

Microbiological beach sand quality in Gaza Strip in comparison to seawater quality

Abdelraouf A. Elmanama^{a,*}, Mona Ishaq Fahd^b, Samir Afifi^a,
Soad Abdallah^b, Salah Bahr^a

^aMedical Technology Department, Islamic University-Gaza, P.O. Box 108, 00972 Gaza, Gaza Strip, PNA

^bAin Shams University, Cairo, Egypt

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Abstract

Gaza beach is the only recreational area available for the local inhabitants. It is heavily polluted with treated, partially treated, and untreated sewage from point and nonpoint sources. The majority of the population is below the age of 15 years. This age group is vulnerable to gastrointestinal diseases and usually restricts their activities to beach sand at the swash zone. Five sampling points along the Gaza beach were selected and monitored for 1 year (fortnightly). Microbial sand content was evaluated for fecal coliforms (FC), fecal streptococci (FS), *Salmonella*, *Shigella*, and *Vibrio*. Seawater samples were subjected to similar evaluation. *Pseudomonas*, yeast, and mold counts were performed for all sand samples as possible sand pollution indicators. Higher fecal indicators (both FC and FS) were obtained in sand than in water in most locations. The frequency of *Salmonella* and *Vibrio* isolation was also higher in sand than in water despite the fact that only 10 g of sand were used while 1 L of seawater was collected. Statistically significant correlations between FC and streptococci and between *Salmonella* and *Vibrio* were found. Similar correlation was also detected between *Pseudomonas* and *Salmonella* in sand samples.

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1. Introduction

Recreational water generally contains mixtures of pathogenic and nonpathogenic microbes derived from sewage effluents, industrial processes, farming activities, wild life, and indigenous microorganisms. This mixture can present a hazard to bathers when an infective dose of pathogen colonizes a suitable growth site in the body and leads to a disease (WHO, 1998).

The extent of seawater pollution varies according to the quantity and quality of pollutants. However, the problem of seawater pollution is acknowledged world-

wide. As a result of recreational activities, many individuals may contract diseases that may range from self-limiting gastrointestinal disturbances to severe and life-threatening infections. The disease incidence is dependent on several factors; the extent of water pollution, the time and type of exposure, the immune status of users, and other factors (Bartram and Rees, 2000).

Seawater and beach quality monitoring and assessment are considered vital parts of any integrated coastal management program (Afifi et al., 2000). Extensive research with the aim of establishing guidelines and standards for recreational water quality has been conducted all over the world. In this context, social, cultural, environmental, and economic factors should be taken into consideration because of the great variation from one area to another.

*Corresponding author.

E-mail addresses: elmanama@mail.iugaza.edu (A.A. Elmanama), saffi@mail.iugaza.edu (S. Afifi), sbahr@mail.iugaza.edu (S. Bahr).

The microbiological context of sediments at the sediment–water interface in bathing waters is receiving increased attention (Arakel, 1995). There is evidence that fecal indicator and pathogenic bacteria survive for longer periods in sediments than in the overlying water and it has been proposed that sediments serve as sinks for fecal bacteria with the potential to pollute the overlying bathing waters (Ashbolt et al., 1993; Nix et al., 1993; Ghinsberg et al., 1994; Howell et al., 1996).

Stream sediments have been shown to contain fecal coliform (FC) at concentrations higher than those observed in the overlying water column. Van Donsel and Geldreich (1971) and Ashbolt et al. (1993), e.g., indicated that sediments may contain 100–1000 times the number of fecal indicator bacteria contained in the overlying water.

Crabill et al. (1999) analyzed FC in water and sediment samples from Oak Creek, Arizona, USA. They found sediment samples with up to 2200 times the FC counts of the water column. Results showed that resuspension of sediments due to agitation by recreational activities and storm events during the summer season negatively impacted the water quality.

Studies on the survival of bacteria indicate that sediments present an environment favorable for growth. Fecal bacteria have been shown to survive and, to a certain extent, even to grow in sediments. Hood and Ness (1982) reported on the survival of *Vibrio cholerae* and *Escherichia coli* in sediments and work by Gerba and McLeod (1976) and LaLiberte and Grimes (1982) showed evidence of survival and growth of *E. coli* in sediments.

Laws and legislations have emphasized the use of microbiological indicator levels in seawater and have almost ignored the fact that many beach visitors may not use water but would use sand, most especially children. The swash zone of a bathing beach is the interface area that is washed over by waves. This region is a popular play area for young children. Bacteria capable of causing human diseases may contaminate the sand in this part of the beach shoreline. Accordingly, concern that the beach sand or similar materials may act as reservoirs or vectors of infection has been expressed (Roses Codinachs et al., 1988).

Gaza beach is considered the only recreational site for a population of more than one million inhabitants of the Gaza Strip. It is usually very crowded in the summer season, mostly with local inhabitants. At the local level, a few studies have been conducted by the Environmental and Rural Research Center (Afifi, 1999); these showed heavily contaminated recreational seawater along the seashore of Gaza Strip.

This study provides the first original data on the microbiological content of sand along the Gaza Strip beach and the seawater microbiological quality. This work addresses the sand contamination issue which was

long neglected by researchers and policy makers in assessing the quality of beaches. Hence, it is expected to assist local authorities in developing plans and policies and in implementing actions to reduce the pollution to acceptable levels. It may prove helpful in setting standards and guidelines. Finally, this work will add to the accumulating literature on sand and seawater which could change our global view of beach monitoring policies.

2. Material and methods

2.1. Sample site selection

Five sampling locations were selected based on visual inspection of the beach and the amount of sewage disposed. These sites were identified by landmarks and by global positioning system (see Tables 1 and 2 and Fig. 1).

2.2. Sample collection

Sampling was performed according to the World Health Organization Manual for Recreational water and Beach Quality Monitoring and Assessment (1995). The sampling frequency was fortnightly. Sample collection lasted from May 2002 to May 2003; 500-mL polyethylene bottles were used to collect water samples

Table 1
Sampling site identification information

Location	City/address	Prominent mark	GPS locations	
			N	E
1	South Deir Elbalah	Resort	31.25.03.6	34.19.43.0
2	North Deir Elbalah	Elementary school	31.25.50.6	34.20.34.7
3	Al-Zawida	Resort	31.26.37.8	34.21.22.2
4	South Wadi Gaza	Army Station	31.27.40.7	34.22.23.7
5	North Wadi Gaza	Life guard station	31.27.56.2	34.22.37.9

Table 2
Reasons for selecting the sampling locations

Location	Reasons of selection
1	Rocky area, a favorite place for children, relatively clean
2	Polluted with raw sewage, usually populated during bathing season, no infrastructure or installations
3	No obvious source of contamination, a cafe shop
4	Near Wadi Gaza (major source of sewage), situated south
5	Near Wadi Gaza, situated south, usually populated during bathing season, several café shops



Fig. 1. Map of Gaza Strip showing the sampling locations.

while 100-mL sterile bottles were used to collect sediments. For *Salmonella* isolation from water, 1-L bottles were used.

2.3. Seawater

Water samples were collected while the sampler stood in water at chest level (about 1.3 m); the lid of the bottle was removed without touching the mouth of the bottle. The bottle was turned upside down and lowered approximately 20–30 cm below the surface with a smooth movement (to avoid collecting sediments). The bottle was then turned so that the mouth was pointing upward, and when the bottle was approximately 2/3 filled, it was lifted above the surface and the lid was placed back on the bottle (WHO, 1995).

2.4. Sediments

Sterile, wide-mouthed, 100-mL disposable plastic bottles were used to collect sediment samples from the

swash zone. The lid of the bottle was carefully removed, and the bottle was inverted and forced into the sand. To ease the removal of the bottle with the sample, a large spatula was used to remove the surrounding sand. The bottle was then pulled together with the samples. Samples were stored on ice until analyzed.

2.5. Fecal coliform and fecal streptococci (FS)

A sterile forceps was used to transfer a 0.45- μ m Millipore membrane on the filter support assembly. The funnel portion was placed and fitted (magnetic fitting). Several volumes of the sample (100, 10, 1, 0.1, and 0.01 mL) were filtered. The membrane filter was aseptically removed and placed in the center of a pre-labeled mFC (Difco) culture plate which was then sealed and incubated in a water bath incubator at 44.5 ± 0.2 °C for FC or in Slanetz and Bartely (Oxoid) medium incubated at 37 °C for 4 h and at 44 °C for 44 h for fecal streptococci (FS). Three–five colonies from each plate were picked and biochemical tests were performed to confirm the identity (APHA, 1995). API 20E was used to confirm FC identity, gram stain, catalase, and bile esculin tests were used to confirm FS.

The membrane filtration technique was modified for use with sediments. A suspended sediment (SS) fraction was produced by adding 100 mL of 0.85% (w/v) sterile saline to each sample, vigorously shaking for 30 s, and then allowing brief settlement of the larger particles. Two 10-mL aliquots of the resultant supernatant (the SS fraction) were collected for analysis. The first aliquot was placed in a graduated centrifuge tube and allowed to settle overnight at 25 °C to measure the volume of sediment in an individual sample (sediment load) to allow accurate comparison between samples. The second 10-mL SS fraction was added to a Warring blender containing 90 mL of sterile saline and mixed slowly for 5 min; appropriate dilutions were then enumerated according to the technique used for water samples (Crabill et al., 1999).

2.6. Salmonella and Shigella

Seawater samples: 1 L of seawater sample was filtered through a 0.45- μ m membrane filter. The membrane filter was placed in enrichment medium (selenite-F broth) overnight. Subcultures were made to xylose lysine dextrose agar (XLD) and *Salmonella Shigella* agar (SSA) plates (Oxoid) (APHA, 1995).

Sand samples: 10 g of sand sample was inoculated into 90 mL of selenite-F broth, incubated at 37 °C overnight, and subcultured onto XLD and SSA plates. Suspect colonies were identified biochemically using API 20E strips (Baron and Finegold, 1990).

2.7. *Pseudomonas* counts

A 10^{-1} w/v suspension of sand sample based on wet weight was prepared in 0.1% buffered peptone water and thoroughly mixed, and serial dilutions were made. Counts were estimated using the plate count method (spread plate method), with PseudoSel agar. Colonies were identified biochemically using API 20E strips.

2.8. *Vibrio*

Seawater samples: 1 L of seawater sample was filtered through a 0.45- μ m membrane filter. The membrane filter was placed in enrichment medium (alkaline peptone water) overnight. Subcultures were made to thiosulfate citrate bile sucrose (TCBS) agars (Difco) (Dumontet et al., 2000).

Sand samples: 10 g of sand sample was inoculated into 90 mL of alkaline peptone water (Oxoid) the pH of which was adjusted to 8.6. After incubation at 37 °C for 24 h, cultures were streaked onto TCBS and further incubated for 24 h at 37 °C. Yellow or blue colonies growing on TCBS were picked for identification. The exact identity was identified using API 20E stamps (API system, France) (Dumontet et al., 2000).

2.9. *Yeast and mold counts*

A 10^{-1} w/v suspension of sand sample based on wet weight was prepared in 0.1% buffered peptone water and thoroughly mixed, and serial dilutions were made. Counts were estimated using the plate count method with dichloran rose Bengal chloramphenicol agar (Oxoid) (Mendes et al., 1993).

2.10. *Statistical analysis*

Data obtained from both seawater and sand samples were tabulated as Microsoft Excel sheets, uploaded to SPSS software and analyzed using Pearson correlation coefficient and paired sample *t* test. χ^2 was also used to correlate the levels of microbial indicators with *Salmonella* and *Vibrio*.

3. Results

All locations exhibited variation in both sand and seawater content of FC and streptococci; however, comparing the ratio of sand/seawater, it is clear that the ratio is below 1.0 in locations of low pollution (1 and 3), and high (ranging from 1.2 to 29.1) in locations with high pollution levels (e.g., 2, 4, and 5). This may suggest the accumulation of these indicators on the sand surface. The ratio of FS is much higher than that of FC in such locations. This may be due to the longer

survival rates exhibited by FS. Table 3 illustrates the difference in FC and FS content of both seawater and sand during the monitoring period.

All locations were evaluated using the European Community (EU) standards for FC and FS and Table 4 includes the number of failures of a location to meet the required criteria.

It can be observed from Table 4 that, in general, the failure rates (percentage) of the five locations are greater due to failure to comply with FS than with FC. Failure of both contaminated and relatively clean locations was mainly during winter (no bathers were observed). Location 2 showed the highest percentage failure during the monitoring period. This was actually expected because it receives raw sewage.

Although locations 4 and 5 are at similar distances from the discharging point of Wadi Gaza, the failure percentage was higher at location 5. This may be due to the dominant current direction, which is usually from south to north, carrying more pollutants toward location 5 situated north of Wadi Gaza. Location 3 passed the mandatory standards for FC and failed the

Table 3

Comparison of the levels of fecal coliform and fecal streptococci in both seawater and sand (cfu/100 mL seawater and 100 g sand) ($N = 26$)

Location no.		Average count sand	Std. dev.	Average count seawater	Std. dev.	Sand/sea ratio
Location 1	FC	74	83	37	46	2
	FS	133	159	181	489	0.7
Location 2	FC	13,996	53,385	2585	5096	5.4
	FS	94,566	385,678	8216	18,602	11.5
Location 3	FC	37	63	71	135	0.52
	FS	152	105	184	374	0.83
Location 4	FC	1134	1728	73	73	15.4
	FS	4080	15,977	140	216	29.1
Location 5	FC	5455	14,839	4742	16,324	1.1
	FS	1813	2857	1502	4883	1.2

Table 4

Percentage failure of the studied location compared to the EU bathing directive (76/160/EEC)

Location	% Failure compared to EU standards ($N = 26$)		
	FC mandatory (2000/100 mL)	FC guidelines (100/100 mL)	FS guidelines (100/100 mL)
1	0	8	23
2	31	73	85
3	0	19	31
4	0	42	35
5	15	53	58

Guidelines five times during the monitoring period while it failed eight times to comply with the guidelines for FS.

3.1. *Salmonella*, *Shigella*, and *Vibrio*

Salmonella and *Shigella* are pathogens that are distributed worldwide and transmitted mainly through food and water ingestion. Their presence in all types of water, including recreational waters, render that water unfit for human use. *Salmonella* was isolated one time each from water samples taken from both location 4 and location 5. *Shigella* was not isolated from any sample during the monitoring program (Table 5).

Vibrio includes several species, the most important of which is *V. cholera*. Noncholera species were isolated on several occasions at locations 2, 4, and 5. Seawater samples from locations 1 and 3 were free from any of these three pathogens.

Salmonella was isolated from locations 2, 4, and 5 during the period 10/10/2002 to 27/2/2003 while *Vibrio* spp. were isolated during the period 26/9/2002 to 27/2/2003 at locations 4 and 5. *Vibrio* isolation showed no seasonality in location 2.

χ^2 tests were used to detect possible significant correlations between the presence of *Salmonella* and the level of fecal indicators (FC and FS), the *Pseudomonas* count, and the yeast and mold counts. A similar test was used for *Vibrio*. Table 6 shows that at all instances when the levels of FC and FS were below 200 cfu/100 g, no *Salmonella* was isolated. All incidences of *Salmonella* isolation were associated with counts higher than 200 cfu/100 g. χ^2 test results showed very high significance for both FC and FS. With regard to *Pseudomonas* counts, only in 3 of 95 instances when the level was lower than 100 cfu/100 g, was *Salmonella* isolated. *Pseudomonas* also correlated significantly with *Salmonella*. Yeast and mold count did not show significant correlation with *Salmonella*.

All measured microbiological parameters correlated significantly with *Vibrio* isolation. Table 7 presents the

Table 5
Number of isolated *Salmonella*, *Shigella*, and *Vibrio* from sand and seawater

Location no.	Number of isolation incidences of 26 sampling occasions for each location					
	<i>Salmonella</i>		<i>Shigella</i>		<i>Vibrio</i>	
	Sand	Seawater	Sand	Seawater	Sand	Seawater
1	0	0	0	0	1	0
2	1	0	0	0	10	2
3	0	0	0	0	0	0
4	4	1	0	0	9	3
5	4	1	0	0	9	5
Total	9/130	2/130	0/130	0/130	29/130	10/130

Table 6
Statistical analysis (χ^2) for the relation between microbial indicators and *Salmonella* ($N = 130$)

Indicator parameter	<i>Salmonella</i>		Significance level
	Negative	Positive	
Fecal streptococci range			
0–200	54	0	0.009
> 200	67	9	
Fecal coliform range			
0–200	62	0	0.003
> 200	59	9	
<i>Pseudomonas</i> range			
0–100	92	3	0.005
> 100	29	6	
Yeast and mold range			
0–100	68	3	0.184
> 100	53	6	

Correlation is significant at the 0.05 level.

Table 7
Statistical analysis (χ^2 test) for the relation between microbial indicators and *Vibrio* in sand ($N = 130$)

Indicator parameter	<i>Vibrio</i>		Significance level
	Negative	Positive	
Fecal coliform range			
0–200	60	2	0.000
> 200	41	27	
Fecal streptococci range			
0–200	51	3	0.000
> 200	50	26	
<i>Pseudomonas</i> range			
0–100	85	10	0.000
> 100	16	19	
Yeast and mold range			
0–100	61	10	0.013
> 100	40	19	

Correlation is significant at the 0.05 level.

statistical analysis of the studied microbial indicators and *Vibrio* in sand.

3.2. *Pseudomonas*, yeast and molds

Table 8 presents the minima, maxima, medians, geometric means, and averages of sand sample contents of *Pseudomonas* and yeast and molds in the monitored locations, while Fig. 2 shows the average values of FC, FS, *Pseudomonas*, and yeast and molds of the studied locations.

Fungi were implicated as a cause of skin infection and thought to be transmitted from contact with infested sands and soil. In this research, as shown in Table 8, locations 2 and 5 showed the highest levels of yeast and mold. *Pseudomonas* could be a good indicator of fecal pollution in sand since it correlated with *Salmonella* (Table 6) and with *Vibrio* (Table 7). Yeast and mold showed a statistically significant correlation with *Vibrio* (Table 7) but not with *Salmonella* (Table 6).

4. Discussion

The wide differences between locations 4 and 5 with regard to the percentage of compliance failure despite the fact that the two locations are only about 300 m apart could be interpreted by the fact that the current direction was toward location 5 most of the monitoring period. Vieira et al. (2001) provided a similar interpretation. This finding suggests that any future monitoring program should take into consideration a daily

record of current direction and, if possible, current speed.

The highest concentration of fecal indicators was found in an area receiving land runoff during the rainy season. In another study and during the summer period, no *E. coli* were isolated from all sampling points, whereas, in autumn, the organism was isolated in most of the sampling points used in the study (Divizia et al., 1997).

In a study by Vidal and Lucena (1997), the impact of heavy rains on the microbiological quality of water persisted a few days and depended on the amount and intensity of rain and weather conditions after the rain episode.

With regard to the EU standards, a general higher compliance failure percentage was associated with FS than with FC. With respect to failure to comply with the FS guidelines, locations 2 and 5 exhibited the highest percentages (84.6% and 57.7%, respectively). A similar pattern, yet with lower percentages (73% and 53%, respectively), of failure to comply with the FC guideline was obtained for locations 2 and 5. Location 3 complied with the mandatory standards of FC and failed the FC guidelines four times; it failed the FS guidelines eight times. Kinzelman et al. (2003) found similar results with a higher failure frequency using enterococci compared with *E. coli*. Noble et al. (2003) found that 99% of failures were due to enterococci during storm periods with 60% of failures occurring during summer periods.

One possible explanation for the consistently higher rate of FS standard failures is that they survive longer in the marine environment than total coliform (TC) or FC. Hanes and Fragala (1967) found that *E. coli* survival in marine water was 0.8 day while enterococci (a specific group of FS) survival was 2.4 days. Sieracki (1980) found that *E. coli* degraded more rapidly with increased sunlight intensity than did enterococci, a finding that

Table 8
Summary results of sand *Pseudomonas* and yeast and molds (cfu/100 g)

Location no.		Mini	Max	Median	Geometric mean	Average
1	Pseudo	1	200	10.5	8	21
	Yeast	8	800	24.5	32	72
2	Pseudo	1	900	160	106	245
	Yeast	1	2300	330	243	444
3	Pseudo	1	21	1	2	5
	Yeast	1	42	12	9	14
4	Pseudo	11	400	30.5	44	80
	Yeast	11	820	121	86	162
5	Pseudo	15	310	65	74	110
	Yeast	12	420	205	173	225

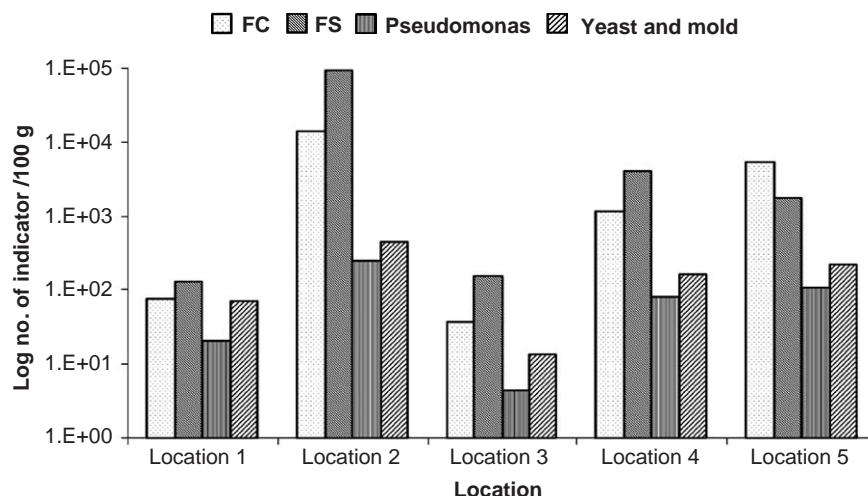


Fig. 2. Average sand microbiological parameters in all locations.

was recently confirmed for bacterial samples from southern California (Noble et al., 2001).

Generally, fecal indicator concentration was higher in sand than in the corresponding water column. The highest ratio obtained in this study was for FS in location 4 (1:29.1). This is considered a small ratio compared to the finding of Crabill et al. (1999); they identified sites with high FC counts, averaging 2200 times the FC counts in the water column. This may be due to different types of samples; they obtained bottom samples while the authors obtained intertidal sand.

In this study, it was found that the increase and decrease in FC numbers in sand is almost always associated with a corresponding increase or decrease in their number in seawater. This may lead to the conclusion that polluted seawater increases the number of FC, which has a longer survival in sand, and sand mixes with the sea during rough weather and high tide where sand is resuspended and shaken, releasing the bacteria on its surface. This assumption is supported by the findings of several investigators that the rate of die off is low in sediments as they provide a favorable, attached, and nonstarvation environment for bacteria (Davies et al., 1995; Howell et al., 1996). Some fecal pathogens have been shown to survive for longer in sediments than in surface water (Ashbolt et al., 1993; Nix et al., 1993; Ghinsberg et al., 1994; Howell et al., 1996).

Observations of lower enteric bacteria survival rates in natural seawater than in sterile seawater (Gauthier et al., 1987; Gonzalez et al., 1992) suggest involvement of biological processes. Le Guyader et al. (1991) provided support for this hypothesis by measuring earlier and faster declines of *E. coli* viability (cfu) in seawater and sediment during a 13-day period when indigenous seawater flora were present as compared to sterile conditions. Predation (Davies et al., 1995; Gonzalez et al., 1992; Greenberg, 1956; Mitchell and Nevo, 1965; Mitchell et al., 1967; Guelin et al., 1967; Mitchel and Morris, 1969; Enzinger and Cooper, 1976; Barcina et al., 1992), competition (Jannasch, 1968; Le Guyader et al., 1991; Greenberg, 1956; Mitchell, 1968), and bacteriophages (Guelin et al., 1967; Carlucci and Pramer, 1960a–c) have been implicated in reducing enteric bacterial concentrations in seawater.

Variation in the numbers of bacteria in sand and seawater from the five locations in the middle camps beach may be a result of varying sources of pollution. Another factor that may have produced a dramatic effect at location 4 was the height of the sand at the intersection between water and land where intertidal waves have a shorter contact time than in other locations.

Salmonella is commonly present in sewage effluent that can contaminate recreational waters. Water microbiology quality standards for recreational waters are based on coliform indicators as predictors of the

presence of pathogenic microorganisms (EEC, 1976; Cabelli et al., 1982). While epidemiological studies constituted the basis of water quality standards in the USA (Cabelli et al., 1982), such studies were not used for standard development in Europe. The objectives of the European Community Bathing Water Directive are to protect the environment and public health (EEC, 1976). Their principal microbiological parameters are total and FC.

The reliability of the indicator-based standards to predict the presence of pathogenic microorganisms, such as *Salmonella*, is still a matter of debate (Morinigo et al., 1993). There is general agreement that *Salmonella* is present at high densities of indicator organisms (Morinigo et al., 1993). However, detection of *Salmonella* in the absence of indicators of fecal pollution has also been reported (Morinigo et al., 1993; Gales and Baleux, 1992). Several epidemiological studies have demonstrated an increasing relative risk of gastroenteritis for bathers exposed to as few as 20 cfu/100 mL (Ferley et al., 1989) or 35 cfu/100 mL (Cabelli et al., 1982; Kay et al., 1994) FS.

The highest incidence of *Salmonella* isolation (15.4%) was obtained from sand samples at locations 4 and 5 in comparison to seawater samples with only 3.8% at those locations. This finding agreed with that of Martinez-Manzanares et al. (1992), where all studied microorganisms were found to be higher in sediments than in the overlying water. Obiri-Danso and Jones (2000), during a 12-month survey, failed to isolate any *Salmonella* from sand samples collected from northwest England. This may be attributed to variations in climate and quantity and quality of sewage. *Salmonella* was isolated with a frequency of 3.8% from seawater samples from locations 4 and 5. This percentage agrees with findings by Morinigo et al. (1990, 1993).

The isolation of *Salmonella* on several occasions during this study from sand and the failure to do so from seawater favors the use of sand samples rather than seawater samples in cases in which *Salmonella* is suspected. *Salmonella* is clearly associated with contaminants from human origin. This assumption was due to the isolation of *Salmonella* from locations that are exposed to sewage (e.g., 2, 4, and 5).

The aim of the present study was to assess possible associations between the presence of *Salmonella* and the concentrations of indicator organisms in relation to the established standards. A significantly higher incidence of *Salmonella* isolation ($P = 0.003$) was found in samples containing FC levels higher than the recommended standards (>200 cfu/100 mL). Similar findings were reported by Polo et al. (1998). A similar significant correlation ($P = 0.009$) was found between *Salmonella* and FS levels higher than 200 cfu/100 mL. *Pseudomonas* levels were grouped into less than 100 cfu and higher than 100 cfu and when tested using χ^2 a significant

($P = 0.05$) correlation was obtained. No significant correlation between *Salmonella* and counts of yeast and mold were obtained when they were subjected to grouping in a manner similar to that of *Pseudomonas*.

Geldreich (1970) had concluded that 200 FCs per 100 mL is the limit above which a sharp increase in the frequency of *Salmonella* detection incidents is expected. Gallacher and Spino (1968) emphasized that a correlation between the levels of total and FCs and the possibility of isolating pathogens would be enormously valuable in setting reliable bacteriological standards, particularly for recreation and fishing uses.

Several studies were conducted for the purpose of correlating the densities of fecal indicators with the presence of *Salmonella*. In the Mediterranean Sea, Papadakis et al. (1998) recorded a better correlation of the coliform group with *Salmonella* isolation, compared with the other indicators. Morinigo et al. (1990) showed that FC and *Clostridium perfringens* were most closely related to *Salmonella* spp. Borrego et al. (1991) found that in polluted marine areas FC, FS, and coliphage correlated well with *Pseudomonas aeruginosa*, *Aeromonas hydrophila* and *Candida albicans*. Yoshpe-Purer and Golderman (1987) showed that total and FC correlated well with the presence of *P. aeruginosa* in bathing beaches. Solic and Krstulovic (1994) described a moderate positive relationship between the three indicators and the pathogen *Staphylococcus aureus* in Croatia. Hoi et al. (1998) demonstrated significant correlations between the occurrence of coliform bacteria and that of FS on the one hand and between coliform bacteria and *Vibrio vulnificus* on the other. Efstratiou et al. (1998) demonstrated that FCs were good predictors of *C. albicans* in moderately polluted areas. Furthermore, studies have often found poor correlations between *E. coli* and particular pathogens (Borrego et al., 1987; Carter et al., 1987; Chauret et al., 1995; Dutka et al., 1987; Sinton et al., 1993a, b). Yet, one may expect that *E. coli* may still serve as an indicator of health risk, rather than as an indicator of particular pathogens.

The isolation of *P. aeruginosa* from almost all sand samples should be considered an alarming factor. *P. aeruginosa* showed a die off rate similar to that of FS and slower than that of FC (De Vicente et al., 1998). A probability of correspondence between the presence of this bacterium and the occurrence of secondary gastrointestinal infection was found indicating a need for the inclusion of other microorganisms, one of which may be *P. aeruginosa*, as indicators of health risk associated with drinking waters in Mexico (De Victorica and Galván, 2001).

References

Affi, S., 1999. Identification and evaluation of seawater and beach quality state in Gaza Governorate. Final Report, Environmental and Rural Research Center, Islamic University, Gaza.

- Affi, S., Elmanama, A., Shubair, M., 2000. Microbiological assessment of beach quality in Gaza Strip. Egypt. J. Med. Lab. Sci. 9.
- American Public Health Association (APHA), 1995. Standard Methods for the Examination of Water and Wastewater, 19th ed. American Public Health Association, Washington DC.
- Arakel, A.V., 1995. Towards developing sediment quality assessment guidelines for aquatic systems: an Australian perspective. Aust. Earth Sci. 42, 335–369.
- Ashbolt, N., Grohmann, G., Kueh, C., 1993. Significance of specific bacterial pathogens in the assessment of polluted receiving waters of Sydney. Water Sci. Technol. 27, 449–452.
- Barcina, I., Gonzalez, J., Iriberry, J., Egea, L., 1992. Role of protozoa in the regulation of enteric bacteria populations in seawater. Marine Microb. Food Webs 5, 179–188.
- Baron, E., Finegold, S., 1990. Diagnostic Microbiology, eighth ed. The C.V. Mosby Company, Philadelphia.
- Bartram, J., Rees, G., 2000. Monitoring Bathing Water. E & FN SPON.
- Borrego, J., Morinigo, M., Vicente, A., Cornax, R., Romero, P., 1987. Coliphages as an indicator of fecal pollution in water. Its relationship with indicator and pathogen microorganisms. Water Res. 21, 1473–1480.
- Borrego, J., Romero, P., Mariano, F., 1991. Epidemiological study on bathers from selected beaches in Malga. MAP Tech. Rep. Ser. nBO 53, 1–27.
- Cabelli, V., Dufour, P., McCabe, L., 1982. Swimming-associated gastroenteritis and water quality. Am. J. Epidemiol. 115, 606–616.
- Carlucci, A., Pramer, D., 1960a. An evaluation of factors effecting the survival of *Escherichia coli* in seawater. Experimental procedures. Appl. Environ. Microbiol. 8, 243–247.
- Carlucci, A., Pramer, D., 1960b. An evaluation of factors effecting the survival of *Escherichia coli* in seawater: salinity, pH, and nutrients. Appl. Environ. Microbiol. 8, 247–250.
- Carlucci, A., Pramer, D., 1960c. An evaluation of factors effecting the survival of *Escherichia coli* in seawater. Bacteriophages. Appl. Environ. Microbiol. 8, 254–256.
- Carter, A., Pacha, R., Clark, G., Williams, E., 1987. Seasonal occurrence of *Campylobacter* spp. in surface waters and their correlation with standard indicator bacteria. Appl. Environ. Microbiol. 53, 523–526.
- Chauret, C., Armstrong, N., Fisher, J., Sharma, R., Springthorpe, V.S., Sattar, S.A., 1995. *Cryptosporidium* and *Giardia* in water in the Ottawa (Canada) region: correlation with microbial indicators of water quality. J. Am. Wat. Works Assoc. 87 (11), 76–84.
- Crabill, C., Donald, R., Snelling, J., Foust, R., Southam, G., 1999. Impact of sediment fecal coliform reservoirs on seasonal water quality in Oak Creek, Arizona. Water Res. 33 (9), 2163–2171.
- Davies, C., Long, J., Donald, M., Ashbolt, N., 1995. Survival of fecal microorganisms in marine and freshwater sediments. Appl. Environ. Microbiol. 61 (5), 1888–1896.
- De Vicente, A., Aviles, M., Borrego, J.J., Romero, P., 1998. Die-off and survival of *Pseudomonas aeruginosa* in seawater. Z. Bakteriell. Mikrobiol. Hyg. [B] 186 (3), 261–272.
- De Victorica, J., Galván, M., 2001. *Pseudomonas aeruginosa* as an indicator of health risk in water for human consumption. Water Sci. Technol. 43 (12), 49–52.
- Divizia, M., Ruscio, V., Donia, D., ElGhazzawi, E., Elcherbini, E., Gabrieli, R., Gamil, F., Kader, O., Zaki, A., Renganthan, E., Pana, A., 1997. Microbiological quality of coastal seawater of Alexandria, Egypt. Ann. Igo. 9 (4), 289–294.
- Dumontet, S., Krovacek, K., Svenson, V., Baloda, S., Figliuolo, G., 2000. Prevalence and diversity of *Aeromonas* and *Vibrio* spp. in coastal waters in Southern Italy. Comp. Immun. Microbiol. Infect. Dis. 23, 53–57.

- Dutka, B., Shaarawi, A., Martins, M., 1987. North and south American studies on the potential of coliphages as a water quality indicator. *Water Res.* 21, 1127–1135.
- EEC (European Economic Community), 1976. Council directive of 8 December 1975 concerning the quality of bathing water. *Off. J. Eur. Commun.* 19 (L/31), 1–7.
- Efstratiou, M., Mavridou, A., Richardson, S., Papadakis, J., 1998. Correlation of bacterial indicator organisms with *Salmonella* spp. *Staphylococcus aureus* and *Candida albicans* in seawater. *Lett. Appl. Microbiol.* 26, 342–346.
- Enzinger, R., Cooper, R., 1976. Role of bacteria and protozoa in the removal of *Escherichia coli* from estuarine waters. *Appl. Environ. Microbiol.* 31, 758–763.
- Ferley, J.P., Zmirou, D., Balducci, F., Baleux, B., Fera, P., Larbaigt, G., Jacq, E., Moissonnier, B., Blineau, A., Boudot, J., 1989. Epidemiological significance of microbiological pollution criteria for river recreational waters. *Int. J. Epidemiol.* 18, 198–205.
- Gales, P., Baleux, B., 1992. Influence of the drainage basin input on a pathogenic bacteria (*Salmonella*) contamination of a Mediterranean lagoon (the Thau lagoon-France) and the survival of this bacteria in brackish water. *Water Sci. Technol.* 25, 105–114.
- Gallacher, T., Spino, D., 1968. The significance of numbers of coliform bacteria as an indicator of enteric pathogens. *Water Res.* 2, 169–175.
- Gauthier, M., Munro, P., Mohajer, S., 1987. Influence of salts and sodium chloride on the recovery of *Escherichia coli* from seawater. *Curr. Microbiol.* 15, 5–10.
- Geldreich, E., 1970. Applying bacteriological parameters to recreational water quality. *J. Am. Water Works Assoc.* 62, 113–120.
- Gerba, C., McLeod, J., 1976. Effects of sediments on the survival of *Escherichia coli* in marine waters. *Appl. Environ. Microbiol.* 32, 114–120.
- Ghinsberg, R., Leibowitz, P., Witkin, H., Mates, A., Seinberg, Y., Bar, D., Nitzan, Y., Rogol, M., 1994. Monitoring of selected bacterial and fungi in sand and seawater along the Tel-Aviv Coast. *MAP Tech. Rep. Ser.* 87, 65–81.
- Gonzalez, J., Iriberry, J., Egea, L., Barcina, I., 1992. Characterization of culturability, protistan grazing, and death of enteric bacteria in aquatic ecosystems. *Appl. Environ. Microbiol.* 58, 998–1004.
- Greenberg, A., 1956. Survival of enteric organisms in sea water. *Public Health Rep.* 71, 77–86.
- Guelin, A., Lepine, P., Lamblin, D., 1967. Pouvoir bactéricide des eaux polluées et rôle de *Bdellovibrio bacteriovorus*. *Ann. Inst. Pasteur Paris* 113, 660–665.
- Hanes, N., Fragala, C., 1967. Effect of seawater concentration on the survival of indicator bacteria. *J. Water Pollut. Control Fed.* 39, 97.
- Hoi, L., Larsen, J., Dalsgaard, I., Dalsgaard, A., 1998. Occurrence of *Vibrio vulnificus* biotypes in Danish marine environments. *Appl. Environ. Microbiol.* 64, 7–13.
- Hood, M., Ness, G., 1982. Survival of *Vibrio cholerae* and *Escherichia coli* in estuarine waters and sediments. *Appl. Environ. Microbiol.* 43, 578–584.
- Howell, J., Coyne, M., Cornelius, P., 1996. Effect of sediment particle size and temperature on fecal bacteria mortality rates and the fecal coliform/fecal streptococci ratio. *J. Environ. Qual.* 25 (6), 1216–1220.
- Jannasch, H., 1968. Competitive elimination of Enterobacteriaceae from seawater. *Appl. Microbiol.* 16, 1616–1618.
- Kay, D., Fleisher, J., Salmon, R., Jones, F., Wyer, M., Godfree, A., Zelenauch-Jacqotte, Z., Shore, R., 1994. Predicting likelihood of gastroenteritis from sea bathing: results from randomized exposure. *Lancet* 344 (October 1), 905–909.
- Kinzelman, J., Ng, C., Jackson, E., Gradus, S., Bagley, R., 2003. Enterococci as indicators of lake Michigan recreational water quality: comparison of two methodologies and their impacts on public health regulatory events. *Appl. Environ. Microbiol.* 1 (69), 92–96.
- LaLiberte, P., Grimes, D., 1982. Survival of *Escherichia coli* in Lake bottom sediments. *Appl. Environ. Microbiol.* 43, 623–628.
- Le Guyader, F., Pommepuy, M., Cormier, M., 1991. Implantation of *Escherichia coli* in pilot experiments and the influence of competition on the flora. *Can. J. Microbiol.* 37, 116–121.
- Martinez-Manzanares, E., Morinigo, M., Castro, D., Balebona, M., Sanchez, J., Borrego, J., 1992. Influence of the fecal pollution of marine sediments on the microbial contents of shellfish. *Mar. Pollut. Bull.* 24 (7), 342–349.
- Mendes, B., Nascimento, M., Oliveira, J., 1993. Preliminary characterization and proposal of microbiological quality standards of sand beaches. *Water Sci. Technol.* 27 (3–4), 453–456.
- Mitchell, R., 1968. Factors affecting the decline of non-marine organisms in sea water. *Water Res.* 2, 535–543.
- Mitchel, R., Morris, J., 1969. The fate of intestinal bacteria in the sea. In: Jenkins, S.H. (Ed.), *Advances in Water Pollution Research. Proceedings of the Fourth International Conference, Prague.* Pergamon Press, New York, pp. 811–817.
- Mitchell, R., Nevo, Z., 1965. Decomposition of structural polysaccharides of bacteria by marine micro-organisms. *Nature* 205, 1007–1008.
- Mitchell, R., Yankofsky, S., Jannasch, H., 1967. Lysis of *Escherichia coli* by marine microorganisms. *Nature* 215, 891–893.
- Morinigo, M.A., Cornax, R., Munoz, M.A., Romero, P., Borrego, J.J., 1990. Relationship between *Salmonella* spp. and indicator microorganisms in polluted natural waters. *Water Res.* 24, 117–120.
- Morinigo, M.A., Martnez-Manzanares, E., Munoz, M.A., Balebona, M.C., Borrego, J.J., 1993. Reliability of several microorganisms to indicate the presence of *Salmonella* in natural waters. *Water Sci. Technol.* 27, 471–474.
- Nix, P., Daykin, M., Vilkas, K., 1993. Sediment bags as an integrator of fecal contamination in aquatic systems. *Water Res.* 27 (10), 1569–1576.
- Noble, R., Ackerman, D., Lee, I., Weisberg, S., 2001. Impacts of various types of anthropogenic inputs on coastal waters of Southern California: an integrated approach. In: *American Society for Limnology and Oceanography. ASLO Press, Albuquerque, NM.*
- Noble, R., Moore, D., Leecaster, M., McGee, C., Weisberg, S., 2003. Comparison of total coliform, fecal coliform, and enterococcus bacterial indicator response for ocean recreational water quality testing. *Water Res.* 37, 1637–1643.
- Obiri-Danso, K., Jones, K., 2000. Intertidal sediments as reservoirs for hippurate negative Campylobacters, Salmonellae and Fecal Indicators in three recognized bathing waters in North West England. *Water Res.* 34 (2), 519–527.
- Papadakis, J., Mavirdou, A., Richardson, S.C., Lambiri, M., Velonakis, E., 1998. Relation between densities of indicator organisms and microbial pathogens in seawater. *Rapports, Commission Internationale pour l'Exploration Scientifique de la Mer Mediterranee* 31(2), M-II9, p. 177.
- Polo, F., Figueras, M.J., Inza, I., Sala, J., Fleisher, J.M., Guarro, J., 1998. Relationship between presence of *Salmonella* and indicators of faecal pollution in aquatic habitats. *FEMS Microbiol. Lett.* 160, 253–256.
- Roses Codinachs, M., Isern Vins, A., Ferrer Escobar, M., Fernandez Perez, F., 1988. Microbiological contamination of the sand from the Barcelona city beaches. *Rev. Sanidad e Higiene Publ.* 62 (5–8), 1537–1544.
- Sieracki, M., 1980. The effects of short exposures of natural sunlight on the decay rates of enteric bacteria, coliphage in a simulated sewage outfall microcosm. *M.Sc. Thesis, Department of Biological Sciences, University of Rhode Island, Providence, RI.*

- Sinton, L., Donnison, A., Hastie, C., 1993a. Faecal streptococci as faecal pollution indicators: a review. Part I: Taxonomy and enumeration. NZ. J. Mar. Freshwater Res. 27, 101–115.
- Sinton, L., Donnison, A., Hastie, C., 1993b. Faecal streptococci as faecal pollution indicators: a review. Part II: Sanitary significance, survival, and use. NZ. J. Mar. Freshwater Res. 27, 117–137.
- Solic, M., Krstulovic, N., 1994. Presence and survival of *Staphylococcus aureus* in the coastal area of Split (Adriatic Sea). Mar. Pollut. Bull. 28, 696–700.
- Van Donsel, D., Geldreich, E., 1971. Relationship of salmonellae to fecal coliforms in bottom sediments. Water Resour. 5, 1079–1087.
- Vidal, J., Lucena, F., 1997. Effect of the rains on microbiological quality of bathing waters in Mediterranean areas. Technical feasibility of an a priori measurement approach for managing bathing water quality. Report of the Workshop Held in Sitges (Spain) on April 26–29.
- Vieira, R., Rodrigues, D., Menezes, E., Evangelista, N., Dos Reis, E., Barreto, L., Gonçalves, F., 2001. Microbial contamination of sand from major beaches in Fortaleza, Ceará state, Brazil. Braz. J. Microbiol. 32 (2).
- WHO, 1995. Manual for Recreational water and Beach Quality Monitoring and Assessment. Draft. WHO, Regional Office for Europe, European Centre for Environment and Health.
- WHO, 1998. Draft Guidelines for Safe Recreational Water Environment: Coastal and Fresh Water. World Health Organization, Geneva.
- Yoshpe-Purer, Y., Golderman, S., 1987. Occurrence of *Staphylococcus aureus* and *Pseudomonas aeruginosa* in Israeli coastal water. Appl. Environ. Microbiol. 53, 1138–1141.