ISSN 2321–340X

Journal of Aquatic Biology & Fisheries | Vol. 5 | 2017 | pp. 54-64

SPATIAL DIFFERENCES IN BACTERIAL AND WATER QUALITY PARAMETERS IN SEAGRASS MEADOWS OF TUTICORIN COAST, GULF OF MANNAR, SOUTHEASTERN INDIA

Arasamuthu, A.,* Mathews, G. and Patterson Edward, J.K.

Suganthi Devadason Marine Research Institute, 44- Beach Road, Tuticorin- 628001, India
(Affiliated to Manonmaniam Sundaranar University, Abishekapatti, Tirunelveli - 627012. Tamilnadu, India)
**Email: kingpearl28@gmail.com*

Abstract: Comparative analyses of bacteriological and water quality parameters were carried out in the coastal waters of Tuticorin, Gulf of Mannar. Samples of sediment, water and seagrass were collected from three stations; the sites were identified on seagrass meadows, based on their spatial differences from the shore. Of the three sites, Site1, lying close to the untreated sewage outlet, was found to be the most polluted one with fecal coliform, THB, *E. coli* and *Vibrio* bacteria. Statistically the three stations had significant differences among themselves ($P<0.001$) in all the microbial parameters examined. Reduced levels of dissolved oxygen, increased values of total suspended solids and high turbidity were also recorded at Site 1, which is evidently due to its proximity to the sewage outlet. In order to conserve the fragile seagrass ecosystem and the dependant biodiversity, immediate action should be taken to treat the domestic waste water before allowing it to mix with the seawater.

Keywords: Seagrass, Bacteria, *E. coli*, waste water, Gulf of Mannar

INTRODUCTION

Seagrasses are marine flowering plants which grow over a large geographic area worldwide (Short *et al*., 1995). A typical seagrass, like any terrestrial plant, has root, stem and leaves; only it lives submerged under the water. Seagrasses belong to the families, Hydrocharitaceae and Potamogetonaceae, and they are distinctively different from the terrestrial grasses of Poaceae. Seagrasses make one of the most dynamic ecosystems of the marine environment, and provide food and shelter to a wide range of ecologically and commercially important marine organisms. They are the main primary producers of the marine ecosystem. Seagrass meadows provide spawning, nursery, and refuge habitats for a wide variety of fishes and crustaceans. Habitat complexity structured by seagrasses is considered one of the major factors responsible for differences in habitat use. Thousands of people around the world depend exclusively on seagrass meadows for their livelihood.

Though seagrasses constitute one of the most predominant and specialized groups of marine flora, they are poorly known in India, when compared to such other related ecosystems as mangroves. Some of the major seagrass meadows in India exist along the southeast coast (Gulf of Mannar and Palk Bay), in the lagoons of islands in the Lakshadweep group in the Arabian Sea, and near the Andaman and Nicobar islands in the Bay of Bengal. The largest area of sea grass occurs along the Gulf of Mannar and Palk Bay. The regions of India that are colonized by seagrasses support rich and diverse fauna. In Tamil Nadu, Gulf of Mannar and Palk Bay have rich seagrass meadows and the fishermen of the hundreds of coastal villages depend on them for their livelihood. The islands and the coast of Gulf of Mannar have dense growth of seagrass meadows, mainly between the mainland and the islands (shoreward from island), as well as towards the seaward sides of the islands. In the seaward sides of the islands, seagrasses are seen as patches. They are found upto 2 to 3 km from the Island shores towards the open sea (Mathews 2007). Diverse groups of animals such as sea horses, sea turtles, sea cucumbers,

sea urchins, star fishes, gastropods, bivalves, ascidians, sponges, crustaceans etc. are abundant in the seagrass meadows making Gulf of Mannar a zone rich in biodiversity.

Distribution of seagrasses depends on a variety of parameters of water quality including temperature, salinity, nutrient availability, substratum characteristics, turbidity and submarine irradiance (Abal and Dennison, 1996; Dennison and Kirkman, 1996). The availability of nutrients affects the growth, distribution, morphology and seasonal cycle of seagrass communities (Short et al., 1995). Most of the nutrients derived from the seagrasses are internal and are the resultant of decomposed seagrass leaves (Erftemeijer *et al.*, 1993). External supply of nutrients is from domestic sewage and agricultural runoff. Seagrass species are highly susceptible to the attack of microbial pathogens (d'Avack *et al.*, 2014). Wasting disease in seagrasses has caused severe damage to the seagrass meadows in Europe and North America (Muehlstein, 1989). Distribution, diversity and activities of bacteria in the marine environment are controlled by many hydro biogeochemical and physicochemical factors present in the environment (Azam *et al.*, 1983). The present study was undertaken with the intentions to assess the bacterial distribution in the water and sediment of seagrass meadows in Tuticorin region of Gulf of Mannar. The other related physicochemical parameters were also assessed to understand the health of the seagrass ecosystem.

MATERIALS AND METHODS

The present study was carried out during March 2017 at three different seagrass sites in Tuticorin region. Site 1 (depth 1.2 meter) is on the shore at Thirespuram fishing village near the domestic waste outlet (08°48'41.82"N 78° 9'54.07"E). Site 2 (depth 2.4 meter) is 2 km off Tuticorin shore into the sea (08°48'51.45"N 78°11'0.78"E). Site 3 (depth 4.2 meter) occurs 5 km away from Tuticorin shore near Vaan Island (08°49'3.74"N 78°12'31.10"E).

Samples of sediment, water and seagrass leaf blades were collected from the three sites for the microbial analysis. THB (total heterotrophic bacteria), fecal coliform, *E. coli* and *Vibrio* populations were estimated. Sample collection involved scuba diving and the samples were collected in sterile screw-capped bottles. The collected samples were brought to the laboratory in a portable ice box within 2 hours. Seagrass samples were rinsed with sterile seawater and scraped off with a sterile knife. To remove the associated organisms, the surfaces of the leaf blades were rinsed with 70% alcohol, and the inner parts were cut with a sterile knife. The resultant tissues were serially diluted, spread on ½ strength Zobell marine agar medium and incubated at room temperature for 48 hours. On the basis of morphological features, colonies were randomly picked and purified by making streak plates (Madigan et al., 2000). 1 ml of water sample and 1 g of sediment sample were separately added to 99 ml of 50% aged

Fig. 1. Map showing the study sites in seagrass meadows of Tuticorin coast, Gulf of Mannar

sea water and then the mixtures were serially diluted. 0.1 ml of serially diluted sample was inoculated in to the Zobell marine agar and some selective media (TCBS, EMB) to enumerate the THB and also to isolate the specific pathogens. After inoculation, the plates were incubated in an inverted position at a temperature of 28^0 C for 24 to 48 hours. Heterotrophic bacterial populations were expressed as colony forming units in water (CFU/ml) and sediments (CFU/g). Fecal coliform count was calculated by Most Probable Number (MPN) technique in water and sediment samples (APHA, 2015).

Temperature of the water samples was measured using a digital stem thermometer; salinity was measured by hand held refractrometer; pH was measured using pH meter and turbidity was analyzed by water quality analyzer (ELICO PE 138); Total Suspended Solids (TSS) was calculated following APHA (2015); Dissolved Oxygen (DO) was estimated using Winkler's method; nutrients such as nitrate, nitrite, calcium, magnesium, phosphate and silicate were measured spectrophotometrically (ELICO SD 164) following Strickland and Parsons, 1982. The data collected were subjected to statistical analysis using complete randomized design and to ANOVA using the SPSS analysis program (Version 17.0, 2008).

RESULTS AND DISCUSSION

At Site 1, the THB load in sediment sample ranged from $164x10^{-4}$ to $287x10^{-2}$ cfu/g and in water sample it ranged from 150 $x10^{-4}$ to 276 $x10^{-2}$ cfu /ml while in the seagrass leaf sample, it ranged from 142 $x10^{-2}$ to $279x10^{-2}$ cfu/g. At Site 2, the THB load in sediment ranged from $94x10^{-4}$ to 211 $x10^{-2}$ cfu/g; in water, the range was $80x10^{-4}$ to $174x10^{-2}$ cfu/ml; in seagrass leaf, the THB load ranged between 65 $x10^{-2}$ and $143x10^{-2}$ cfu/g. At Site 3, the THB load in sediment ranged from $65x10^{-4}$to 133 $x10^{-2}$ cfu/g; in water it ranged from 56 $x10^{-4}$ to 113 $x10^{-2}$ cfu/ml; in sea grass leaf, the THB load ranged from $17x10^{-2}$ to 43 $x10^{-2}$ cfu/g (Fig.2, 3 and 4). One way ANOVA showed a significant difference ($p<0.001$) in THB load between the sites (Table 1).

Many researchers have reported on the marine bacterial diversity from various parts of the world. Helan et al (2014) reported that the total heterotrophic bacterial load in Poovar estuary ranged between $18.87x10^5$ cfu/g and $38.46x10^5$cfu/g dry sediment. Similar findings were made by Kannan (1996) from Palk Bay, Karuppasamy and Perumal (2000) from Pichavaram waters and Rajasekar (2003) from Vellar estuary. Rajagopal et al (2013) reported in Andaman the density of THB in water samples as varying from $43x10^5$ cfu/ml to $182x10^5$ cfu/ml, and in the sediment samples as varying from $79x10^5$ cfu/g to $259x10^4$ cfu/g. Jayanth et al (2002) reported the THB load in seawater of Therespuram region at a minimum of $5x10^4$ and a maximum of $1.06x10^6$ cfu/ml, and the sediment THB load at $5.10x10^4$ cfu/g and $4.90x10^7$ cfu/g. Site 1 in the present study was found to be affected by the untreated sewage, and hence it may be inferred that the bacterial diversity was higher in it than in the other 2 sites. At Site 1, the microbial load was higher as expected because the wastes of human and animal origin are discharged into the sea

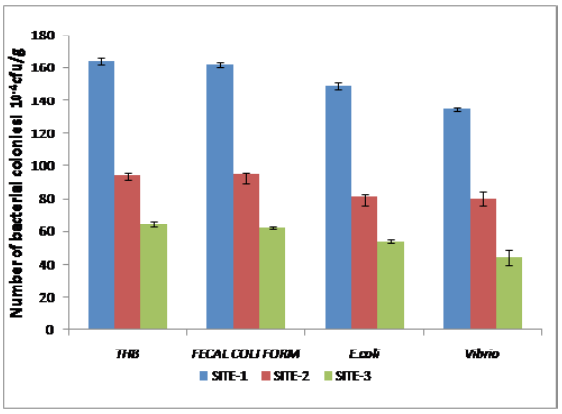

Fig. 2. Range of microbial parameters in sediment samples

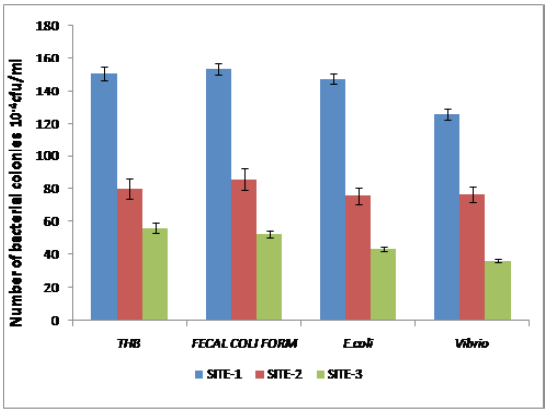

Fig. 3. Range of microbial parameters in water samples

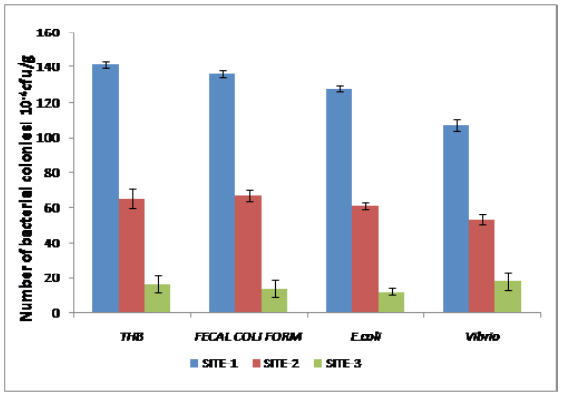

Fig. 4. Range of microbial parameters in seagrass leaf samples

(Metcalf, 1982). The higher densities of THB population (Hatha *et al.*, 2008) and fecal coliform (Shehane *et al.*, 2005) are attributed to the land run off from various sources after the rainfall. The distribution values of heterotrophic bacteria studied in the samples of water, sediment and seagrass leaf were in the order: sediment >water >seagrass leafs; and the site-wise order is: Site1 > Site 2 > Site 3.

Fecal coliform load in the sediment sample of Site 1 ranged from $162x10^{-4}$ cfu/g dry weight to $294x10^{-2}$ cfu/g dry weight; and in the water sample it ranged from $153x10^{-4}$ cfu/ml to 271 $x10^{-2}$ cfu/ml; and in seagrass leaf it ranged from $136x10^{-2}$ cfu/g dry weight to $299x10^{-2}$ cfu/g dry weight. At Site 2, fecal coliform load in sediment ranged from $95x10^{-4}$ cfu/g dry weight to $188x10^{-2}$ cfu/g dry weight; in water it ranged from $86x10^{-4}$ cfu/ml to $164x10^{-2}$cfu/ml; and in seagrass leaf it ranged from $134x10^{-2}$ cfu/g dry weight to $67x10^{-2}$ cfu/g dry weight. At Site 3, fecal coliform load in sediment ranged from $62x10^{-4}$ cfu/g dry weight to $117x10^{-2}$ cfu/g dry weight; in water the range was $53x10^{-4}$ cfu/ml to $105x10^{-2}$cfu/ml; and in seagrass leaf it was from 35 $x10^{-2}$ cfu/g dry weight to $14x10^{-2}$ cfu/g dry weight (Fig.2, 3 and 4). One way ANOVA showed highly significant difference on fecal coliform count ($p< 0.001$) between sites (Table 1).

At Site 1, *E. coli* load in seagrass sediment ranged from $149x10^{-4}$ cfu/g to $292x10^{-2}$ cfu/g (Fig.2); in water, it ranged from $147x10^{-4}$ cfu/ml to 255 $x10^{-2}$ cfu/ml while in seagrass leaf *E. coli* load ranged from $241x10^{-2}$ cfu/g to $128x10^{-2}$ cfu/g (Fig.4). At Site 2, *E. coli* load in sediment ranged from $81x10^{-4}$ cfu/g to $153 x10^{-2}$ cfu/g; in water it ranged from $76x10^{-4}$ cfu/ml to $140x10^{-2}$ cfu/ml while in seagrass leaf *E. coli* load ranged from 113 $x10^{-2}$ cfu/g to 61 $x10^{-2}$ cfu/g. At Site 3, *E. coli* load in sediment ranged from $120x10^{-2}$ cfu/g to 54 $X10^{-4}$ cfu/g; in water, it ranged from 100 $x10^{-2}$ cfu/ml to 43 $X10^{-4}$ cfu /ml while in seagrass leaf, *E. coli* ranged from $38x10^{-2}$ cfu/g to 12 $x10^{-2}$ cfu/g (Fig. 2, 3 and 4). One way ANOVA showed highly significant difference ($p<0.001$) in E. *coli* count between the sites (Table 1).

Fecal coliform bacteria have been used as indicators of the sanitary quality of water for many years (Siva Kumar et al., 1986; Edberg et al., 1989). Of the 3 sites studied, Site 1 recorded the maximum fecal coliform and *E. coli* counts in sediments. At Site 3 the coliform bacteria count recorded was less and this could be because of the diffusion and dilution of sewage in sea water. Laboratory studies (Gerba and McLeod, 1976) showed longer survival of *E. coli* in sand compared to water due to higher levels of organic content. Fecal coliform bacteria survive longer in the bottom deposits as physiologically varied forms when suitable nutrients are supplied (Lopez – Torres et al. 1988). Similar findings of higher bacterial counts from the land source have been made by Hatha *et al.* (2008) and Shehane *et al.* (2005).

At Site 1, *Vibrio* load in seagrass sediment ranged from$134x10^{-4}$ cfu/g to $291x10^{-2}$ cfu/g; in water, it ranged from $125x10^{-4}$ cfu/ml to $257x10^{-2}$ cfu/ml (Fig.3); while in seagrass it ranged from $107x10^{-2}$ cfu/g to $190x10^{-2}$ cfu/g. At Site 2, *Vibrio* in seagrass sediment ranged from 80 $x10^{-4}$ cfu/g as to $187x10^{-2}$ cfu/g; in water it was from $77x10^{-4}$ cfu/ml to $178x10^{-2}$ cfu/ml; in seagrass leaf, *Vibrio* load ranged from $133x10^{-2}$ cfu/g to $53x10^{-2}$ cfu/g. At Site 3, *Vibrio* load in sediment ranged from 44 $x10^{-4}$ cfu/g to $131x10^{-2}$ cfu/g; in water it ranged from $36x10^{-4}$ cfu/ml to $178x10^{-2}$ cfu/ml; while in seagrass leaf, *Vibrio* load ranged from $32x10^{-2}$ cfu/g to $18x10^{-2}$ cfu/g (Fig. 2, 3 and 4). One way ANOVA showed highly significant difference ($p<0.001$) in *Vibrio* count between sites (Table 1).

Baker *et al.* (2010) suggested that *Vibrio* species along with other pathogenic bacteria mostly appear in the warm surface water of the sea and it is also attributed to climate change. In our study *Vibrio*

Table 1. Results of one way ANOVA for microbial parameters

Samples		Sum of Squares	df	Mean Square	F	Sig. P value
THB in water	Between sites	83412.963	2	41706.48	23.083	0
	Within sites	43363.333	24	1806.806		
	Total	126776.296	26			
THB in sediment	Between sites	99985.407	2	49992.7	20.797	0
	Within sites	57691.778	24	2403.824		
	Total	157677.185	26			
THB seagrass leaf	Between sites	149035.63	2	74517.82	44.337	0
	Within sites	40337.556	24	1680.731		
	Total	189373.185	26			
Fecal coli form	Between sites	82624.074	2	41312.04	27.814	0
In water	Within sites	35647.556	24	1485.315		
	Total	118271.63	26			
Fecal coli form	Between sites	85633.852	2	42816.93	23.125	0
Sediment	Within sites	44437.333	24	1851.556		
	Total	130071.185	26			
Fecal coli form	Between sites	150057.556	2	75028.78	36.374	0
Seagrass leaf	Within sites	49504.444	24	2062.685		
	Total	199562	26			
Vibrio in water	Between sites	69938.296	2	34969.15	15.415	0
	Within sites	54443.556	24	2268.481		
	Total	124381.852	26			
Vibrio in sediment	Between sites	78845.407	2	39422.7	14.247	0
	Within sites	66410.444	24	2767.102		
	Total	145255.852	26			
Vibrio in seagrass leaf	Between sites	78408.296	2	39204.15	42.216	0
	Within sites	22287.556	24	928.648		
	Total	100695.852	26			
E.coli in water	Between sites	86725.852	2	43362.93	34.261	0
	Within sites	30376.222	24	1265.676		
	Total	117102.074	26			
E.coli in sediment	Between sites	90104.519	2	45052.26	23.297	0
	Within sites	46412	24	1933.833		
	Total	136516.519	26			
E.coli in seagras sleaf	Between sites	113865.407	2	56932.7	54.325	0
	Within sites	25152.222	24	1048.009		
	Total	139017.63	26			

counts were higher in sediment than in water and seagrass leaf. Bacteria found on seagrass leaves include the genera *Vibrio, Alteromonas, Moraxella, Pseudomonas, Marinobacter* and *Brochothrix* (Kurilenko *et al.*, 2001) as well as several nitrogen fixing bacteria (Pereg et al., 1994). Some microbial pathogens in coastal environments are indigenous to the sea and they include *Vibrio*, while the others such as *Escherichia coli*, *Salmonella* sp., and *Shigella* sp., are introduced through urban surface run off, agricultural discharge, and waste water inflow (Mahalakshmi *et al.*, 2011).

Wahbeh and Mahasneh (1984) reported the difference in heterotrophic association of bacteria seen attached to the leaves, rhizomes and roots of three seagrass species (*Halophila ovalis H. stipulacea* and *Halodule uninervis*) in Jordan. There is great concern for the global decline of the ecologically important seagrass

beds. Epiphytic biofilms of bacteria and algae may contribute to the decline of seagrasses by shading them from the light thereby affecting photosynthesis (Michael et al 2008). Eutrophication and other environmental changes may affect the overall quantity, diversity and species richness of these epiphytes (Balata et al 2008). Because microbial populations can be indicators of biogeochemical conditions, characterization of the epiphytic organisms, particularly bacteria, will provide an insight into the environmental conditions.

Marine pollution has now become a matter of worldwide environmental concern. Continuous discharge of untreated industrial and domestic wastes into sea has been a serious threat to marine habitats and the dependant diversity. The bacterial counts of the samples collected from different sites in Tuticorin indicate that the seagrass ecosystems are disturbed by the discharge of waste water through sewage disposal in Thirespuram region. Hence immediate steps has to be taken for the treatment of the sewage water before letting it into sea, and this is the only way to protect the seagrass ecosystems and the dependant livelihood.

Temperature of water in the present ranged between 30.4 ± 0.10^0C and 30.8 ± 0.06^0C, and it had highly significant difference (P>001) between the three sites (Table 2 and 3). Orton (1920) suggested that sea temperature is the most important environmental factor controlling reproduction in marine invertebrates. Salinity is one of the most important factors which exert various effects on the vitality of marine organisms (Abdo et al 2005). Salinity varied between 31.4 and 33.5 ppt at Site 1; between 34 and 35 ppt at Site 2; while it was 35 ppt at Site 3. The difference in the salinity between the three stations was statistically significant (P >0.001) (Table 2 and 3). Bruckner and Burrows (2005) suggested that the salinity of coastal and offshore environments is influenced by a number of environmental factors such as runoff, precipitation, evaporation, surface current patterns, and upwelling. Earlier, Jayaraman (1954) and Bapat (1955) have also reported such unimodal fluctuation in salinity in Gulf of Mannar. Not much variation was observed in the pH between the three stations and the value ranged between 7.94 ± 0.05 and 8.22 ± 0.06, and the differences were also found to be much significant statistically between the three stations (P >0.001) (Table 2 and3). The DO content was lower at Site 1 and ranging from 1.2 to 1.4 mg/l; at Site 2 it ranged from 3.8 to 4.5 mg/l and at Site 3 from 4.9 to 5 mg/l; the differences were statistically significant between the three sites (P>0.00) (Table 2 and 3). These are comparatively lower values for DO and this is because of sewage mixing in a site (Site 1). A very high statistical difference was also observed in the dissolved oxygen content between the three sites (Santhanam and Venkataramanujam 1996). The temporal variation of DO level depends on temperature, light penetration, rain fall, and climate. Similar findings were reported by Padmini and Kavitha (2003) at Ennore and Kovalam estuaries and Eswari and Ramanibai (2004) at Chennai coast.

The value of turbidity was the highest in Site 1and it ranged from 27.5 to 28.4 NTU; at Site 2 it ranged from 5.65 to 6.85 NTU, and at Site 3 it ranged from 4.6 to 4.7 NTU. The differences were also observed to be statistically significant between the three stations (P >0.001) (Table 2 and 3). Comparatively Site 1 recorded less light penetration than the other sites. Turbidity value in marine environment is mainly influenced by wave action, tides, wind and fresh water inflow and sewage discharges. Easterson *et al.,* (2000) have reported turbidity values between 13.4 to 45.6 NTU in Tuticorin coastal waters which are similar to the estimates of the present study. The values of total suspended solid level in Sites 1, 2 and 3 were 265, 93 and 77 mg/l respectively and the differences were observed to be statistically significant between the stations (P >0.001) (Table 2 and 3). The physical factors such as temperature, salinity, waves, currents, depth, nature of substrate and day length regulate the physiological activity of seagrass; and natural phenomena such as light, nutrients, epiphytes and diseases limit the photosynthetic activity of the plants; and anthropogenic inputs such as nutrient and sediment loading inhibit the access to available plant resources (Twilley et al.,1985; Wetzel and Neckles, 1986; Dennison et al., 1993; Abal and Dennison, 1996).

On comparison, Site 1 had higher concentrations of phosphate, magnesium, nitrite and silicate which could cause adverse effects on seagrass diversity. The maximum phosphate concentration in Sites 1, 2 and

Table 2. water quality parameters in the study sites

Water quality parameter	SITE-1	SITE-2	SITE-3
Temperature ° C	30.8 ± 0.06	30.4 ± .0.1	30.8 ± 0.03
Salinity (ppt)	32.4 ± 0.61	34.3 ± 0.3	34.7 ± 0.33
PH	8.2 ± 0.06	8.03 ± 0.04	7.9 ± 0.05
Turbitity (NTU)	27.9 ± 0.26	6.44 ± 0.4	4.65 ± 0.03
TSS (mg/l)	265.3 ± 5.05	92.6 ± 3.9	77.3 ± 3.93
DO (mg/l)	1.26 ± 0.07	4.16 ± 0.2	4.9 ± 0.03
Calcium (mg/l)	391.6± 4.41	410 ± 5.8	452 ± 6.02
Magnesium (mg/l)	1250 ± 8.67	1203.6 ± 39.6	1271 ± 10.14
Nitrates (¼g/L)	6.83 ± 0.36	1.70 ± 0.1	1.09 ± 0.04
Nitrites (¼g/L)	22.5 ± 1.27	0.69 ± 0.1	0.24 ± 0.03
Phosphate (¼g/L)	5.34 ± 0.06	3.01 ± 0.2	0.063 ± 0.01
Silicate (¼g/L)	14.25 ± 0.58	3.85 ± 0.1	1.82 ± 0.10

3 were 5.34mg/l, 3.01mg/l and 0.06mg/l respectively. ANOVA showed significant difference in phosphate levels between the sites. Asha *et al.,* (2009) have recorded in Gulf of Mannar levels of phosphates higher than in the present study. The maximum silicate concentrations in Sites 1, 2 and 3 were 15.26, 3.35 and 1.83 mg/l respectively. A significant difference was also observed in the silicate content between the three sites. The comparatively high level of silicate in Site 1 may be due to freshwater inflow. The high silicate concentration may also be due to heavy load of sewage. Jansi et al., (2009) have recorded higher concentration of silicate in Manakudi estuary than in the present investigation.

The maximum nitrite concentrations in Sites 1, 2 and 3 were 24.35, 0.89 and 0.28 mg/l respectively; and the differences were also statistically significant between the stations (P>0.00) (Table 1). The enrichment of nitrites and nitrates could be attributed to various factors. The unpolluted waters have nominal quantity of nitrates (Jaji *et al.,* 2007). The maximum nitrate concentrations in Sites 1, 2 and 3 were 7.25, 1.86 and 1.14 mg/l respectively; and the differences were statistically significant between the stations (P >0.001) (Table 2 and 3). According to Short *et al*. (1995) long-term nitrate additions cause severe decline of seagrass, and enriched levels of ammonia and phosphate lead to reduction in shoot density and biomass. The maximum magnesium levels in Sites 1, 2 and 3 were 1250, 1204 and 1271mg/l respectively, and the difference between the sites were not statistically significant. (P>0.001) (Table.1). The maximum calcium level was 452 mg/l in Site 3, 410 mg/l in Site 2, 392 mg/l in Site 1. The differences were statistically significant between the three stations (P >0.001) (Table 2 and 3).

Nutrients stimulate bacterial growth in water systems (Patrick et al 2012). Thirespuram water has high nutrient load (nitrate, nitrite, phosphate and silicate) which could be the factor that facilitates the large presence of bacteria in water and sediment. Large populations of coliforms have been reported from Andhra coast, because of high the amounts of sulphate, nitrate and ammonium ions (Swamy *et al.,* 2006). Sediment sample had more number of fecal coliform than water samples because of its higher capacity for binding with organic matter and nutrients that easily attract the pathogenic organism especially fecal coliform bacteria. Earlier studies in India too have reported higher concentration of faecal coliform bacteria in sediment than in water (Vaidya *et al.,* 2001; Mohandass and Bharathi, 2003).

Globally, it has been accepted that nutrient loading has been the most detrimental factor for seagrass decline (Carruthers et al., 2006). The other causes of seagrass degradation are sewage enrichment (Johansson and Lewis, 1992); enrichment of groundwater supplies (Short et al., 1993); and runoff from agricultural lands (Philips and Menez, 1988). Impact of sewage fallout on macro benthic diversity at Long reef and Bungan Head was studied by Roberts (1996). Tuticorin is an industrial town with a large population and it also has a seaport. The town generates an estimated 17.5 MLD of sewage annually,

Table 3. Results of one way ANOVA for water quality parameters

		Sum of Squares	df	Mean Square	F	Sig.
Temperature	Between Sites	0.202	2	0.101	11.375	0.009
	Within Sites	0.053	6	0.009		
	Total	0.256	8			
Salinity	Between Sites	11.056	2	5.528	7.37	0.024
	Within Sites	4.5	6	0.75		
	Total	15.556	8			
PH	Between Sites	0.122	2	0.061	8.869	0.016
	Within Sites	0.041	6	0.007		
	Total	0.163	8			
Turbitity	Between Sites	1010.268	2	505.134	2.25E+03	0
	Within Sites	1.348	6	0.225		
	Total	1011.617	8			
Tss	Between Sites	65392.89	2	32696.444	580.41	0
	Within Sites	338	6	56.333		
	Total	65730.89	8			
DO	Between Sites	22.442	2	11.221	240.452	0
	Within Sites	0.28	6	0.047		
	Total	22.722	8			
Calcium	Between Sites	5672.222	2	2836.111	31.906	0.001
	Within Sites	533.333	6	88.889		
	Total	6205.556	8			
Magnesium	Between Sites	7092.167	2	3546.083	2.037	0.211
	Within Sites	10445.83	6	1740.972		
	Total	17538	8			
Nitrates	Between Sites	59.667	2	29.833	219.668	0
	Within Sites	0.815	6	0.136		
	Total	60.482	8			
Nitrites	Between Sites	978.155	2	489.078	302.242	0
	Within Sites	9.709	6	1.618		
	Total	987.864	8			
Phosphate	Between Sites	42.002	2	21.001	576.418	0
	Within Groups	0.219	6	0.036		
	Total	42.22	8			
Silicate	Between Sites	275.157	2	137.579	378.31	0
	Within Sites	2.182	6	0.364		
	Total	277.339	8			

and there are no facilities for the treatment of sewage; all the sewage is disposed off in canals that eventually reach the sea. Industries around Tuticorin include refinery, aquaculture, chemicals and fertilizers, caustic soda and thermal power plants. The total volume of waste discharge from these industries, other than aquaculture, is about 10.4 MLD The effluent from these industries contain suspended solids, ammonia, nitrate, BOD compounds, oil and grease, and trace quantities of heavy metals such as chromium (Asha et al 2007). Since the 1940s the total area of seagrass has declined by more than thirty percent, and the loss rate per year in recent times is about 7% (Nellemann and Fonseca, 2009). This is largely due to the fact that seagrasses are a shoreline benthic species subject to degradation in response to natural and human activity (Caccia et al., 2007). Growing populations, fisheries, and tourism affect the shoreline benthic ecosystems. It is especially the growing populations and the waste associated with it that affect the nutrient loading of seagrass beds and other marine ecosystems. Site 2 and 3 of the present study are comparatively less affected because they occur farther from sewage pollution.

The present investigation brings to light the higher abundance of THB and pathogenic bacteria in the

water, sediment and seagrass samples of Site 1 than in the other two sites, which is due to the influx of untreated sewage into sea at Site 1. THB and pathogenic bacteria load were higher in sediments than in the water samples. This is due partly to the rich organic content of the sediment and partly to the lesser residential time of the microorganisms in the water column than the sediments. Humans have resided in close association with Site 1 for the past many years. A continuous monitoring of the marine environment (water and sediments) is necessary to detect the presence of THB and pathogenic bacteria. This will directly improve the hygiene of the coastal people, and will also help in creating awareness on health management in them. The likely impact of increased sewage pollution at Site 1 warrants immediate research attention. Installation of proper sewage treatment plant, draining of treated industrial effluence and creation of public awareness about the pollution are the solutions to protect the marine habitats of Tuticorin region.

ACKNOWLEDGEMENT

The authors are thankful to Ministry of Environment, Forests and Climate Change, Government of India; to the Principal Chief Conservator of Forests and to the Chief Wildlife Warden, Government of Tamil Nadu; to the Wildlife Warden, Gulf of Mannar Marine National Park for research permissions; and to the authorities of Suganthi Devadason Marine Research Institute for the research equipments and other facilities.

REFERENCES

Abal, E.G. and Dennison W.C.1996. Seagrass depth range and water quality in Southern Moreton Bay: Queensland, Australia. Mar Freshwater Res., 47:763–771

Abdo, M.H. 2005. Physicochemical characteristics of AbuZa'baal ponds, Egypt: Egyptian J. of. Aqua. Research.,31:(2)2-3

APHA., 2015.Compendium of methods for the microbiological examination of foods, 5 th Ed. Lloyd M, Jeffrey LK (Eds.) Washington

Asha, P.S. and Diwakar K. 2007. Hydrobiology of the inshore waters off Tuticorin in the Gulf of Mannar : J.Mar. Biol. Ass India., 49: (1)07-11

Azam, F., Fenchel, T., Field, J.G, Gray,T.S, Meyer-Reil, L.A and Thingsted, F.1983. The ecological role of water column microbes in the sea. *Mar. Ecol. Prog. Ser*., 10: 257-263

Baker-Austin, C., Stockley, L., Rangdale, R. and Martinez-Urtaza J. 2010. Environmental occurrence and clinical impact of *Vibrio vulnificus* and *Vibrio parahaemolyticus*: a European perspective. *Environ. Microbiol Rep*., 2(1): 7-18

Balata,D.A., Bertocci B.L., Piazzi L.P., Nesti B.U. 2008. Comparison between epiphyte assemblages of leaves and rhizomes of the seagrass Posidonia oceanica subjected to different levels of anthropogenic eutrophication: Estuarine, Coastal and Shelf Science., 79 : 533–540

Bapat, S.V. 1955. A preliminary study of the pelagic fish eggs and larvae of the Gulf of Mannar and Palk Bay.Indian J. Fish., 2: 231-255

Bruckner, A. and Burrows, F.M. 2005. Restoration moniring of coral reefs. Chapter3 in Science based restoration monitoring of coastal habitats, Volume:2, Tools for monitoring coastal habiotats (eds) Thayer, G.W. McTigue, T.A., Salz, R.J.Merkey, D.H.Burrows, F.M.and Gayaldo, P.F., p3.1-3.72

Caccia, M.2007. Vision-based ROV horizontal motion control: near-seafloor experimental results. Control Engineering Practice., 15(6):703–714

Carruthers, T.J.B., Dennison,W.C., Kendrick, G.A., Waycott, M., Walker,D.I. and Cambridge, M.L. 2007. "Seagrasses of south-west Australia: A conceptual synthesis of the world's most diverse and extensive seagrass meadows: "Journal of Experimental Biology and Ecology., 350: 21-45

Correll, D. L. 1998. The role of phosphorus and eutrophication of receiving waters: A review. J. Environ. Qual., 27: 261-266

d'Avack E.A.S., Tillin, H., Jackson, E.L. and Tyler-Walters, H. 2014. Assessing the sensitivity of seagrass bed biotopes to pressures associated with marine activities. JNCC Report No. 505 Joint Nature Conservation Committe, Peterborough. Available from www. Marlin .ac.uk/puplications

Dennison, W.C and Kirkman, H. 1996. Seagrass survival model. In: J.Kuo, R. C. Phillips, D. I. Walker and K. Kirkman (Eds.), Proceedings of an International Workshop on Seagrass Biology, Rottnest Island, Western Australia., P: 341-344

Dennison, W.C., Orth, R.J., Moore, K.A., Stevenson, J.C., Carter, V., Kollar, S., Bergstrom, P.W., Batiuk, R.A. 1993. Assessing water quality with submersed aquatic vegetation: Bio Sci .,43: 86–94

Easterson, D.C.V., Asha, P.S. and Selvaraj, M. 2000. Effect of Thermal power plant effluent on the hydrological conditions in the Tuticorin Bay: J. Mar. Biol. Ass.

India., 42: 135 - 138

Edberg, S.C., Allen, M.J., Smith, D.B. 1989. National field evaluation of a defined substrate method for the simultaneous detection of total coliforms and Escherichia coli from drinking water. Comparison with presence absence techniques: Appl. Environ. Microbiol., 55: 1003-1008

Erftemeijer, P., Djunarlin, and Moka, W. 1993. Stomach content-analysis of a dugong (*Dugong dugon*) from south Sulawesi, Indonesia: Australian Journal of Marine and Freshwater Research., *44*: 229-233

Eswari, Y.N.K. and Ramanibai, R. 2002. Distribution and abundance of phytoplankton in the estuarine waters of Chennai, southeast coast of India: J. Mar. Biol. Ass. India., 44, 205-211(2002).

Gerba,C.P. and John,S.M. 1976.Effect of sediments on the survival of *Escherichia coli* in marine waters: App. Environ. Microbiol July., P. 114-120

Hatha, M., chandran, A. and Varghese, S. 2008. Increased prevalence of indicator and pathogenic bacteria in the Kumarankom Lake: A function of salt water regulator in Vembandu Lake, A Ramsar site, along west coast of India. The 12th World Lake Conference. pp. 250-256

Jaji, M.O., Bamgbose, O., Odukoya, O.O. and Arowlo, T.A. 2007. Water quality assessment of Ogun River, south southwest Nigeria: Environ. Monit. Assess., 133 (1-3) : 447-482.

Jansi, M. and Ramadhas, V. 2009. Effect of salinity and dissolved nutrients on the occurrence of some seaweeds in Manakkudy estuary: Indian Journal of Marine Sciences., 38(4) : 470-473

Jayanth, G., Jeyasekaran, G. and Jeya Shakila, R. 2001. Isolation of marine bacteria, antagonistic to human pathogens: Indian J. Mar. Sci., 31(1): 39-44

Jeyaraman, R. 1954. Seasonal variations in salinity, dissolved oxygen and nutrient salts in the inshore waters of the Gulf of Mannar and Palk bay near Mandapam (S.India): *Indian* J. Fish., pp: 345-364

Johansson, J.O.R. and Lewis, R.R. 1992. Recent improvements of water quality and biological indicators in Hillsboro Bay, a highly impacted subdivision of Tampa Bay, Florida, USA: Science Total Environment Supplement., p.1199-1215

Kannan, R. and Kannan, L. 1996. Physico-chemical characteristics of sea weed beds of the Palk Bay, Southeast coast of India: Ind. J. M ar. Sci., 25: 358-362

Karuppasamy, P.K. and Perumal, P.2000. Biodiversi ty of zooplankton at Pichavaram mangroves, South India: Adv. Biosci., 19, 23-32

Kurilenko, V.V., Ivanova, E.P. and Mikhailov, V.V. 2001. Zonal distribution of epiphytic microorganisms on the eelgrass Zostera marina: Microbiology., 07: 372–373

Mahalakshmi, M., Srinivasan, M., Murugan, M., Balakrishnan, S. and Devanathan, K. 2011. Isolation and identification of total heterotrophic bacteria and human pathogens in water and sediment from Cuddalore fishing Harbour after the Tsunami: *Asian. J. Boil. Sc.*, 1-9

Mary Helen, H., Premjith, S. and Jaya, D.S. 2014. Bacteriological studies on water, sediment and fish samples of poovar estuary, south india: *Journal of Aquatic Biology and Fisheries* ., 2: 337 to 343

Mathews, G. 2007.Studies on the coral reef restoration in Tuticorin region of the Gulf of Mannar, South east coast of India. Ph.D thesis. Manonmaniam Sundaranar University, Tirunelveli., p 310

Metcalf, T.G.1982. Virus in shellfish-growing waters: Environ. Int., 7(1): 21-27.

Michael, S.T., Shin, H.W., Hanna,R. and Spafford, C.S. 2008. A review of epiphyte community development: Surface interactions and settlement on seagrass: J. of .Env. Biology., 29(4) 629-638

Mohandass, C. and Bharathi, L.P.A. 2003 Representation, dispersion and variation of bacteria indicators in the coasta waters of nagore (east coast of india): Water Environ. Res., 75: 66-72

Mudryk, Z.J. 2005. Occurrence and distribution antibiotic resistance of heterotrophic bacteria isolated from a marine beach: Mar. Pollut. Bull., 50: 80-86

Muehlstein, L.K. 1989. Perspectives on the wasting disease of eelgrass Zostera marina: Dis. Aquat. Orgs.,7:2ll-221.

Nellemann, C., Corcoran, E., Duarte, C.M., Valdés, L., DeYoung, C. and Fonseca., L . 2009. Grimsditch., G. (Eds) Blue Carbon. A Rapid Response Assessment,United Nations Environment Programme: GRID-Arendal, www.grida.no, 80

Orton, J.H. 1920 Sea-temperature, breeding and distribution in marine animals: J mar biol Ass., 12:339-366

Padmini, E., and M. Kavitha. 2003. Seasonal Pollution Assessment through comparative hydrobiological studies in Ennore and Kovalam estuaries: Indian Hydrobiology., Vol. 6, 139-144

Patrick, A.S, Roland, A., Chris A., Samuel B.D. 2012. Nutrient-Induced Growth of Coliform and HPC Bacteria in Drinking-Water Pipes: Journal of Environmental Protection.,3:508-517

Phillips, R.C. and Menez, E.G. 1988. Seagrasses. Smithsonian Contribution to the Marine Sciences: Washington, D.C., Smithsonian Institute Press. 34: 1-104

Rajasegar, M. 2003. Physico-chemical characteristics of the Vellar estuary in relation to shrimp farming: J.

Environmental Biol., 24: 95-101

Roberts, D.E. 1996. Effect of the North head deep water sewage outfall on Near shore coastal reef macrobenthic assemblages: Marine Poll Bull., 33(7- 12): 303-308

Santhanam, R. and Venkataramanujam, V. 1996. Impact of industrial pollution and human activities on the coral resources of Tuticorin (South India) and method for conservation: In: Proceedings of Symposium of 8 thInternational Coral Reef, Panama, 176pp.

Shehane, S.D., Harwood, V.J., Whitlock, J.E. and Rose, J. B. 2005. The influence of rainfall on the incidence of microbial fecal indicators and the dominant sources of fecal pollution in a Florida river: J. *Appl.*Microbiol., 98: 1127-1136

Short, F.T. and Wyllie-Echeverria, S. 1996. Natural and human-induced disturbances of seagrass: Environ. Conserv., 23: 17–27

Short, F.T., Burdick, D.M and Kaldy, J.E. 1995. Mesocosm experiments quantify the effects of eutrophication on eelgrass, Z & era matins: Limnol. Oceanogr., 40:740-749

Short, F.T., Montgomery, J., Zimmermann, C.F., Short, C.A. 1993. Production and nutrient dynamics of a *Syringodium filiforme* Kuetz. seagrass bed in Indian RiverLagoon, Florida: *Estuaries.*, 16, 323–334

Siva Kumar, V., Thangaraj, G.S., Chandran, R., Lakshman Perumalsamy, P., and Ramamoorthy, K.1986. Studies on faecel coliform in an Esturine environment of Porto Novo. Proc. Symp: Coastal Aqua culture., 4: 1330 - 1338

Swamy, Y.V., Roy Chaudhary, G., Das, S.N. and Muduli, R. 2006. Assesment of water quality in tsunami affected Andhra coast: Current Sciences., 91: 1409-1412

Twilley, R.R., Kemp, W.M., Staver, K.W., Stevenson, J.C., Boyton, W.R. 1985. Nutrient enrichment of estuarine submersed vascular plant communities. Algal growth and effects on production of plants and associated communities: Mar. Ecol. Prog. Ser. 23, 179–191.

Vaidya, S.Y., Vala, A.K. and Dube, H.C.2001. Bacterial indicators of faecal pollution at Bhavnagar coast: Ind. J. Microbiol., 41, 37-39

Wahbeh, M.I. and Mahasneh, A.M. 1984. Heterotrophic bacteria attached to leaves, rhizomes and roots of three seagrass species from Aqaba (Jordan): Aquat. Bot. 20:87–96.

Wetzel, R.L. and Neckles, H.A. 1986. A model of Zostera marina L. photosynthesis andgrowth: simulated effects of selected physical– chemical variables and biological interactions: *Aqua. Bot.*, 26, 307–323