Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Annals of Biological Research, 2011, 2 (1) :76-82
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0976-1233
CODEN (USA): ABRNBW

Biodiversity of actinomycetes in Manakkudi mangrove ecosystem, Southwest coast of India

Ravikumar. S[1], Fredimoses. M[2*] and Gokulakrishnan. R[1]

[1]*School of Marine Sciences, Department of Oceanography and Coastal Area Studies, Alagappa University, Tamilnadu, India*
[2]*Department of Biotechnology, K. S. Rangasamy College of Arts & Science, Tiruchengode*

ABSTRACT

The diversity of actinomycetes in the Manakkudi mangrove ecosystem were analysed by the present study reveals that, the diversity of actinomycetes are found maximum in the rhizosphere soil than the non-rhizosphere soil that too mangrove associate of Achrostichum aureum *harbours maximum counts than true mangrove plants. The diversity of actinomycetes is found maximum between the soil depth of 10-20cm are not correlated with the maximum level of nutrients between the soil depth of 0-10cm. The presence of actinomycetes in the Manakkudi mangrove ecosystem could pave the way for the establishment of disease free mangrove seedlings in the nursery and in the field. Efforts have been undertaken to find out the bioactive potential of mangrove sediment actinomycetes for the treatment dreadful human diseases.*

Key words: Actinomycetes, Diversity, Enumeration, Mangrove ecosystem.

INTRODUCTION

Microbes from the extreme environments have attracted considerable attention in recent years. Almost 80% of the world's antibiotics are known to come from actinomycetes, mostly from the genera Streptomyces and Micromonospora [1]. Actinomycetes play a major role in recycling of organic matter [2], production of novel pharmaceuticals, nutritional materials, cosmetics, enzymes, antitumour agents, enzyme inhibitors, immune-modifiers and vitamins. Streptomyces are especially prolific and can produce a great diversity of antibiotics (around 80% of the total antibiotic production) and active secondary metabolites [3, 4].

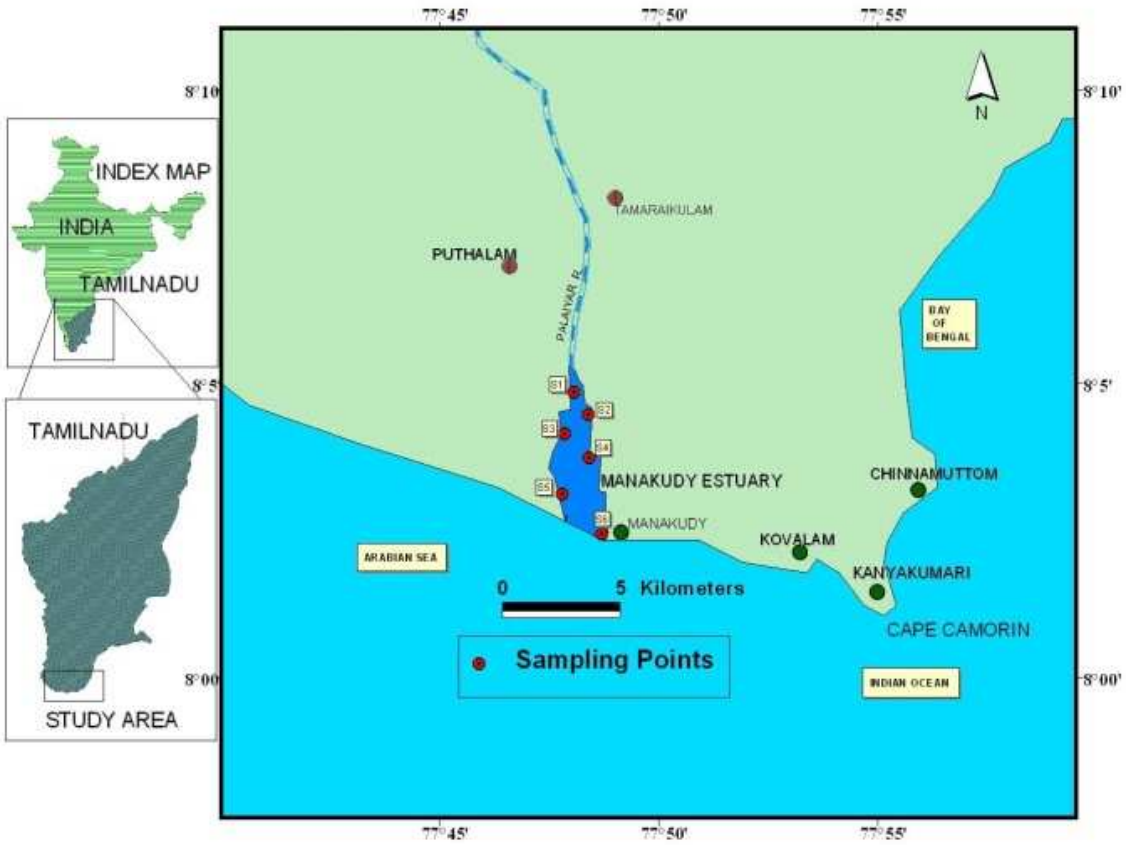
The authors [5] reported a bimodal distribution of actinomycetes in near shore tropical marine environments. The existence of an autochthonous actinomycete from marine deep oceanic sediments were reported by several authors [5-7]. Although, recent studies [8] have focused on moderately or extremely halophilic microorganisms but very little information is available concerning the taxonomic distribution of actinomycetes from mangrove environment. Therefore, the present study was designed to evaluate the distribution of actinomycetes in mangrove sediment.

MATERIALS AND METHODS

Collection site description

The present study area is situated in the South West coast of Tamil Nadu (Fig.1). The Manakkudi estuary extends over a distance of 5.0 KM in length and the coastline of Kanyakumari is nearly 68 KM in length. The perennial rivers empty along the coastline. The river Pazhayar is one of the most important surface water resources of Kanyakumari district. At its confluence with the Arabian Sea near Manakkudi village it forms the Manakkudi estuary. In the upper reaches of the estuary there are small islands. One of the islands has artificially regenerated mangroves. Besides the mangrove plants viz; Rhizophora mucronata (Family: Rhizophoracea) and Avicennia officinalis (Family:Avicenniaceae) some associated plant species viz; Acrostichum aureum (Family: Pterdaceae; Order: Pteridales), Panicum repens (Family: Poaceae; Order: Poales) also found beyond the mangrove environment.

Fig.1. Map Showing the Collection Sites



Collection and processing of samples

Rhizosphere soil samples from 2 mangrove viz., Rhizophora mucronata and Avicennia officinalis and 3 mangrove associated plants viz., Acrostichum aureum, Panicum repens and Hymenachene acutigluma were collected using peat corer along the roots of mangrove up to a depth of 1M from August 2005 to July 2006. The soil and water samples away from the mangrove vegetation representing the non-rhizosphere region were also collected from five different sites (S I to S V) at different depths along the Manakkudi estuary.

Station-I (Lat.8^{o}05'N; Long.77 ^{o} 46`E) represents where the river water entered into the mangrove environment with dense vegetation of Acrostichum aureum. Station-II (Lat 8 $^{o}$ 04`N; Long.77^{o} 46`E) represents the middle portion in between the mangrove and freshwater entry point. Station III (Lat.8$^{0}$ 03`N; Long.77^{0} 47`E) represents the true mangrove area vegetated with mangrove and associated plant species. Station IV (Lat 8$^{0}$03`N; Long.77^{0} 48`E) represents the area in which coir-retting process has been done. Station V (Lat.8$^{0}$ 02`N; Long.77^{0} 48`E) represents the area in which the mouth of the sea where mangrove water and seawater mix together. Collected samples were kept in sterile plastic containers and brought to the laboratory in an iced chests and immediately subjected for the microbiological analysis [9].

Chemical analysis

Samples were analyzed in triplicates for pH, electrical conductivity, inorganic phosphate, nitrate, nitrite, particulate organic carbon [10], chloride, sulphate, potassium, calcium and magnesium [11]. Atmospheric temperature was measured using a mercury centigrade thermometer with 0.5^{0}C accuracy. Rainfall data was obtained from Meteorological Department, Nagercoil. Statistical analysis was carried out to find out the level of significance.

Bacteriological analysis

The sediment samples were processed as described by [12]. Serial dilution were made by aseptically removing 1g of wet sediment and adding it to 5ml of sterilized filtered sea water (dilution 2x 10 $^{-1}$), mixing and further diluting (1:10) with sterilized filtered sea water (dilutions 2x 10 $^{-4}$). The dilution procedure was repeated in triplicate for each sample. One milliliter of each sample was spread over the surface of Starch casein agar plates (Starch-10g. l^{-1}; Casein-1g. l^{-1}, Agar-1.8g.l^{-1}) were prepared by using 50% of sea water and distilled water. The medium was supplemented with 20 µg.ml^{-1} Nalidixic acid, 25 µg.ml^{-1} Nystatin and 100 µg.ml^{-1} cycloheximide to minimize the gram negative bacteria, fungal and yeast contaminations respectively [6]. The incubation was carried out at 28^{o}C for 7-10days.

RESULTS AND DISCUSSION

Microorganisms in the mangrove environment has much role in the growth and development of mangrove plants through nutrient regeneration by biological processes either decomposition or by fixing the nitrogen and phosphorous. Manakkudi mangrove is one of the artificially regenerated mangroves in South west coast of India, which is of 8 years old. Assessment of microbial diversity in the Manakkudi mangroves ecosystem are not attempted so far. Hence, the present study has been undertaken to find out the diversity and function of actinomycetes. Rhizosphere soil and non-rhizosphere soil samples from different depths at various stations were collected monthly for a period of one year and all the samples were subjected for the analysis of both the actinomycetes diversity and nutrient concentrations.

The actinomycetes counts were found maximum in the rhizosphere soil sample of Avicennia officinalis and Acrostichum aureum during the month of April. The counts were found maximum during the month of May in Rhizophora mucronata, October in Hymenachene acutigluma, August in Panicum repens (Table 1).

Table 1. Monthly variation in the counts of Actinomycetes in the rhizophere soil sample

Plant species	Counts of Actinomycetes (CFUx 10^3 g^{-1})											
	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July
Avicennia officinalis	60.8	78.9	49.8	6.1	36.5	3.4	24.5	40.6	80.5	63.2	4.1	25.1
Panicum repens	85.1	45.6	49.2	5.6	7.8	4.8	3.2	70.3	39.5	42.3	4.7	4.5
Rhizophora mucronata	25.2	15.8	98.5	19.1	5.2	15.6	16.7	19	10.9	106.7	19.1	0.0
Hymenachene acutigluma	95.2	35.9	168.9	162.1	146.2	138.9	125.8	24.5	54.3	179.5	4.7	166.3
Acrostichum aureum	98.5	498.6	315.6	16.8	89.4	45.6	46.8	103.2	506.8	372.7	13.8	99.3

Values are found significant between months (P>0.01) and found insignificant between plant species

The statistical analysis reveals that, the counts were found significant among months (P>0.01) and found insignificant between the plant species. Actinomycetes counts at different depths from selected five stations reveals that, the counts were found maximum in the month of December between the soil depths of 0-10cm (330.6 CFU X $10^3 g^{-1}$) in Station II when compared to the other stations (Table 2). However, the counts of actinomycetes in all the six stations seem to have more in number between the soil depths of 10-20cm. In general, the counts of actinomycetes were increasing with the increasing depths up to 10-20cm and further decreasing trend was noticed between the soil depths of 20-30cm. The statistical analysis reveals that, the counts were found significant (P>0.05) between the stations and soil depths.

Table 2. Station wise variations of actinomycetes in the sediments samples of different depths at different months

Soil depths	Counts of actinomycetes (CFU x $10^3 g^{-1}$)											
	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	June	July
0-10 cm	59.3	41.8	33.8	12.4	66.7	72.8	91.8	79.3	35.03	323.8	14.0	61.8
10-20 cm	49.3	98.1	31.9	29.4	112.3	49.7	62.8	81.2	70.5	12.7	5.7	45.1
20-30 cm	29.1	47.1	72.9	121.3	142.7	134.9	147.1	14.5	132.8	39.7	9.9	17.5
Stations II												
0-10 cm	91.3	142	130	140.1	330.6	298.5	198.5	79.0	328.6	191.1	25.3	87.0
10-20 cm	62.8	33.5	49.5	62.9	34.1	39.5	67.8	14.0	132.1	56.1	4.7	44.9
20-30 cm	71.8	113	198	135.8	145.1	298.3	110.1	43.4	163.1	6.8	8.5	78.1
Stations III												
0-10 cm	33.1	22.8	45.8	31.9	41.8	52.1	62.4	31.9	81.1	59.4	10.6	44.5

10-20 cm	121.8	141	138	78.1	44.9	33.3	97.5	29.5	50.8	96.4	24.6	115
20-30 cm	92.1	44.8	66.4	88.3	141.5	128.5	192.5	44.2	29.4	92.3	7.0	41.8
Stations IV												
0-10 cm	62.9	44.5	92.5	112.5	133.5	142.8	155.4	68.1	95.7	161.9	8.8	71.5
10-20 cm	115.2	15.8	69.7	55.5	43.1	72.1	68.5	20.2	41.2	5.0	12.2	118
20-30 cm	29.5	30.1	29.5	27.7	28.3	30.3	48.1	18.6	21.0	246.7	5.3	10.1
Stations V												
0-10 cm	21.8	41.8	52.3	92.8	111.8	121.8	141.8	10.6	22.5	173.0	14.3	15.9
10-20 cm	52.9	62.8	71.9	118	220.1	189.1	45.8	13.3	47.8	39.1	2.8	41.9
20-30 cm	54.5	12.1	88.5	112.6	34.8	14..8	62.5	33.6	93.6	14.8	9.1	49.8

Values are found significant between the stations and between the soil depths (P>0.05)

The results clearly reveals that, the counts of actinomycetes were found maximum in the rhizosphere soil. The maximum counts reported in the plant species might be due to the exhaustive root system of mangrove associates which provides more space for the colonisation of actinomycetes and also depends upon the availability of maximum level of aminoacids, sugars and minimum concentration of tannins in the rhizosphere soil.

The physico and chemical parameters clearly indicates that, important nutrients such as nitrogen and potassium were found maximum between the soil depths of 0-10cm (Table 3 & 4). The distribution and abundance of actinomycetes were found maximum between the depths of 10-20cm. The authors [13] reported that, the counts of actinomycetes were found maximum in the upper sediment layers (0-20cm). In a few instances, the number of actinomycetes population showed bimodal maxima in 0-20cm and 60-100cm soil depth which can be accounted for by the taxon-specific distributions of the suprageneric groups and hence the physico chemical properties are not influenced the diversity of mangrove actinomycetes [7, 12].

Table 3. Annual mean value of physico-chemical parameters in Manakkudi mangrove rhizophere soil samples

Plant species	Temp (C^o)	Rain fall (mm)	pH	EC	C (%)	N(µg)	K(µg)	P(µg)
Avicennia officinalis	24.5	87.08	3.7	4.8	2.8	32.86	24.18	1.36
Acrostichum aureum	24.5	87.08	4.2	4.6	3.7	28.95	21.35	1.48
Rhizophora mucronata	24.5	87.08	4.8	4.9	2.9	34.74	20.67	1.88
Panicum repens	24.5	87.08	4.7	4.7	2.6	31.89	22.97	1.67
Hymenachene acutigluma	24.5	87.08	4.5	4.8	3.8	33.52	23.59	2.46

The present results also shows that, actinomycetes are evenly distributed according to the microenvironments governed by intermediates factors. The present investigation proved that, the Manakkudi mangrove ecosystem in the South west coast of India is eminently a suitable ecosystem for the diversity of actinomycetes which could thus enhance the multiplication of disease free mangrove seedlings.

Table 4. Annual mean value of physico-chemical parameters in the Non- rhizophere soil samples collected from Manakkudi mangrove

Sampling stations / Depths	Temp(C°)	Rain fall (mm)	pH	EC	C (%)	N(µg.g^{-1})	K(µg.g^{-1})	P(µg.g^{-1})
Station I 0-10cm	24.5	87.08	6.4	3.5	1.5	252.08	38.54	0.13
10-20cm	23.5	87.08	6.7	3.2	1.3	169.21	36.25	0.12
20- 30cm	21.1	87.08	6.9	3.1	1.2	138.56	34.67	0.09
Station II 0-10cm	23.8	87.08	6.8	4.2	1.9	269.21	36.67	1.52
10-20cm	23	87.08	6.4	3.9	2.1	173.65	32.97	1.98
0- 30cm	20.7	87.08	6.7	3.4	1.7	144.25	33.59	2.28
Station III 10cm	24	87.08	6.9	3.8	1.3	270.36	41.32	0.08
10-20cm	23	87.08	6.8	4.1	1.4	245.26	38.25	0.16
20- 30cm	21	87.08	6.4	3.6	1.6	400.32	37.26	0.25
Station IV 0-10cm	24.5	87.08	6.5	3.9	1.7	275.21	62.54	0.13
10-20cm	23.6	87.08	6.4	4.2	1.4	183.65	53.49	0.11
20- 30cm	22	87.08	6.8	3.6	1.2	164.25	49.26	0.19
Station V 0-10cm	24.5	87.08	7.2	3.8	1.6	125.63	65.32	0.21
10-20cm	23	87.08	6.9	3.5	1.4	145.24	35.24	0.19
20- 30cm	22	87.08	6.8	3.6	1.3	132.65	27.85	0.23

Values are found significant between months (P>0.01) and insignificant between depths

Acknowledgement

The authors are greatfully thanks to the authorities of Alagappa University, Karaikudi and K.S.Rangasamy college of Arts & Science College, Tiruchengode for providing facilities and Ministry of Environment and forests for financial assistance. .

REFERENCES

[1] Pandey B, Ghimire P, Agarwal VP Studies on the antibacterial activity of actinomycetes isolated from the Khumbu region of Mt. Everst. A paper presented in the International Conference on the Great Himalayas: Climate, Health, Ecology, Management and Conservation, Kathmandu. January 12-15. **2004**, Organized by Kathmandu University and the Aquatic Ecosystem Health and Management society, Canada.

[2] M.C. Srinivasan, R.S. Laxman, M.V. Deshpande, *World J. Microbiol Biotechnol*., **1991,** 7, 171-184.

[3] Y. Okami, K. Hotta Search and discovery of new antibiotics. In: Good Fellow, M., S. T. Williams and M. Mordarski (eds), Actinomycetes in Biotechnology. Acadamic press Inc .San Diego. **1988,** pp, 33-67.

[4] J. Berdy, The discovery of new bioactive microbial metabolites: screening and identification. In: Bushell ME, and Graefe U. (eds), Bioactive Microbial Metabolites. Elsevier Amsterdam, **1989**, 27, 3–25.

[5] M. Takizawa, R.R. Colwell, R.T. Hill, *Appl Environ Microbiol*, **1993**, 59, 997–1002.

[6] J. Ravel, H. Schrempf, R.T. Hill, *Appl Environ Microbiol*, **1998,** 64, 3383-3388.

[7] H. Weyland, E. Helmke, *Int J Syst Bacteriol*, **1988,** 35, 127-138.
[8] A. Ventosa, Taxonomy and phylogeny of moderately Halophilic bacteria. In Bacterial Diversity and Systematics, ed. by Priest FG, Plenum Press, New York, **1994**, pp, 231–242.
[9] R. A. Holt, J.F. Conrad, J.L. Fryer, *Prog Fish Culturist*, **1975,** 37, 137-139.
[10] J.D.H. Strickland, T.R. Parsons, A Practical Handbook of Seawater Analysis, ed. by J.C. Stevenson, L.W. Billingsley, R. H. Wigmore, Fish Res Bd, Ottawa, **1968**, pp, 167.
[11] *Anon*, Image analysis. Principles and Practice. A Technical Handbook produced by Joyce-Loebl, A Vickers Company. Short Run Press, Exeter, **1985**, pp, 250.
[12] P.R. Jensen, R. Dwight, W. Fenical, *Appl Environ Microbiol*, **1991**, 57, 1102–1108.
[13] B. Ghanem Nevine, A. Sabry soraya, M. El-sherif zeinab, A. El-ela geha., *J Gen Appl Microbiol.*, **2000**, 46, 105-111.