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Diversity of driftwood associated marine fungi of the Muthupet mangrove of Tamilnadu, India

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ABSTRACT

Marine mycology is one of the major emerging areas or research in tropics. It is gaining importance not only due to taxonomic and ecological perspective, but also for unique metabolites biochemical and enzymes from the sea. Majority of the studies on marine fungi are confined to the temperate regions. Studies in tropics and subtropics gained importance only in last two decades. So in this present study addresses the marine fungi undertaken in tropical/subtropical habitats especially driftwoods in muthupet mangroves. Diversity of marine fungi colonizing the drift wood samples was assessed and the common and dominant driftwood inhabiting marine fungus were grown on seven different media. A total number of 23 species (11 genera) of fungi were recorded from the driftwood samples collected from five different stations of Muthupet mangrove (Chief corner, Koraiyar River, Saradi, Sethuguda, Xavier Munai). They were assignable to ascomycotina (3 species), basidiomycotina (1 species) and Deuteromycotina (19 species). The common and dominant marine fungus was showed fast growth in SWPDA.

Introduction

Mangroves represent one of the most productive ecosystems in tropical environment and are characterized by efficient turnover of nutrients. (Kotari., 2002).Mangrove forests are the hot spots of biodiversity and also for marine fungi (Rani., 2009) Mangroves are highly productive ecosystem next to the coral reefs and provide energy to marine habitats through production and decomposition of plant detritus (Ananda.k., 2004, Lugo.a., 1974). Marine filamentous fungi have been reported on a variety of detritus such as, decaying wood, leaves, seaweeds, and sea grasses, calcareous and chitinous substrates. A variety of allochthonous and autochthonous substrata are available in sea for fungal colonization. The substrata include leaves, roots, sea weeds, leaves, seedlings, prop roots and wood of mangroves and terrestrial habitats, dead animals, faecal pellets, chitinous exoskeleton, and corals (K.R.Srithar et al, 2001)

Marine fungi play an important role in nutrient regeneration cycles as decomposers of dead and decaying organic matter in the estuaries.(Hyde and lee, 1995).,Although mangroves are the dominant features of Indian coastline and provides niches and habitats for many marine organisms.Noverthless very little is known about the fungi associated with them till recently . More than 500 species of marine fungi have been described all over the world (Jones, 1993 b). The marine mycota is represented by lower fungi (Haplomastigo mycotina and Diplomastigomycotina) and higher fungi (Ascomycotina, Basidiomycotina, and Deuteromycotina). Marine fungi are adapted to the saline environment; most of them require seawater for the successful completion of their lifecycle. The spores of marine fungi show adaptation to the marine ecosystem in the production of appendages, which facilitate buoyancy in water, entrapment and adherence to substrates. Pioneering studies of Barghoorn and Linder 1944, on the fungi on submerged wood in the sea made the greatest impact on the development of marine mycology. The progress becomes rapid following the publication of the book “Fungi in oceans and Estuaries” by T.W.Johnson and F.K.Sparrow (1961). Among the plant detritus, intertidal driftwood are one of the important and interesting objects for fungal diversity study, as they are unknown origin of plant and drifted to any part of the coast by wind, wave action and water movement. They get exposure to sunlight and atmosphere at a frequent interval due to the tidal variations. Further, the study will give clues on the fungi responsible in the precise role of break down of driftwood and thereby take part in the nutrient cycles of marine environment. The marine fungi occurring on the driftwood samples collected from muthupet mangrove environment are deal with this article.

Materials and methods

Muthupet mangrove forest situated in Thiruvarur district, Tamil nadu, South India is covering an area of about 20,000ha.The mangrove always is an association of halophyte trees, shrubs and other plants growing in brackish to saline tidal water of tropical and subtropical coastlines.Muthupet mangrove is a reserve forest.Avicenia marina (95%) is the dominant element of the forest and the trees to the height of 20 feet.

Collection of samples

Small wood pieces of mangrove were collected from five places (Saradi, Xavier munai, Chief corner, Sethukuda and Koraiyar River).

Mycological examinations

After collection, the samples were kept in new polythene bags, tied with a string and transported to the laboratory. From this, some wood pieces were taken and their surfaces were

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scrabbed with the help of a new blade and particles were used for plating technique using PDA. The remaining wood pieces were incubated with 50% sterilized distilled water in order to maintain the moisture condition at room temperature for seven days. After incubation, all the culture was examined under dissection microscope for the observations of ascocarps, basidiocarps, and conidia. Their morphology and septation were used for identification. The reproductive structures were transferred with a needle to a microscopic slide, add a drop of water to expose the spore and carefully squeezed under a cover glass. Photomicrographs were taken using Nikon microscope. (Nikon, Japan).

Identification

As the plating method yield mostly terrestrial (facultative to marine) fungi were identified referring the standard manuals. Raper and Thom (1949), Raper and Fennell (1965), Ainsworth *et al.* (1973a and b) and Ellis (1976). As the direct examination (after incubation) yield obligate marine fungi were identified referring the works of Kohl Meyer and Kohl Meyer (1979:1991).

Effect of different media on the growth rate of some isolated fungi

After identification dominant fungal species pure culture were prepared by single spore isolation technique (Raghukumar and Bhat, 1994). The isolated culture was maintained as stock culture in potato dextrose agar slants. They were grown at 30°C for 5 days and stored at 4°C for regular sub culturing.

The growing margin of the colony was cut with the help of a sterilized cork borer and the inoculums was transferred to Petri plates containing differed media, such as, SWPDA (Sea water potato dextrose agar), SWMEA (Sea water malt extract agar), SWRBA (Sea water Rose Bengal Agar), SWCMA (Sea water corn meal agar), SWYEA (Seawater Yeast extract agar), SWYPGA (Sea water yeast peptone glucose agar) and SWCDA (Sea water Czapek-Dox agar). The plates were incubated at room temperature (28 ± 2°C) for a period of five days. The radial growth of fungi (colony diameter) was measured using a centimeter scale.

Result

Investigation of filamentous higher fungi inhabiting on intertidal driftwood samples collected from five places of Muthupet mangrove environment were recorded. Species composition of filamentous fungi showed presence of 23 species belonging to 11 genera. Out of 23 species, 3 species were ascomycetes, 1 species was basidiomycetes and 19 were deuteromycetes (Table 1).

From the chief corner station 17 species belonged to 7 genera were recorded. Among the total isolates 6 genera/13 species belonged to Deuteromycetes, 1 species/1 genera was belong to basidiomycetes and 3 species/3 genera were ascomycetes.

Altogether 14 species/4 genera were recorded from the station Koriyar River. Among them 13 species/4 genera were assignable to deuteromycetes and 1 species/1 genera were belonged to ascomycetes and basidiomycetes was not recorded.

Totally 14 species/7 genera, 12 species/ 3 genera were recorded in both Saradi and Sethuguda respectively. Among them,11 species/7 genera belongs to deuteromycetes,1 species/1 genera were belonged to basidiomycetes,2 species/2 genera belongs to ascomycetes in saradi.In sethukuda 11 species/ 3 genera were belongs to deuteromycetes, Basidiomycetes was not recorded.

In Xavier munai station,14 species/8 genus were recorded, among them 12 species/ 6 genus were belonged to Deuteromycetes,1 species/1 genus were belonged to Basidiomycetes and 1 species/ 1 genera were belonged to ascomycetes.

The total number of 67.17% colonies were obtained from the samples collected from Muthupet.The total number of colonies were observed high in saradi (84.6) and low in Xavier munai (46.59).Mean percentage contribution of fungi isolated by plating technique showed variations between from 0.4 to 9.3 percent.(Table 2)

Growth of the different fungal species was measured by radial growth. The radial growth 0.5 cm of culture plug of the fungal species viz, *Aspergillus ochraceous, A. niger, A. luchuensis, A. flavus, A. terreus, Halocyphina villosa, Helicascus kanaloanus, Lignicola longirostris, Cystospora rhizophorae* and *A. sydowi* were studied for a period of 5 days at 24 hours intervals. All the fungal species showed fast growth in SWPDA medium (Table 3).

Discussion

Marine fungi are distributed from the oceans all over the world.Biogeographical distribution of marine fungi in different substratum was first attempted by Hughes (1974). A detailed analysis of distribution of marine fungi have been made by Hughes (1986) and Booth and Kenkel (1986).

Mangrove is a unique habitat where there are diverse niches for fungi.The driftwoods was one of the substratums for marine fungi which colonize the woods because of their genetics potentiality, a battery of enzyme producing abilities and adaptability to the changing physiochemical condition of the environment. In the present investigations totally 23 species belonging to 11 genera were encountered ascomycetes (3), basidiomycetes (1) deuteromycetes (19). A wide range of fungi belonged to 3 groups were recorded. Deuteromycotina were the most prevalent group of fungi. The abundance of this group of fungi on marine and mangrove substrates has been reported by Hyde and Jones 1988.

All the deuteromycetes isolated by plating method are reported from mangrove environment from many parts of Tamilnadu, covering Kanniyakumari (Upadhyay, 1978), Gulf of Mannar (Nadimuthu, 1998), Pichavaram (Venkatesan, 1984) and Madras Coast (Subramanian and Raghukumar, 1974). Dominant occurrence of deuteromycetes as facultative marine forms was already reported by these workers, coinciding with the present study.Dominent occurrence of ascomycetes in muthupet mangrove wood samples reported by Rani.2009. Ravikumar and Purushothaman 1988 a, 1988, b (vellar estuary), Ravikumar and Vittal, 1996(Pichavaram mangrove). Ravikumar and Vittal 1987, Ragukumar 1973(Tamil nadu coast) reported that investigation of marine fungi on different substratum in mangrove habitats coinciding with present study. Period of driftwood on the occurrence of marine fungi (Prasannarai and Sridhar, 1997), occurrence of higher fungi on marine animal substrate (Ananda *et al.*, 1998), intertidal occurrence of fungi along west coast (Prasannarai *et al.*, 1999). Common occurrence of *A. terreus* and *A. niger* was reported in the sites from Madras coast by Subramanian and Raghukumar, 1974).

Raghukumar and Bhat (1994) pointed out that no method is available for the study of entire mycoflora assemblage of any of the habitat / substratum as there are different fungal groups occupy as niche and therefore several methods has to be adopted to obtain the wholes tic picture of the fungal diversity.

Accordingly, the present investigation was carried out by (I) plating method and (ii) direct examination method. Both the method yielded distinct group of fungal population's viz., facultative marine fungi and obligate marine fungi, respectively.Ragukumar 1996, pointed out that a through understanding of various niches occupied by marine fungi should be given newer thrust in the areas of Indian mycological research. In this context, the present investigations were also new record on the distributional pattern of marine fungi.

Requirement of nutrient and other physical, chemical environment vary greatly among the marine fungi which in turn affects the distribution of the fungi. Therefore, Byrne and Jones (1975b) stressed the need for the analysis of fungi under laboratory so as to understand their distribution and successful colonization on different substratum. Among the different media studied, the fungus did not sporulate in any of the culture media. However, a good growth of 37 mm (dia) was observed on 7^{th} day on sea water corn meal agar medium. Poor growth rate was observed in SWCDA medium with 15 mm (dia) on 7^{th} day. Hyde *et al.* (1987) lasted a number of culture media so far been described as suitable for isolation and culture of different marine fungi. Further, it is stated that seawater potato dextrose agar as a best medium besides the other media described so far, as most of the fungi grew well in this medium as *Aspergillus ochraceous, A. niger, A. luchuensis, A. flavus, A. terreus, Halocyphina villosa, Helicascus kanaloanus, Lignicola longirostris, Rhizopus nigricans, A. sydowi* observed (Saravanan, 2002).

In the present study also, growth of the different fungal species on different fungal media was measured by radial growth. The radial growth 0.5 cm of culture plug of the fungal species viz, *Aspergillus ochraceous, A. niger, A. luchuensis, A. flavus, A. terreus, Halocyphina villosa, Helicascus kanaloanus, Lignicola longirostris, Cystospora rhizophorae A. sydowi* were studied for a period of 5 days at 24 hours intervals. All the fungal species showed fast growth in SWPDA medium.

Conclusion

Higher marine fungi due to their unique adaptation to marine habitats become an ideal group for biogeography study. Besides recording the species richness and diversity of the geographic location in substrate. These surveys strengthen the mycogeographic and mycodiversity studies. In addition this information helps to monitor the status of marine and marine influenced ecosystems

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Table 1. Occurrence of fungi on driftwood in different stations of Muthupet mangrove by plating technique

S.No.	Name of the species	Taxa	Chief corner	Koraiyar river	Saradi	Sethuguda	Xavier Munai
1.	*Aspergillus awamori*	Deuteromycetes	+	+	-	+	+
2.	*A. terreus*	Deuteromycetes	+	+	+	+	+
3.	*A. flavus*	Deuteromycetes	+	+	+	-	-
4.	*A. fumigatus*	Deuteromycetes	+	+	+	+	-
5.	*A. luchuensis*	Deuteromycetes	+	+	+	+	-
6.	*A. niger*	Deuteromycetes	+	+	+	+	+
7.	*A. nidulans*	Deuteromycetes	+	+	-	+	+
8.	*A. nigricans*	Deuteromycetes	-	-	+	+	-
9.	*A. harzianum*	Deuteromycetes	+	+	-	-	-
10.	*A. ochraceous*	Deuteromycetes	+	+	+	+	+
11.	*A. sydowi*	Deuteromycetes	+	+	+	+	+
12.	*A. sulphureus*	Deuteromycetes	+	+	-	+	-
13.	*Fusarium oxysporum*	Deuteromycetes	-	-	-	-	+
14.	*F. moniliforme*	Deuteromycetes	-	-	-	-	+
15.	*Curvularia lunata*	Deuteromycetes	+	-	-	-	+
16.	*Halocyphina villosa*	Basidiomycetes	+	-	+	-	+
17.	*Helicascus kanaloanus*	Ascomycetes	+	-	+	-	+
18.	*Ligniocola longirostris*	Ascomycetes	+	-	+	+	-
19.	*Cystospora rhizophorae*	Deuteromycetes	-	+	+	-	+
20.	*Penicillium* sp.	Deuteromycetes	-	-	+	+	+
21.	*Trichoderma viride*	Deuteromycetes	-	-	+	-	+
22.	*Neurospora crasse*	Ascomycetes	+	+	-	-	-
23.	*Alternaria* sp.	Deuteromycetes	+	-	-	-	-

Table 2. Percentage contribution of fungi on driftwood in different stations of muthupet mangrove

S.No.	Name of the species	Percentage contribution in different stations					Percentage contribution
		Chief corner	Koraiyar river	Saradi	Sethukuda	Xavier Munai	
1.	*Aspergillus awamori*	1.5	2.5	-	5.0	2.83	1.96
2.	*A. terreus*	3.8	2.33	10.5	3.0	2.9	4.50
3.	*A. flavus*	3.1	4	9.3	-	-	3.28
4.	*A. fumigatus*	3.1	3.6	3.8	3.0	-	2.7
5.	*A. luchuensis*	6.5	15.0	14.5	10.5	-	9.3
6.	*A. niger*	10.8	5.2	12.8	7.5	2.5	7.76
7.	*A. nidulans*	1.2	2.33	-	6.0	1.0	2.10
8.	*A. nigricans*	-	-	3.8	7.5	-	2.26
9.	*A. harzianum*	1	1	-	-	-	0.4
10.	*A. ochraceous*	8.6	9.0	2.0	6.5	12.5	6.12
11.	*A. sydowi*	4.3	5	3.0	6.0	2.16	4.09
12.	*A. sulphureus*	7	8	-	9	-	4.8
13.	*Fusarium oxysporum*	-	1.0	-	-	1.5	0.5
14.	*F. moniliforme*	-	-	-	-	1.0	1.0
15.	*Curvularia lunata*	2	-	-	-	3.5	1.1
16.	*Halocyphina villosa*	2.8	-	2.9	-	4	1.94
17.	*Helicascus kanaloanus*	4.2	-	7.0	-	5.7	3.38
18.	*Ligniocola longirostris*	5	-	6.0	4	-	3
19.	*Cystospora rhizophora*	-	4	4.0	-	2.0	1.6
20.	*Penicillium* sp.	-	-	3.0	2.9	4	1.98
21.	*Trichoderma viride*	-	-	2.0	-	1.0	0.6
22.	*Neurospora crasse*	2	1.9	-	-	-	0.78
23.	*Alternaria* sp.	2	-	-	-	-	0.4
	Total no. of colonies	68.9	64.86	84.6	70.9	46.59	67.17
	Total no. of species	17	14	14	12	14	

Table 3. Radial growth rate of fungi on different media

S.No.	Name of the organism	Growth rate (after 5 days) (cm)						
		SWMEA	SWPDA	SWCMA	SWYEA	SWRBA	SWCDA	SWYPGA
1.	*Aspergillus niger*	7.0	8.9	7.4	5.9	7.1	7.5	6.9
2.	*A. ochraceous*	4.7	7.4	6.7	6.1	7.0	7.0	6.1
3.	*A. flavus*	5.2	8.5	7.1	7.0	6.4	6.6	6.8
4.	*A. luchuensis*	6.2	7.1	6.0	5.8	6.7	6.7	5.9
5.	*A. terreus*	6.0	8.1	5.8	5.7	6.0	5.9	6.0
6.	*A. sydowi*	5.0	8.4	5.4	7.2	6.4	6.0	6.4
7.	*Halocyphina villosa*	5.9	6.7	6.6	4.8	6.0	6.4	5.8
8.	*Helicascus kanaloanus*	7.1	7.8	5.0	6.4	7.7	6.1	6.0
9.	*Lignincola longirostris*	7.2	8.2	6.9	6.1	7.9	6.2	5.2
10.	*Cystospora rhizophora*	6.7	8.3	7.1	7.5	7.8	6.0	7.1