rhizosphere soils samples in various medicinal plant species at Ponnuthu hills, Western Ghats of Coimbatore district, Tamilnadu, India. Root and rhizosphere soil samples were collected during the month of August, 2018-March, 2019 from the surface to 30 cm depth as well as pH were also recorded. Totally 36 plant species belonging to 21 families were collected and identified. The present result showed arbuscular mycorrhizal spore population in the rhizosphere soil and root colonization of all the plant species. A total of 21 AM fungal spores were recovered from the rhizosphere soil samples in this study region. The *Glomus* was dominant and found in rhizosphere soil samples in all the medicinal plant species. The maximum spore population was found in the rhizosphere soil samples of *Hemidesmus indicus* (573/100g of soil) which belongs to the family Asclepiadaceae and the lowest spore population was observed in the *Abutilon indicum* 145/100g of soil) which belongs to Malvaceae family. The highest 81% AM fungal colonization was found in roots of *Gymnema sylvestre* which belongs to the family Apocynaceae. While the lowest 16 % AM fungal colonization was found in the root of *Tridax procumbens* which belongs to the family Asteraceae.

Keywords: *Glomus aggregatum***,** Medicinal plants, Ponnuthu hills.

1. INTRODUCTION

India is recognized as one of the seventeen mega biodiversity zones of the world. The forest of Western Ghats, in view of their floristic diversity and numerous multipurpose species, are considered as a varietal storehouse of economically important plants and beneficial microbial communities. AM fungi are geographically ubiquitous in occurrence that have a broad range of dissimilar environments (1,2) from the arctic to the tropics and occupy a wide range of ecological niches (3). The fossil record suggests that AM were also present in the subterranean parts of the earliest land regions (4). Mycorrhizas are one such examples of a plant-fungal association that is found in plants under a range of abiotic conditions. Mycorrhizal symbiosis occurs in a vast majority of vascular plants except for members of a few families, including Cruciferae, Brassicaceae and Zygophyllaceae (5,6).

Arbuscular mycorrhizal symbiosis that appeared with the first land plants more than 400 million years ago, is still formed by the large majority of extent plant species with no host specificity (7). Occurrence of AM fungi has been reported from an exceptionally wide range of plant and different ecosystems and plays a major role in better nutrition, species diversity and survival. (8). Almost all higher plants in the terrestrial ecosystems are known to be associated with mycorrhizal fungi (9).

Associations between plants and arbuscular mycorrhizal fungi are common in natural and agricultural ecosystems. Soil microorganisms play important role in plant-soil interactions. Microbes alter nutrient availability, immobilize heavy metals in soils, and bind soil particles into stable aggregates (10). Of the several types of mycorrhizal fungi, the arbuscular mycorrhizal fungi (AMF) form important symbiosis with the flora prevalent in serpentine grasslands.

In developing countries and rural societies, the use of medicineal plants is both a valuable resource and necessity and furthermore it provides real alternative for primary health care systems (11). Globally about 85% of the traditional medicine used for primary healthcare are derived from plants. In many countries scientific investigations of medicinal plants have been initiated because of their contribution to healthcare. Herbal medicines have good values in treating many diseases including infectious diseases, etc.

2. MATERIALS AND METHODS

2.1. Study area Description

Kurudi Malai is the hill at the base of which Ponnuthu Amman temple is situated. It lies between 11.1186° N, 76.8923° E. Few even call it as Ponnuthu Malai. The summit is at a rough elevation of 1200m (3900ft). The terrain is rocky at the beginning and an

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abrupt rise in elevation arises as tracks of salt could be clearly seen on those rocks indicating the past traces of waterfalls. After which thick forest cover exists. Thorny shrubs can be found in abundance. Other smaller amphibians like newt were also spotted.

Fig. 1. Study area of Ponnuthu hills.

2.2. Sample collection

In this present study, root and rhizosphere soil samples of 36 plant species were collected for the duration of August, 2018- March, 2019. The collected soil and root samples were placed in the polyethylene bags, labelled 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 in study species.

2*.3. Estimation of AM fungal root colonization*

The root samples were cleared and stained in tryphan blue with a modified version of following method by Philips and Hayman's). The collected roots samples were cut into 1-2 cm pieces, heated at 90ºC 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 percentage of AM fungal infection was calculated using the formula:

2.4. Identification of AM fungi

The present study isolation and identification of AM fungal spores based upon their morphological characters such as spore size, color, hyphal attachement, cell wall layer characters, were identified in addition with nomenclature, keys of the following manual authors were used: Raman and Mohankumar (12) Schenk and Perez (13) and Schubler and Walker (14). The Photomicrographs were taken with the help of a Magnus Olympus Microscope.

2.5. Soil pH

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

3. RESULTS AND DISCUSSION

AM fungal colonization and spore population of 36 plant species belongs to 21 families and also analysis of the pH ranges between 4.8 and 6.9 are represented in the (Tables--1 & 2). The maximum temperature was recorded 36.5^{0}C while the maximum rain fall were noted in August, 390 mm.

The maximum spore population was observed in the plant species of *Hemidesmus indicus* (573/100g of soil) belonging to Asclepiadaceae and minimum was observed in *Abutilon indicum* (145/100g of soil) belonging to Malvaceae. In the present investigation the highest AM fungal infection was recorded in *Gymnema sylvestre* (81%) belonging to Apocynaceae and minimum was noticed in *Tridax procumbens* (16%) belonging to Asteraceae.

The plant species like *Acorus calamus* 28% (Acoraceae), *Barleria prionitis* 24% (Acanthaceae), *Catharanthus pusillus* 22% (Apocynaceae), *Commelina benghalensis* 28% (Commelinaceae), *Plectranthus barbatus* 38% (Lamiaceae), *Corchorus aestuans* 33% (Tiliaceae), *Capparis zeylanica* 30% (Capparidaceae), showed 20 to 40 % of infection.

The other plant species like *Areva lanata* 59% (Amaranthaceae), *Azadirachta indica* 55 % (Meliaceae), *Anisomeles malabarica* 43% (Lamiaceae), *Achyranthes aspera* 44 % (Amaranthaceae), *Abutilon indicum* 52 % (Malvaceae), *Argemone mexicana* 58 % (Papaveraceae), *Blepharis maderaspatensis* 48% (Acanthaceae), *Barleria cristata* 46% (Acanthaceae), *Catharanthus roseus* 41% (Apocynaceae), *Datura innoxia* 50% (Solanaceae), *Evolvulus alsinoides* 53% (Convolvulaceae), *Hibiscus micranthus* 46 % (Malvaceae), *Hibiscus vitifolius* 42 % (Malvaceae), *Ipomoea obscura* 58% (Convolvulaceae), *Leucas aspera* 43% (Lamiaceae), *Mimosa pudica* 44% (Mimoseaceae), *Oxalis corniculata* 52% (Oxalidaceae), *Ocimum sanctum* 49% (Lamiaceae) showed above 41 to 60% of infection.

The rest of the species like *Bacopa monnieri* 61% (Plantaginaceae), *Euphorbia hirta* 67% (Euphorbiaceae), *Ficus benghalensis* 64 % (Moraceae), *Oldenlandia umbellate* 69% (Rubiaceae),

Portulaca oleracea 65% (Portulacaceae), showed above 61 to 80% of infection. The plant family like Amaranthaceae and Juncaceae were thought to be mycorrhiza free, most of the species were found to be infected under natural stressed rangeland conditions (15). The plants that do not form mycorrhizas may be related to the presence of fungi toxic compounds in root cortical tissue or in root exudates. It may also be due to interactions between the fungus and the plant at the cell wall and (or) middle lamella level (16). High concentrations of salycilic acid have been found to reduce mycorrhization. But in the in the present study revealed that the plant family Amarathaceae showed the mycorrhizal infection. The research clearly showed that AMF enhanced nutrient uptake and growth of endangered plants (17,18).

Many studies conducted in different ethnic communities, have reported frequently the use of leaves was widely accepted for traditional therapies may be due to large quantity of biologically active components present inside them. Apart from leaves, almost all the other parts of medicinal plants such as flower, bark, stem, seed, fruit are also used. The utilization of leaves in traditional medication may also be due to their easy availability.

Table: 1. List of plants species collected from the Ponnuthu hills, and their medicinal uses.

S. No	Plant species	Family	Habit
1.	*Areva lanata* (L.) Juss. Ex schult.	Amaranthaceae	Herb
2.	*Azadirachta indica* A. Juss.	Meliaceae	Tree
3.	*Anisomeles malabarica* (L.) R.Br.ex Sims	Lamiaceae	Herb
4.	*Achyranthes aspera* L.	Amaranthaceae	Herb
5.	*Abutilon indicum* L.	Malvaceae	Shrub
6.	*Acalypha indica* L.	Euphorbiaceae	Herb
7.	*Acorus calamus* L.	Acoraceae	Herb
8.	*Argemone mexicana* L.	Papaveraceae	Herb
9.	*Blepharis maderaspatensis* (L.) Heyne ex Roth.	Acanthaceae	Herb
10.	*Bacopa monnieri* (L.)	Plantaginaceae	herb
11.	*Barleria cristata* L.	Acanthaceae	Shrub
12.	*Barleria prionitis* L.	Acanthaceae	Shrub
13.	*Crotalaria retusa* L.	Fabaceae	Herb
14.	*Corchorus aestuans* L.	Tiliaceae	Herb
15.	*Capparis zeylanica* L.	Capparaceae	Shrub
16.	*Catharanthus pusillus* (Murray) G. Don	Apocynaceae	Herb
17.	*Catharanthus roseus* (L.) G.Don	Apocynaceae	Shrub
18	*Commelina benghalensis* L.	Commelinaceae	Herb
19	*Datura innoxia* Mill.	Solanaceae	Shrub
20	*Evolvulus alsinoides* (Linn)	Convolvulaceae	Herb
21	*Euphorbia hirta* L.	Euphorbiaceae	Herb
22	*Ficus benghalensis* L.	Moraceae	Tree
23	*Gymnema sylvestre* R.Br.	Apocynaceae	Shrub
24	*Hibiscus micranthus* L.f.	Malvaceae	Shrub
25	*Hibiscus vitifolius* L.	Malvaceae	Herb
26	*Hemidesmus indicus* (L.)	Asclepiadaceae	Shrub
27	*Ipomoea obscura* (L.) Ker Gawl.	Convolvulaceae	Herb
28	*Leucas aspera* Linn.	Lamiaceae	Herb

29	*Mimosa pudica* L.	Fabaceae	shrub
30	*Oxalis corniculata* L.	Oxalidaceae	Herb
31	*Ocimum sanctum* L.	Lamiaceae	Herb
32	*Oldenlandia umbellate* L.	Rubiaceae	Herb
33	*Plectranthus barbatus* Andrews	Lamiaceae	Herb
34	*Phyllanthus amarus* Schumach & Thonn.	Phyllanthceae	Herb
35	*Portulaca oleracea* L.	Portulacaceae	Herb
36	*Tridax procumbens* L.	Asteraceae	Herb

Table 2. Arbuscular Mycorrhizal fungal spore population and root colonization in the plant species of Ponnuthu hills, a part of Western Ghats, Coimbatore district, Tamilnadu, during 2018-2019.

S. No	Plant Species	pH	Types of infection			Spore Population (100g/soil)	(%) root colonization
			Hyphae	Arbuscule	Vesicles		
1.	*Areva lanata* (L.) Juss. Ex schult.	5.2	+	-	+	320	61
2.	*Azadirachta indica* A. Juss.	4.8	+	+	-	280	55
3.	*Anisomeles malabarica* (L.) R.Br.ex Sims	6.2	+	+	-	410	43
4.	*Achyranthes aspera* L.	5.5	+	-	+	185	44
5.	*Abutilon indicum* L.	5.7	+	+	-	387	52
6.	*Acalypha indica* L.	6.0	+	-	+	156	39
7.	*Acorus calamus* L.	5.8	+	+	-	270	28
8.	*Argemone mexicana* L.	5.1	+	-	+	355	58
9.	*Blepharis maderaspatensis* (L.)	4.9	+	+	-	190	67
10.	*Bacopa monnieri* (L.)	5.3	+	-	+	177	65
11.	*Barleria cristata* L.	6.7	+	+	-	445	74
12.	*Barleria prionitis* L.	6.6	+	-	+	390	24
13.	*Crotalaria retusa* L.	5.9	+	+	-	365	43
14.	*Corchorus aestuans* L.	5.4	+	-	+	402	33
15.	*Capparis zeylanica* L.	5.3	+	-	+	339	30
16.	*Catharanthus pusillus* (Murray) G. Don	6.1	-	-	-	130	22
17.	*Catharanthus roseus* (L.) G.Don	6.4	+	-	+	470	60
18.	*Commelina benghalensis* L.	5.2	+	+	-	293	22
19.	*Datura innoxia* Mill.	6.3	+	-	+	245	51
20.	*Evolvulus alsinoides*	5.5	+	+	-	310	53

	(Linn)						
21.	*Euphorbia hirta* L.	6.8	+	-	+	360	67
22.	*Ficus benghalensis* L.	5.7	+	-	+	387	68
23.	*Gymnema sylvestre* R.Br.	6.9	+	-	+	422	81
24.	*Hibiscus micranthus* L.f.	5.6	+	+	-	126	46
25.	*Hibiscus vitifolius* L.	6.1	+	-	+	405	63
26.	*Hemidesmus indicus* (L.)	6.5	+	-	+	573	55
27.	*Ipomoea obscura* (L.) Ker Gawl.	5.8	+	+	-	280	58
28.	*Leucas aspera* Linn.	5.0	+	+	-	410	43
29.	*Mimosa pudica* L.	5.1	+	-	+	185	44
30.	*Oxalis corniculata* L.	5.4	+	+	-	387	52
31.	*Ocimum sanctum* L.	5.2	+	-	+	156	49
32	*Oldenlandia umbellate* L.	5.5	+	+	-	270	49
33.	*Plectranthus barbatus* Andrews	6.2	+	-	+	355	38
34.	*Phyllanthus amarus* Schumach & Thonn.	6.3	+	+	-	190	47
35.	*Portulaca oleracea* L.	5.7	+	-	+	420	65
36.	*Tridax procumbens* L.	5.3	+	+	-	445	16

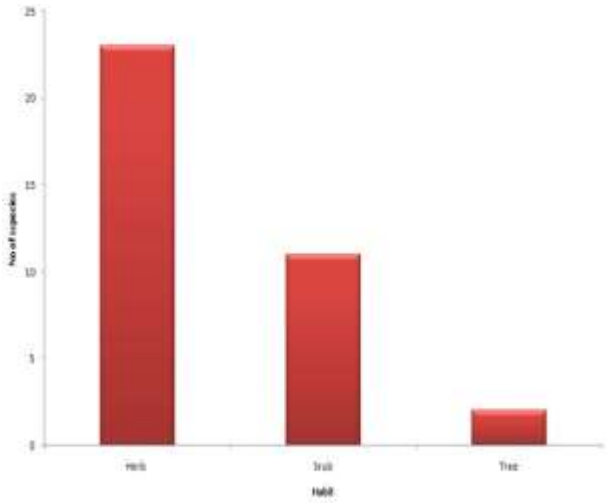

Fig. 2. Habit wise distribution of plant species in Ponnuthu hills.

From the rhizosphere soils sample of Ponnuthu hills, totally 21 AM fungal species were isolated and identified. Of these 4 species of *Acaulospora*, *Aca. delicata, Aca. denticulatum*, *Aca. gdanskensis Aca.levies*, 1 species of *Ambispora*, *Amb. appendicula*, 1 species of *Gigaspora, Gig.candida*, 13 species *of Glomus*, *Gl. aggregatum*, *Gl. albidum*, *Gl. ambisporum*, *Gl. arborense*, *Gl. australe*, *Gl. canadense*, *Gl.citricola*, *Gl. delhiense*, *Gl. deserticola*, *Gl. dimorphicum*, *Gl. radiatum*, *Gl. segmentatum*, *Gl. versiforme*, 1 species of *Redeckera*, *Red*. *fulvum*, 1 species of *Rhizophagus*, *Rhi*. *fasciculatus* were observed.

The names of the species are represented in Table 3. In addition with Santhoshkumar and Nagarajan (19) were studied on AM spore population in the plants species at Sirumalai hills, Eastern Ghats of Dindugul district. Totally 39 AM fungi species belongs to 6 genera were isolated and identified. To isolate and identification of the Am fungal spores in rhizosphere soils samples in different regions such as Yellanahalli hills reported by (20), Kondranghi hills (21), Bargur hills (22).

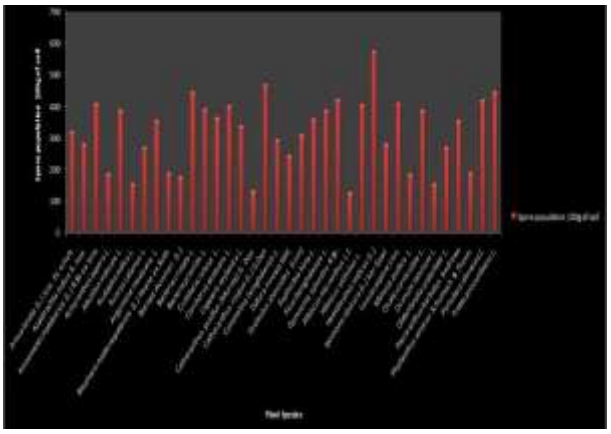

Fig. 3. AM fungal spore population of the plant species of Ponnuthu hills.

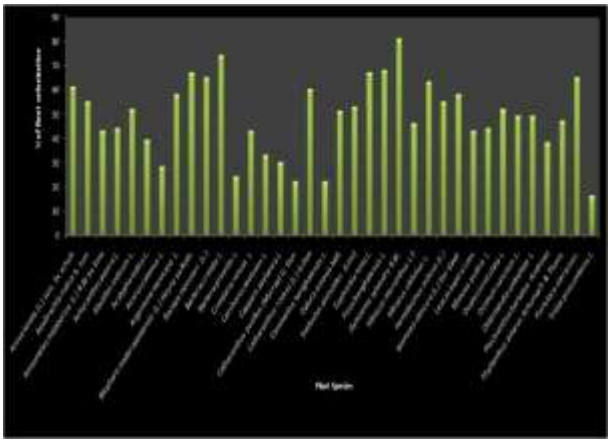

Fig. 4. AM fungal root colonization in collected plant species from Ponnuthu hills.

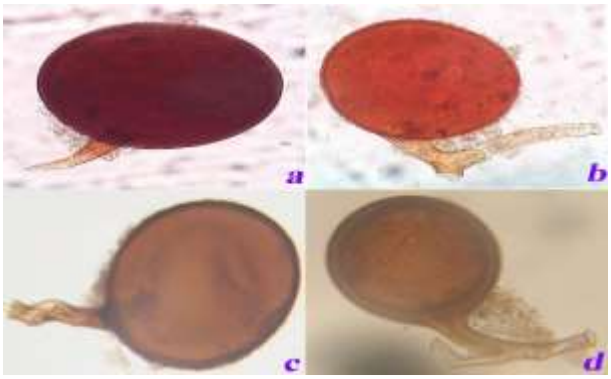

Fig. 5. Isolation and Identification of AM fungal spores in rhizosphere soils of Ponnuthu hills, Western Ghats of Coimbatore district.

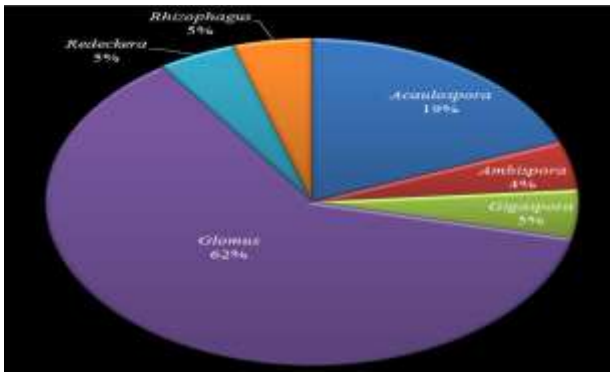

Fig. 6. Dominant genus was recovered from the rhizosphere soils samples in Ponnuthu hills.

4. CONCLUSION

From above results we concluded that the present study the AM fungal root colonization and spore population in all the plant species in Ponnuthu hills. In this symbiotic association of AM fungi in the plant species to absorb the soil nutrients, zinc, copper especially phosphorous and also increased plant resistance to various stresses like drought, salt and heavy metal. In future, the AM fungal spores were cultured under *in vitro* condition for raise agricultural crops plant species growth and development.

REFERENCES

1. Gerdemann, J.W. and J.M. Trappe, (1974). The Endogonaceae in the Pacific Northwest. *Mycological Memoirs* **5**: 1–76.
2. Koske, R.E. (1987). Distribution of VA mycorrhizal fungi along a latitudinal temperature gradient. *Mycologia* **79**: 55-68.
3. Srivastava, D., R. Kapoor, S.K. Srivastava, and K.J. Mukerji, (1996). Mycorrhizal research A priority in agriculture. In: K.G. Mukerji (ed.), Concepts in Mycorrhizal Research. Kluwer Academic Publishers, Dordrecht, The Netherlands.
4. Butler, E.J. (1939). The occurrences and systematic position of the Vesicular arbuscular type of mycorrhizal fungi. *Trans. Br. Mycol. Soc.* **22**: 274-301.
5. Newman, E.I. and P. Reddell, (1987). The distribution of mycorrhizas among families of vascular plants. *New Phytol.* **106**: 745-751.
6. Dhillon, S.S. and C.F. Friese, (1994). The occurrence of mycorrhizas in prairies: application to ecological restora-tion. Thirteenth North American Prairie Conference.

7. Redecker, D., R. Kodner and L.E. Graham, (2000). Glomalean fungi from the Ordovician. *Science* **289:** 1920-1921.
8. Bergelson, J.M., and J.M. Crawley, (1998). Mycorrhizal infection and plant species diversity. *Nature* **334**: 202-204.
9. Smith, S. and D.J. Read (1997). Mycorrhizal symbiosis. Second edition Academic Press, London. 605 pp.
10. Shetty, K.G., B.A.D. Hetrick, D.A.H. Figge and A.P. Schwab, (1994). Effects of mycorrhizae and other soil microbes on revegetation of heavy metal contaminated mine spoil. *Environ. Pollut*. A **86**: 181-188.
11. Alexiades, M.N., and D. Lacaze, (1996). Fenamad program in traditional medicine: an integrated approach to health care in the Peruvian Amazon. In: Balick, M.J., Elisabetsky, E., Laird, S.A. (Eds.), Medicinal Resources of the Tropical Forest. Columbia University Press, New York, pp. 341–365.
12. Raman, N., and V. Mohan Kumar, (1988). Techniques in Mycorrhizal Research. University of Madras, Madras. 279p.
13. Schenck, N.C., and Y. Perez, (1987). A Manual for the Identification of VA-mycorrhizal Fungi, University of Florida, Synergistic Publications, Gainesville, Florida, U.S.A.: 245.
14. Schubler, A., and C. Walker, (2010). The Glomeromycota: a species list with new families and new genera. Online: **1**(58): 41-75.
15. Neeraj, A., J. Shauker Mathew, and A. Varma, (1991). Occurrence of vesicular-arbuscular mycorrhizae with Amaranthaceae in soils of the Indian semi-arid region. *Biol. fertile soils* **11**: 40-144.
16. Tester, M., S.E. Smith, and F.A. Smith, (1987). The phenomenon of "nonmycorrhizal" plants. *Can. J. Bot*. **65**: 419-431.
17. Barroetavena C., S.D. Gisler, D.L. Lumoa and R.J. Meinke, (1998). Mycorrhizal status of the endangered species Asragalus applegatei Peck as determined from a soil bio assay. *Mycorrhiza*, **8**: 117-119.
18. Panwar J., and A. Vyas, (2002). AM fungi: A biological approach towards conservation of endangered plants in Thar desert, India. *Curr. Sci*. **82** (5): 576-578.
19. Santhoshkumar, S., and N. Nagarajan, (2017). Arbucular Mycorrhizal Fungal Association in the Rhizosphere Soil and Root Colonization of Some Medicinal Plant Species in Sirumalai Hills Eastern Ghats of Dindugul District Tamilnadu. American-Eurasian *J. Agric. & Environ. Sci.* **17** (3): 206-212.
20. Santhoshkumar, S., N. Nagarajan and K. Santhoshkumar (2018). Studies on the Arbuscular Mycorrhizal Fungal biodiversity in the Plant species of Kondranghi hills, Dindugul district, Tamil Nadu, India. Kong. Res. J. **5**(2): 34-40.
21. Santhoshkumar, S., N. Nagarajan, R. Prema, R. Kowsaliya, F. Amjath Alikhan and P. Aishwarya (2018). Studies on the Arbuscular Mycorrhizal Fungal biodiversity in the plant species of Yellanahalli hills, Valley View of Nilgiris, Udhagamandalam, Tamil Nadu, India. *Kong. Res. J.* **5**(2): 27-33.
22. Santhoshkumar, S., N. Nagarajan and S. Naveen (2019). Association of Arbuscular Mycorrhizal Fungal species in the plant species of bargur hills, Erode District, Tamil nadu, India. *Kong. Res. J.* **6**(1): 60-65.