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An assessment of potential public health risk associated with the extended survival of indicator and pathogenic bacteria in freshwater lake sediments

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ABSTRACT

Microcosm studies were performed to evaluate the survival of *Escherichia coli*, *Salmonella paratyphi* and *Vibrio parahaemolyticus* in water and sediment collected from the freshwater region of Vembanad Lake (9°35'N 76°25'E) along the south west coast of India. All three test microorganisms showed significantly ($p < 0.01$) higher survival in sediment compared to overlying water. The survival in different sediment types with different particle size and organic carbon content revealed that sediment with small particle size and high organic carbon content could enhance their extended survival ($p < 0.05$). The results indicate that sediments of the Lake could act as a reservoir of pathogenic bacteria and exhibit a potential health hazard from possible resuspension and subsequent ingestion during recreational activities. Therefore, the assessment of bacterial concentration in freshwater Lake sediments used for contact and non contact recreation has of considerable significance for the proper assessment of microbial pollution of the overlying water, and for the management and protection of related health risk at specific recreational sites. Besides, assessment of the bacterial concentration in sediments can be used as a relatively stable indicator of long term mean bacterial concentration in the water column above.

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Introduction

Constant microbial pollution by the discharge of untreated or only partly treated wastewaters including industrial, agricultural and domestic wastes are the major pathways of enteric bacterial pathogens into the natural surface water resources. The long term survival of these organisms in natural waters constitutes a significant public health concern because of the dangers they pose to humans through consumption and recreation. Similarly the association of microorganisms with sediment particles are the primary complicating factors in assessing microbiological quality of water because sediment associated bacteria has the potential to contaminate the overlying water column through microbial resuspension during human and boat activity or natural turbulence (An et al., 2002; Craig et al., 2004). This is especially critical for water bodies where recreational activities and human contact takes place. On the other hand microbial adsorption and sedimentation may pose

an indirect public health risk through the consumption of contaminated shellfish due to settlement of particles carrying pathogens to the bottom water layers (West et al., 1985). It is also demonstrated that contact with bathing water subject to faecal contamination increases the risk of disease (Kay et al., 1994; Fleisher et al., 1993).

In India, almost three-quarters of a billion people live in rural areas do not have access to safe drinking water and water-borne infections are a major cause of morbidity (Patil et al., 2002). Diseases such as enteric fever and diarrhoeal diseases are highly endemic to India and are major public health problems among the children under the age of five years. The Planning Commission in its report 'India Assessment 2002 – Water Supply and Sanitation' acknowledges that mortality and morbidity levels due to water borne diseases in the country are unacceptably high (www.cseindia.org/programme/health/pdf/conf2006/a1water.pdf).

Generally *Escherichia coli* is considered as a typical faecal indicator bacteria and its presence in natural waters indicates the possible presence of enteric pathogens. However, certain pathogenic serotypes of *E. coli* such as enterotoxigenic *E. coli* (ETEC), enteroinvasive *E. coli* (EIEC), enterohemorrhagic *E. coli* (EHEC), enteropathogenic *E. coli* (EPEC) enteroaggregative *E. coli* (EAaggEC) and enteroadhesive *E. coli* (DAEC) have been reported as an emerg-

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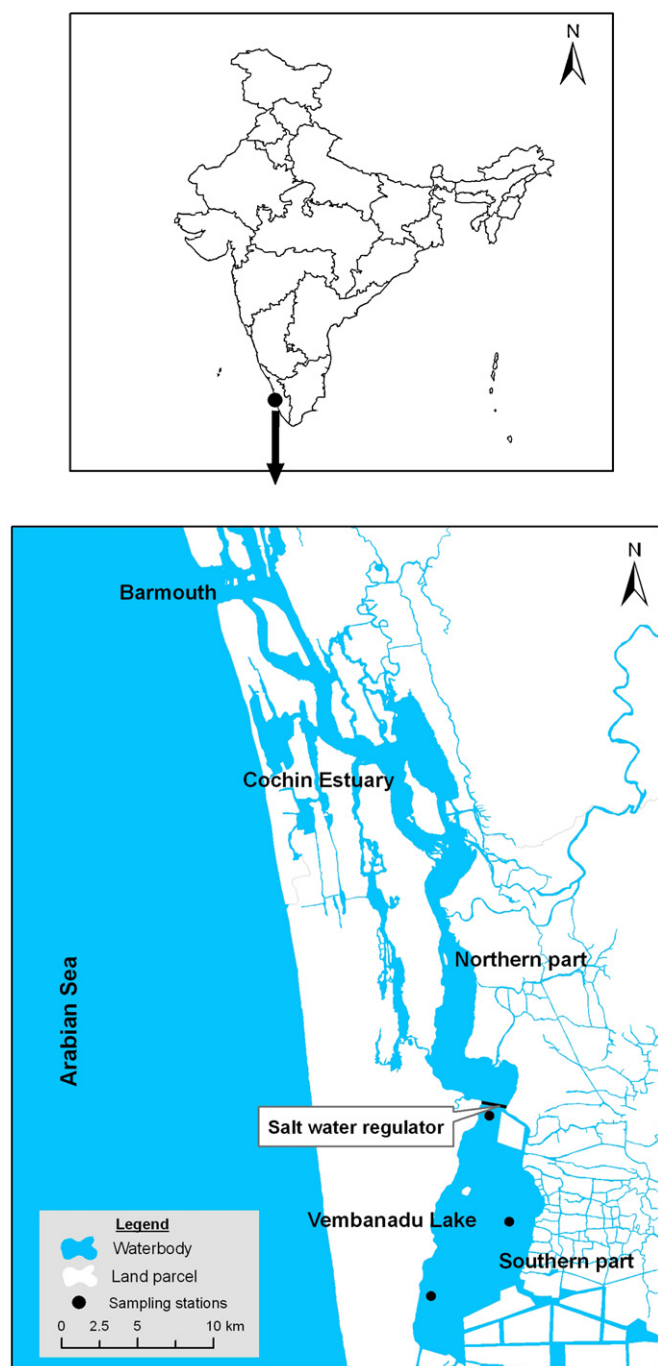


Fig. 1. Map showing Vembanadu Lake and sampling stations.

ing public health concern for children in developing countries (Kaper et al., 2004) including India where frequent outbreaks have been reported (Kahali et al., 2004). *Vibrio parahaemolyticus* is a natural inhabitant of marine and estuarine environments which has been recognized as an important cause of food borne illness in Asia and United States and most of the outbreaks are associated with consumption of raw or undercooked shellfish (Daniels et al., 2000).

The present study has been carried out in Vembanadu Lake that lies 0.6–2.2 m below mean sea level (MSL) along the west coast of India (9°35'N 76°25'E) and has a permanent connection with the Arabian Sea at Barmouth region (Fig. 1). As the north-east monsoon recedes, the area is exposed to tidal incursion of saline water from the Arabian Sea. In order to prevent the saline incursion during certain periods of the year, a salt water regulator is constructed in the

Lake. It divides the Lake into a freshwater region on the southern part and a saline lagoon on the northern part. When the regulator is closed there is virtually no flow of water beyond it making the southern region as a static pool. The periodic tidal inflow, which use to flush the water body is completely prevented with the result that the drained water from the surrounding rice fields and human dwellings with heavy load of pollutants remains stagnant in the water body. On the other hand over 1.6 million people directly or indirectly depend on it for various purposes such as agriculture, fishing, transportation and recreation. A large population of the area is facing severe scarcity of drinking water because of the saline nature of the ground water and the Lake serves as the only source of freshwater. As a result water related diseases are very common in this region particularly in young children but none of them were reported officially.

The U.S. Environmental Protection Agency, the European Union and Central Pollution Control Board (CPCB) recommend the use of *E. coli* to assess the hygienic safety of recreational waters (USEPA, 2000; EU, 2006). As hygienic quality of the water is of utmost importance to the society, regular water quality monitoring is necessary because prolonged survival and persistence of pathogenic bacteria in natural water resources used as a common source of water by humans would be a public health concern. Usually the hygienic quality of the surface water is assessed only in terms of the density of faecal indicator bacteria (*E. coli*) in overlying water alone and sediment is often overlooked. Thus the potential of sediment bound bacteria to pollute the overlying water during recreational activities and the subsequent health risks posed by pathogenic microorganisms are not often considered while assessing the hygienic safety of water.

The southern region of Vembanadu Lake is being used for contact recreation and other domestic purposes because of the availability of freshwater. However, no regular water quality monitoring programmes have been instituted and the only published work available on the microbial pollution of Vembanadu Lake is by Abhirosh et al. (2008) where we recorded and isolated high microbial pollution and pathogenic bacteria such as *Salmonella paratyphi* A, B, C, *Salmonella newport*, *Salmonella bareilly*, *V. parahaemolyticus*, *Vibrio cholerae* and several diarrhegenic serotypes of *E. coli*. Prolonged survival and persistence of these pathogenic bacteria in sediment could be a public health concern. To the best of our knowledge no published works are available on the survival of pathogenic bacteria in Vembanadu Lake sediments. Hence in the present investigation microcosm experiments have been carried out to evaluate the survival of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* in water and sediments collected from the southern freshwater region of the Vembanadu Lake.

Materials and methods

Survival experiments

Test microorganisms

Confirmed cultures of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* isolated (Abhirosh et al., 2008) from the Vembanadu lake and maintained in the culture collection of the School of Environmental Sciences of Mahatma Gandhi University were used for survival studies. *V. parahaemolyticus* is an indigenous bacterium, which has been selected because of its importance as a leading shellfish borne pathogen in the developed and developing world. *E. coli* and *Salmonella* were selected because they are allochthonous to aquatic environments as well as water borne and food borne pathogens.

Preparation of inocula

The inocula were prepared as previously described by Abhirosh and Hatha (2005). Briefly, *E. coli*/*S. paratyphi*/*V. parahaemolyticus*

were grown in Tryptone Soy Broth (TSB) and incubated at 37 °C for 24 h. After incubation, the cells were concentrated by centrifugation at 1400 × g for 15 min and washed twice with sterile isotonic saline. After the final wash, the cells were resuspended in sterile isotonic saline for inoculation into the microcosms.

Collection of core sediment microcosm

In our earlier study on the prevalence of indicator and pathogenic bacteria on either sides of the saltwater regulator, we have recorded high incidence of bacteria on the southern part of the Lake compared to northern part (Abhirosh et al., 2008). Considering the public health risk associated with the increased use of southern region for contact recreation and other domestic purposes, survival experiments were conducted in water and sediment collected from southern freshwater part of the Vembanadu Lake. Therefore intact sediment cores along with overlying water were collected in perspex columns from three locations on the southern freshwater part of the Vembanadu Lake (Fig. 1) as described by Craig et al. (2004) during the period when the salt water regulator was closed. Prior to sampling, microcosm equipment was treated with ethanol and rinsed with sterile water to remove any microorganisms that may have been present. Perspex columns (60 mm diameter, 300 mm length) were inserted into sediment to a depth of 100 mm. The top of the column was capped with a rubber bung to aid in removing the core from the sediment. The sediment core was kept in place by inserting a combination of neoprene (5 mm thick) and closed-cell foam (20 mm thick) bungs into the bottom of the core. This prevented the movement of sediment and water from the column.

Inoculation and enumeration of bacteria from sediment microcosms

Each column (microcosm) was inoculated separately by adding 1 ml of washed *E. coli*/*S. paratyphi*/*V. parahaemolyticus* suspension into the overlying water. The initial concentration of the microorganisms was around 10⁸ CFU per ml. After inoculation the columns were kept at 25 °C (to imitate the mean sediment temperature) without disturbing the upper layer of sediment. For the enumeration of bacteria, water and sediment samples from the microcosm were taken and assayed at specific time interval for 27 days. One ml water sample was collected carefully from the overlying water column using a sterile pipette without disturbing the sediment. Sediment samples were obtained by first aseptically removing the overlying water. The top 10 mm of sediment was removed with the aid of a sterile spatula and added to a sterile beaker. Of this sediment, 10 g (wet weight) was added to 90 ml sterile isotonic saline water and mixed for 10 min with the help of a magnetic stirrer. After sampling, the water was replaced over the sediment. The enumeration of bacteria was done after appropriate dilution by spread plate technique using selective media. Eosin Methylene Blue (EMB) agar was used as selective medium for *E. coli*, Xylose Lysine Deoxycholate (XLD) agar for *S. paratyphi* and Thiosulfate Citrate Bile Salt Sucrose (TCBS) agar for *V. parahaemolyticus* as previously described by Abhirosh and Hatha (2005). Sterilised (autoclaved) sediment and water inoculated with test organisms were also used as controls. All media used were purchased from Hi-Media laboratories, Mumbai, India.

Survival in sediment with different particle size and organic carbon content

To study the survival in different sediment types, sediments were collected with the aid of a grab sediment sampler from the same locations on the southern fresh water part of the Lake. Sediments were characterised as % sand, silt and clay using the pipette method (Sheldrick and Wang, 1993). The percentage of organic car-

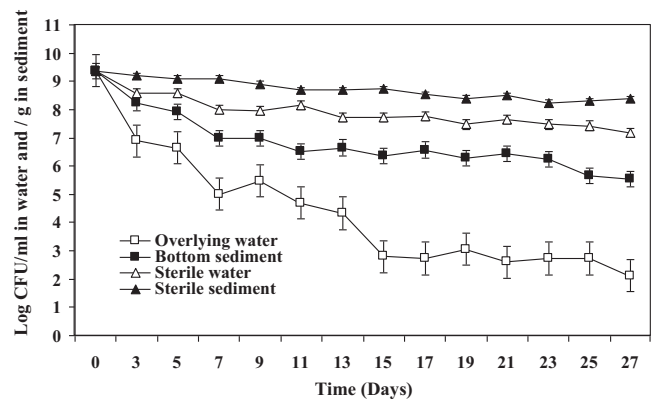


Fig. 2. Survival curves of *E. coli* in overlying water and bottom sediment at 25 °C (Mean ± SD, n = 4).

bon present in the sediment was determined by the dichromate method (Tiessen and Moir, 1993).

Closed bottle sediment microcosms were prepared according to Davies et al. (1995) by taking 100 g of sediment (wet weight) into sterile conical flasks (250 ml) with 50 ml of autoclaved lake water. In the laboratory 1 ml of the washed cell suspension of *E. coli*/*S. paratyphi*/*V. parahaemolyticus* (10^{8–9} CFU per ml) were inoculated separately into each microcosm. After inoculation, the sediment was mixed thoroughly to evenly distribute the bacteria and incubated at 25 °C. From the microcosms, sediment samples were collected aseptically after removing the overlying water. The enumeration of the bacteria was done as mentioned above.

Statistical analysis

The difference in the survival of the test organisms in sediment and overlying water as well as in different sediment types were compared using two way analysis of variance (ANOVA). The inactivation rates were calculated using the following formula. $\text{Log}_{10}(N_t/N_0)$ where N_t is the number of bacteria at time t and N_0 is the number of bacteria at time 0.

Results and discussion

Survival of indicator bacteria and enteric pathogens in sediment and overlying water

The survival curves of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* in water and sediment at 25 °C are illustrated in Figs. 2–4, respectively. The results revealed that the survival of the test microorganisms were much lower in the overlying water column

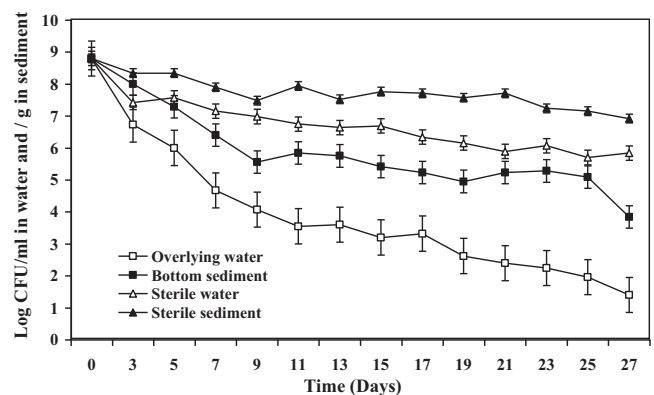


Fig. 3. Survival curves of *S. paratyphi* in overlying water and bottom sediment at 25 °C (Mean ± SD, n = 4).

Table 1
Inactivation rates of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* in overlying water and bottom sediment.

Days	Water			Sediment			Sterile water			Sterile sediment		
	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>
0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
3	-2.47	-2.07	-1.19	-1.13	-0.8	-0.44	-0.77	-1.38	-0.22	-0.17	-0.46	-0.31
5	-2.72	-2.80	-3.02	-1.45	-1.51	-1.49	-0.77	-1.23	-0.83	-0.27	-0.46	-0.8
7	-4.37	-4.13	-3.52	-2.39	-2.4	-2.15	-1.37	-1.65	-1	-0.28	-0.9	-0.63
9	-3.90	-4.73	-5.15	-2.39	-3.24	-2.61	-1.41	-1.82	-1.64	-0.47	-1.32	-1.05
11	-4.68	-5.25	-4.68	-2.84	-2.96	-3.37	-1.21	-2.05	-1.6	-0.68	-0.86	-0.88
13	-5.04	-5.20	-5.50	-2.72	-3.05	-3.17	-1.66	-2.16	-2.15	-0.68	-1.28	-1.24
15	-6.57	-5.60	-6.06	-3.00	-3.38	-3.73	-1.66	-2.11	-1.9	-0.64	-1.04	-0.99
17	-6.64	-5.48	-5.85	-2.8	-3.57	-3.45	-1.61	-2.46	-2.3	-0.84	-1.09	-1.19
19	-6.31	-6.18	-6.16	-3.08	-3.85	-3.73	-1.9	-2.65	-2.36	-0.97	-1.23	-1.45
21	-6.77	-6.40	-6.62	-2.92	-3.57	-3.45	-1.74	-2.91	-2.65	-0.88	-1.09	-1.14
23	-6.64	-6.56	-7.13	-3.13	-3.52	-3.62	-1.9	-2.73	-2.51	-1.13	-1.56	-1.39
25	-6.64	-6.84	-7.33	-3.7	-3.71	-3.93	-1.94	-3.1	-2.45	-1.08	-1.65	-1.34
27	-7.25	-7.40	-7.85	-3.82	-4.96	-3.85	-2.19	-2.96	-2.61	-1.00	-1.88	-1.24
<i>T</i> ₉₀	3.06	3.12	2.97	6.13	5.25	5.46	10.75	7.39	8.5	22.11	14.25	15.87

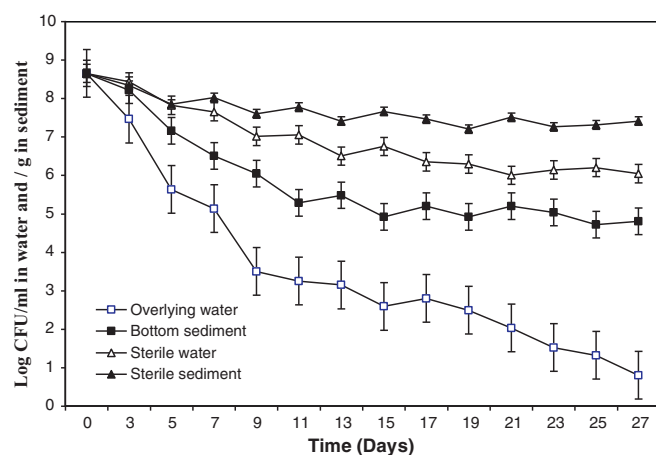


Fig. 4. Survival curves of *V. parahaemolyticus* in overlying water and bottom sediment at 25 °C (Mean ± SD, n = 4).

than in the sediment. At the end of the 27th day the decline of *E. coli* was almost 7 logs in the overlying water while it was only 3 logs in the sediment and the *T*₉₀ (the time for bacterial concentration to decrease by 1 log) reached in 3.06 and 6.13 days, respectively in water and sediment. However in sterile water (*T*₉₀ = 10.75) and sediment (*T*₉₀ = 22.11 days) they required even more time to reduce the bacterial abundance by 1 log and showed enhanced survival. Almost similar reduction patterns were obtained for *S. paratyphi* and *V. parahaemolyticus* in overlying water and sediment (see Table 1 for *T*₉₀). In the present study the *T*₉₀ observed in the overlying water (almost 3 days) and sediment (almost 6 days) was higher than those reported by Craig et al. (2004) in different recreational waters where the *T*₉₀ was almost 2 days in water and 5 days in sediment. However a higher *T*₉₀ (4.5 days) was reported for faecal bacteria in freshwater by Menon et al. (2003) and *T*₉₀ ranged from 1.5 to 9 days were reported for *Vibrio* species in freshwaters (Feachem et al., 1981). These variation found in *T*₉₀ value in different study might be due to the difference in the strain of organism used and type of environment etc.

Relatively higher decay of the test microorganisms in overlying water column may be due to the active role of biological factors such as predation by protozoans (Hahn and Hofle, 2001) and bacteriophages (Ricca and Cooney, 1999). In our earlier studies on the factors affecting the survival of *E. coli*, *V. parahaemolyticus* and *S. paratyphi* in estuarine water, we have noticed that biological factors such as protozoans, bacteriophages and competing autochthonous bacteria cause considerable inactivation of the microorganisms in estuarine water (Abhirosh et al., 2009). This was further strengthened by our previous observation in Cochin estuary that protozoans and bacteriophages exert considerable inactivation of these organisms in estuarine water (Abhirosh and Hatha, 2005). However, when biological factors were removed from the water and the sediment by autoclaving, *E. coli*, *S. paratyphi* and *V. parahaemolyticus* showed enhanced survival clearly indicating the dominant role of biological factors on the survival of the test organisms in aquatic environment.

The comparative survival in water and sediment revealed that the survival of the test microorganisms were significantly higher in sediment (*p* < 0.05). Slower reduction rate of the test organisms in sediment suggest that sediment offer some sort of protection to the test microorganisms. This includes protection from protozoan bacteriophage (Davies and Bavor, 2000), attack by bacteriophage (Roper and Marshall, 1979), rich supply of nutrients (Craig et al., 2004) and substratum for adherence (Davies et al., 1995). Results indicate that enteric microorganisms could survive and accumulate in sediments at levels higher than those in the overlying waters and thus

Table 2
Particle size and organic carbon content of surface sediment layers.

Sediment Sample	% Clay	% Silt	% Sand	% Organic content (OC)
OC high	17.53	1.75	80.713	4.96
OC medium	9.395	15.655	74.95	2.24
OC low	9.654	0.509	89.837	1.84

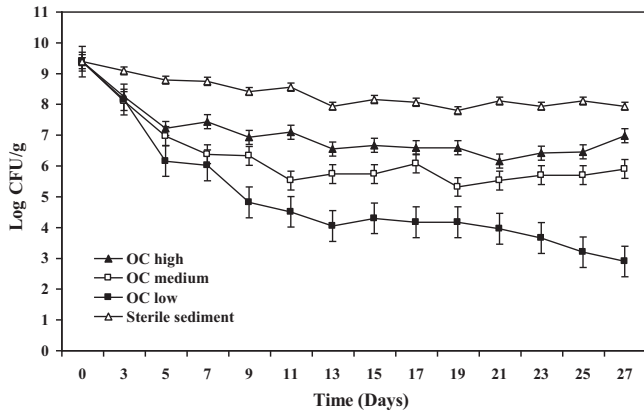


Fig. 5. Survival curves of *E. coli* in sediments with varying particle size and organic carbon (OC) content at 25 °C (Mean ± SD, n = 4).

serves as sinks for faecal indicator bacteria (Ashbolt et al., 1993). Our results support the view of previous researchers (Burton et al., 1987; Davies et al., 1995; Craig et al., 2004) that *E. coli*, *Salmonella* and *V. parahaemolyticus*, can survive better in the sediments than in overlying waters.

Survival in sediments with different particle size and organic carbon content (OC)

Particle size analysis and organic carbon content of surface sediment layers are given in Table 2. The sediments were designated as OC high, OC medium and OC low based on the amount of organic carbon content. The survival of *E. coli*, *S. paratyphi*, and *V. parahaemolyticus* in different bottom sediments is illustrated in Figs. 5–7, respectively and the inactivation rates and T_{90} are given in Table 3. All test organisms showed better survival in sediments with small particle size and high organic content. The results indicated that survival was influenced by sediment characteristics (sand, silt, clay and organic carbon), which might reflect intrinsic differences among sediments. The survival of the test microorganisms were varied among themselves as well as in different sediment types and the survival were correlated well with the particle size and organic carbon content

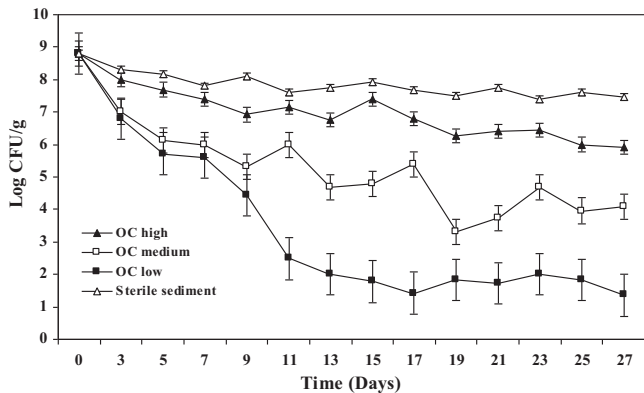


Fig. 6. Survival curves of *S. paratyphi* in sediments with varying particle size and organic carbon (OC) content at 25 °C (Mean ± SD, n = 4).

Table 3
Inactivation rates of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* in sediments with varying particle size and organic carbon (OC) content.

Days	OC High			OC Medium			OC Low			Sterile sediment		
	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>	<i>E. coli</i>	<i>S. paratyphi</i>	<i>V. parahaemolyticus</i>
0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
3	-1.13	-0.80	-0.44	-1.8	-1.23	-1.28	-1.23	-2	-1.48	-0.3	-0.5	-0.35
5	-2.17	-1.11	-1.67	-2.42	-2.66	-2.51	-3.23	-3.08	-1.88	-0.6	-0.62	-0.72
7	-1.95	-1.40	-1.28	-3.01	-2.8	-2.65	-3.37	-3.2	-3.01	-0.64	-1.00	-0.51
9	-2.46	-1.88	-2.06	-3.06	-3.47	-3.38	-4.57	-4.36	-4.14	-0.97	-0.69	-1.24
11	-2.29	-1.64	-1.28	-3.86	-2.8	-3.63	-4.88	-6.31	-4.34	-0.83	-1.2	-1.05
13	-2.84	-2.05	-1.82	-3.65	-4.13	-3.58	-5.34	-6.8	-4.6	-1.45	-1.05	-0.99
15	-2.72	-1.40	-2.50	-3.65	-4.00	-3.42	-5.09	-7.02	-5.21	-1.23	-0.87	-1.19
17	-2.80	-2.00	-2.65	-3.31	-3.42	-4.19	-5.22	-7.38	-6.29	-1.32	-1.11	-1.58
19	-2.80	-2.53	-2.89	-4.07	-5.49	-4.09	-5.22	-6.97	-5.98	-1.59	-1.29	-1.33
21	-3.23	-2.40	-2.99	-3.86	-5.07	-4.75	-5.43	-7.08	-6.44	-1.28	-1.05	-1.19
23	-2.97	-2.35	-2.85	-3.69	-4.13	-4.19	-5.73	-6.8	-6.21	-1.45	-1.4	-1.33
25	-2.93	-2.80	-2.94	-3.69	-4.84	-4.34	-6.19	-6.97	-6.55	-1.28	-1.24	-1.24
27	-2.41	-2.88	-2.75	-3.49	-4.72	-4.7	-6.49	-7.44	-6.41	-1.45	-1.34	-1.09
T_{90}	6.91	8.40	7.47	5.25	4.42	4.66	3.48	2.84	3.34	14.75	16.49	15.89

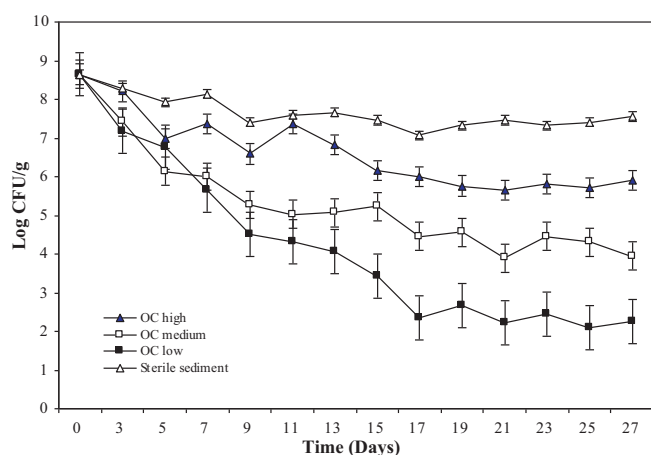


Fig. 7. Survival curves of *V. parahaemolyticus* in sediments with varying particle size and organic carbon (OC) content at 25 °C (Mean \pm SD, $n=4$).

of the sediment. The persistence of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* was significantly ($p < 0.05$) higher in OC high sediment (% clay 17.53, % silt 1.75; % sand 80.713; % organic carbon 4.96, $T_{90} = 7-8$ days) followed by OC medium (% clay 9.395; % silt 15.6555; % sand 74.95; % organic carbon 2.24, $T_{90} = 4-5$ days) and OC low sediment (% clay 9.654, % silt 0.509; % sand 89.837; % organic carbon 1.84, $T_{90} = 2-3$ days). In agreement with our observation Craig et al. (2004) also reported high survival of enteric bacteria in organically rich sediment compared to low organic levels; however the T_{90} (2–4 days) he observed was much lower compared to our study. It has also been evidenced from the experiments conducted by Burton et al. (1987) that the T_{90} of different pathogenic bacteria was ranged between 3 and 5 days in various freshwater sediments depending on the characteristic of the sediments (sand, silt clay). Jeng et al. (2005) reported that the T_{90} of enteric microorganisms was less than 1 day in estuarine water and more than 7 days in sediment. Even though there are variations in T_{90} values in different studies, in general, the longer period required for the enteric microorganisms to reach T_{90} in the sediments of different aquatic environments clearly indicating their higher survival capacity in sediments than in overlying water.

Bottom sediment of Vembanadu Lake is characterised by high organic load and clay content. While the organic load is from the dense human habitation surrounding the Lake, clayey nature of the sediment is due to relatively stagnant rather slow flow patterns. Construction of a salt water regulator is the main reason for relatively stagnant nature of the Lake. In our previous studies we have recorded high load of faecal indicator bacteria on the southern part of the Lake (Abhirosh et al., 2008). When the regulator is closed the natural flow is prevented which results in the accumulation of organic load in the southern part of the Lake, giving proper environmental conditions for the multiplication of bacteria. While clayey sediment offers more surface area than silty and sandy sediment, and organic load provides better nutrition. These results are in agreement with the findings of other workers that the survival of microorganisms has correlation with particle size and organic carbon content of the sediment. For instance, extended survival of *E. coli* and *S. newport* in coastal sediment with high clay content and nutrient availability has been reported (Burton et al., 1987; Craig et al., 2004). In agreement with our observation Gerba and McLeod (1976), Chan et al. (1979) reported that nutrient rich, fine-grained sediments may have a significant effect on extended microbial survival.

In our earlier investigation, we have recorded high prevalence of pathogenic bacteria such as *S. paratyphi* A, B, C, *S. newport*, *S. bareilly*, *V. parahaemolyticus* and different pathogenic serotypes of *E. coli*



Photo 1. Mechanical dredging for molluscan shell for cement production.

such as *ETEC*, *EPEC*, *EHEC* and *UPEC* from the present study area as well as from Cochin region of Vembanadu Lake (Hatha et al., 2004; Abhirosh et al., 2008). These strains are considered as a major public health problem in developing countries including India (Nataro and Kaper, 1998; Gupta et al., 2009). This indicates that the sediment of Vembanadu Lake could also act as a repository for these pathogenic bacteria as it showed extended survival in lake bottom sediments which in turn would be a real public health concern.

The resuspension of sediment bound bacteria and subsequent pollution of overlying water exceeding regulatory standards has been reported in recreational water (Obiri-Danso and Jones, 1999). The risks associated with swimming in microbiologically polluted Lakes or rivers during recreational activities are usually not life threatening but could take a substantial toll in children and immune-compromised individuals (Clark et al., 2003). The resuspension and subsequent deterioration of water quality is very likely to occur in different parts of Vembanad Lake because mechanical dredging for molluscan shell for cement production is a regular activity in every morning and evening (Photo 1). Well developed house boat based tourism in the study area also results in the disturbance of lake bottom since house boats are very often propelled with the help of long wooden poles. The Lake also supports rich shellfish resource, which is being harvested on a daily basis and marketed in the nearby areas, where it forms a cheap, popular and protein rich food item of the local people. The manual collection of shellfish could also disturb the bottom sediments.

Erkenbrecher (1981) suggested that sediments in shellfishing areas could serve as a reservoir for high densities of indicator bacteria and that, potentially, pathogens could pose a health hazard. High concentration and prolonged survival of pathogenic bacteria in sediments from the study area coupled with the filter feeding nature of the shellfishes pose direct or indirect health risk through the consumption of contaminated shellfish. Shellfish associated food borne illness has been reported from India and other Asian countries (Deepanjali et al., 2005). The results of the present study are of particular significance to public health because Vembanadu Lake is the life line of the people in that area as they use this water body for agriculture, fishing, transport and contact recreation. The availability of drinking water is very low and some times the people are forced to drink this water which may pose serious health risk. As pathogenic bacterial survival was higher in small particle size sediment an epidemiological study indicated that the risk of gastrointestinal illness appears to correlate well with the number of *E. coli* associated with small particle size of the sediment (Dufour, 1983). Therefore, the period of pathogen survival and associated particle transport into water used for recreation and fisheries is of concern to regulatory authorities.

Conclusions

Results of the microcosm study demonstrated prolonged survival of *E. coli*, *S. paratyphi* and *V. parahaemolyticus* in sediments compared to overlying water. The results also indicated that small particle size and high organic carbon content enhanced the survival of indicator and pathogenic bacteria in the sediments. Therefore the sediments of Vembanadu Lake could act as a reservoir of pathogenic bacteria and exhibit increased risk of infection to the sensitive population due to the possible resuspension and subsequent contamination of overlying water during recreational activities. Therefore assessment of bacterial concentration in freshwater Lake sediments used for contact and non contact recreation is of considerable significance for the proper assessment of microbial pollution of the overlying water and the management and protection of related health risk at specific recreational sites. In addition, assessment of the bacterial concentration in sediments can be used as a relatively stable indicator of long term mean bacterial concentration in the water column above.

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