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Distribution of Heterotrophic Bacteria in the near shore waters and Intertidal Sediments of Visakhapatnam, east coast of India

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Abstract

Heterotrophic bacteria density in the near shore waters and intertidal sediments was studied from May 2014 to May 2015 at fortnightly intervals from five stations. The mean values of water temperature, salinity, dissolved oxygen, pH and sediment organic matter were 28.6°C, 34.9 ppt, 4.73 mg/l, 7.7 and 0.73% respectively. The total bacteria density in the near-shore waters ranged from a low density of 213 x 10^3 cfu/ml (Station 2; May II) to a high density of 463 x 10^3 cfu/ml (Station 3; November II) and averaged to 334 x 10^3 cfu/ml. The total bacteria density in the intertidal sediments fluctuated between 172 x 10^3 cfu/g (Station 1; March II) and 475 x 10^3 cfu/g (Station 3; December II) and averaged to 316 x 10^3 cfu/g. Significant negative correlations are observed for salinity, DO and pH at Station 2. Significant positive correlations are recorded for water salinity and sediment temperature at Station 4. The paper discusses the impact of physico-chemical parameters on the density distribution of bacteria along Visakhapatnam coast.

Keywords: Heterotrophic bacteria, Density, Near-shore water, Intertidal sediment, Visakhapatnam coast.

Introduction

Heterotrophic bacteria play an important role in food web. They survive in the marine environment by obtaining nourishment from decaying organic materials and dissolved organic matter. They represent a major component of marine organisms and need nutrients like carbohydrates, proteins, lipids etc. for growth and metabolism. Marine heterotrophic bacteria are also a potential source of many commercially important bioactive compounds. Their bioremediation capabilities are also remarkable. Earlier investigators studied the distribution of heterotrophic bacteria in relation to ecological parameters from coastal and mangrove habitats[1-15]. Studies on the heterotrophic bacteria from Visakhapatnam coast are limited. They include the heterotrophic bacteria from Fishing Harbour waters and from Meghadri mangroves of Visakhapatnam[16, 17]. The present study focuses on the density of different categories of heterotrophic bacteria in relation to the physico-chemical parameters in near shore waters and intertidal sediments of Visakhapatnam, east coast of India.

Materials and Methods

The investigations were carriedout along Visakhapatnam coast (Lat 17°42'N; Long 83°18'E). The coast of is principally formed by sandy shores. The rocky shores are also present here and there. Five stations (St1: Coastal Battery, St 2: VUDA Park, St 3: Kailasagiri, St 4: Sagar nagar and St 5: Rishikonda) were selected for the present study. An open sewer enters the Bay between St 2 and St 3. Rocky shores are present between St 1 and St 2 and also between St 3 and St 4. All the sampled shores are gradient shores. Samples of seawater and sediment were collected during low tide time at fortnightly intervals from five stations from May 2014 to May 2015. At each station, shore water samples (five replicates) were aseptically collected into five sterile polythene bottles. Sediment core samples (five replicates) were aseptically collected into five sterile polythene bags. Samples of shore water and sediment water were collected for salinity and dissolved oxygen. Data on temperature were collected for air, seawater and sediment using hand-held thermometer (0.1°C sensitivity). Dissolved oxygen was determined by standard Winkler's method. Salinity was determined using a digital salinometer. pH of the water samples was recorded in the field using a digital pH meter (Elico). Sediment organic matter was determined by chromic acid digestion method[18]. One ml of water sample was used for culture purposes from each replicate water samples. Each sediment replicate sample (10 g) was diluted in 100 ml aged sterile seawater (25% distilled water and 75% seawater). Inoculations were prepared using 10 fold dilutions. The heterotrophic bacteria were cultured on Zobell Marine Agar (Himedia) at 33°C for 36 hours. The bacterial colonies were counted using a bacteriological colony counter and the densities were expressed as x10^3 cfu per ml or per g. The density data were presented as mean data of the station. Bacteria were identified upto genus level using cultural, morphological and biochemical characters using Bergey's manual 2010. Pearson correlations were calculated to determine the relation between physicochemical parameters and bacterial densities. The obtained correlations were tested for their significance using't' test.

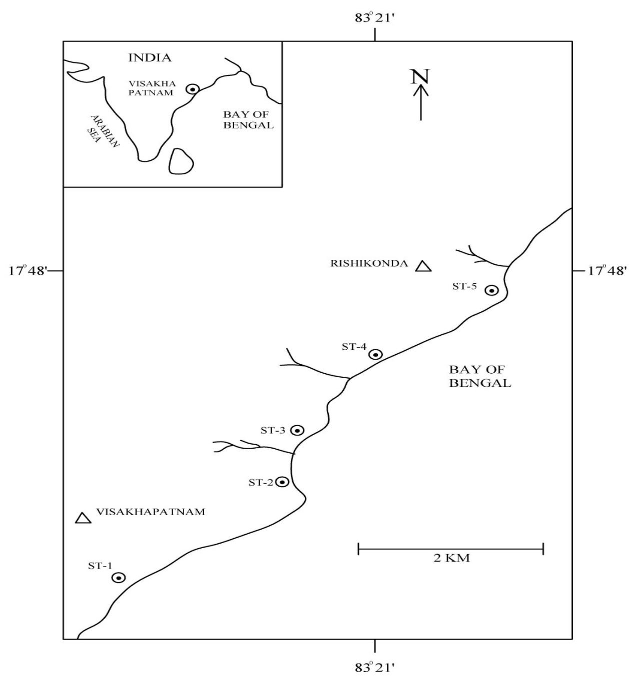

Figure-1
The location of sampling Stations 1 to 5 along Visakhapatnam coast

Results and Discussion

The locations of sampling Stations 1 to 5 along Visakhapatnam coast are shown in Figure-1. Figure-2 gives the fortnightly data on temperature of air (2a), near-shore water (2b), and sediment (2c) and sediment water (2d) at Stations 1 to 5 from May 2014 to May 2015. Figure-3 presents the fortnightly data on near-shore water salinity (3a), near-shore water dissolved oxygen (3b), near-shore water pH (3c), sediment water salinity (3d), sediment water dissolved oxygen (3e), sediment water pH (3f) and sediment organic matter (3g) at Stations 1 to 5 during the study period. Table-1 shows the mean (n=5) density distribution ($x10^3$ cfu/ml) of heterotrophic bacteria in the near-shore waters at Stations 1 to 5 from May 2014 to May 2015. The mean (n=5) density distribution ($x10^3$ cfu/g) of heterotrophic bacteria in the sediment at Stations 1 to 5 from May 2014 to May 2015 is presented in Table-2. Table-3 presents Pearson correlation coefficients (p=0.05) between physico-chemical parameters and bacterial densities of near-shore water and intertidal sediments at Stations 1 to 5 during the study period.

The temperature distribution of air, near-shore water, sediment and sediment water showed in general similar trends during study period. High temperatures were recorded during April and May months. Low temperatures were observed during January month. The mean values of temperature for the air, near-shore water, sediment and sediment water were 31.5, 28.6, 29.2 and 28.6°C respectively. The salinity distribution of near-shore water and sediment water exhibits minor fluctuations between stations. High salinities were recorded in April and May months and the low values were observed from August to January. The mean salinity values for near-shore water and sediment water 34.9 and 35.1 ppt respectively and they were more or less similar for all the five stations investigated. The dissolved oxygen (D.O.) values showed irregular fluctuations and they did not represent any trend either in the near-shore water or in the sediment water in all the five stations. The D.O. values for the near-shore waters were slightly higher than the values of sediment water. The mean values of D.O. for the near-shore and sediments waters were 4.73 and 4.44 mg/l respectively. Minor irregular fluctuations were observed for pH, both in the near-shore and sediment waters, in all the stations. The mean values of pH for near-shore and sediment waters were 7.7 and 7.7 respectively. The sediment organic matter is relatively low in all the stations and did not exhibit any seasonal trend. The mean value of sedimentary organic matter was 0.73%.

The qualitative analyses of the bacterial samples, both in the water and sediment, revealed the occurrence of five genera along Visakhapatnam coast and their density distribution will be discussed elsewhere. Quantitatively, the total bacteria in the near-shore waters exhibited irregular fluctuations in all the stations investigated during the study period. The total bacteria density in the near-shore water ranged from a low density of $213x10^3$ cfu/ml (Station 2; May II) to a high density of $463x\ 10^3$ cfu/ml (Station 3; November II) and averaged to $334x10^3$ cfu/ml. Among the stations, the mean total bacterial densities were lowest ($322\ x10^3$ cfu/ml) and highest ($356x10^3$ cfu/ml) at St. 4 and St. 3 respectively and averaged to $334\ x\ 10^3$ cfu/ml. The quantitative distribution of total bacteria in the intertidal sediments also showed irregular fluctuations, throughout the study period, in all the five stations. The total bacteria density in the sediment fluctuated between $172x10^3$ cfu/g (Station 1; March II) and $475\ x10^3$ cfu/g (Station 3; December II) and averaged to $316x10^3$ cfu/g. Among the stations, the mean lowest ($286x\ 10^3$ cfu/g) and highest ($353\ x10^3$ cfu/g) total bacteria densities were observed in St.4 and St. 5 respectively and averaged to $316\ x\ 10^3$ cfu/g. Variations in bacterial density fluctuations at St. 3 and St.5 were relatively low in the near-shore waters when compared with the intertidal sediments. The Pearson correlation analyses between physico-chemical parameters and bacterial densities revealed both the negative and positive correlations. Majority of these correlations were insignificant (p=0.05). At Station 2, significant negative correlations were observed in the intertidal sediments for salinity, dissolved oxygen and pH. At Station 4, the total bacteria densities showed significant positive correlations for near-shore water salinity and sediment temperature. At Station 3, a significant negative correlation was observed between near-shore water temperature and total bacteria density. Sediment organic matter, which was present in low quantities during the present study, showed insignificant negative correlations at all stations except at St.1.

Table-1
The mean (n=5) density distribution (x10^3 cfu/ml) of heterotrophic bacteria at fortnightly intervals in the near-shore waters at Stations 1 to 5 from May 2014 to May 2015

S1 WATER			S2 WATER		S3 WATER		S4 WATER		S5 WATER	
FORTNIGHTS	T.B.C in 10^3cfu/ml	SD	T.B.C in 10^3cfu/ml	SD	T.B.C in 10^3cfu/ml	SD	T.B.C in 10^3cfu/ml	SD	T.B.C in 10^3cfu/ml	SD
M-2	243	142.618	213	69.62	296	76.702	239	84.583	292	16.000
J-1	283	45.021	334	143.60	328	131.567	340	74.287	303	139.039
J-2	245	146.293	272	112.37	253	148.928	434	85.431	346	131.373
J-1	379	140.520	306	134.14	356	72.772	422	87.460	338	89.968
J-2	350	156.090	410	78.99	393	122.469	285	141.656	259	87.243
A-1	274	106.214	334	99.09	356	83.242	364	128.332	285	93.859
A-2	335	110.646	337	160.85	419	220.714	375	70.235	292	181.293
S-1	288	147.098	290	162.39	362	180.283	256	85.587	367	103.833
S-2	332	176.382	355	88.20	305	127.248	347	88.519	337	110.350
O-1	344	121.106	322	107.35	301	107.670	233	111.747	364	102.307
O-2	428	200.766	289	73.37	403	262.809	353	108.146	347	203.177
N-1	347	132.926	342	93.25	403	82.707	302	143.954	376	84.206
N-2	284	108.787	330	65.40	463	173.447	271	87.426	408	105.791
D-1	242	126.379	343	164.56	411	246.127	291	71.143	311	153.462
D-2	382	59.781	389	150.79	409	80.739	345	95.366	358	91.212
J-1	405	161.214	287	81.37	361	84.769	462	76.292	297	146.114
J-2	366	128.778	260	102.71	418	60.632	233	112.713	392	72.178
F-1	321	96.831	279	99.60	348	100.979	300	103.640	342	62.400
F-2	314	120.020	296	106.98	431	221.301	252	105.398	366	69.262
M-1	351	143.278	392	120.26	349	137.393	368	178.806	370	168.367
M-2	347	35.473	408	113.80	328	88.990	361	156.857	295	74.248
A-1	327	128.632	415	219.48	243	81.509	266	122.453	298	59.663
A-2	322	116.622	328	105.90	250	121.826	262	95.578	325	94.261
M-1	385	109.017	409	87.83	366	83.697	370	53.505	291	124.935

Table-2
The mean (n=5) density distribution (x10³ cfu/g) of heterotrophic bacteria at fortnightly intervals in the intertidal sediments at Stations 1 to 5 from May 2014 to May 2015

S1 Sediment			S2 Sediment		S3 Sediment		S4 Sediment		S5 Sediment	
FORT NIGHTS	T.B.C 10^3cfu/ml	SD	T.B.C 10^3cfu/ml	SD	T.B.C 10^3cfu/ml	SD	T.B.C 10^3cfu/ml	SD	T.B.C 10^3cfu/ml	SD
M-2	206	74.159	335	5.380	249	86.549	191	51.160	280	60.7580
J-1	225	138.895	207	23.825	275	86.131	231	119.086	321	148.9040
J-2	215	108.073	314	16.702	379	88.754	444	84.780	277	167.9230
J-1	307	171.676	288	22.075	246	111.064	310	186.202	306	70.6940
J-2	383	139.708	254	13.739	400	241.255	352	76.383	257	18.2130
A-1	283	116.573	271	29.114	297	155.024	256	120.527	403	39.3000
A-2	288	99.991	332	26.293	414	195.884	278	105.179	420	113.7400
S-1	260	183.357	271	20.694	349	88.190	198	77.450	343	123.3560
S-2	364	186.233	414	10.947	322	83.898	344	61.732	393	90.8140
O-1	344	127.621	305	24.143	253	65.161	281	83.344	334	47.7700
O-2	336	97.266	357	7.454	281	72.947	312	52.376	307	125.7110
N-1	336	180.088	384	11.331	272	61.634	224	83.589	461	74.9050
N-2	376	91.253	285	9.331	318	164.234	394	90.954	417	158.1540
D-1	309	142.566	323	4.216	403	78.729	305	93.233	298	97.2910
D-2	212	50.413	333	17.027	475	80.104	238	107.720	258	59.0400
J-1	318	107.482	337	22.437	358	167.339	312	82.500	410	66.4150
J-2	342	138.846	332	27.853	370	112.810	193	102.632	463	61.0550
F-1	392	198.279	397	18.496	342	48.910	213	56.260	397	63.2190
F-2	349	162.531	287	30.575	286	142.345	292	79.939	468	128.0100
M-1	327	131.051	348	18.458	311	88.976	278	61.188	341	68.2220
M-2	172	101.967	303	31.107	351	67.489	195	60.230	286	58.5340
A-1	285	152.982	299	26.091	255	43.557	414	82.270	272	85.1360
A-2	290	139.933	309	22.109	321	150.830	265	145.589	400	98.3450
M-1	342	111.760	296	9.159	300	167.127	351	57.191	365	156.5250

Table-3a
Pearson correlation coefficients (p=0.05) between physico-chemical parameters and bacterial densities of near-shore water at Stations 1 to 5 during the study period

	S1	S2	S3	S4	S5
Temperature	-0.333	0.144	-0.401*	0.005	-0.245
DO	-0.027	0.321	-0.0148	0.166	0.083
pH	-0.228	-0.148	-0.306	-0.122	-0.172
Salinity	-0.090	0.213	-0.0492	0.366*	-0.420

Table-3b
Pearson correlation coefficients (p=0.05) between physico-chemical parameters and bacterial densities of intertidal sediment at Stations 1 to 5 during the study period

	S1	S2	S3	S4	S5
Temperature	-0.298	-0.194	-0.229	0.340*	-0.260
DO	0.085	-0.360*	0.297	0.185	0.063
pH	-0.264	-0.477*	-0.157	0.254	-0.290
Salinity	-0.128	-0.469*	-0.214	0.055	-0.007
OM	0.113	-0.214	-0.066	-0.279	-0.172

* indicates significant correlation

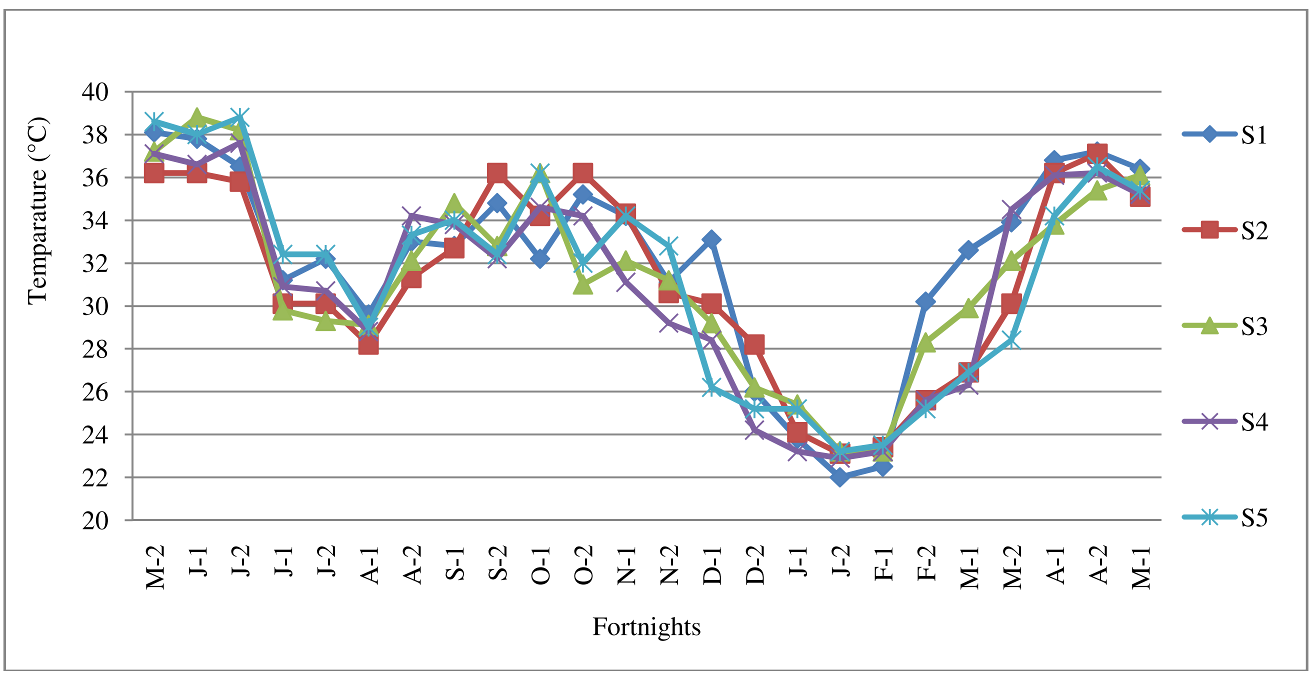

Figure-2a
Distribution of Air Temperature (°C) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

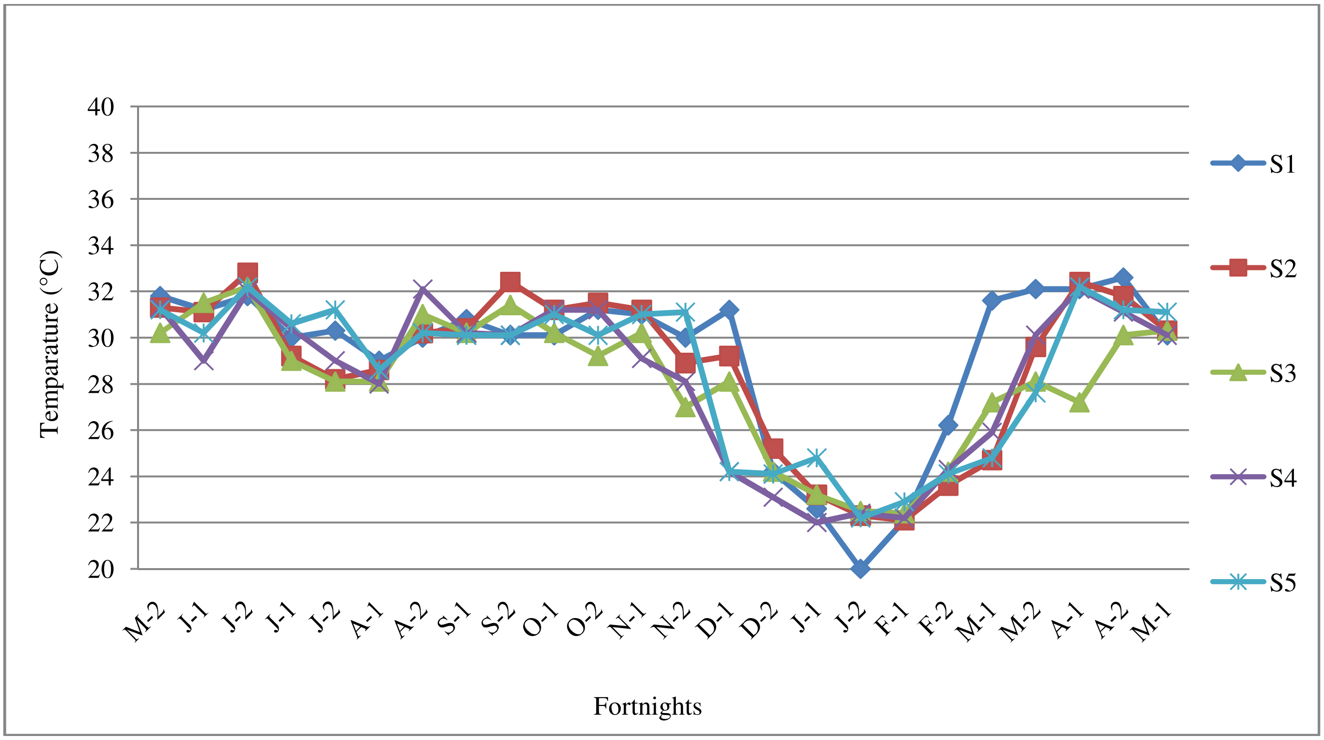

Figure-2b
Distribution of near shore water Temperature (°C) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

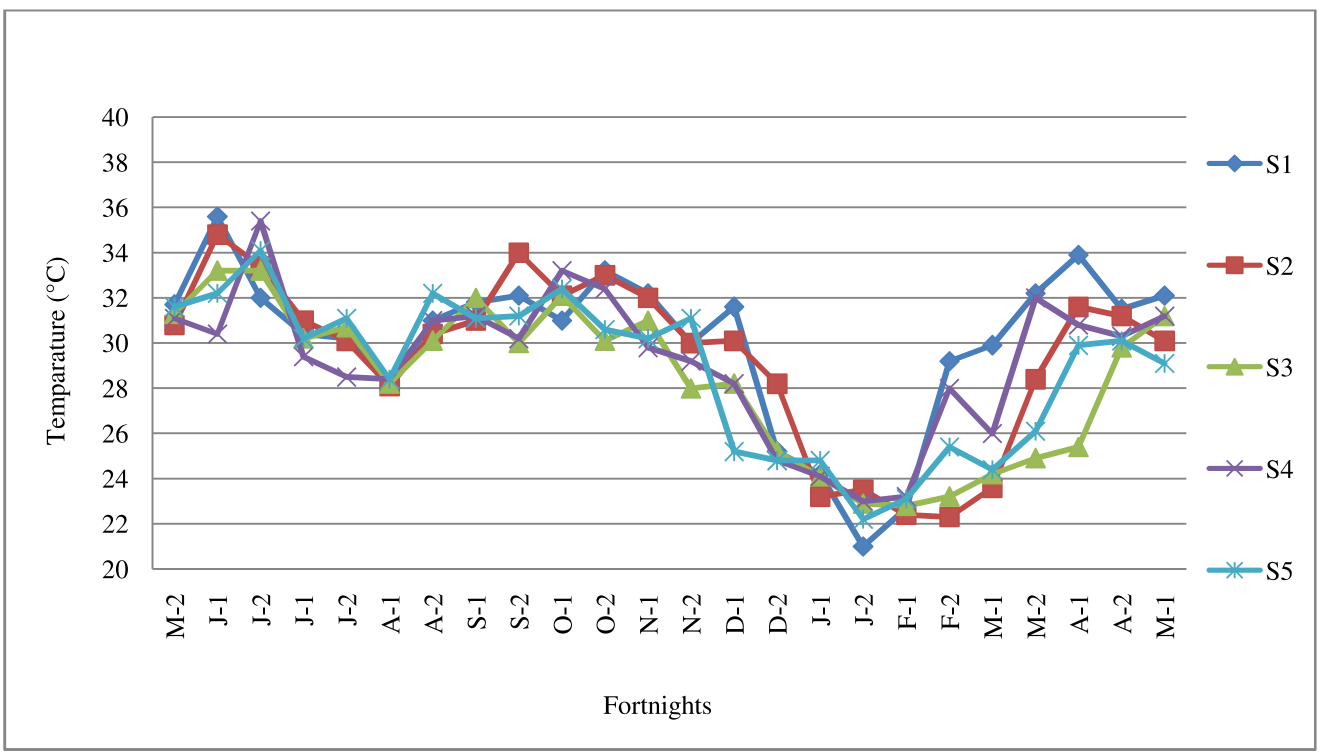

Figure-2c
Distribution of sediment Temperature (°C) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

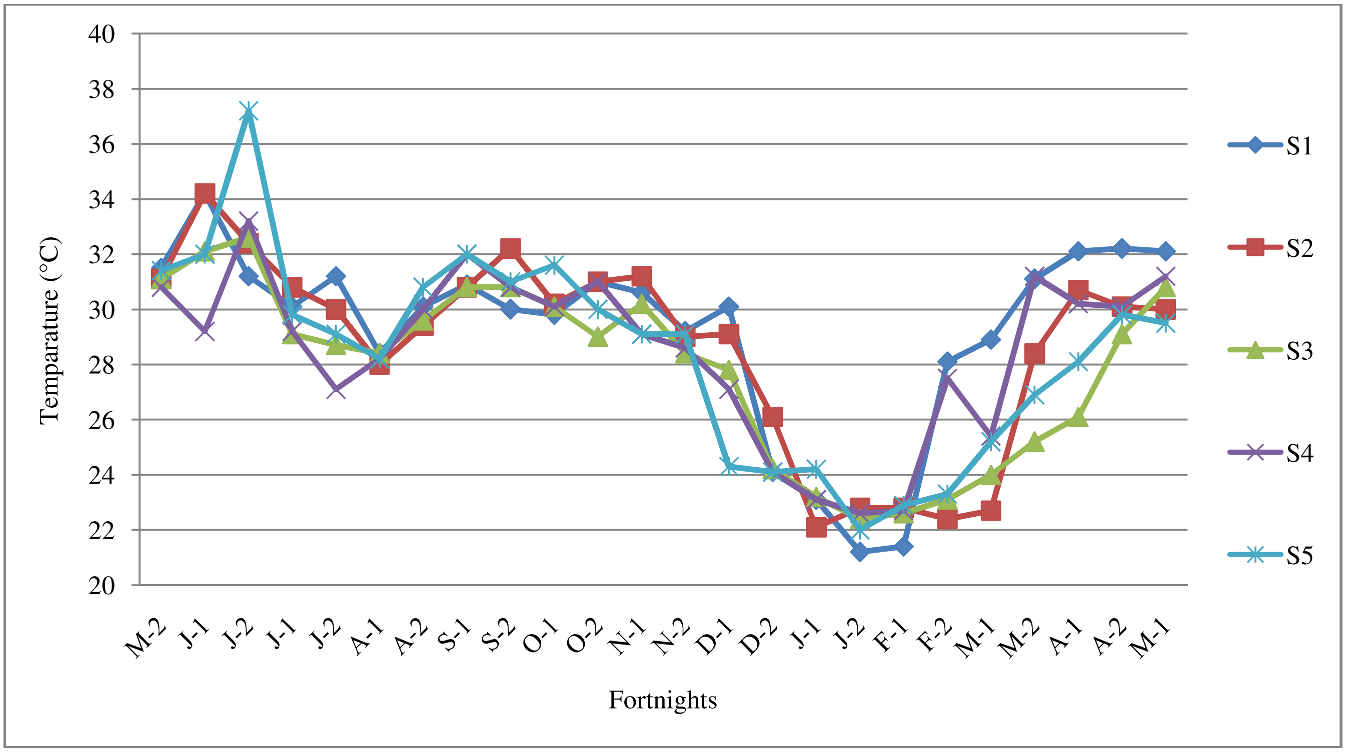

Figure-2d
Distribution of sediment water Temperature (°C) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

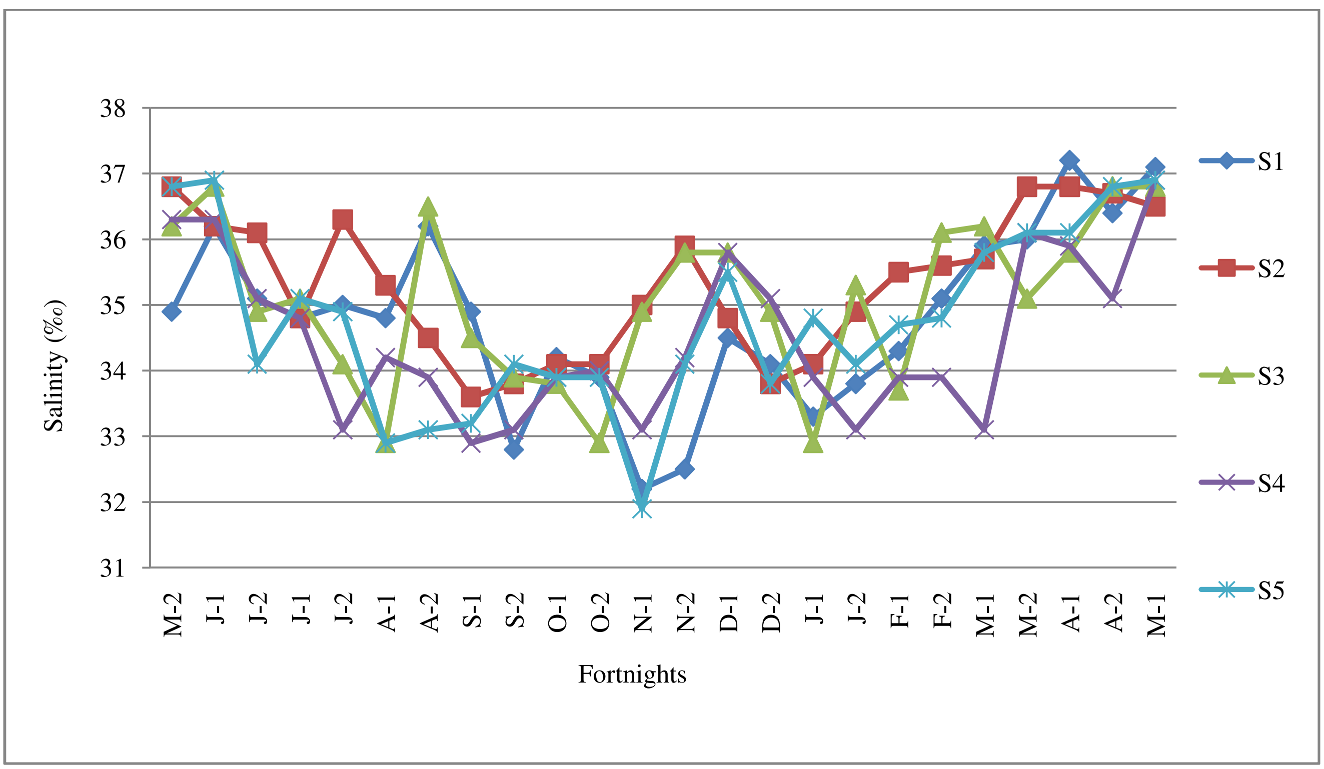

Figure-3a
Distribution of near shore water salinity (‰) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

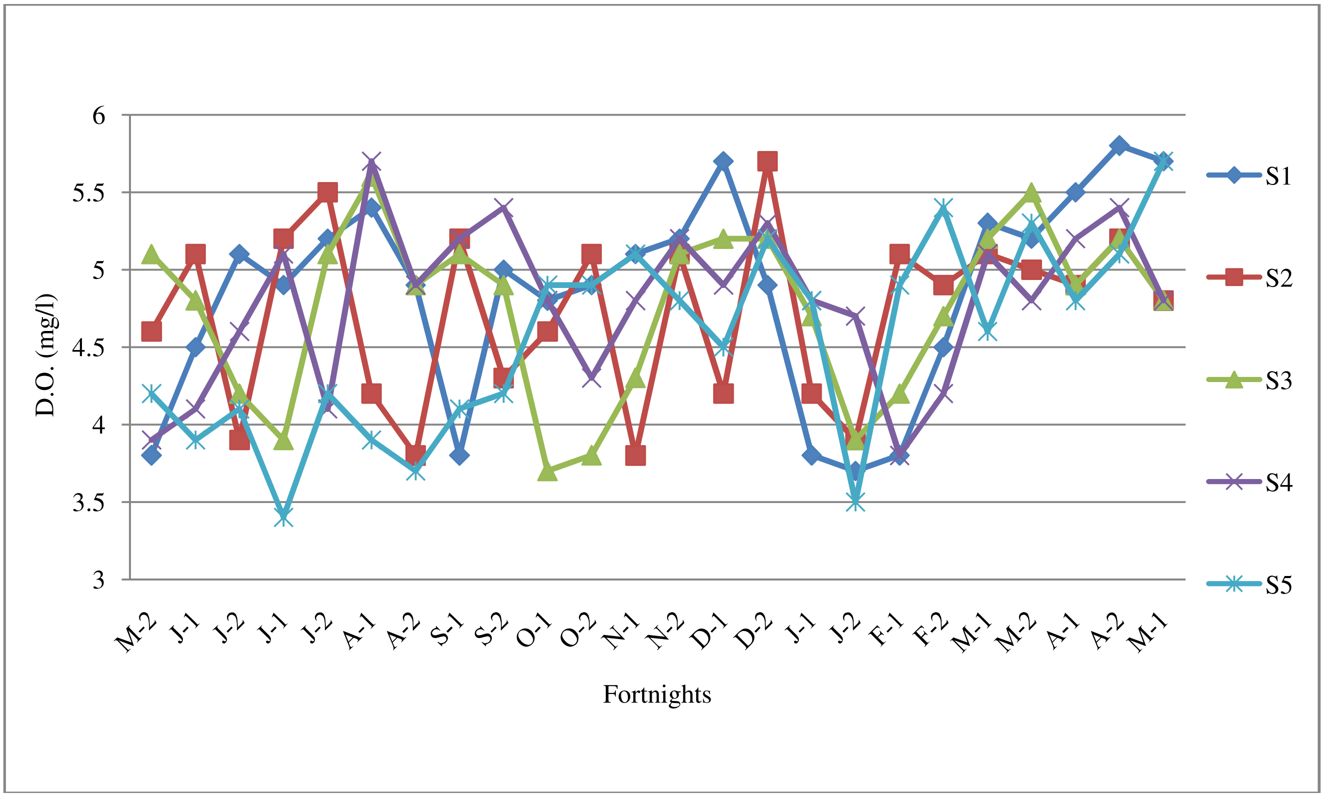

Figure-3b
Distribution of surface waters dissolved oxygen (mg/l) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

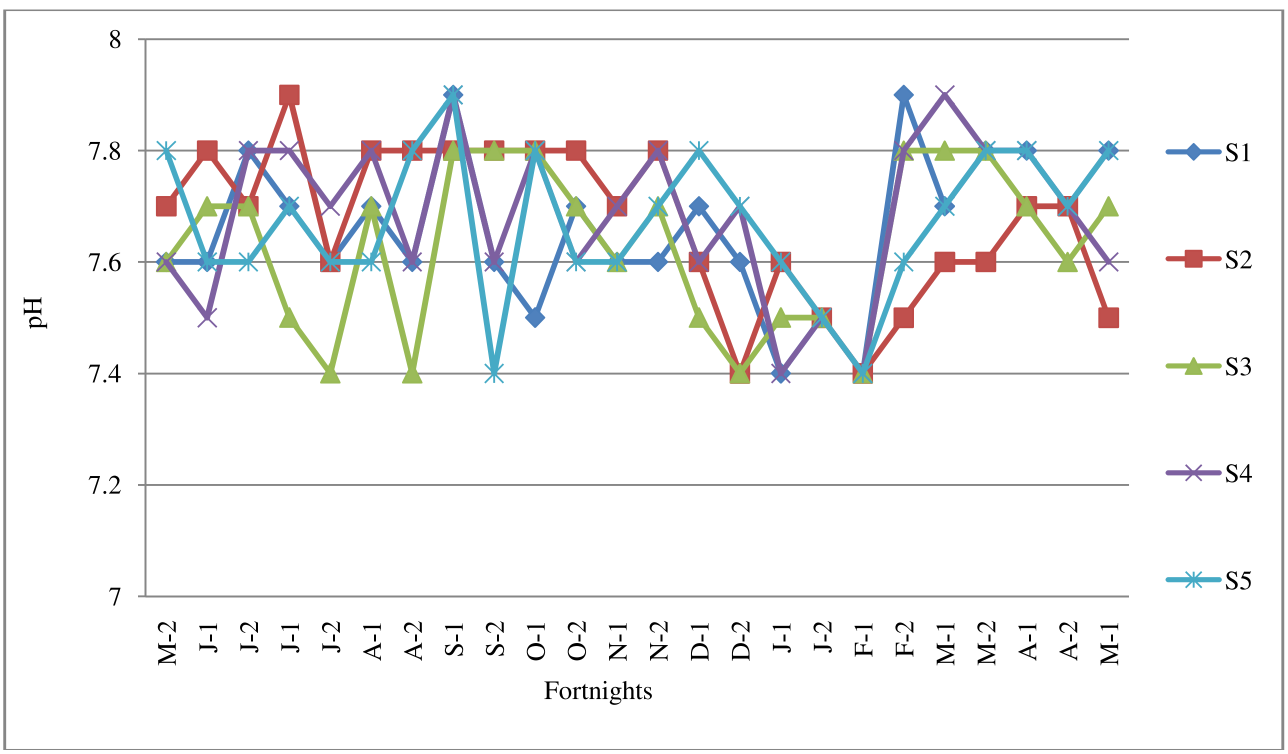

Figure-3c
Distribution of surface waters pH (‰) at fortnightly intervals at Stations 1 to 5 from May 2014to May 2015

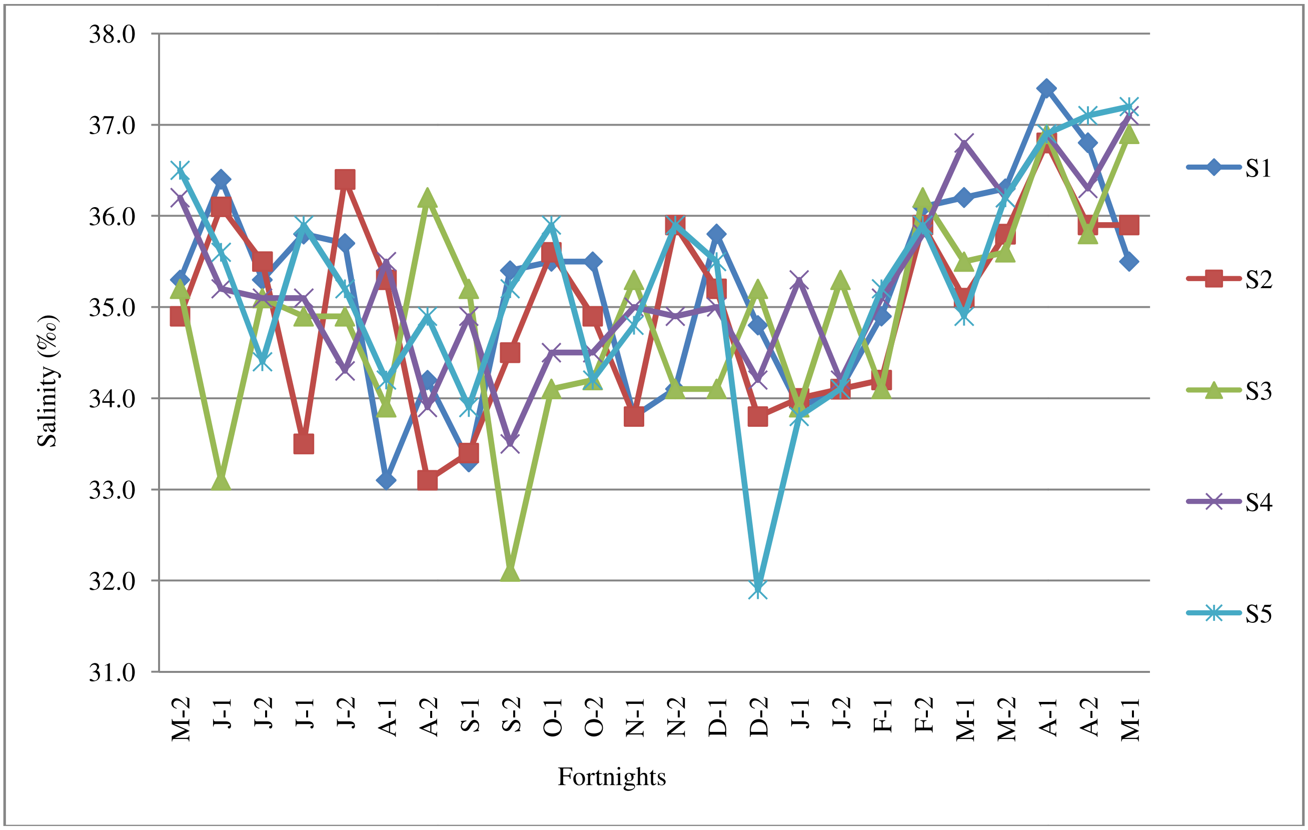

Figure-3d
Distribution of sediment water salinity (‰) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

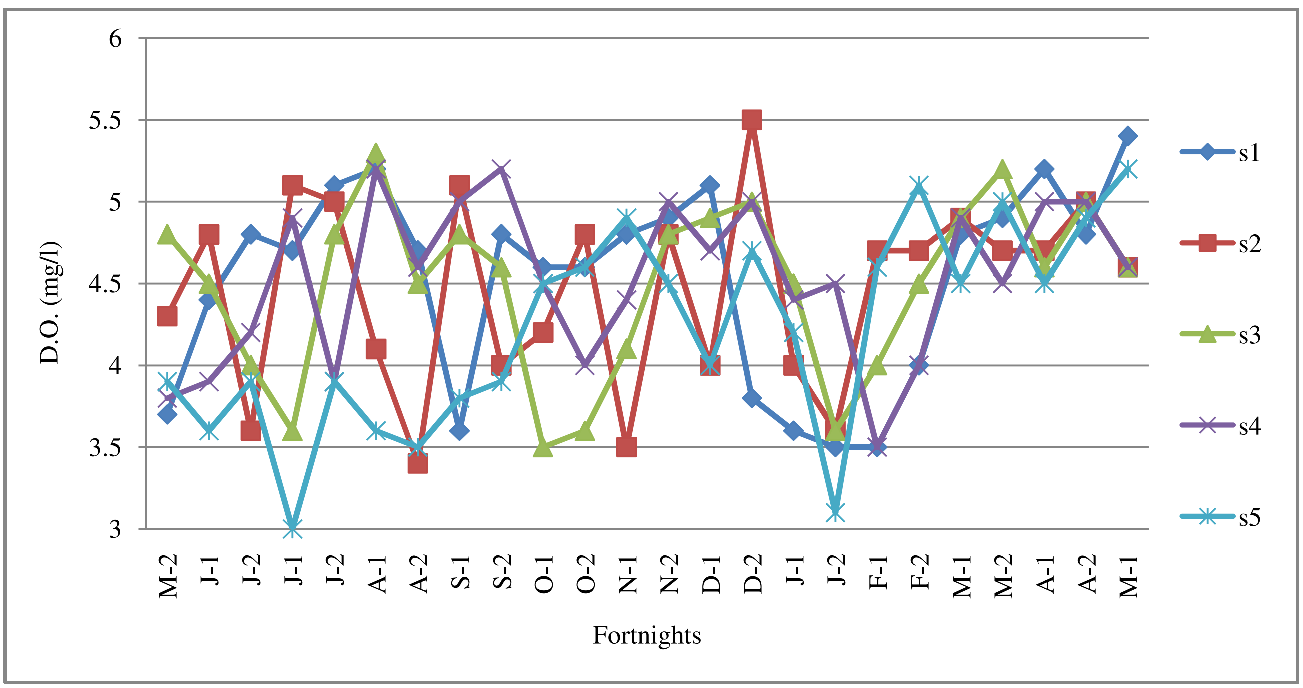

Figure-3e
Distribution of sediment waters dissolved oxygen (mg/l) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

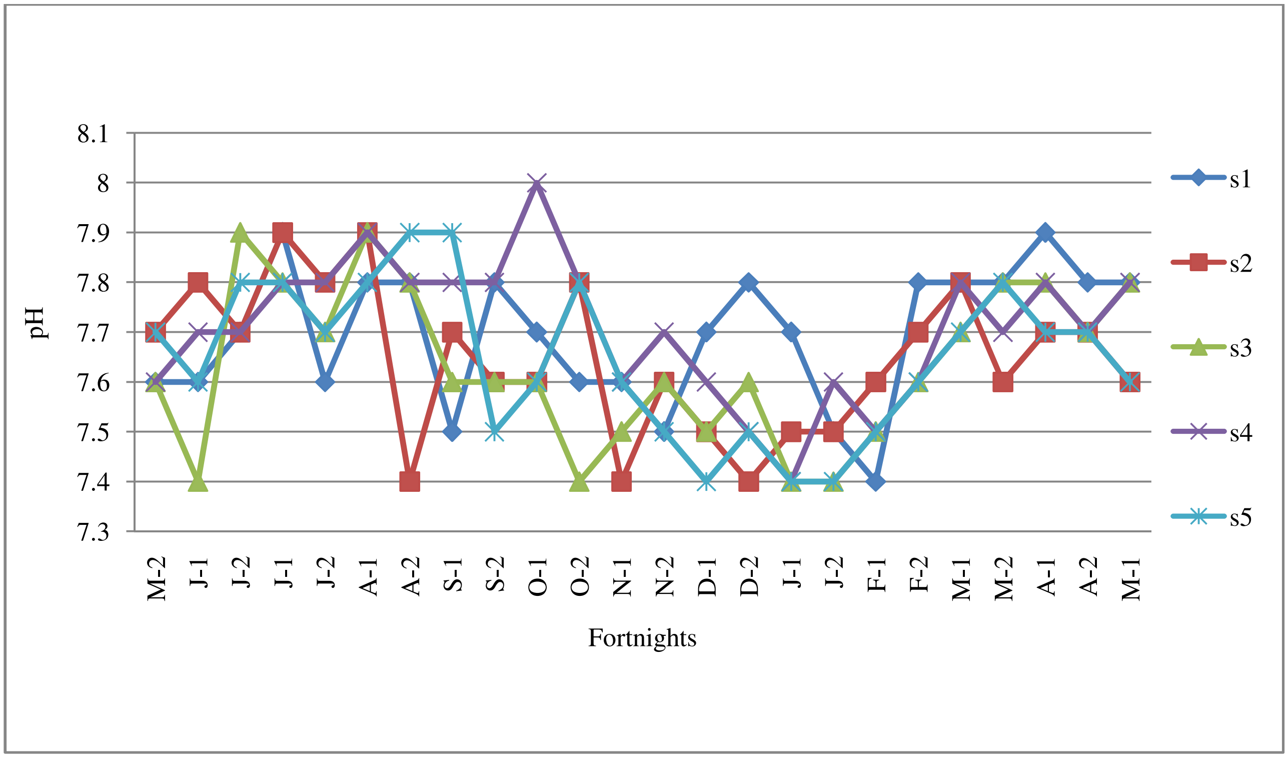

Figure-3f
Distribution of sediment waters pH at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

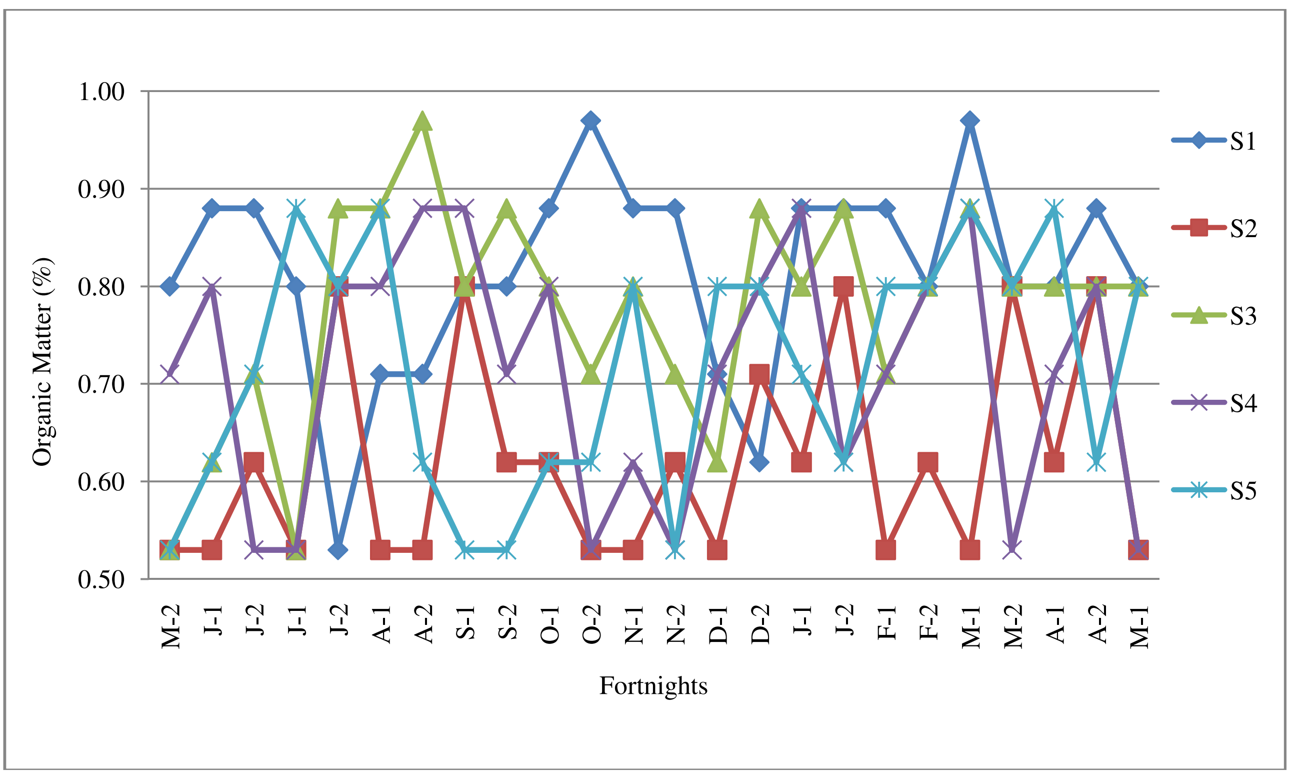

Figure-3g
Distribution of sediment organic matter (%) at fortnightly intervals at Stations 1 to 5 from May 2014 to May 2015

Discussion: It is observed that, in all the stations investigated, the total bacteria densities are relatively higher in shore water than in intertidal sediments. The mean total bacterial densities for the near-shore water (334 x10³ cfu/ml) and intertidal sediment (316x10³ cfu/g) also indicate that shore water support *ca.* 6% high load of bacterial density than the intertidal sediments. The high bacterial density in the shore water may due to land drainage, which enters the Bay waters between St. 2 and St. 3. The occurrence of high bacterial densities at Station 3 (356x10³ cfu/ml; near-shore water) and at Station 5 (352 x 10³ cfu/g; intertidal sediment) may be due to anthropogenic factors as these two stations are used by tourists.

The correlation analyses (Table 3) between physico-chemical parameters and heterotrophic bacteria reveal that majority of the correlations are negative though insignificant. It is interesting to note that at Station 4 the bacterial densities exhibited significant positive correlations with near-shore water salinity and sediment temperature. At Station 2, the sediment bacteria showed significant negative correlations with dissolved oxygen, pH and salinity. At Station 3, the near-shore water bacteria density recorded a significant negative correlation with water temperature.

The correlation data between total heterotrophic bacteria and four environmental parameters during 2014-15 (Table 3) indicate that the heterotrophic bacteria density fluctuations are governed by several other factors, which ultimately determine the types of correlations between densities and parameters.

Heterotrophic bacteria densities in the marine zones of Vellar estuary and Killai were reported as 10.9 x 10³ cfu/ml and 100 x 10³cfu/ml respectively[5,7]. In Cochin coastal waters, 750 x 10³ cfu/ml of heterotrophic bacteria were recorded[9].

He registered two peaks: the major one in January and February and the minor one in August and September. He attributed the evaporation of surface water, suitable temperature and low variations in salinity as the reasons for a major peak during summer. The minor peak is attributed to the DOM-exudates of microalga Noctiluca. The *Noctiluca* releases DOM-containing exudates, which favour the growth of bacteria. Prabhu *et al* recorded highest density 26.8x10³ cfu/ml of bacteria in the coastal waters of Madras[19]. He observed very minimal fluctuations in the monthly density distribution of bacteria.

He attributed the high density distribution to sewage pollution. Even though anthropogenic impact is noted, the present study did not cover on the pollution aspect. From Dandi coastal waters, bacterial density of 720 x 10³ cfu/ml was recorded[20]. The coastal waters of Tamilnadu supported very low (1.0 x 10³ cfu/ml) bacterial densities[21]. Mandovi-Zuari estuaries harboured 18.9 x 10³ cfu/ml[11]. Visakhapatnam fishing harbour registered 2.23 x 10³ cfu/ml[16]. Meghadri estuary at Visakhapatnam recorded 7.93 x 10³ cfu/ml[17].

Conclusion

The heterotrophic bacterial densities recorded in the near-shore waters of the present study are relatively higher than the earlier reports except from Cochin coastal waters[9]. The sediment bacterial density recorded in Madras coastal sediments (2460 x 10³ cfu/g) is relatively higher than the present study report[19]. In the present investigation, only some of the observed fluctuations in the bacterial densities could be explained with the observed temperature, salinity, dissolved oxygen and sedimentary organic matter. The recorded variations in bacterial densities may be due to some other physico-chemical and biological factors besides regional variations in the habitats.

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