

Arbuscular Mycorrhizal fungal diversity in Coastal region of Manapaadu near Tiruchendur, Tamil Nadu

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ABSTRACT

The diversity of Arbuscular Mycorrhizal fungal distribution was studied in the coastal region of Manapaadu near Tiruchendur of Tamil Nadu. AM-Fungi are ubiquitous and form a mutual relationship with roots of most plant species. Spores and sporocarps present in the coastal sand were isolated by using the decanting and wet-sieving technique. Totally 36 species of AM spores belonging to genera viz. Acaulospora, Diversispora, Entrophosphora, Gigaspora, Glomus, Rhizophagus, Sclerocystis and Septaglomus were isolated from the rhizosphere soil of Manapaddu coastal region. The distribution of AM spores in soil is governed by edaphic factors like soil pH and E.C. were measured under laboratory conditions. The occurrence of AM fungi in soil favours the growth of plants and also which were mainly involved in the soil aggregation.

Key words: Arbuscular Mycorrhiza, AM diversity, AM-spores, soil pH and E.C.

INTRODUCTION

Arbuscular mycorrhizal (AM) fungi are common in sand dune systems throughout the world and are known to significantly contribute to the development of plant community structure and sand dune stabilization^{13,14,22,24}. AM fungal interaction in rhizosphere facilitates adequate nourishment of dune vegetation and their establishment¹³.

Arbuscular Mycorrhizal (AM) fungi are ubiquitous in occurrence⁸. The diversity of AM fungi studied in different soils of cultivated cereal crops and medicinal plants in TamilNadu^{17,21,25,29,31} and in the coastal regions of Kongan and Shervaroyan hills of Tamil Nadu⁷, south east coast of Tamil Nadu²³, Western coast of India³ and Western Ghats of Goa¹².

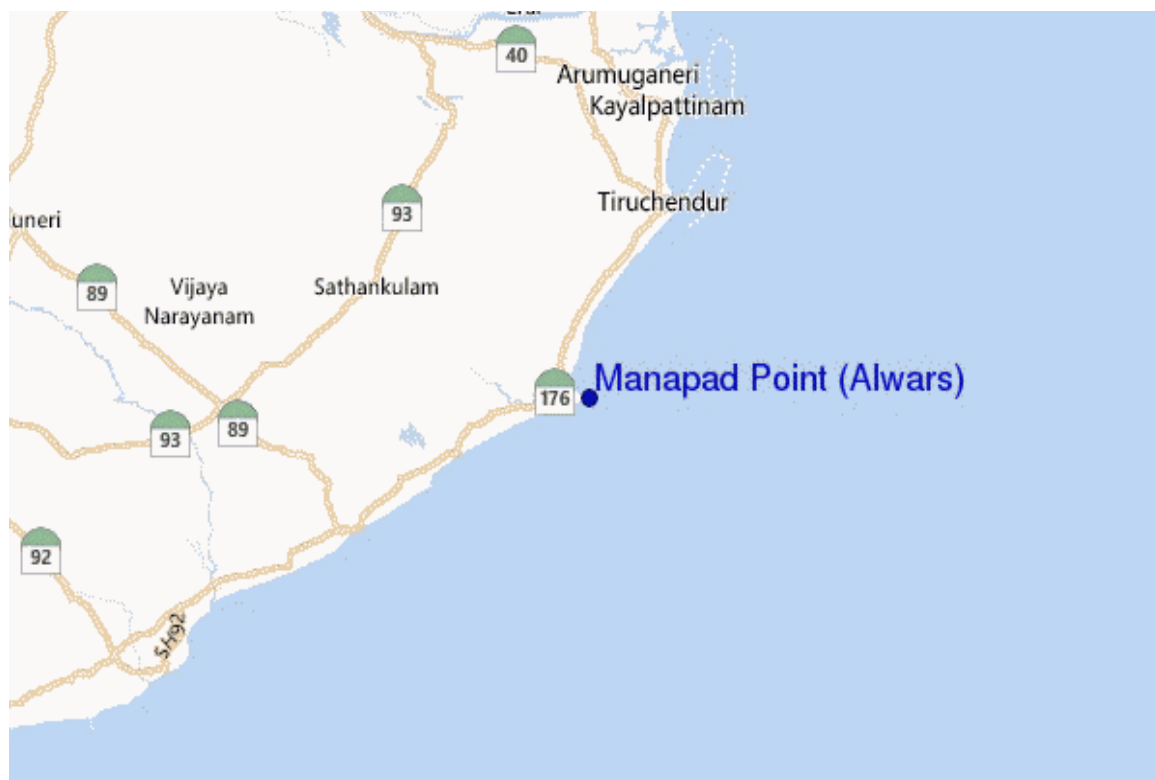
Mycorrhizae are also known to improve soil structure and stability by forming aggregates¹⁰. It is evident that hyphae of AM fungi bind sand grains forming sand aggregates, which remain intact even after the death of the root and hyphae¹³. Sand aggregates of different sizes resist strong winds and storms^{13,22}. Most studies on AM fungi of coastal dunes are confined to temperate regions, only a few are available from the Indian coast.

The coastal soil examined in this study was collected from Manapaadu near Tiruchendur of Tamil Nadu. The present study analysed the diversity of AM fungi and its distribution and soil characteristics of Manapaadu coastal region.

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MATERIALS AND METHODS

Soil sampling: The rhizosphere soil of *Distichlis distichophylla*, *Cucumis humifructus*, *Tephrosia purpurea* and *Prosopis juliflora* samples were collected from coastal region of Manapaadu region nearer to Tiruchendur (Plate: 1& 2). The soil samples were collected randomly after removing 5-10cm of top layer from the selected locality.



Soil pH:

Ten grams of air dried soil were added to 100 ml of distilled water and made to a suspension of 1: 10 (w/v) solution. Then the pH of the suspension was determined using pH meter.

EC (Electrical conductivity)¹¹

Ten grams of air dried soil were added to 100 ml of distilled water to get a suspension of 1:10 (w/v) solution. The electrical conductivity of the suspension was measured in a digital electrical conductivity meter.

Isolation of AM spores from soil samples

Spores and sporocarps present in the root zone soil were isolated following the decanting and wet-sieving technique. Five grams of soil samples were suspended in water and were allowed to settle down for few minutes. The suspension was passed through a series of sieves with 250, 206, 90 and 40 μm pore size. The spores in the soil suspension were collected.

The spores collected were placed on the filter paper and examined under a binocular microscope, transferred to a clean microscopic slide with the help of a fine needle and mounted in lacto phenol. Semi-permanent slides were made by sealing the edges of the cover slip with nail polish. Microscopic observations were made under high magnification for qualitative and quantitative characters of spores.

AM spore identification

The isolated AM fungal spores were identified using the key's proposed by Gerdemann and Trappe⁶, Trappe³², Schenck and Perez^{26,28} and INVAM⁹ (www.invam.caf.wvu.edu).

Key to AM Genera¹⁹

A. Azygospores/ zygozspores produced on the apex of a sporogenous cell of a fertile hypha Auxillary cell formed with a single family - Gigasporaceae

- B.** Germ tube produced directly through the spore wall; inner flexible wall group absent; auxillary cells finely papillate or echinulate - *Gigaspora*
- BB.** Germ tubes formed from germination shield inner flexible wall group present auxillary cells knobby, broadly papillate or smooth - *Scutellospora*
- AA.** Intercalary or terminal chlamydo spores formed singly and or in sporocarps. Auxillary cells not produced - Glomineae
- C.** Chlamydo spores born on fertile hyphae - Glomaceae
- D.** Fruiting body, the sporocarp composed of spores with lateral walls adherent to one another connecting hyphae embedded in a central hyphal plexus, chlamydo spores in a single layer except at a base composed of sterile hyphae - *Sclerocystis*
- DD.** Spores produced single or loose to tight aggregates in soil, less commonly in roots: fruiting body is a sporocarp but not formed as in D above - *Glomus*
- CC:** Chlamydo spores formed from or within the “neck” of sporogenous saccule - Acaulosporaceae
- C:** Spores arise laterally from the neck of sporogenous saccule - *Acaulospora*
- EE.** Spores formed in the neck of sporogenous saccule - *Entrophospora*

Glomus

Sporocarpic and non-sporocarpic, chlamydo spores formed at a hypha tip, usually one per tip. Two or more hyphae may be attached to spores of some species. Spores of most *Glomus* species are borne singly in soil, but some of the same species may also form in the cortex of roots or in sporocarps. A few species are known only form sporocarps. Spores globose- verily shaped 200-500µm in diameter, wall smoothly, warty rods. Single wall layer or double layered rarely 3 or more. Wall thickness 20µm spores of some *Glomus* species are wrapped in interwoven hyphae.

Acaulospora

Azygospores bud laterally from the funnel shaped stalk of a large inflated hypha terminus. Spores are sessile, single, no subtending hyphae, globose, 100 to 400µm in diameter. Multiple walls layered or single wall layered.

Gigaspora

Azygospores bud from the bulbous suspensor like tip of hyphae. As the spore expands and approaches maturation, one to several septa form below the suspensor like bulb, which usually remains attached to the spore and produces one to several narrow out growing hyphae. The spores are borne singly in soil, 200-600µm in diameter, 20µm wall thickness, multiple wall layers and also single wall layer. Spores have no flexible inner walls presence of direct germination and with echinulate auxillary cells *Gigaspora* species can generally be identified by spore characters and auxillary cells.

Scutellospora

One or two flexible inner walls, germination through germination compartments and with knobby auxillary cells produce azygospores terminally on a large swollen suspension like cells.

Entrophospora

Azygospores are formed within the funnel-shaped stalk of a large, inflated hypha terminus. The spores are borne singly in soil. Only one species has been described but second has been recently discovered.

Sclerocystis

Chlamydo spores form in sporocarps as a single, crowded layer of erect spores that surrounds the sides and top of a spore-free central mass of tightly interwoven hyphae. The sporocarps may be borne singly in soil or fused together in crusts on organic debris or moss at the soil surface.

RESULT AND DISCUSSION

Naturally soil provides the physical support needed for the anchorage of the root system of plant and also serves as the reservoir of air, water and nutrients which are essential for plant growth. Soil aggregation is a vital factor in plant growth, since movement of air; water and transfer of energy are interlinked with the porosity of soil. The stability of soil aggregates depends on the organic matter content of individual soils

and the nature of microbial products which binds the particles together. The organic content in soil samples was considered as one of the key determinants driving the microbial community structure³³.

The characteristics of rhizosphere soils of coastal grown plant species from Manapaadu locality were shown in Table: 1. Soil pH and EC played an important role in distribution of AM fungal spores in coastal zones of Manapaadu. The soil pH was lightly alkaline in all samples. The EC was higher ($464 \pm 2.65\mu\text{s}$) in *Distichlis distichophylla* and low EC ($174.82 \pm 0.84\mu\text{s}$) was observed in *Cucumishumofructus* sample. The diversity of AM fungal species (36 species) were isolated (Plate:3) from the soils of different study sites belong to the genera *Acaulospora*, *Diversispora*, *Entrophospora*, *Gigaspora*, *Glomus*, *Rhizophagus*, *Sclerocystis* and *Septoglomus*. Among them fourteen species species belongs to *Glomus*, ten species belongs to *Acaulospora*, three species belongs to *Diversispora* and *Rhizophagus*, two species belongs to *Sclerocystis* and *Septoglomus* and one species from *Entrophospora* and *Gigaspora*. The number of AM fungal spores ranged from 12.0 ± 1.00 (*Entrophospora schenckii*) to 48.0 ± 1.00 (*Glomus versiformae*) / 10g soil in rhizosphere of Manapaadu coastal plant species (Table:2).

In the present study, the AM spores were identified up to species level with the help of VAM fungi identification manual^{26,27,28} under binocular microscope. The following AM spores were identified as *Acaulosporabireticulata*, *A. colombiana*, *A. delicata*, *A. denticulata*, *A. dilatata*, *A. elegans*, *A. foveata*, *A. morrawae*, *A. rehmi*, *A. scrobiculata*, *Diversispora epigaea*, *D. eburnea*, *D. sparca*, *Entrophospora schenckii*, *Gigaspora rosea*, *Glomus ambisporum*, *G. aggregatum*, *G. botryoides*, *G. clarum*, *G. clavisporum*, *G. geosporum*, *G. hoi*, *G. macrocarpum*, *G. mosseae*, *G. pansihalos*, *G. leptodicum*, *G. verruculosum*, *G. versiformae*, *G. sinuosum*, *Rhizophagus aggregatus*, *R. fasciculatus*, *R. intraradices*, *Sclerocystis sinuosum*, *Sclerocystis pakistanica*, *Septaglomus constrictum* and *S. viscosum*. The *Acaulospora* and *Glomus* was recorded predominant species in all rhizosphere soil of coastal grown plants.

The spores of AM fungi were recovered at a depth of 130 cm from the rhizosphere of the perennial grass, *Elmus mollis* in Japanese coast¹. It has been demonstrated that sand dune AM fungal spores withstand immersion in seawater and get dispersed to new sites through seawater medium¹⁵.

A survey of 53 sand dune species along the Chennai coast showed colonization of most of them²⁰. Another survey of 10 sand dune species at four locations in south-east Tamil Nadu revealed high AM spore density during summer season⁵, whereas 12 plants of someshwara dunes on the west coast showed the presence of 16 AM fungi¹⁶. However, with increased disturbance at locations like Kaup, the AM fungal population has been decreased drastically².

The distribution of AM spores in rhizosphere soil was governed by edaphic and certain climatic factors. The physical factors in soil affecting plant growth are pH, electrical conductivity, macro nutrients and moisture holding capacity. Retention of water in soil is related to pore space and capillary action of soil particles while pH is dependent on the chemical conditions of soil. The soil pH did not influence the mycorrhizal spore density and frequency⁴.

The water holding capacity of a soil is governed by the porosity or soil moisture. The field capacity of a soil is the amount of water held in the soil after the excess gravitational water has drained away. This can be measured by saturating the soil with water followed by draining for 2-3 days under normal conditions which is then expressed as the percentage of water in the dry weight volume of the soil. Soil moisture plays a significant role on mycorrhizal development and colonization³⁰.

Table 1: Soil characteristics of rhizosphere soil from Plants growing in the Coastal region of Manapaadu

S.No	Plant Name	pH	EC (μs)
1	<i>Distichlis distichophylla</i>	7.66 ± 0.02	464 ± 2.65
2	<i>Cucumis humofructus</i>	7.48 ± 0.40	174.82 ± 0.84
3	<i>Tephrosia purpurea</i>	7.75 ± 0.03	232.2 ± 3.19
4	<i>Prosopis juliflora</i>	7.72 ± 0.03	223.4 ± 1.14

Table: 2 AM fungal diversity in the Coastal region of Manapaadu

S.No	AM spore	Number of spores /10g soil
1	<i>Acaulospora bireticulata</i>	13±1.00
2	<i>A. colombiana</i>	16±1.00
3	<i>A. delicata</i>	18±2.00
4	<i>A. denticulata</i>	23±1.00
5	<i>A. dilatata</i>	27±1.00
6	<i>A. elegans</i>	24±2.00
7	<i>A. foveata</i>	20±2.00
8	<i>A. morrawae</i>	30±1.00
9	<i>A. rehmi</i>	32±1.00
10	<i>A. scrobiculata</i>	38±3.00
11	<i>Diversispora epigaea</i>	21±1.00
12	<i>D. eburnea</i>	24±1.00
13	<i>D. sparca</i>	19±1.00
14	<i>Entrophospora schenckii</i>	12±1.00
15	<i>Gigaspora rosea</i>	36±2.00
16	<i>Glomus ambisporum</i>	39±2.00
17	<i>G. aggregatum</i>	40±1.00
18	<i>G. botryoides</i>	21±1.00
19	<i>G. clarum</i>	23±3.00
20	<i>G. clavisorum</i>	27±1.00
21	<i>G. geosporum</i>	14±1.00
22	<i>G. hoi</i>	29±2.00
23	<i>G. macrocarpum</i>	37±1.00
24	<i>G. mosseae</i>	35±1.00
25	<i>G. pansihalos</i>	32±2.00
26	<i>G. leptodicum</i>	21±1.00
27	<i>G. verruculosum</i>	26±2.00
28	<i>G. versiformae</i>	48±1.00
29	<i>G. sinuosum</i>	41±2.00
30	<i>Rhizophagus aggregatus</i>	34±1.00
31	<i>R. fasciculatus</i>	38±1.00
32	<i>R. intraradices,</i>	32±2.00
33	<i>Sclerocystis sinuosum</i>	27±1.00
34	<i>Sclerocystis pakistanica,</i>	29±1.00
35	<i>Septaglomus constrictum</i>	21±2.00
36	<i>S. viscosum</i>	20±1.00

Plate: 1 Manapaadu Coastal zone site for Sampling

Plate:2 Specific coastal grown plants in the Manapaadu site (a-f)

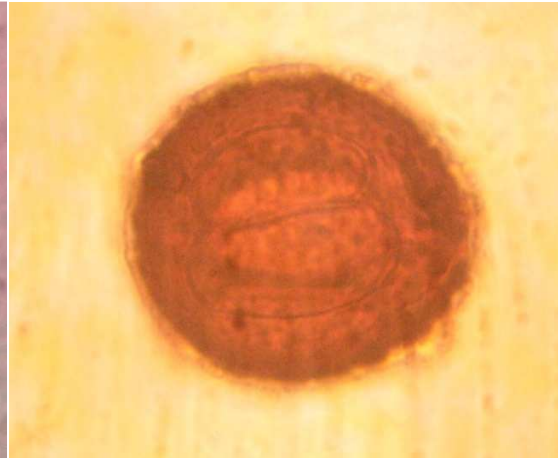


Plate: 3AM fungal spores isolated from rhizosphere soil from plants grown in coastal region of Manapaadu, near Tiruchendur

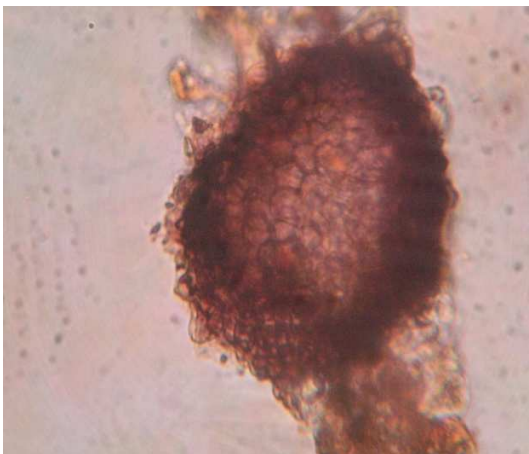
Acaospora dentiulculata



A. dilatata



A. elegans



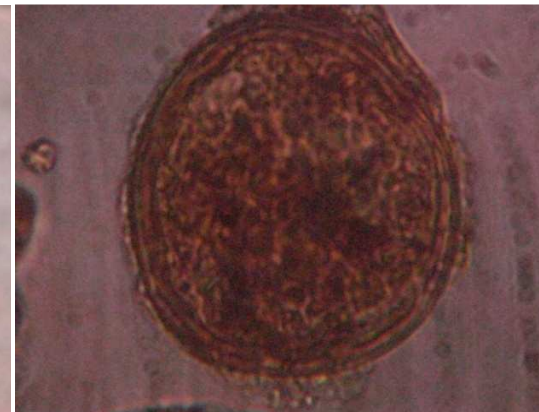
A.rehmii



Diversispora epigaea



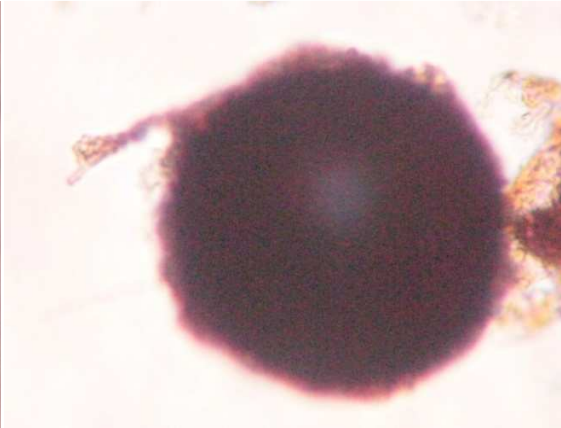
Entrophospora schenckii



Gigaspora rosea



Septoglomus constrictum



Glomus aggregatum



Glomus ambisporum



G. macrocarpum

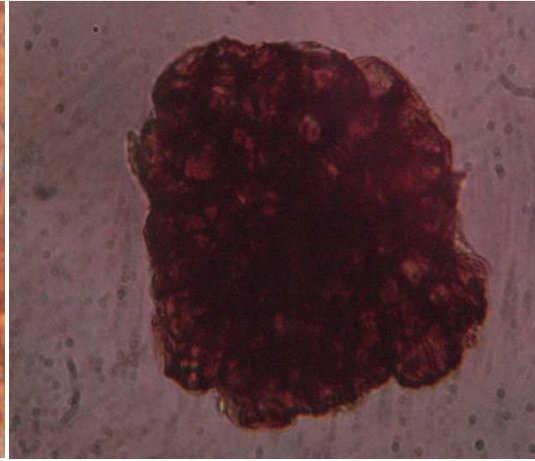
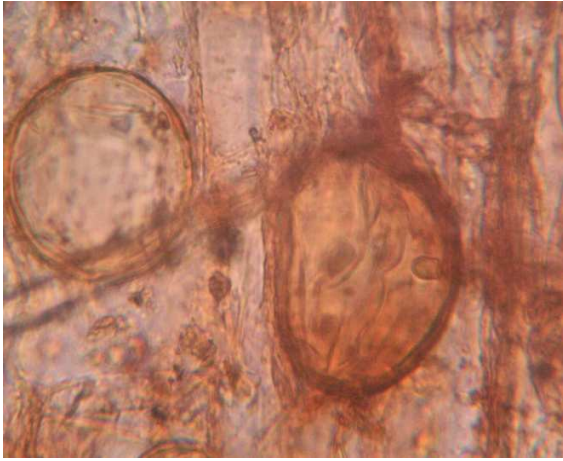


G. versiformae



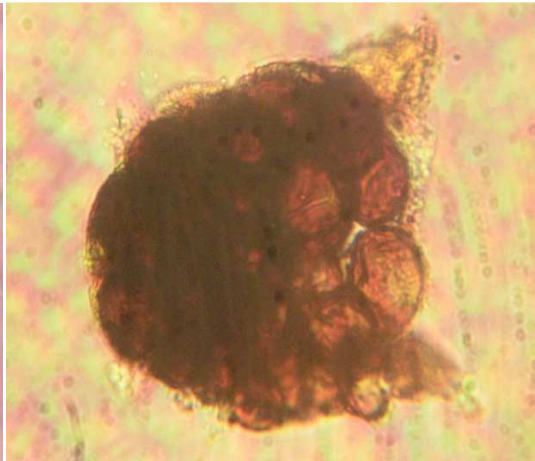
Glomus versiformae and *Rhizophagus intraradices*

Sporocarp of Glomus sinuosum



Funneliformis mosseae

Sporocarp of Sclerocystis sinuosum



G. hoi

G. Verruculosum



CONCLUSION

Coastal sand dunes are common in different parts of the world. These are natural structures which protect the coastal environment by absorbing energy from wind, tide and wave action¹⁸. Plants establishing on coastal sand dunes are subjected to several environmental fluctuations which affect their growth, survival and community structure. The present study concluded that the huge distribution of AM fungal diversity in coastal region of Manapaadu region. It was lightly alkaline and show various measuring of electrical conductivity. The AM Fungal distribution and physical properties of such soil favours the coastal vegetation and also maintain sand dune stabilization.

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