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BACTERIOLOGICAL STUDIES ON WATER, SEDIMENT AND FISH SAMPLES OF POOVAR ESTUARY, SOUTH INDIA

Mary Helen, H[1]*., Premjith, S[2]. and Jaya, D.S[3].
[1]*P.G. and R research Department of Chemistry, S.T.Hindu College, Nagercoil, K.K. District, Tamilnadu, India.*
[2]*Department of Zoology, St.Judes College, Thoothoor, K.K, District, Tamilnadu, India.*
[3]*Department of Environmental Sciences, University of Kerala, Kariavattom, Thiruvananthapuram, Kerala, India.PIN-695581*
**Email: helencolombus@gmail.com*

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Abstract: The present investigation deals with the bacteriological analysis of water, sediment and fish samples of Poovar estuary of Kerala coast, South India during the period 2004 – 2005. The major objectives of the study were to analyse the incidence of microbial population in water, sediment and commercially important fishes collected from the study stations and to suggest the management measures to reduce the pollution load of the water bodies in Poovar-Vizhinjam area which is always under the influence of tourism. The water, sediment and fish samples were collected from three selected sampling stations of the estuary in the premonsoon, monsoon and post monsoon seasons of the study period. The fish samples (*Mugil cephalus*) were collected using a cast net and the skin, gill and intestine of fish samples were refrigerated for microbiological analysis. The bacteriological parameters were determined using the standard microbiological techniques. The bacterial strains isolated from water, sediment and fish samples were identified up to the generic level by employing the scheme of Gunasekaran (1995) and Holt *et al*. (1961). The present study showed that bacterial population in sediment was found to be higher than in fish and water. The distribution of heterotrophic bacteria studied in water, sediment and fish samples were in the order: sediment >water >fish. The maximum bacterial count was recorded during pre monsoon season and minimum during monsoon season. The important bacterial genera encountered were *Escherichia coli*, *Salmonella* and *Shigella*. Among the identified bacterial genera, *Escherichia coli* were found to be the dominant one followed by *Salmonella* and *Shigella*. The coliform bacteria of the estuary may affect the fish production and deteriorates the quality of water in the estuary. The data obtained from this study could therefore serve as base line information for future environmental impact assessment of the effluents discharged from tourist resorts in this region. The bacterial diversities of samples collected from different stations in Poovar estuary indicates that the estuary and ecosystems are highly disturbed by waste water discharge through sewage disposal, beach resort effluents and retting activities. Man requires a clean ecologically well balanced environment to promote healthy living. It is the responsibility of the society to ensure minimum damage to the water resources.

Key words: Estuary, Environment, Bacterial strain, Fish, Sediment, Tourist resorts

INTRODUCTION

Most of India's finest beaches are in Kerala. The eco-tourism traffic to Kerala shows an increasing trend over the past few years due to natural geographic advantages. The total tourism inflow to Kerala is about 50 lakhs during 1997 and which 13% is more than the previous year. Foreign tourist arrival is about 1.82 lakhs. Kerala Government is working on a strategy to increase the foreign tourist inflow to the tune of 5 lakhs by the turn of the century. The earnings from tourism industry in Kerala are roughly around Rs. 250 crores at present. Kerala has 12 wildlife sanctuaries and two national parks. These may form base for planning ecotourism activities. Apart from these, the rich biological diversity can be an added advantage.

Thiruvananathapuram district in Kerala state is predominantly an agricultural land. The river Neyyar that originates in the Western Ghats at an altitude of 1500 m above Mean Sea Level (MSL) is one of the largest rivers and mixes with Arabian Sea, forming an estuary at Poovar. Many tourist resorts exist on the northern side of the estuary. The drainage channels from the tourist resorts, paddy field and AVM Canal (Ananda Victoria Marthandavarma Canal) are the main sources for the flow of sewage and

domestic wastes into the estuary. The villagers in and around the locality are using this estuary for their day to day activities. Coconut farms fringe the estuary. AVM canal water is used for retting activities and for washing purposes round the year. Thus the estuarine environment of Poovar estuary is subjected to pollution through sewage disposal, waste water discharge and retting activity.

Study area

The present study was carried out in the coastal waters with special reference to Poovar estuary adjacent to the beach resorts of Poovar – Vizhinjam area in Thiruvananthpuram District, Kerala State. Poovar estuary is a bar built estuary of area 30.93 hectare and length 15 km and it is situated between N. Lat. 8° 18' 32" to 8° 18' 6" and E. Long. 77° 4' 32" to 77° 5' 14", and is formed by the confluence of river Neyyar. Poovar-Vizhinjam area is occupied by 10 villages. The villages are Poovar, Karumkulam, Kochuthura, Puthiyathura, Pallam, Pulluvila, Adimalathura, Chowara, Vizhinjam North and Vizhinjam South. The nearest village to Poovar estuary is Poovar (Statistical Cell Report, 2007). The estuarine environment of Poovar estuary is subjected to pollution through different anthropogenic activities like sewage disposal, coir retting and beach resort effluents. The location map of the study area is given in Fig. 1.

MATERIALS AND METHODS

The present study was carried out in the Poovar estuary over a period of twelve months from October 2004 to September 2005, covering pre-monsoon (February – May), the monsoon (June – September) and post monsoon (October – January) seasons. Three stations were selected for the present study. Station I: Poovar Bar mouth, the Pozhimugam; Station II: Poovar-Bridge and Station III: Mavalakadavu.

The collection of samples was done between 6 a.m. to 8 a.m. The surface and bottom water samples were collected separately in sterile glass bottles and the sediment samples were collected in sterile polythene bags and brought to the laboratory in ice box for bacteriological analysis. The fin fish species, *Mugil cephalus* were collected by cast net. Skin, gill, intestine and muscle of fish samples were separated and refrigerated for microbiological analysis. The isolates were maintained on seawater nutrient agar slants and identified up to generic level using the method adopted by Surendran and Gopakumar (1981); Oliver (1982).

RESULTS AND DISCUSSION

Variations of total viable count in Surface water samples: The monthly and seasonal variation of total viable count (TVC) in surface and bottom water samples is given in Table 1.

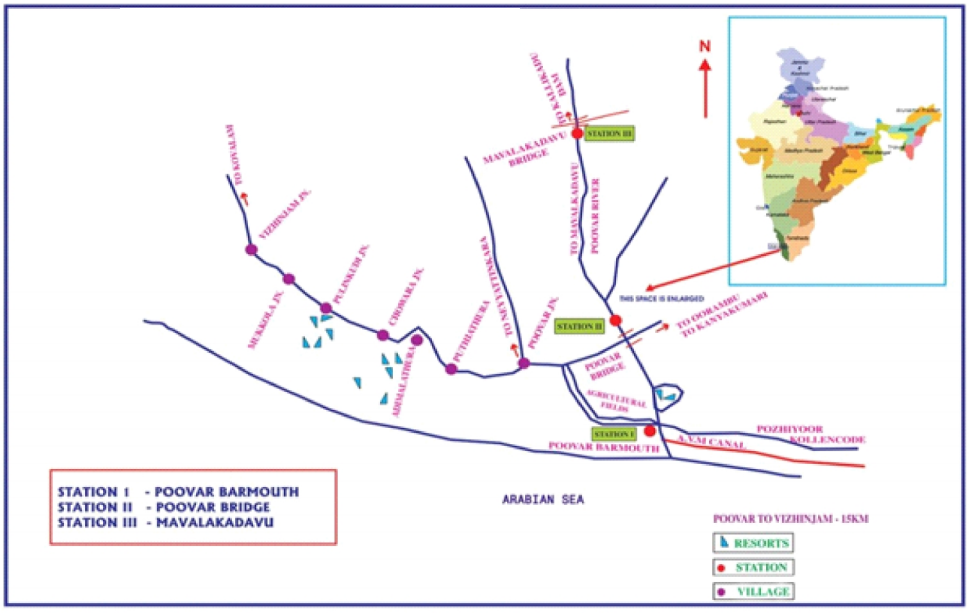

Fig. 1. Study area showing sampling location

Table 1. Monthly distribution (TVC x 10^5 CFU/ml) of total heterotrophic bacterial count in the water samples of Poovar estuary

Station	Samples	Post monsoon				Pre monsoon				Monsoon			
		Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
I	Surface water	30.26	32.10	30.40	31.60	34.20	29.30	35.60	31.43	22.00	28.16	27.83	24.66
	Bottom water	27.20	30.40	28.10	29.13	32.60	23.41	26.03	31.76	17.46	26.33	22.26	22.00
II	Surface water	22.40	20.32	23.20	25.46	27.40	24.60	23.20	26.00	17.30	19.10	21.66	16.37
	Bottom water	18.30	20.32	19.40	20.60	23.14	19.90	20.70	25.43	13.40	17.67	18.00	12.36
III	Surface water	16.60	18.20	16.40	11.20	21.70	14.60	18.80	21.66	11.53	13.43	12.46	10.16
	Bottom water	16.10	19.20	15.34	9.10	20.50	12.10	18.20	20.60	10.20	11.80	12.40	10.10

The monthly bacterial counts ranged from 10.16 to 35.60 x 10^5 CFU/ml and maximum bacterial count was recorded during April at station I and minimum value was recorded during September at station III. Seasonal variations in the pooled data showed the maximum bacterial counts of 32.63 x 10^5 CFU/ml which was recorded during pre monsoon season at station I and minimum of 11.90 x 10^5 CFU/ml which was recorded during monsoon season at station III.

Variations of total viable count in Bottom water samples: The maximum TVC value was observed during February at station I (32.60 x 10^5 CFU/ml) and minimum value was observed during January at station III (9.10 x 10^5 CFU/ml). The maximum bacterial counts of 28.71 x 10^5 CFU/ml recorded during post monsoon season at station I and the minimum of 11.13 x 10^5 CFU/ml recorded during monsoon season at station III.

Variations of bacterial strains in Surface water: Data on monthly and seasonal variation of bacterial strains in surface and bottom water samples is presented in Table 2. Among the total bacterial strains, a maximum of 125 CFU/ml strains were observed in April at station I and a minimum of 38 CFU/ml strains were observed in September at station III. The bacterial strain

Table 2. Total bacterial strains (CFU/ml) isolated from the water samples of Poovar Estuary

Station	Samples	Post monsoon				Pre monsoon				Monsoon			
		Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
I	Surface water	97	103	95	91	104	116	125	114	109	86	98	84
	Bottom water	81	92	83	96	110	103	112	106	97	91	93	71
II	Surface water	66	74	66	86	70	77	74	88	71	60	55	62
	Bottom water	71	68	58	74	63	72	81	90	65	51	69	53
III	Surface water	60	63	51	55	67	50	64	49	48	40	46	38
	Bottom water	67	58	25	47	64	59	56	61	41	32	55	30

was maximum during premonsoon season at station-I (114.75 CFU/ml) and minimum during monsoon season at station-III (43.00 CFU/ml).

Variations of bacterial strains in Bottom water: The highest bacterial strain was detected during April at station I (112 CFU/ml) and lowest was detected during December at station III (25 CFU/ml). The present study reveals that the total bacterial strain was found to be highest in pre monsoon season at station I (107.75 CFU/ml) and lowest in monsoon season at station III (39.50 CFU/ml). Microorganisms like bacteria were widely distributed in the water, sediment and fish samples and they are playing a vital role in aquatic ecosystem. The interactions of the microorganisms with the aquatic biota are unique and diverse. Sediment and water are excellent culture media for the growth of many types of microorganisms. The majority of the bacterial population was found in surface level and their number decreases with depth.

In the present study, the data on the seasonal variations showed maximum bacterial count during premonsoon season and minimum during monsoon season. Similar results were also observed by Prema (2000) in Rajakka-mangalam estuary in Tamil Nadu. The bacterial counts recorded in the present study were of similar magnitudes as given by Zobell (1948) in southern California coast, Civic (1955) in the Adriatic Sea and Kriss (1961) in the Black sea. High bacterial count was recorded during summer by Brown (1964) in the waters of Sydney. Chandrika and Nair (1994) reported maximum counts in the Cochin water during premonsoon and post monsoon. Iriberri *et al.* (1987) noticed higher counts during hot season and lower counts during the cold season. The microorganisms living in particular water and favourable conditions multiply quickly in summer season (Reheinheimer, 1965).

Variations of total viable count in Sediment samples: The monthly and seasonal variation of TVC in sediment samples are presented in Table 3. The value ranged between 18.87 x 10^5 CFU/g and 38.46 x 10^5 CFU/g dry sediment. The maximum value was observed in May at station I and minimum value in August at station III. Seasonal variations showed the maximum bacterial counts of 35.51 x 10^5 CFU/g dry sediment during monsoon season at station I and the minimum of 20.90 x 10^5 CFU/g dry sediment during monsoon season at station III.

Variations of bacterial strains in Sediment samples: The data on bacterial strains recorded in the sediment samples collected from the experimental zones of Poovar estuary is presented in Table 4. A maximum of 135 CFU/g strains were isolated and identified at station I in April and a minimum of 42 CFU/g bacterial

Table 3. Monthly distribution of total heterotrophic bacterial count (TVC x 10^5 CFU/g) in the sediment samples of Poovar estuary

Station	Post monsoon				Pre monsoon				Monsoon			
	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
I	34.27	36.07	33.14	30.46	36.17	35.68	31.72	38.46	37.19	29.33	26.62	28.27
II	31.96	30.63	32.94	28.35	33.63	30.91	28.87	34.08	32.36	28.70	24.45	26.16
III	23.46	26.32	24.62	20.27	27.45	23.87	20.94	28.44	22.39	20.62	18.87	21.70

Table 4. Total bacterial strains (CFU/g) isolated from the sediment samples of Poovar estuary

Station	Post monsoon				Pre monsoon				Monsoon			
	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
I	110	117	124	115	103	120	135	128	97	114	106	95
II	75	84	78	96	84	92	96	113	84	76	67	71
III	67	74	61	65	78	67	74	61	59	42	58	62

strains were identified at station III in July. The highest value of sediment bacterial strain was observed from the study area was 121.50 CFU/g during pre monsoon season at station I and lowest value observed was 52.55 CFU/g during monsoon season at station III.

The total viable heterotrophic bacterial content in an area depends on the availability of growth supporting organic matter and micronutrients (Sayler *et al.*, 1975). Due to the abundance of micronutrients and organic matter, high counts of total heterotrophic bacteria were detected in sediment samples. High correlation between bacterial counts and temperature supported the fact that the distribution of nutrients and temperature played a dominant role in the distribution of heterotrophic bacteria. A positive correlation between temperature and heterotrophic bacterial distribution was noted by Nedwell and Floodgate (1971) in intertidal sediment. In the present study also, there was positive correlation between temperature and heterotrophic bacterial population.

In Poovar estuary, high bacterial count was observed during premonsoon season at station I. The total heterotrophic bacterial density was found to be higher in the sediment than that of water. Total heterotrophic bacterial population increased probably due to the continuous availability of substrate, nutrients as unconsumed feed, plankton and other organic and inorganic matters in the bottom of the water. In Poovar estuary, the data on the seasonal variations showed maximum bacterial count during pre monsoon season and minimum during monsoon season. Anand *et al.* (1996) noticed an increasing trend regarding the total heterotrophic bacterial content in estuarine. The total heterotrophic bacterial count observed was in the range from 1.82 x 10^6 to 4.72 x 10^6 CFU/g (Sharmila *et al.,* 1996) and from 9.30 x 10^5 to 31 x 10^7 CFU/g (Anand *et al.*, 1996). Simillar range (10^5 CFU/g) was obtained in the present study also.

Variations of total viable count in Fishes: The result on total viable count (TVC) in fish samples (Mugil *cephalus)* of Poovar estuary ranged from 18.40 x 10^5 CFU/g to 34.10 x 10^5 CFU/g. The maximum value was noted during April at station I and minimum value was detected during September at station III (Table 5). The average bacterial counts recorded with the maximum of 30.58 x 10^5 CFU/g during premonsoon season at station I and a minimum of 20.92 x 10^5 CFU/g during monsoon season at station III.

Variations of bacterial strains in Fish (*Mugil Cephalus*): The monthly and seasonal changes of bacterial strains in fish samples are presented in Table 6. A maximum of 120 CFU/g strains were isolated and identified in the fish samples

Table 5. Monthly distribution of total heterotrophic bacterial count (TVC x 10^5 CFU/g) in fish samples of Poovar estuary

Station	Post monsoon				Pre monsoon				Monsoon			
	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
I	28.13	30.26	26.20	23.40	27.60	29.20	34.10	31.40	25.64	27.45	21.17	20.60
II	25.40	23.72	28.40	21.34	29.78	20.54	28.44	30.07	20.67	24.91	19.47	21.32
III	20.31	24.45	22.74	23.10	21.71	24.60	29.80	26.54	19.62	26.04	19.60	18.40

Table 6 . Total bacterial strains (CFU/g) isolated from the fish samples of Poovar estuary

Station	Post monsoon				Pre monsoon				Monsoon			
	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
I	94	98	116	101	84	110	120	114	93	81	91	86
II	83	71	64	88	73	91	101	96	84	67	70	64
III	74	60	33	67	76	64	71	58	46	51	43	34

during the month April at station I and a minimum of 33 CFU/g strains were isolated during the month of December at station III. The total bacterial strains of fish samples recorded with maximum of 107 CFU/g in pre monsoon season at station I and recorded minimum of 43.50 CFU/g in monsoon season at station III. The eutrophication status of the estuary was related to microbial activities. The estuarine waters uptake the inorganic nutrients and developed pico planktonic microbial communities. So the inorganic nutrient concentrations were low but microbial activities were high.

The higher bacterial load of fish may due to its bottom dwelling behavior. The viable bacterial counts in pond fishes are higher than those reported from the fishes collected from the sea. Gram-negative bacteria are predominant in the marine environment, usually constituting the majority in the normal micro flora of wild Prawns.

Fluctuations of temperature, salinity, pH, dissolved oxygen, high levels of ammonia, nitrate, nitrite, organics, heavy metals, pesticides and nutritional factors induce stress in fishes. The bacterial population in sediment was found to be higher than in water. In the water samples total heterotrophic bacterial count was higher than the fish samples. The distribution of heterotrophic bacteria studied in water, sediment and fish samples were in the order sediment > water > fish. The findings on the percentage occurrence of micro flora in water, sediment and fish samples in Poovar estuary during the study period shows that the Escherichia coli was the most dominant group highest percentage. Next to Escherichia coli, the identified genera were Salmonella and Shigella. In most of the cases the composition of bacterial genera was in the order: *Escherichia coli* > *Salmonella* > *Shigella.*

CONCLUSIONS

In the present study, the data obtained on the seasonal variations of bacteriological contaminants showed that maximum bacterial count was during premonsoon season and minimum during monsoon season. The important bacterial genera encountered were *Escherichia coli, Salmonella* and *Shigella*. Among the identified bacterial genera, *Escherichia coli* were found to be the dominant one. Next in abundance, were *Salmonella* and *Shigella*. The bacterial contamination of sediment samples was found to be higher than that in the water samples of Poovar estuary. Fishes also found to be with higher bacterial contamination in their intestine than in their gills and skin.

RECOMMENDATIONS

Tourism is one of the world's largest and fastest growing industries. A clean environment is a key pre requisite for the continued prosperity of the tourism industry. The extent and nature of the environmental and ecological damage done by tourists is related to the magnitude of the development and the number of visitors. To limit the extent of the impacts, a monitoring mechanism must be developed for Impact Assessment of ecotourism. Initiation must be taken to develop an awareness campaign to highlight economic and conservation benefits of ecotourism. The data obtained from this study could therefore serve as base line information for future environmental impact assessment of the effluents discharged from tourist resorts in this region. Man requires a clean ecologically well balanced environment to promote healthy living. Society produces a vast array of waste products and the water provides an effective means of disposing many of these. It is the responsibility of the society to ensure minimum damage to the water resources

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