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RESEARCH ARTICLE

POLYETHYLENE - A THREAT TO UNIQUE BIODIVERSITY OF PICHAVARAM MANGROVES

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ABSTRACT

Mangroves provide a unique ecological environment for diverse microbial communities. Hence the present study revealed to analyze the diversity of the bacterial and fungal communities of a mangrove in Pichavaram of south India. Six samples of throwed and partially buried in soils were collected from various sites in Pichavaram, Samples were serially diluted and using pour plate technique, colonies were enumerated. Colony morphology, Biochemical studies were used to identify the microbes. This work will help to know about the diversity and ecological function of the bacterial communities found in mangrove ecosystems, especially in biodegradation of polyethylene.

INTRODUCTION

Mangrove wetlands are prominent features of the coastal zone of tropical countries. A mangrove wetland consists of a mangrove forest and its associated water bodies. A mangrove forest harbors a group of plant species that grow well in the estuarine areas-where salinity undergoes constant changes due to freshwater flow and where the substratum is composed of accumulated deposits of river-borne sediment. According to the Forest Survey of India (1999), the total area of the Indian mangrove wetland is about 4,87,100 ha of which 56.7% (2,75,800 ha) is on the east coast, 23.5% (1,14,700 ha) on the west coast and the remaining 19.8% (96,600 ha) on the Andaman and Nicobar islands. Mangrove ecosystem has long been a natural resource of importance to mankind by virtue of its utility and aesthetic value. This eco system is one of the most important productive ecosystems. The primary productivity from mangroves is enormous and various organisms such as wood borers, fungi and bacteria are involved in recycling the detritus. Such nutrient recycling helps in maintaining ecological balance in the estuarine environment. The dynamic mangrove ecosystem supports numerous diversified soil micro and macro organisms. Mangrove species diversity is well known for animals and plants, but poorly known for other organisms such as Bacteria and fungi. Mangroves provide a unique ecological environment for diverse microbial communities. Microbes from the extreme environments have attracted considerable attention in recent years. Mangrove ecosystems are rich in bacterial flora. Fertility of the mangrove waters results from the microbial decomposition of organic matter and recycling of nutrients. Most of bacteria belong to gram-negative. Gram-positive bacteria are less than 10% of the total bacterial population and higher percentage in sediments. With the exception of a few recent studies, little is known about the diversity and ecological function of the bacterial communities found in mangrove ecosystems (Hewson and Fuhrman, 2004; Zhang *et al*., 2008; Zhou *et al*., 2009; Gomes *et al*., 2010; Santos *et al*., 2011; Silveira *et al*., 2011). Hence the present study revealed to analyze the diversity of the bacterial and fungal communities of a mangrove in Pichavaram of South India.

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MATERIALS AND METHODS

Study Area

All the samples were collected from of various sites Pichavaram mangroves and the associated wetlands .It lies between Latitude 11°25′N; and longitude 74°47′E it situated at the mouth of the Vellar, Coleroon and Uppanar rivers in Cuddalore District, Tamil Nadu, India. This area is extensive covering of mangrove, an area of about 1100 ha. About 50% is covered by forest, 40% by water-ways and the remaining filled by sand-flats and mud-flats (Krishnamurthyand Prince Jayaseelan, 1983).

Sample collection

Six samples of throwed and partially buried polyethylene sheets with soil were collected from various sites in Pitchavaram, were collected in the clean polyethylene bags with the help of sterile spatula. Immediately transported to the laboratory and processed. Sediments of this place are acidic in nature. Clay soil was found and it is black in colour. Sediment becomes loose because of the presence of the decaying organic matter.

Enumeration of microorganisms

The soil samples collected from various places were serially diluted up to 10^{-6} dilution to determine the population of fungi and bacteria. One ml of 10^{-4}, 10^{-6} were used for enumerating fungi, and bacteria respectively. The appropriate dilutions were aseptically transferred to petriplates containing and Potato Dextrose Agar and Nutrient Agar respectively and mixed well Triplicates were maintained in each case. The plates were incubated for 24hrs for bacteria, for 3-4 days for fungi at 28 ± 2°C. After incubation the numbers of fungal and bacterial colonies in the respective plates were counted and the population was expressed in terms of cfu g^{-1} soil.

Identification of Total Heterotrophic Bacteria (thb) and fungi

The samples were diluted and plated in Nutrient agar medium and Potato Dextrose Agar. Triplicate plates from each dilution were incubated at 37^0C and 28^0Cfor 24 hrs, 3-4 days respectively. After incubation the colonies were counted by colony forming unit (CFU) and subculture by colony morphology. The different morphological and biochemical characterization of the isolates were investigated according to the Bergey's Manual of determinative bacteriology.

Purification and Maintenance

The isolated bacteria were purified by streak plate technique. Once in a month the fungal were purified by single hyphal tip method maintained on Nutrient agar medium and Potato Dextrose Agar at 4°C and subculture once in a month. The isolates were subjected to various biochemical tests and identified according to "Bergey's Manual of Determinative Bacteriology" (Volume 4) for bacteria were characterized. Fungi were identified by using "A Manual of soil fungi-(Gilman, 1975).

RESULTS AND DISCUSSION

The mangrove forest soil samples of Pichavaram was collected and analyzed for their microbial diversity. Total bacterial, fungal populations of these soils were enumerated. The abundance of Bacteria and fungi were shown in Table 1. Six locations showed a marginal variation of the Bacterial count. In rhizosphere region the presence of Bacterial population are high. Among these three regions PMC shows highest count of 35.66 x 10^{-6}. Other 2 region shows the count of 36 x 10-^{6} in PMB and 33 x 10^{-6} in PMA respectively. The mangrove sediment harbors larger bacterial population than the water column (Ravikumar, 1995). This is attributed to nutrient accumulation, precipitation of inorganic compounds and settlement of dead organic matter in the sediments.

Table 1. Microbial population found in partialy buried polyethylene sheets of mangroves of pichavaram

S.No	Places	Heterotrophic Bacteria	Fungi
1.	Rhizosphere -I (PMA)	33 x 10-6	13.66 x 10-3
2.	Rhizosphere –II(PMB)	36x 10-6	14.33 x 10-3
3.	Rhizosphere –III(PMC)	35.66x 10-6	15.33 x 10-3
4.	Non – rhizosphere I(PMD)	25x 10-6	7.33 x 10-3
5.	Non – rhizosphere II(PME)	26x 10-6	7.33 x 10-3
6.	Bank of the mangrove(PMF)	23.33 x 10-6	3.33 x 10-3

Non-rhizosphere region have the less counts of 25 x10^{-6}(PMD) and 26x 10^{-6} (PME). All the study sites PMF exhibit very low count of 23.33 x 10^{-6}. The rhizosphere soil sample contained higher microbial populations compared to non- rhizosphere soil (Lakshmipriya and Sivakumar 2012). Mangrove forests are biodiversity hotspots" for marine fungi (Shearer *et al*., 2007). These fungi play an important role in the nutritive cycle and support the mangrove ecosystem. The fungal load was found to be considerably lower when compared to THB load (Shristi Kumar *et al*., 2007). In our study fungal population ranges from 3.33 to 15.33 x 10^{-3} CFU/g. Higher counts found in rhizosphere region of PMC. Subsequent higher values are14.33 x $10^{-3,}$ 13.66x 10^{-3} also found in other rhizosphere regions of PMB, PMA respectively. The lower counts of fungi in mangrove ecosystem than in fresh water also buttressed (Okpowasili *et al*., 2012) He also revealed that the fungal count also high in Rhizosphere region than other areas. Fungal populations of Non- rhizosphere region are 7.33 x 10^{-3} in both areas of PMD and PME.

Table 2. Diversity of soil heterotrophic bacteria found in partialy buried polyethylene sheets from mangroves of pichavaram

BACTERIA	PMA	PMB	PMC	PMD	PME	PMF
Bacillus sp.	+	+	+	-	+	-
Escherichia coli	+	+	+	+	+	+
Micrococcus sp.	+	+	+	-	+	-
Pseudomonas sp.	+	+	+	+	-	+
Staphylococcus sp.	+	+	+	-	+	-
Vibrio sp.	-	+	-	-	+	+

Table 3. Diversity of soil fungi found in partialy buried polyethylene sheets from mangroves of pichavaram

FUNGI	PMA	PMB	PMC	PMD	PME	PMF
Aspergillus sp.	+	+	+	+	+	+
Pencillium sp.	-	+	+	+	-	-

Heterotrophic bacteria are very important in mangrove habitats as the bacteria decompose the mangrove litter, recycle the nutrients and produce the detritus food for many fishes (Odum and Heald, 1972; Agate *et al*., 1988). Diverse and dense bacterial community inhabiting the mangrove sediment. (Armando C. F. Dias *et al*., 2009).The study resulted in six bacterial isolates It indicated *Bacillus, E.coli, Micrococcus, Pseudomonas, Staphylococcus, Vibrio* were abundant in the mangrove samples. Similar reports existed in Pichavaram

mangrove, southeast India common Bacterial genera are *Vibrio, Bacillus, Micrococcus, Pseudomonas, Aeromonas, Flavobacterium* etc. (Sathiyamurthy *et al*., 1990). Characterization of bacterial isolates from Suva mangrove soil revealed Bacillus as the dominant genera .Other genera such as *Micrococcus, Listeria, Vibrio* were also presented (Kumar *et al*., 2007). *Aspergillus* and *Pencillium* sp. were observed in our study. The predominance of *Aspergillus* and *Pencillium* in Pichavaram mangrove of south India has been reported. (Venkatesan and Natrajan, 1986) 16 species of fungi belonging to various groups especially the Ascomycetes were dominated (Okpokwasili *et al.*, 2012). The degradation of mangrove vegetative material produces detritus, which can be defined as organic matter in the active process of decomposition. It rich in energy and contains a large microbial population both attached and living free (Odum and Heald 1975a and Bano *et al.*, 1997).

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