Heliyon 5 (2019) e01893

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.heliyon.com

Heliyon

Recreational water quality status of the Kidd's Beach as determined by its physicochemical and bacteriological quality parameters

Oluwaseun O. Adeniji [a,b,*], Timothy Sibanda [c], Anthony I. Okoh [a,b]

[a] SAMRC Microbial Water Quality Monitoring Centre, University of Fort Hare, Alice, 5700, South Africa
[b] Applied and Environmental Microbiology Research Group (AEMREG), Department of Biochemistry and Microbiology, University of Fort Hare, Private Bag X1314, Alice, 5700, Eastern Cape, South Africa
[c] Department of Biological Sciences, University of Namibia, Private Bag 13301, 340 Mandume Ndemufayo Ave, Pioneers Park, Windhoek, Namibia

ARTICLE INFO

Keywords:
Environmental science
Public health
Microbiology
Kidd's beach
Physicochemical
Recreation
Enterococcus

ABSTRACT

Coastal water resources are habitually exposed to indiscriminate anthropogenic pollution. However, due to their negative consequences to the public health, recreational waters require continuous monitoring for disease-causing organisms as a way of preventing ailments associated with swimming. As a result, the present study assessed the physicochemical parameters and microbial loads of water samples collected from six different sampling points on Kidd's Beach using standard analytical procedures. Generated data were analysed with One-way ANOVA and spearman correlation (at 95%). The physicochemical qualities varied as follows: pH (7.21–8.23), temperature (18.46–27.63 °C), turbidity (0–25.67 NTU), electrical conductivity (22723–62067 μS/cm), total dissolved solids (7662–31037 mg/L), and salinity (8.95–41.84 PSU). All these measured parameters were significantly different ($P < 0.05$) with respect to the sampling sites. Presumptive *Enterococcus* counts ranged from 64 – 168 CFU/100 mL of water samples. Out of 409 presumptive *Enterococcus* isolates obtained from the culture-based method, 67 were confirmed to be *Enterococcus* by PCR-techniques. From the 67 confirmed isolates, 19(*E. faecalis*) and 40(*E. feacium*) while 8(other species that were non-targeted). Findings from this study shown that Kidd's Beach water samples contain some pathogenic bacteria that pose high risk to the public health and make it to be unfit for recreational use when compared to DWAF and US EPA guidelines. Therefore, effort should be made to strictly control all activities contributing to the level of pollution in the marine environment.

1. Introduction

Seawater pollution has been recognized as a global problem that requires an important public health and environmental concern and the quality of beach water is commonly evaluated in comparison to national and/or international standards which vary from one country to another (Davies et al., 1995; Andraus et al., 2014; Velonakis et al., 2014; Halliday et al., 2014; DEA, 2012). Improper garbage disposal and coastal discharge of untreated domestic sewage, urban storm water runoff and animal waste-rich river discharges all contribute to the contamination of beach sand and coastal seawater (Odonkor and Ampofo, 2013; Boehm, 2014). Recreational coastal waters have previously been reported to be harbouring high density of indicator microorganisms (Ashbolt et al., 2001; Noble et al., 2003; Pachepsky et al., 2016) that could cause health problems such as gastrointestinal, skin, eye and ear infections among swimmers and beach goers (Sabino et al., 2014; Andraus et al., 2014). Indicator organisms are bacteria that are used as a signal of quality/hygienic condition in the environment (air, food and water). To determine the potential presence of enteric pathogens in beach sand and/or coastal water, indicator organisms are currently the functional standard, given their association with fresh faecal pollution (NRC, 2004; Harwood et al., 2017) and hence pathogens (WHO, 1998; Graciaa et al., 2018). *Enterococcus* and *E. coli* are currently regarded as indicator organisms of choice for the determination of the microbiological quality of marine water (WHO, 2003; Boehm and Sassoubre, 2014). This is because, under an extensive range of environmental conditions such as high temperature and salt concentration, intestinal enterococci have been noted to be able to mimic the fate and dissemination of some of the most environmentally persistent pathogens such as viruses (Wade et al., 2006). *Enterococcus* species such as *E. sulphurous, E. casseliflavus* and *E. mundtii* are mostly found in aquatic milieu. However, *E. faecalis* and *E. faecium* are predominant in human faeces and urban sewage, making

* Corresponding author.
E-mail address: seunola2012@gmail.com (O.O. Adeniji).

https://doi.org/10.1016/j.heliyon.2019.e01893
Received 1 April 2019; Received in revised form 23 May 2019; Accepted 31 May 2019

them faecal indicators of choice (Manero et al., 2002; Tyrell et al., 2002; Boehm, 2014).

Several studies have reported the occurrence of potentially pathogenic *Enterococcus* spp., *Aeromonas* spp. *Staphylococcus* spp., *Pseudomonas* spp., Viruses, *Salmonella, campylobacter*, and *E. coli*, isolated either from beach sand, seawater or in both samples (Elmanama et al., 2005; Curiel-Ayala et al., 2012; Ahmad et al., 2013). Sewage pollution causes more threats to recreational water users than animal faecal contamination (Soller et al., 2010), making it important to detect their sources in recreational water so that proper mitigation approaches can be put into practice, and hazards precisely measured. When *Enterococcus* densities were higher than 35 CFU/100 mL of recreational sea water, Arnold et al. (2016) estimated the risk for contracting diarrhoea at 35 per 1000 for non-swimmers, and 59 per.

Recreational waters with a concentration of $\leq$35 colony forming units (CFU) of culturable enterococci per 100 mL are safe for recreational purposes (Smeets et al., 2010; USEPA, 2012). Therefore, in the interest of public health, beach quality monitoring and assessment based on microbiological and physicochemical parameters are considered a vital part of coastal management programs as recreational activities increase (Bordalo, 2003; Ariza et al., 2007; DEA, 2012). Nutrients in water can also lead to unfavourable fluctuations in physicochemical conditions of water such as turbidity, pH, oxygen availability, biological oxygen demand (BOD), conductivity and total dissolved solids (WHO, 2011). Even slight changes in water physicochemical parameters may affect the quality of water and therefore its usability for recreational purposes (Caissier, 2006; Kolawole et al., 2011; Singh et al., 2013). Against this background, it is noteworthy that no studies have been undertaken to characterise the bacteriological and physicochemical qualities of Kidd's Beach till date. Hence, this study assessed the physicochemical parameters and the bacteriological qualities of Kidd's Beach water to ascertain its fitness for recreational purposes.

2. Materials and methods

2.1. Description of study area and sample collection

Kidd's Beach is situated in Centani in the Eastern Cape Province of South Africa, with geographical coordinates; 33° 9′ 0″ S and 27° 41′ 0″ E. It is a small coastal resort, located on the Mkhanzi River, about 28 km from East London with total area of 2.13 square kilometers with population of 499 (217 males and 282 females as indicated by 2011 Census Report). It is an extremely common holiday destination that has a growing population of permanent residents and tourists, and is mostly used during summer period. Water samples were collected between November 2017 and April 2018 from six sites situated 200 m apart, where swimming activity usually takes place. Sampling sites: KP-1, KP-2 and KP-3 represented collection sites for beach water while KP-4 and KP-5 were canals and site KP-6 was a tidal pool. The six (6) sampling sites were located within the following geographical coordinates: S 33° 08′ 35″ E 27° 42′ 11″, S 33° 08′ 45″ E 27° 42′ 08″, S 33° 08′ 32″ E 27° 42′ 11″, S33° 08′ 48″ E 27° 41′ 59″, S 33° 08′ 46″ E 27° 42′ 21″ and S 33° 08′ 53″ E 27° 42′ 11″ for KP-1, KP-2, KP-3, KP-4, KP-5 and KP-6, respectively.

The samples were collected at 15–30 cm below the surface on the seaward side of a recently broken wave, following the descriptions of DEA (2012). *On-site,* the physicochemical analyses were carried out according to the recommended Standard Methods (APHA, 2005). These parameters tested include; temperature, TDS (total dissolved solids) pH, salinity and electrical conductivity were conducted in triplicates using a portable Hanna multi-parameter device (HI 98185, ROMANIA). A HACH turbidimeter (HACH Company, model 2100P) was used to measure the turbidity. Subsequently, water samples were transported on ice to the Applied and Environmental Microbiology Research Group (AEMREG) laboratory at the University of Fort Hare, Alice, South Africa and bacteriologically examined within 4–6 h of collection.

2.2. Microbiological analysis

2.2.1. Isolation and identification of presumptive enterococci

Samples were serially diluted following which 100 mL of each dilution was filtered through nitrocellulose membrane filters (0.45-μm pore size, Millipore, Ireland) for the isolation and enumeration of the faecal coliforms as described in APHA (2005). Following filtration, the membrane filters were aseptically transferred with sterile forceps onto plates containing Bile Aesculin Azide agar (acc. to ISO 7899-2) and incubated for 48 h at 37 °C. After the incubation period, all brown to black colonies were enumerated as presumptive faecal enterococci and reported as CFU/100 mL surface water. The presumptive isolates were stored at –80 °C in 20% glycerol stock for further assays.

2.2.2. Extraction of DNA and molecular confirmation of presumptive *Enterococcus spp*.

Bacterial DNA was extracted using the boiling method as described by Maugeri et al. (2006). Briefly, colonies were picked and suspended in 200 μL sterile distilled water, vortex and boiled at 100 °C for 15 min. The cell suspension was centrifuged at 15,000 rpm for 15 min, after which the supernatant containing the DNA was collected and stored at -20 °C for downstream processes. Polymerase chain reaction (PCR) amplification of the *Enterococcus* specific *tuf*-gene (amplicon size 112 bp) was then used to confirm the identities of the presumptive enterococci isolates as described by Ke et al. (1999). *Enterococcus feacalis* (DSM 20478) was used as a positive control. PCR amplification was carried out in 25 μL reaction mixture consisting of 12.5 μL of PCR master mix (Thermo Scientific, (EU) Lithuania), 0.5 μL each of the forward and reverse primer (Inqaba Biotech, SA), (Ent1 F-5'TACTGACAAACCATTCATGATG-3' and Ent2 R-5'AACTTCGTCACCAACGCGAAC-3'), 5 μL of DNA template and 6.5 μL of nuclease-free water. The cycling conditions were set as follows: initial denaturation at 94 °C for 3 min, followed by 30 cycles of melting at 94 °C for 30 s, annealing at 53 °C for 45 s, and extension at 72 °C for 60 s. The process was then followed by a final extension step at 72 °C for 7 min. PCR products were resolved by electrophoresis in 2% agarose gel incorporated with 5 μL of ethidium bromide (Sigma-Aldrich, USA) at 100 V for 45 min in 0.5× TBE buffer, and visualized under a UV transilluminator (ALLIANCE 4.7, France).

2.2.3. Speciation of confirmed enterococci isolates

All PCR confirmed enterococci isolates were subjected to another round of PCR to determine their species using species specific primers as described by Jackson et al. (2004). The PCR cycling conditions, primers sets, product size and targeted species are as shown in Table 1. The PCR reaction mixture was constituted as previously described.

2.3. Statistical analysis

Accrued data was subjected to descriptive statistical analysis (95% confidence interval) using IBM SPSS version 23 (IBM, Armonk, NY, USA). One-way analysis of variance (ANOVA) was done to test for differences among the parameters measured with respect to sampling sites. Relationships between groups was determined through correlation analysis and described as significant when P values were less than 0.05. Correlations between the physicochemical properties and *Enterococcus* were observed using a 2-tailed Pearson's correlation analysis.

3. Results and discussion

The mean monthly values recorded for each of the determined physicochemical parameter and presumptive *Enterococcus* counts at each of the 6 sampling sites are presented in Supplementary Table 1. The readings at each sampling site were also combined to give the overall monthly water quality at Kidd's Beach and the results are presented in Fig. 1. Water pH was largely in the neutral range, and ranged between 7.6 in February 2018 to 7.9 in April 2018 whereas water temperature ranged

Table 1
Primers for the speciation of *Enterococcus* species. (are there no genes used to target their strains?).

Target strains	Primer	Sequences (5′–3′)	PCR cycling conditions	Product Size(bp)	Reference
E. faecalis ATCC 19433	FL1	ACTTATGTGACTAACTTAACC	95 °C (4 min), 30 cycles of {94 °C (30 sec), 52 °C (1 min), 72 °C (1 min)}, 72 °C (7 min)	360	Jackson et al. (2004)
	FL2	TAATGGTGAATCTTGGTTTGG			
E. durans ATCC 19432	DU1	CCTACTGATATTAAGACAGCG	95 °C (4 min), 30 cycles of {94 °C (30 sec), 52 °C (1 min), 72 °C (1 min)}, 72 °C (7 min)	295	Jackson et al. (2004)
	DU2	TAATCCTAAGATAGGTGTTTG			
E. casseliflavus ATCC 25788	CA1	TCCTGAATTAGGTGAAAAAAC	95 °C (4 min), 30 cycles of {94 °C (30 sec), 52 °C (1 min), 72 °C (1 min)}, 72 °C (7 min)	288	Jackson et al. (2004)
	CA2	GCTAGTTTACCGTCTTTAACG			
E. faecium ATCC19434	FM1	GAAAAAACAATAGAAGAATTAT	95 °C (4 min), 30 cycles of {94 °C (30 sec), 48 °C (1 min), 72 °C (1 min)}, 72 °C (7 min)	215	Jackson et al. (2004)
	FM2	TGCTTTTTTGAATTCTTCTTTA			
E. hirae ATCC 8043	HI1	CTTTCTGATATGGATGCTGTC	95 °C (4 min), 30 cycles of {94 °C (30 sec), 48 °C (1 min), 72 °C (1 min)}, 72 °C (7 min)	187	Jackson et al. (2004)
	HI2	TAAATTCTTCCTTAAATGTTG			

between 20 °C and 25 °C throughout the sampling period. When compared by sampling sites, the pH values at different sampling sites did not significantly differ in the month of November 2017 though they were observed to be significantly different ($P < 0.05$) from site to site in the remainder of the sampling period (December 2017 to April 2018) (Supplementary Table 1). Recreational water with very low or very high values pH may cause problems to the skin and eyes of swimmers (WHO, 2003).

However, both the recorded pH and temperature in this study conformed to the no observable effect level (NOEL) recreational water quality standards set by the Environmental Protection Agency (EPA), ranges of which stand at 6.5–8.5 for pH and 15–35 °C for temperature respectively (DEA, 2012; USEPA, 2015). There was a noticeable trend between the concentration of total dissolved solids (TDS) and electrical conductivity (EC) which were both highest in November 2018 and lowest in April 2018, with concentration ranges of 20 000–25 000 mg/L and 40 000–50 000 μS/cm, respectively. This was confirmed by correlation analysis which showed a positive relationship between these two parameters ($P < 0.05$; Table 2). This result agrees with the report of Sunitha et al. (2005) that EC tends to have higher levels of correlation with many of the water quality parameters.

While there is no precise recommendation obtainable for the EC levels of recreational coastal waters, very high values are likely in ocean water on the basis of the level of fluctuations in temperature (FEI, 2014) and quantity of dissolved solids (Yilmaz and Koç, 2014). River discharges containing domestic sewage effluents have also been noted to increase EC (Verma et al., 2012) while freshwater influx as during rainy seasons may actually lower it (Solanki, 2012). Water salinity ranged between 25 PSU to 33 PSU for the entire six-month sampling period.

The turbidity of the water samples for the month of November 2017 exceeded the acceptable range of 0.5–10 NTU for coastal water as set by the Department of Environment and Conservation (New South Wales)

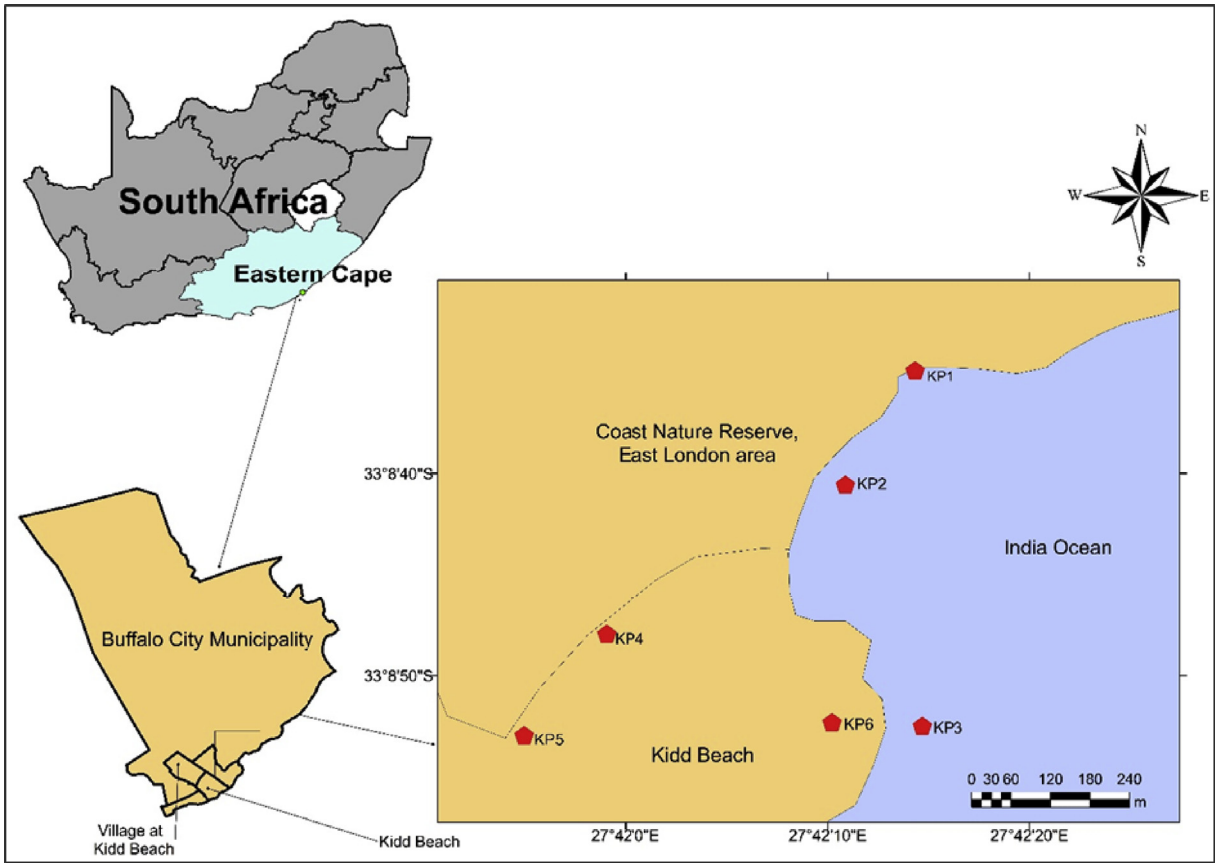

Fig. 1. A map showing the sampling points in Kidd's Beach.

Table 2
Relationship among the physicochemical parameters and *Enterococcus* spp at Kidd's Beach (November 2017–April 2018).

EC	pH	salinity	TDS	Temp	enterococci Count	Turbidity	Statistic	Parameter
1	-0.01478	.930**	1.000**	-.466**	-.377**	0.028715	Pearson Correlation	EC
	0.849237	4.99E-74	0	2.04E-10	4.64E-07	0.711763	Sig. (2-tailed)	
	168	168	168	168	168	168	N	
	1	-0.02242	-0.01491	0.059861	0.015574	-.375**		pH
		0.773006	0.847879	0.440835	0.841193	5.44E-07	Sig. (2-tailed)	
		168	168	168	168	168	N	
		1	.930**	-.450**	-.278**	0.002895		salinity
			4.8E-74	9.73E-10	0.000264	0.970291	Sig. (2-tailed)	
			168	168	168	168	N	
			1	-.466**	-.378**	0.02834		TDS
				1.88E-10	4.36E-07	0.715365	Sig. (2-tailed)	
				168	168	168	N	
				1	.389**	-0.10193		Temp
					1.93E-07	0.18859	Sig. (2-tailed)	
					168	168	N	
					1	-.152*		Count
						0.048785	Sig. (2-tailed)	
						1		Turbidity
							Sig. (2-tailed)	
							N	

N = sample size. EC: Electrical conductivity, TDS total dissolved solids.
*. Correlation is significant at the 0.05 level (2-tailed). **. Correlation is significant at the 0.01 level (2-tailed).

(ANZECC, 2000). However, since this increase in turbidity coincided with the summer season in this part of South Africa, it was attributed to increased river discharges and muddy runoff water that entered into the ocean during that time. Highly turbid water impedes light penetration, reducing the microbial-killing effect of the sun's UV light (EPA, 2009), so that such water is associated with higher levels of pathogenic microorganisms such as bacteria, viruses and parasites (Momba and Kaleni, 2002).

Data for site-specific monthly prevalence of presumptive *Enterococcus* in Kidd's Beach water over the sampling period is presented in Fig. 2. Presumptive *Enterococcus* counts ranged between 64 and 168 CFU/100 mL across the sampling sites. Sampling sites KP4, KP5 and KP6 had significantly higher counts than other sites ($P < 0.05$) this is likely due to the flow of Mcantsi River into the canals which impacts the K4 and K5 sites, and municipal sewage discharge impacting KP6. The presence of these indicators in water connotes faecal contamination, which could be detrimental to public health, effects of which can cascade down to the economy (Gourmelon et al., 2007; Abdelzaher et al., 2010; MacIntyre and de Villiers, 2010). Going by USEPA guidelines, recreational waters with microbial densities greater than the maximum contaminant limit of 33 or 35 CFU/100 mL for *Enterococcus* could depict a serious threat of waterborne infections (USEPA, 1986; USEPA, 2012; Abdelzaher et al., 2010). Reports that correlate the occurrence of bacterial indicators and the occurrence of gastrointestinal illness at swimming beaches suggest that the best indicators of health risk from recreational water contact are *E. coli* (fresh water) and enterococci (marine and fresh water) (Wu et al.,

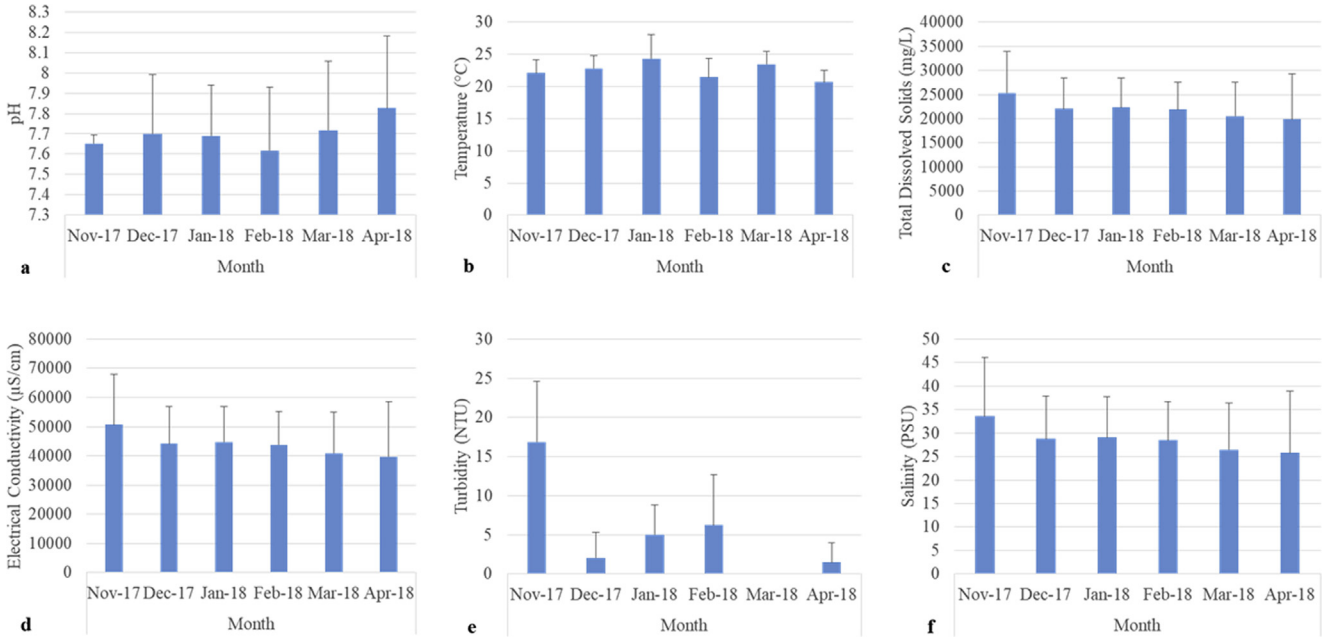

Fig. 2. The physicochemical parameters of coastal water at the Kidd's Beach, November 2017 to April 2018.

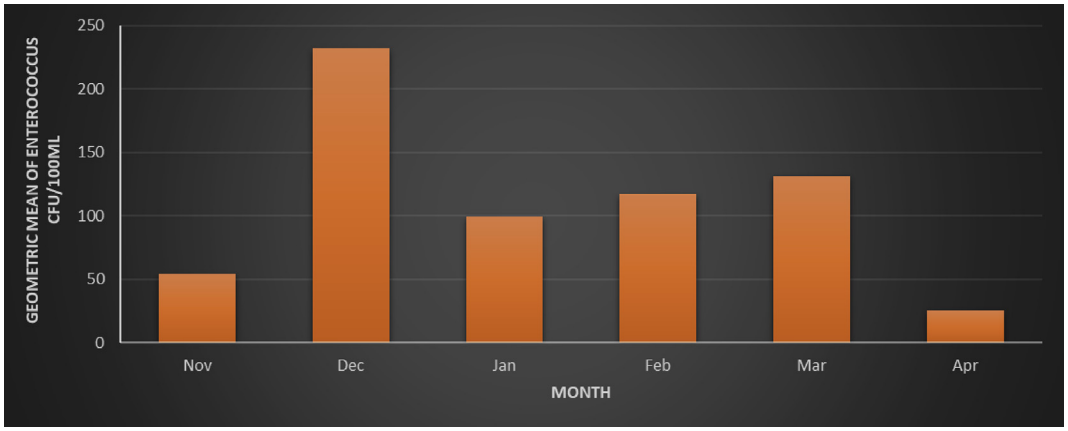

Fig. 3. Geometric mean of *Enterococcus* (CFU/100mL).

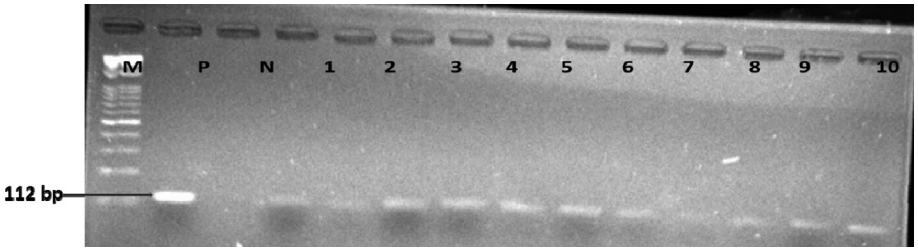

Fig. 4. PCR products of the amplification of tuf-gene. Lane M: molecular weight marker (100 bp); lane N: negative control; lane P: positive control (DSM 20478) lanes 1–10: positive isolates.

2011; Abdelzaher et al., 2010; EPA, 2011). The basis for the use of enterococci as quality indicators for marine recreational water stems from its higher correlation to human pathogens often found in sewage compared to other faecal coliforms (EPA, 2003; Jin et al., 2004). In addition, the enhanced survival of the members of the genus *Enterococcus* in brackish and salty conditions characteristic of marine recreational beaches (Jin et al., 2004), if compared to others. Results of this study indicated that the beach and canal waters were of poor microbiological quality and suggest that swimming or surfing in Kidd's Beach exposes children, elderly persons and people with compromised immune systems to significant health risks. The possible effect is more profound in the Eastern Cape bearing in mind the high number of people with immune systems compromised via HIV/AIDS (TAC, 2018).

Since different water quality parameters are used to determine water quality, correlation analysis was performed to determine possible relationships. Table 2 shows the correlation coefficients of physicochemical water quality parameters and enterococci counts. Some correlations of note included the fact that temperature was positively correlated with *Enterococcus* counts. In addition, there was a highly significant correlation between *Enterococcus* counts and turbidity ($P < 0.01$).

3.1. *Enterococcus confirmation and speciation*

Out of the 409 presumptive *Enterococcus* isolates recovered from the samples, 67 were confirmed to be *Enterococcus* at the genus level. Our findings corroborate with the report of Fergunson et al. (2010) in which about 10% of colonies growing on mEI agar did not belong to *Enterococcus* spp. Therefore, the use of standard procedures such as biochemical tests and molecular characterisations like 16S rRNA gene sequencing for the reassessment of the growing colonies and identification of *Enterococcus* spp at the species level is indispensable. The 67 confirmed enterococci isolates were screened for species level identities targeting the species *E. durans, E. casseliflavus, E. faecalis, E. hirae* and *E. faecium,* due to their re-occurrence in human infections as reported in literature (Murray et al., 1990; Ruoff et al., 1990; Devriese et al., 1995; Tannock and Cook, 2002). A total of 19 (28.4%) isolates were identified to be *E. faecalis* while 40 (59.7%) isolates were identified to be *E. feacium.* However, 8 of the confirmed isolates could not be characterized to species level. Monitoring ratios *of E. faecium* or *E. faecalis* and the dissemination of enterococci in different matrices is considered important in evaluating the value of this group as a faecal indicator in a bid to protect human health (Leclerc et al., 1996). The species distribution of enterococci encountered in recreational waters sampled in the current study showed a higher ratio of *E. faecium* to *E. faecalis* (40:19) as a proportion of the entire species isolated. It is generally accepted that *E. faecium* are more specific to humans (Kühn et al., 2000; de Oliveira and Pinhata, 2008), and compared with other species, they seem to live longer in aquatic milieu which makes them the most common enterococci species found in environmental samples (Leclerc et al., 1996; de Oliveira and Pinhata, 2008). The high frequencies of *E. faecalis* and *E. faecium* in beach water samples collected in the current study therefore indicate possible human faecal contamination.

In addition, these species are often implicated in recreational seawater with enterococci population above acceptable regulatory guidelines (Ferguson et al., 2005). In our findings, *E. faecium* (n = 40) constituted the majority of culturable enterococci isolates, which is similar to reports from France (Łuczkiewicz et al., 2010) and Poland (Leclercq et al., 2013) in which *E. faecium* were the most common isolates among the enterococcal populations with 28.9% (n = 153) of the cases. However, *E. faecalis* was found to be the major enterococcal species in reports from Malaysia (Macedo et al., 2011) and Portugal (Ahmad et al., 2014). The difference could be ascribed to seasonal sample collection as, in a study by Lanthier et al. (2011), *E. faecium, E. faecalis* and *E. durans* were found to be most often isolated in summer, winter and spring respectively. This could be elucidated by the fact that during summer time *E. faecalis* is abundant in livestock faeces while *E. faecium* is the most detected species in winter due to its ability to live longer in aquatic milieu (Dada et al., 2013) (see Figs. 3, 4, 5 and 6).

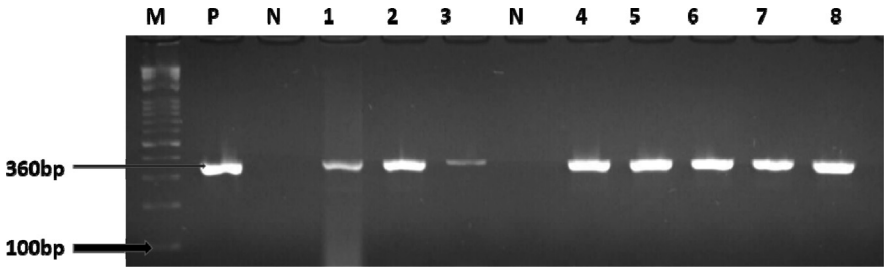

Fig. 5. Gel image showing molecular confirmation of *E. feacalis* (360). Lane M: molecular weight marker (100bp); Lane P: positive control (DSM20478) Lane N: Negative control: Lane 1–3 are positive isolates; N: negative control; Lane 4–8 positive isolates.

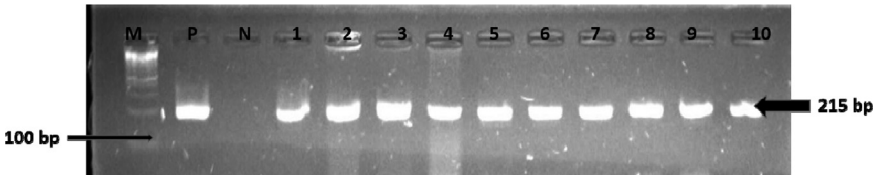

Fig. 6. Gel image showing molecular confirmation of *E. feacium* (215bp). Lane M: molecular weight marker (100bp); Lane P: positive control (DSM 20478) Lane N: Negative control: Lane 1–10 are positive isolates.

4. Conclusions

Pollution of recreational water either by human or animal faecal waste is a major public health hazard. In the present study, the levels of the physicochemical quality parameters of the Kidd's Beach samples complied with set standards for recreational water and as a result may be used for recreational purpose. However, the microbiological quality of the water samples suggests possible health threats if ingested. In like manner, *tuf* and superoxide dismutase (*sodA*) genes are good molecular markers for the identification of *Enterococcus* at the genus and species levels, respectively. High prevalence of *Enterococcus* species may be an indicator of potential health hazards for beach users. Future research should focus on the assessment of beach water for the presence of this bacterium throughout the four season of the year to give room for epidemiological studies to establish the role played by these pathogens.

Declarations

Author contribution statement

Oluwaseun O. Adeniji: Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Anthony I. Okoh: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Tim Sibanda: Analyzed and interpreted the data; Wrote the paper.

Funding statement

This work was supported by the University of Fort Hare and the South African Medical Research Council (SAMRC).

Competing interest statement

The authors declare no conflict of interest.

Additional information

Supplementary content related to this article has been published online at https://doi.org/10.1016/j.heliyon.2019.e01893.

Acknowledgements

The authors are grateful to the University of Fort Hare and the South African Medical Research Council (SAMRC) for financial supports.

References

Abdelzaher, A.M., Wright, M.E., Ortega, C., Solo-Gabriele, H.M., Miller, G., Elmir, S., Newman, X., Shih, P., Bonilla, J.A., Bonilla, T.D., Palmer, C.J., Scott, T., Lukasik, J., Harwood, V.J., McQuaig, S., Sinigalliano, C., Gidley, M., Plano, L.R.W., Zhu, X., Wang, J.D., Fleming, L.E., 2010. Presence of pathogens and indicator microbes at a non-point source subtropical recreational marine beach. Appl. Environ. Microbiol. 76, 724–732.

Ahmad, A., Dada, A., Usup, C.G., Heng, L.Y., 2013. Validation of the *Enterococci* indicator for bacteriological quality monitoring of beaches in Malaysia using a multivariate approach. SpringerPlus 425, 1–18.

Ahmad, A., Dada, A.C., Usup, G., Heng, L.Y., 2014. Occurrence of *Enterococcus* Species with Virulence Markers in an Urban Flow-Influenced Tropical Recreational Beach and Development. U.S. Environmental Protection Agency, Cincinnati, OH. and Ear Infirmary, Boston.

Andraus, S., Pimentel, I.C., Dionísio, J.A., 2014. Microbiological monitoring of seawater and sand of beaches Matinhos, Caiobá e Guaratuba-PR, Brazil. Estud. Biol. 36.

Australian and New Zealand Environment and Conservation Council (ANZECC), 2000. The Guidelines (Paper No 4, Chapters 1–7): Australian and New Zealand Guidelines for Fresh and Marine Water Quality, 1. ANZECC, Wellington, New Zealand, p. 314.

APHA, 2005. Standard Methods for the Examination of Water and Wastewater, twenty-first ed. American Public Health Association, Washington, DC.

Ariza, E., Sardá, R., Jiménez, J.A., Mora, J., Ávila, C.Y., 2007. Beyond performance assessment measurements for beach management: application to Spanish mediterranean beaches. Coast. Manag. 36, 47–66.

Arnold, B.F., Wade, T.J., Benjamin-Chung, J., Schiff, K.C., Griffith, J.F., Dufour, A.P., Weisberg, S.B., Colford Jr., J.M., 2016. Acute gastroenteritis and recreational water: highest burden among young US children. Am. J. Public Health 10, 1690–1697.

Ashbolt, N.J., Grabow, W.O.K., Snozzi, M., 2001. Indicators of microbial water quality. In: Fewtrell, L., Bartram, J. (Eds.), Water Quality: Guidelines, Standards and Health: Risk Assessment and Management for Water Related Infectious Diseases. IWA Publishing, London, pp. 289–316.

Boehm, A., 2014. Enterococci as indicators of environmental fecal contamination. In: non-human sources of fecal contamination. Water Res. 44 (16), 4674–4691.

Boehm, A.B., Sassoubre, L.M., 2014. Enterococci as indicators of environmental fecal contamination. In: Enterococci: from Commensals to Leading Causes of Drug Resistant Infection [Internet]. Massachusetts Eye and Ear Infirmary.

Bordalo, A.A., 2003. Microbiological water quality in urban coastal beaches: the influence of water dynamics and optimization of the sampling strategy. Water Res. 37, 3233–3241.

Caissier, 2006. Regime of rivers. A review. Freshw. Biol. 51, 456–520.

Curiel-Ayala, F., Quinones-Ramirez, E., Pless, R., Gonzalez-Jasso, E., 2012. Comparative studies on *Enterococcus*,*Clostridium perfringens*and *Staphylococcus aureus*as quality indicators in tropical seawater at a Pacific Mexican beach resort. Mar. Pollut. Bull. 10, 2193–2198.

Dada, A.C., Ahmad, A., Usup, G., Heng, L.Y., 2013. Speciation and antimicrobial resistance of Enterococci isolated from recreational beaches in Malaysia. Environ. Monit. Assess. 185, 1583–1599.

Davies, C.M., Long, J.A., Donald, M., Ashbolt, N.J., 1995. Survival of fecal microorganisms in marine and freshwater sediments. Appl. Environ. Microbiol. 5, 1888–1896.
de Oliveira, A.J.F.C., Pinhata, J.M.W., 2008. Antimicrobial resistance and species composition of Enterococcus spp. isolated from waters and sands of marine recreational beaches in South eastern Brazil. Water Res. 8–9, 2242–2250.
Department of Environmental Affairs. (DEA), 2012. Chapter 9: Oceans and Coasts: Ocean and Coasts Ecosystem Services Are Important as They Directly and Indirectly Impact on Human Livelihoods, Food Security and Agriculture; Oceans and Coasts, p. 26. Pretoria, South Africa.
Devriese, L.A., Pot, B., Van Damme, L., Kersters, K., Haesebrouck, F., 1995. Identification of Enterococcus species isolated from foods of animal origin. Int. J. Food Microbiol. 2, 187–197.
Elmanama, A.A., Fahd, M.I., Afifi, S., Abdallah, S., Bahr, S., 2005. Microbiological beach sand quality in Gaza Strip in comparison to seawater quality. Environ. Res. 1, 1–10.
Environmental Protection Department (EPA), 2003. Water Quality Criteria/Standards Adopted in the Asia Pacific Region. Asia Pacific Economic Cooperation Secretariat, Singapore.
Environmental Protection Agency. (EPA), 2009. Technical Support Document for the Endangerment and Cause or Contributes Findings for Greenhouse Gases under Section 202(a) of the Clean Air Act. U.S.
Fondriest Environmental Inc. (FEI), 2014. Conductivity, Salinity and Total Dissolved Solids: Fundamentals of Environmental Measurements, 2014. Available online: http://www.fondriest.com/environmentalmeasurements/parameters/water quality/conductivity-salinity-tds/. (Accessed 23 November 2016). for Brazilian waters. In: Ministry of the Environment (Ed.), Current Conama Resolutions.
Fergunson, D.M., Griffith, J.F., McGee, C.D., Weisberg, S.B., 2010. Comparison of Enterococcus Species Using Enterolert and EPA Method 1600. Southern California Coastal Water Research Project Annual Report.
Ferguson, D.M., Moore, D.F., Getrich, M.A., Zhowandai, M.H., 2005. Enumeration and speciation of enterococci found in marine and intertidal sediments and coastal water in southern California. J. Appl. Microbiol. 3, 598–608.
Gourmelon, M., Caprais, M.P., Ségura, R., Le Mennec, C., Lozach, S., Piriou, J.Y., Rincé, A., 2007. Evaluation of two library-independent microbial source tracking methods to identify sources of fecal contamination in French estuaries. Appl. Environ. Microbiol. 15, 4857–4866.
Graciaa, D.S., Cope, J.R., Roberts, V.A., Cikesh, B.L., Kahler, A.M., Vigar, M., Hilborn, E.D., Wade, T.J., Backer, L.C., Montgomery, S.P., Secor, W.E., 2018. Outbreaks associated with untreated recreational water—United States, 2000–2014. MMWR Morb. Mortal. Wkly. Rep. 67, 701.
Halliday, E., McLellan, S.L., Amaral-Zettler, L.A., Sogin, M.L., Gast, R.J., 2014. Comparison of bacterial communities in sands and water at beaches with bacterial water quality violations. PLoS One 3, e90815.
Harwood, V.J., Shanks, O., Korajkic, A., Verbyla, M.E., Ahmed, W., Iriarte, M., 2017. General and host-associated bacterial indicators of fecal pollution. In: Rose, J.B., Jiménez-Cisneros, B. (Eds.), Global Water Pathogens Project. UNESCO, E. Lans-ing, MI.
Jackson, C.R., Fedorka-Cray, P.J., Barrett, J.B., 2004. Use of a genus- and species-specific multiplex PCR for identification of enterococci. J. Clin. Microbiol. 42, 3558–3565.
Jin, G., Englande, A.J., Bradford, H., Jeng, H.W., 2004. Comparison of *E. coli*, enterococci, and fecal coliform as indicators for brackish water quality assessment. Water Environ. Res. 3, 245–255.
Ke, D., Picard, F.J., Martineau, F., Menard, C., Roy, P.H., Ouellette, M., Bergeron, M.G., 1999. Development of a PCR assay for rapid detection of enterococci. J. Clin. Microbiol. 37, 3497–3503.
Kolawole, O.M., Ajayi, K.T., Olayemi, A.B., Okoh, A.I., 2011. Assessment of water quality in Asa River (Nigeria) and its indigenous Clarias gariepinus fish. Int. J. Environ. Res. Public Health 11, 4332–4352.
Kühn, I., Iversen, A., Burman, L.G., Olsson-Liljequist, B., Franklin, A., Finn, M., 2000. Epidemiology and ecology of enterococci, with special reference to antibiotic resistant strains, in animals, humans and the environment. Example of an ongoing project within the European research programme. Int. J. Antimicrob. Agents 14, 337–342.
Lanthier, M., Scott, A., Zhang, Y., Cloutier, M., Durie, D., Henderson, V.C., Wilkes, G., Lapen, D.R., Topp, E., 2011. Distribution of selected virulence genes and antibiotic resistance in Enterococcus species isolated from the South Nation River drainage basin, Ontario, Canada. J. Appl. Microbiol. 2, 407–421.
Leclerc, H., Devriese, L.A., Mossel, D.A.A., 1996. Taxonomical changes in intestinal (faecal) enterococci and streptococci: consequences on their use as indicators of faecal contamination in drinking water. J. Appl. Bacteriol. 5, 459–466.
Leclercq, R., Oberlï, K., Cattoir, V., Petit, F., 2013. Changes in enterococcal populations and related antibiotic resistance along a medical center-wastewater treatment plant-river continuum. Appl. Environ. Microbiol. 7, 2428–2434.
Macedo, A.S., Freitas, A.R., Abreu, C., Machado, E., Peixe, L., Sousa, J.C., 2011. Characterization of antibiotic resistant enterococci isolated from untreated waters for human consumption in Portugal. Int. J. Food Microbiol. 145, 315–319.
MacIntyre, U.E., de Villiers, F.P.R., 2010. The economic burden of diarrheal disease in a tertiary level hospital, Gauteng, South Africa. J. Infect. Dis. 202, S116–S125.
Manero, A., Vilanova, X., Cerda-Cuellar, M., Blanch, A.R., 2002. Characterization of sewage waters by biochemical figerprinting of Enterococci. Water Res. 36, 2831–2835.
Maugeri, T.L., Carbone, M., Fera, M.T., Gugliandolo, C., 2006. Detection and differentiation of Vibrio vulnificus in seawater and plankton of coastal zone of the Mediterranean Sea. Res. Microbiol. 157, 194–200.
Momba, M.N., Kaleni, P., 2002. Regrowth and survival of indicator microorganisms on the surfaces of household containers used for the storage of drinking water in rural communities of South Africa. Water Res. 3, 3023–3028.
Murray, B.E., Singh, K.V., Heath, J.D., Sharma, B.R., Weinstock, G.M., 1990. Comparison of genomic DNAs of different enterococcal isolates using restriction endonucleases with infrequent recognition sites. J. Clin. Microbiol. 28, 2059–2063.
Noble, R.T., Moore, D.F., Leecaster, M.K., McGee, C.D., Weisberg, S.B., 2003. Comparison of total coliform, fecal coliform, and enterococcus bacterial indicator response for ocean recreational water quality testing. Water Res. 37, 1637–1643.
National Research Council (NRC), 2004. Indicators for Waterborne Pathogens. National Academies Press, Washington, DC.
Odonkor, S.T., Ampofo, J.K., 2013. Escherichia coli as an indicator of bacteriological quality of water: an overview. Microbiol. Res. 4 e2-e2.
Pachepsky, Y., Shelton, D., Dorner, S., Whelan, G., 2016. Can E. coli or thermotolerant coliform concentrations predict pathogen presence or prevalence in irrigation waters? Crit. Rev. Microbiol. 42, 384–393.
Ruoff, K.L., De La Maza, L., Murtagh, M.J., Spargo, J.D., Ferraro, M.J., 1990. Species identities of enterococci isolated from clinical specimens. J. Clin. Microbiol. 28, 435–437.
Sabino, R., Rodrigues, R., Costa, I., Carneiro, C., Cunha, M., Duarte, A., Faria, N., Ferreira, F.C., Gargaté, M.J., Júlio, C., Martins, M.D.L., 2014. Routine screening of harmful microorganisms in beach sands: implications to public health. Sci. Total Environ. 472, 1062–1069.
Singh, K., Singh, B., Singh, R.R., 2013. Effect of land rehabilitation on physicochemical and microbial properties of a sodic soil. Catena 109, 49–57.
Smeets, P.W.M.H., Rietveld, L.C., Van Dijk, J.C., Medema, G.J., 2010. Practical applications of quantitative microbial risk assessment (QMRA) for water safety plans. Water Sci. Technol. 61, 1561–1568.
Solanki, H., 2012. Status of Soils and Water Reservoirs Near Industrial Areas of Baroda: Pollution and Soil; Water Chemistry. Lap Lambert Academic Publishing.
Soller, J.A., Schoen, M.E., Bartrand, T., Ravenscroft, J.E., Ashbolt, N.J., 2010. Estimated human health risks from exposure to recreational waters impacted by human and non-human sources of faecal contamination. Water Res. 4674–4691.
Sunitha, V., Sudarshan, V., Reddy, B.R., 2005. Hydrogeochemistry of groundwater, Gooty area, Anantapur district, Andhra Pradesh, India. Pollut. Res. 24, 217.
Treatment Action Campaign (TAC), 2018. State of Provincial Healthcare System Spotlight on Eastern Cape Province, South Africa.
Tannock, G.W., Cook, G., 2002. Enterococci as members of the intestinal microflora of humans. In: The Enterococci. American Society of Microbiology, pp. 101–132.
Tyrell, G.J., Turnbull, L., Teixeira, L.M., Lefebvre, J., Carvalho, M.G., Facklam, R.R., Loygren, M., 2002. Enterococcus Gilvus Sp. Nov. And Enterococcus Pallens Sp. Nov. Isolated.
United States Environmental Protection Agency (USEPA), 1986. Ambient Water Quality Criteria for Bacteria. EPA440/5-84-002. U.S. Environmental Protection Agency Office of Water, Washington, DC.
United States Environmental Protection Agency (USEPA), 2012. Recreational Water Quality Criteria. EPA 820-F12-058. Office of Researchand Development, U.S. Environmental Protection Agency, Cincinnati, OH.
United States Environmental Protection Agency (USEPA), 2015. Review of Coliphages as Possible Indicators of Fecal Contamination for Ambient Water Quality. United States Environmental Protection Agency, Washington, DC, USA.
Velonakis, E., Dimitriadi, D., Papadogiannakis, E., Vatopoulos, A., 2014. Present status of effect of microorganisms from sand beach on public health. Journal of Coastal Life Medicine 2, 746–756.
Verma, P.U., Chandawat, D., Gupta, U., Solanki, H.A., 2012. Water quality analysis of an organically polluted lake by investigating different physical and chemical parameters. Int. J. Res. Chem. Environ. 2, 105–111.
Wade, T.J., Calderon, R.L., Sams, E., Beach, M., 2006. Rapidly measured indicators of recreational water quality are predictive of swimming-associated gastrointestinal illness. Environ. Health Perspect. 114, 24.
World Health Organization (WHO), 1998. Guidelines for Safe Recreational-Water Environments. Coastal and Fresh-Water, WHO/EOS/98.14, vol. 1. World Health Organization, Geneva, 1998.
World Health Organization (WHO), 2003. Guidelines for Safe Recreational-Water Environments. Coastal and Fresh-Water, vol. 1. World Health Organization, Geneva.
World Health Organization (WHO), 2011. Guidelines for Drinking-Water Quality, fourth ed. WHO Press, Geneva, Switzerland.
Wu, J., Long, S.C., Das, D., Dorner, S.M., 2011. Are microbial indicators and pathogens correlated? A statistical analysis of 40 years of research. J. Water Health 9 (2), 265–278.
Yilmaz, E., Koç, C., 2014. Physically and chemically evaluation for the water quality criteria in a farm on Akcay. J. Water Resour. Prot. 6, 63.
Łuczkiewicz, A., Jankowska, K., Kurlenda, J., Olańczuk-Neyman, K., 2010. Identification and antimicrobial resistance of Enterococcus spp. isolated from surface water. Water Sci. Technol. 62, 466–473.