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Assessment of Microbial Diversity In Relation To Biochemical Constituents along Palk Strait

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Abstract: The present study was made an attempt to enumerate the total heterotrophic bacteria (THB), *E. coli*, actinomycetes and fungi population and to evaluate the biochemical parameters from water and sediment samples. The samples were collected from eight different stations *viz.*, Kodiyakkarai, Mallipattinam, Manora, Manamelkudi, Kottaipattinam, Mimisal, S. P. Pattinam and Thondi along Palk Strait coast of South India. The results suggested that, the maximum THB $(12x10^5 \text{ CFU.mL}^{-1})$ and *E. coli* $(14x10^5 \text{ CFU.mL}^{-1})$ counts were recorded in water sample at Mallipattinam coast during the month of January and February. The maximum $(5x10^5 \text{ CFU.g}^{-1})$ counts of actinomycetes were recorded in sediment at Thondi during the month of March. Moreover, the courts of fungi were found maximum $(12 x10^5 \text{ CFU.g}^{-1})$ in sediment at Mimisal during the month of March. The correlation analysis revealed that, the counts of THB showed positive correlation (p<0.05) with amino acids in both water and sediment samples and showed negative correlation (p<- 0.05) with carbohydrates and proteins. The courts of *E. coli* showed positive correlation (p<0.05) with amino acids in sediment. The fungal counts showed positive correlation (p<0.05) with protein and reducing sugar in sediment.

Keywords: Actinomycetes, E. coli, Fungi, Microbial diversity, Palk Strait coast, Total heterotrophic bacteria.

I. INTRODUCTION

The microbial community constitutes 90% of oceanic biomass, which includes bacteria, archaea, protists and fungi etc. They are responsible for 98% of primary production in the marine ecosystem [2]. They have known to produce potential commercially important bioactive secondary metabolites. Marine microorganisms can be survived in extreme pressure, salinity, temperature and absence of light. The microbial diversity in the marine ecosystem is an important to understand about the microbial biomass, biogeochemical cycling and distribution pattern. The microbial diversity monitoring in marine water and sediment plays a vital role to predict anthropogenic interventions and helps to assess sanitary condition in the coastal environment [15]. But so far it is remains unknown to understand their community distribution patterns in most of the marine ecosystem for future perspectives [24], [3]. The change in the environmental parameters may leads to the microbial diversity fluctuations in the marine ecosystem. In order to assess the microbial diversity in coastal environment, it is essential to determine the relationship between the biochemical parameters and the microbial diversity in the marine environment [5]. Several authors assessed only the microbial diversity in Sagami Bay [16], Pichavaram [21] and Palk Strait [23] etc. However studies on microbial diversity in relation to biochemical parameters in Palk Strait are poorly understood. In this connection, the present study has



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Vol. 2, Issue 11, November 2013

initiated to investigate the microbial diversity in relation to biochemical constituents along the Palk Strait coast of South India.

II. MATERIALS AND METHODS

A. Sample Collection

The water and sediment samples were collected between the months of January 2012-March 2012 from eight different stations *viz*. Kodiyakkarai (Lat. 10° 16' N; Long. 79° 49' E), Mallipattinam (Lat. 10° 16' N; Long. 79° 15' E), Manora (Lat. 10° 15' N; Long. 79° 18' E), Manamelkudi (Lat. 10° 02' N; Long. 79° 15' E), Kottaipattinam (Lat. 09° 59' N; Long. 79° 13' E), Mimisal (Lat. 09° 54' N; Long. 79° 08' E), S.P. Pattinam (Lat. 09° 51' N; Long. 79° 05' E) and Thondi (09° 44' N; Long. 79° 01' E) (Fig. 1). The collected samples were carefully transported to the laboratory in an ice cold condition for further analysis.

Fig.1 Map showing the collection site along the Palk Strait region



B. Microbiological Analysis

The collected samples were serially diluted and 1ml of each dilution was added in sterile petriplate separately. After that, sterile molten media was poured on each triplicate plate containing the sample for the isolation of THB (Zobell Marine Agar-2216e), *E. coli* (Tergitol-7), actinomycetes (Starch Casein agar) and fungi (Rose Bengal agar) and kept for incubation. Colonies appeared on the solid media will be counted.

C. Biochemical Analysis

The biochemical parameters such as total sugar [7], reducing sugar [13], protein [11] and amino acid [14] were estimated by following standard protocols.

D. Statistical Analysis

The correlation analysis was performed by using SPSS (ver. 16.0) software.

III. **RESULTS AND DISCUSSION**

In water, the counts of THB were recorded maximum $(12.5 \times 10^5 \text{ CFU.mL}^{-1})$ during the month of January at Mallipattinam and minimum $(1.0 \times 10^5 \text{ CFU.mL}^{-1})$ was recorded during the month of January at Manora (Fig. 2). The counts of *E. coli* were recorded maximum $(14 \times 10^5 \text{ CFU.mL}^{-1})$ during the month of February at Mallipattinam and minimum $(1.0 \times 10^5 \text{ CFU.mL}^{-1})$ was recorded during the month of January at Mallipattinam and minimum $(1.0 \times 10^5 \text{ CFU.mL}^{-1})$ was recorded during the month of January at Mallipattinam and minimum $(1.0 \times 10^5 \text{ CFU.mL}^{-1})$ was recorded during the month of January at Minisal (Fig. 3). However, none of the collection site showed the counts of actinomycetes and fungi. In sediment, the maximum $(7.8 \times 10^5 \text{ CFU.g}^{-1})$ counts of

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Vol. 2, Issue 11, November 2013

THB were recorded during the month of January at Kodiyakarai and minimum $(1.3x10^5 \text{ CFU.g}^{-1})$ was recorded during the month of February at Mimisal (Fig. 4). The counts of *E. coli* were recorded maximum $(6.0x10^5 \text{ CFU.g}^{-1})$ during the month of February at Mallipattinam and minimum $(1.0x10^5 \text{ CFU.g}^{-1})$ counts were recorded during the month of January at Mimisal and Manamelkudi (Fig. 5). The counts of actinomycetes suggested that, the maximum $(5 x10^5 \text{ CFU.g}^{-1})$ counts were recorded during the month of March at Thondi and minimum $(1.0x10^5 \text{ CFU.g}^{-1})$ was recorded in January at Kodiyakkarai (Fig. 6). The counts of fungi were recorded maximum $(12 x10^5 \text{ CFU.g}^{-1})$ during the month of March at Mimisal and minimum $(1.0x10^5 \text{ CFU.g}^{-1})$ was recorded during the month of March at

In water sample, the total sugar was found maximum (0.38 mg.g^{-1}) during the month of February at Manora and minimum (0.12 mg.g^{-1}) was recorded during the month of March at Kodiyakarai. The protein was found maximum (3.73 mg.g^{-1}) during the month of January at Kottaipattinam and minimum (1.33 mg.g^{-1}) was found at Kodiyakarai during the month of March. The amino acid was found maximum (1.68 mg.g^{-1}) during the month of January at Kottaipattinam and minimum (1.68 mg.g^{-1}) during the month of January at Kodiyakarai and found minimum (1.0 mg.g^{-1}) during the month of February at S. P. Pattinam. The reducing sugar was found maximum $(0.136 \text{ mg.g}^{-1})$ during the month of January at Kodiyakarai and the minimum was recorded by 0.106 mg.g^{-1} during the month of March at Kodiyakarai. The level of non reducing sugar was found maximum (0.27 mg.g^{-1}) at Manora during the month of February and found minimum $(0.014 \text{ mg.g}^{-1})$ in March at Kodiyakarai coastal region.

In sediment, the total sugar was recorded maximum (5.44 mg.g⁻¹) at Minisal during the month February and minimum (1.70 mg.g⁻¹) was recorded during the month of March-2012 at Manamelkudi. The protein was recorded maximum (123.33 mg.g⁻¹) at Kottaipattinam during the month of January and the minimum (38.66 mg.g⁻¹) was recorded during the month of February at Kodiyakarai. The amino acid was recorded maximum (41.27 mg.g⁻¹) during the month of February at Mallipattinam and minimum (14.12 mg.g⁻¹) was recorded during the month of February at Thondi. The reducing sugar was found maximum (1.58 mg.g⁻¹) during the month of March at Manora and minimum (0.72 mg.g⁻¹) was recorded during the month of February at Manamelkudi. The non reducing sugar was found maximum (4.25 mg.g⁻¹) during the month of February at Minisal and found minimum (0.82 mg.g⁻¹) during the month of March at Thondi (Table 1).

The assessment of microbial diversity poses a considerable importance and interesting task. Besides that, it helps to identify the novel secondary metabolites derived from the microbes. Generally, the water and sediments in the marine environment plays a significant role to assess microbial diversity and sanitary condition of the marine environment. In order to understand the feacal pollution, the enumeration of E. coli was highly recommended as a feacal indicator. E. coli has already been used as a feacal indicator in many countries [1]. In view of this, the present study has initiated to enumerate the microbial population including E. coli and to find out the relationship upon the biochemical constituents along the Palk Strait coast of South India. The counts of THB were recorded maximum (12.5x10⁵ CFU.mL⁻ ¹) in water at Mallipattinam and the sediments showed the maximum counts by 7.8x10⁵ CFU.g⁻¹ in Kodiyakarai. Similarly, [10] and [12] reported that, the maximum counts of THB were recorded in Porto Novo and Cuddalore coasts. The counts of E. coli were recorded maximum (14x10⁵ CFU.mL⁻¹) in water and the sediment showed the maximum counts by $6x10^{5}$ CFU.g⁻¹ at Mallipattinam. Similarly, [20] reported that, the maximum (5.9x10⁴ CFU.mL⁻¹) counts of E. coli was recorded in water and showed the counts by 4.7x10⁴ CFU.g⁻¹ in sediments at Porto Novo. Generally, the bacterial groups were found maximum in water during the rainy season. This might be due to the proliferation of nutrients derived from the adjacent river runoff which enhance the maximum growth of bacterial population. Addition of essential nutrients would have stimulated the bacterial counts [6]. However, none of the actinomycetes and fungi counts were recorded in water sample. Likewise, [22] reported that, no actinomycetes and fungi counts were recorded in water sample in Bay of Bengal.

The counts of sediment actinomycetes were recorded maximum (5 $x10^5$ CFU.g⁻¹) at Thondi whereas the maximum (12 $x10^6$ CFU.g⁻¹) counts of sediment actinomycetes were recorded in Andaman Islands [4], [23] reported that, the maximum counts of sediment actinomycetes were recorded in Palk Strait region. Concordantly, the counts of fungi were also recorded maximum (12 $x10^5$ CFU.g⁻¹) in sediment by the present study. The fungal counts in sediment were recorded maximum (17 $x10^5$ CFU.g⁻¹) in Bay of Bengal [22]. Generally, the actinomycetes and fungi counts were higher in sediments than in water. Moreover, the maximum counts of actinomycetes and fungi might be due to the availability of the huge amounts of dissolved and particulate nutrients would have gradually deposited on the bottom sediments which stimulate the counts [25]. [8] Reported that, the actinomycetes can forms only a small fraction of micro flora and they can survive as spores or resting propagules in marine sediment. The bacteria play an important role in the formation of sediments through their metabolic activities and responsible for the biological transformation of organic





(An ISO 3297: 2007 Certified Organization)

Vol. 2, Issue 11, November 2013

matter [18]. So, the level of organic particles and other composition including carbohydrates also diminished gradually due to their metabolic activities. Moreover, they involved in the degradation of organic matter in sediment and water which release the dissolved organic and inorganic substances [26], [9] and [17]. The biochemical constituents of the present study suggested that, the level of total protein and amino acids found maximum than the total sugars and this could be due to either microbial degradation or dissolution of reserve soluble carbohydrate and its derivatives. [19] Reported that, the bacterial groups are the main contributors for the degradation and sedimentation of organic particles. The microbial counts showed statistically significant correlation with the biochemical constituents.

The correlation analysis revealed that, the counts of THB showed significant (p<0.05) positive correlation with amino acids in sediment and water and showed negative (p<-0.05) correlation with carbohydrates and proteins in sediment and water. *E. coli* showed significant (p<0.05) positive correlation with amino acids in sediment and carbohydrates, non reducing sugar in water. The counts of actinomycetes didn't showed positive correlation with none of the biochemical constituents in sediment. The counts of fungi showed significant (p<0.05) positive correlation with protein and reducing sugar in sediment (Table 2).

Name of the	Total sugar		Protein		Amino acid		Reducing sugar		Non-reducing sugar	
Station	sediment	water	sediment	water	sediment	water	sediment	water	sediment	water
Station	(mg.g ⁻¹)	(mg.ml ⁻¹)	(mg.g ⁻¹)	(mg.ml ⁻¹)	(mg.g ⁻¹)	(mg.ml ⁻¹)	(mg.g ⁻¹)	(mg.ml ⁻¹)	(mg.g ⁻¹)	(mg.ml ⁻¹)
Biochemical constituents in water and sediment during January										
Kodiyakarai	2.64	0.31	50.66	2.66	29.45	1.68	1.40	0.136	1.24	0.17
Mallipattinam	3.87	0.24	100.66	1.73	41.27	1.35	1.33	0.132	2.54	0.11
Manora	4.83	0.15	119.33	1.99	32.34	1.06	1.48	0.134	3.53	0.02
Manamelkudi	3.63	0.25	56.66	3.00	30.54	1.23	1.31	0.134	2.32	0.12
Kottaipattinam	4.61	0.36	123.33	3.73	32.48	1.15	1.32	0.133	3.29	0.23
Mimisal	5.21	0.24	97.99	2.33	21.24	0.98	1.33	0.132	3.88	0.10
S. P. Pattinam	4.22	0.13	111.33	2.46	17.14	1.02	1.43	0.118	2.79	0.02
Thondi	4.00	0.18	90.66	3.26	18.36	1.10	1.30	0.126	2.70	0.05
	Biochemical constituents in water and sediment during February									
Kodiyakarai	2.03	0.22	38.66	2.86	22.64	1.02	0.96	0.118	1.07	0.11
Mallipattinam	1.94	0.28	45.33	2.40	27.56	1.08	0.81	0.112	1.13	0.17
Manora	1.95	0.38	49.33	3.26	31.35	1.22	0.87	0.116	1.08	0.27
Manamelkudi	1.83	0.26	43.99	2.59	24.99	1.03	0.72	0.097	1.11	0.17
Kottaipattinam	2.08	0.23	51.99	2.79	24.09	1.05	1.00	0.131	1.08	0.10
Mimisal	5.44	0.25	57.99	2.73	18.13	1.12	1.19	0.123	4.25	0.13
S. P. Pattinam	2.72	0.28	48.00	2.33	14.61	1.01	1.23	0.114	1.49	0.17
Thondi	2.89	0.24	49.99	2.19	14.12	1.03	1.19	0.116	1.70	0.13
Biochemical constituents in water and sediment during March										
Kodiyakarai	2.30	0.12	51.99	1.33	24.58	1.03	1.37	0.106	0.93	0.014
Mallipattinam	2.74	0.26	54.0	2.06	27.02	1.14	1.51	0.108	1.23	0.157
Manora	3.10	0.13	62.66	1.59	23.36	1.31	1.58	0.114	1.52	0.016
Manamelkudi	1.70	0.14	79.33	1.46	24.40	1.17	1.48	0.109	0.22	0.034
Kottaipattinam	2.85	0.13	89.33	2.46	23.81	1.17	1.49	0.119	1.36	0.019
Mimisal	3.08	0.15	84.66	2.79	22.24	1.15	1.40	0.120	1.68	0.033
S. P. Pattinam	2.67	0.13	72.06	1.80	23.63	1.13	1.38	0.113	1.29	0.022
Thondi	2.21	0.13	74.66	1.66	22.96	1.12	1.39	0.115	0.82	0.017

Table 1: Biochemical constituents in water and sediment samples at chosen collection sites



(An ISO 3297: 2007 Certified Organization)

Vol. 2, Issue 11, November 2013

Fig.2. Assessment of THB counts in water along Palk Strait





Fig.5. Assessment of E. coli counts in sediment along Palk Strait

Fig.3. Assessment of *E. coli* counts in water along Palk Strait



Fig.4. Assessment of THB in sediment along Palk Strait



Fig.6. Assessment of actinomycetes counts in sediment along Palk Strait



Fig.7. Assessment of fungi counts in sediment along Palk Strait



Table 2: Significant (p<0.05) correlation between microbial population and biochemical constituents



(An ISO 3297: 2007 Certified Organization)

Vol. 2, Issue 11, November 2013

S. No	Variables	Correlation					
Sediment							
1.	THB Vs carbohydrate	Negative					
2.	THB Vs protein	Negative					
3.	THB Vs amino acid	Positive					
4.	THB Vs reducing sugar	Positive					
5.	THB Vs non reducing sugar	Negative					
6.	E. coli Vs carbohydrate	Negative					
7.	E. coli Vs protein	Negative					
8.	E. coli Vs amino acid	Positive					
9.	E. coli Vs reducing sugar	Negative					
10.	E. coli Vs non reducing sugar	Negative					
11.	Actinomycetes Vs carbohydrate	Negative					
12.	Actinomycetes Vs protein	Positive					
13.	Actinomycetes Vs amino acid	Negative					
14.	Actinomycetes Vs reducing sugar	Negative					
15.	Actinomycetes Vs non reducing sugar	Negative					
16.	Fungi Vs carbohydrate	Negative					
17.	Fungi Vs protein	Positive					
18.	Fungi Vs amino acid	Negative					
19.	Fungi Vs reducing sugar	Positive					
20.	Fungi Vs non reducing sugar	Negative					
Water							
21	THB Vs carbohydrate	Negative					
22	THB Vs protein	Negative					
23	THB Vs amino acid	Positive					
24	THB Vs reducing sugar	Negative					
25	THB Vs non reducing sugar	Negative					
26	E. coli Vs carbohydrate	Positive					
27	E. coli Vs protein	Positive					
28	E. coli Vs amino acid	Negative					
29	E. coli Vs reducing sugar	Negative					
30	E. coli Vs non reducing sugar	Positive					

III. CONCLUSION

In conclusion, the microbial counts vary with their geographical regions and significantly correlated with the biochemical constituents. Among the bacterial groups, the faecal coli form of *E. coli* counts were found maximum in Mallipattinam coast and this might be due to the pollution caused particularly by the anthropogenic activity when compared with other collection sites. Moreover, the present study provides adequate information on the relationship between the microbial population and the biochemical constituents. However, necessary steps to be needed so as to manage the pollution free coastal ecosystem by the manmade activities.

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International Journal of Innovative Research in Science, Engineering and Technology

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Vol. 2, Issue 11, November 2013

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