Dry Sand Quality: The Case Study of a Touristic Beach from Rio de Janeiro, Brazil

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Abstract

Coastal contamination became a growing public health concern. Enteric illness outbreaks, and the occurrence of dermatitis and mycoses during the summer season in leisure areas, were usually related to seawater pollution. Pathogenic microorganisms can reach coastal areas through sewage discharges, compromising marine water, and beach quality. Although sand transmission of enteric illness is still unclear, there is an expressed concern that sand may act as reservoirs or vectors for humans infection. In this context, the main hypotheses of this study were: (i) fecal coliforms density changes within beach compartments; (ii) dry sand is the most contaminated beach compartment; (ii) fecal coliforms densities are within international standard limits for sand. Therefore, this case study quantified and compared within three months total fecal bacteria (TC) and thermotolerant coliforms (TEC) densities in seawater, wet, and dry sand of a chosen touristic beach from Rio de Janeiro, Brazil. Furthermore, to contribute to coastal beach management, sand contamination data surveyed were compared with the standard limits ruling worldwide until April 2018 to check suitableness. Vermelha beach should be considered as a

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reference beach for sand monitoring in Brazil. Despite being a buffer zone from the MONA Pão Açucar conservation unit, it is subjected to intense touristic pressure. Even though, fecal bacteria densities quantified in Vermelha beach were within the standards of "excellent" quality sand and water according to current legislation/guidelines. However, dry sand was the most contaminated compartment, followed by seawater and wet sand. The bacterial density in dry sand was up to 4,600 times higher than wet sand. Except for Rio de Janeiro city, recreational guidelines for beach quality is exclusive for seawater contamination. There are no established parameters for recreational beach sand classification, only recommendations, including Portugal. Monitoring recreational beach dry sand is critical to reducing the risk of beachgoer exposure to pathogens.

Keywords: Rain; Thermotolerant Coliforms; Fecal Bacteria; Guidelines.

It is well established that population growth increases pressure against natural resources (WHO 2003; Dwight et al. 2005; Wagener 2005; Pinto et al. 2012; Pereira et al. 2013; Sabino et al. 2014; Abreu et al. 2016; Testolin et al. 2017). However, in recent years, greater importance has been attributed to the impacts caused by the marked development of coastal cities (Dwight et al. 2005; Pinto et al. 2012; Testolin et al. 2017). The increase in effluent and wastewater discharge leads to environmental degradation. Through discharges, pathogenic bacteria and viruses can reach coastal areas compromising marine water and beach quality (Testolin et al. 2017). Thus, coastal contamination became a growing concern for public health (Dwight et al. 2005; Solo-Gabriele et al. 2016). Several studies have linked exposure to contaminated recreational waters with the occurrence of various enteric and cutaneous diseases (Cabelli 1983; Pruss 1998; Shuval 2003; Wade et al. 2006; Wiedenmann et al. 2006; Yau et al. 2009; Boehm and Soller 2012; Boehm and Sassoubre 2014). Therefore, outbreaks of enteric illness, including gastroenteritis, hepatitis, salmonellosis, viral diseases, and the occurrence of dermatitis and mycoses during the summer season in leisure areas, were usually related to seawater pollution (Cabelli 1983; Mendes, Nascimento, and Oliveira 1993; Mendes et al. 1996; Dwight et al. 2005; Solo-Gabriele et al. 2016).

Although sand transmission of enteric illness is still unclear, there is an expressed concern that beach sand or similar materials may act as reservoirs or vectors of infection in humans (Mendes et al. 1996; WHO 2003; Abreu et al. 2016; Solo-Gabriele et al. 2016). In this direction, despite the absence of public policies associating epidemiological data with the health problems caused by primary contact with contaminated sands, several researchers and organizations highlight the urgent need for the assessment of beach sand quality (Mendes, Nascimento, and Oliveira 1993; WHO 2003; Sabino et al. 2014; Whitman et al. 2010; Solo-Gabriele et al. 2016; Testolin et al. 2017). Public concern is also growing as a response to the increasing number of diseases attributed to contaminated sand contact (Heaney et al. 2012; Abreu et al. 2016). Therefore, correlations between beach sand exposure and infectious disease have been currently identified (Mendes, Nascimento, and Oliveira 1993; Mendes et al. 1996; Phillips et al. 2011; Heaney et al. 2012; Praveena et al. 2016; Solo-Gabriele et al. 2016). Studies have shown that contaminated water can contaminate the sand where children often spend most of their leisure time on the beach (Wade et al. 2006; Solo-Gabriele et al. 2016). Similarly, a possible relationship between sand exposure duration and perceived health symptoms with the microbiological quality of Malaysian tropical beaches was previously reported (Praveena et al. 2016).

Microorganisms, including bacteria, fungi, parasites, and viruses, are noteworthy components of beach sand, and several of them are potential pathogens (WHO 2003; Heaney et al. 2012; Whitman et al. 2010; Solo-Gabriele et al. 2016; Testolin et al. 2017). They can be indigenous biota or originate from anthropogenic contamination, including sanitary discharges and runoff (Testolin et al. 2017). The nutrient input is a microbial growth limiter in beach sand. The scientific literature proposes many factors encouraging the survival and dispersion of fecal microorganisms and pathogens on beach sand (Khiyama and Makemson 1973; WHO 2003; Solo-Gabriele et al. 2016). These include the nature of the beach, tidal phenomena, sewage outlets, the season, the presence of animals, and the number of bathers. Water movement, for example, causes erosion, transportation, and deposition of beach sediment and redistribution of associated microorganisms (WHO 2003; Phillips et al. 2011). Thus, it is essential to monitor the microbes from waters and sands. This measure can support actions to recover the coastal environment and reduce risks to public health (Cabelli 1983; WHO 2003; Wade et al. 2006; Wiedenmann et al. 2006; Boehm and Soller 2012; Sabino et al. 2014; Boehm and Sassoubre 2014; Praveena et al. 2016; Solo-Gabriele et al. 2016; Testolin et al. 2017).

Besides public health risks, clean beaches attract visitants and move the local economy (Dwight et al. 2005; Sabino et al. 2014; Pinto et al. 2012; Pereira et al. 2013; Abreu et al. 2016; Testolin et al. 2017). In higher latitudes, sand beaches are sought after for recreational purposes. Beachgoers spend a significant percentage of time on the beach itself rather than in the water (WHO 2003; Pinto et al. 2012; Pereira et al. 2013; Abreu et al. 2016). Consequently, during the summer season, coastal areas are subjected to intense anthropogenic pressure. Thus, some pathogens can be found in high densities both in seawater and sand beaches, impairing frequenters health (Mendes, Nascimento, and Oliveira

1993). However, dry sand beach quality legislation and guidelines are an issue worldwide. Until 2018 there were no defined parameters for dry sand classification, only recommendations. Portugal was the first country with recommending guidelines for monitoring Recreational beach sand quality, while Rio the Janeiro (Brazil) was the first city in the world with official values for fecal index quantification on dry sand (Mendes, Nascimento, and Oliveira 1993; SMAC 2010). The Portuguese guideline also includes thermotolerant coliforms, *Enterococcus faecalis,* and the yeast *Candida spp.* monitoring in sand samples (Table 01).

In summary, fecal bacterial and several pathogens have been isolated from beach sand (Mendes et al. 1996; WHO 2003; Phillips et al. 2011; Lescreck et al. 2016; Solo-Gabriele et al. 2016; Testolin et al. 2017). However, the capacity of these pathogens to infect beach users remains unproven, and how they threaten public health is unknown (Mendes et al. 1996; WHO 2003; Canada 2012; Solo-Gabriele et al. 2016; Praveena et al. 2016). Although research recommendation, systematic beach surveillance as part of pollution control is relatively limited worldwide, and routine index organisms monitoring of beach sand is generally not justified (WHO 2003). Nevertheless, epidemiological studies investigating the cause-effect of a possible dose-response relationship linking beach sand microbial quality with skin, eye, ear, and gastrointestinal symptoms would improve understanding in this area (WHO 2003; Dwight et al. 2005; Praveena et al. 2016; Solo-Gabriele et al. 2016).

Table 01. The international microbial standards for sand beach quality ruling in April 2018.

Microbiological parameters for sand								
Country	**Quality index**	**TC**	**TEC**	**EC**	**ENT**	**CAND**	**Microbial unit by weight of sand**	**Reference**
Brazil	Excelent	TC < 10,000	ND	EC < 40	ND	ND	MPN/100g	SMAC 2010
(City of Rio de Janeiro)	Good	10,000 < TC > 20,000	ND	40< EC > 400	ND	ND		
	Regular	20,000 < TC > 30,000	ND	400 < EC > 3,800	ND	ND		
	NR	TC > 30,000	ND	EC > 3,800	ND	ND		
Portugal		TC < 10,000	ND	EC < 2,000	ENT <2,000	CAND < 6,000	CFU/100g	Brandão et al. 2011
		TC < 1,000,000	TEC<100,000	ND	ENT<10,000	CAND < 1,000	MPN/100g or CFU/100g	Mendes et al. 1996 Mendes et al. 1993

All values were presented as Most Probable Number in 100 g of sand (MNP/100 g) for comparison. NR=not recommended; TC=Total Coliforms; TEC= Thermotolerant Coliforms; EC= *Escherichia coli*; ENT= *Enterococcus faecalis*; CAND= Yeasts.
Source: The Authors.

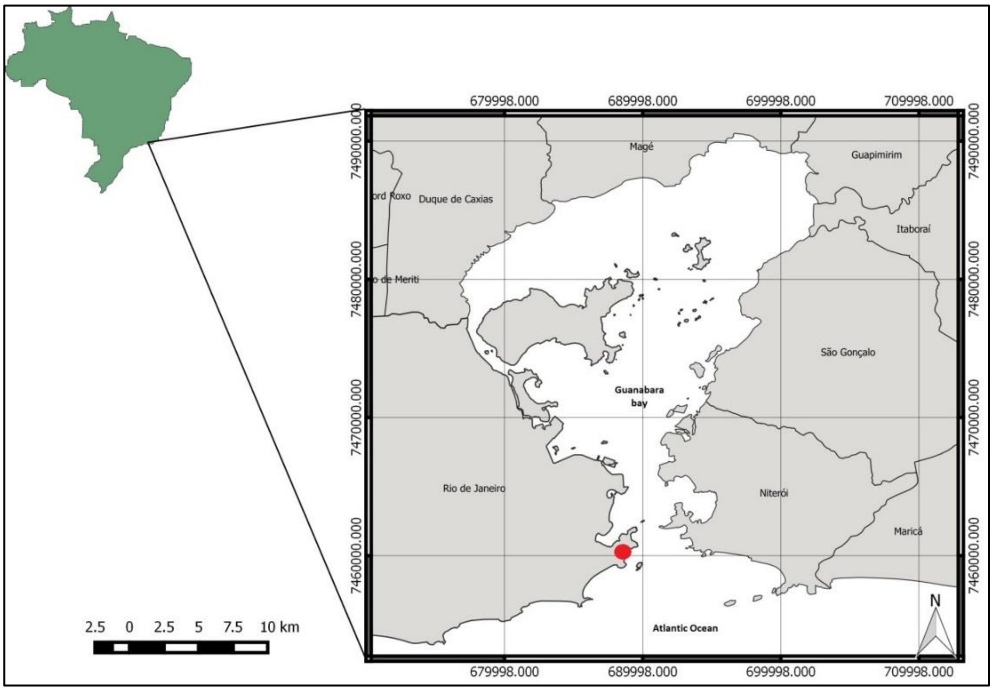
In this context, the main hypotheses of this study were: (i) the density of fecal coliforms contamination changes within beach compartments; (ii) dry sand is the most contaminated beach compartment; (ii) fecal coliforms density in the sand is within international standard limits for this compartment. Therefore, to address those hypotheses, this case study quantified and compared within three months total (TC) and thermotolerant coliforms (TEC) densities in seawater, wet, and dry sand of a chosen touristic beach from Rio de Janeiro, RJ, Brazil. Furthermore, to contribute to coastal beach management, sand contamination data surveyed were compared with the standard limits ruling worldwide until April 2018 (Table 01) to check suitableness.

Materials and Methods

Characterization of the Case Study Area

Vermelha is a tropical beach with 280 m of length located on the foot of Sugar Loaf at Urca neighborhood, Rio de Janeiro, Brazil, at coordinates 22°57'S; 43º9'W (Figure 01). Vermelha beach presents a 17° slope within 40-48 m of width and few morphological variations. Although stable, Vermelha is exposed to the ocean with direct wave incidence. Its sediments are composed of coarse sand consisting of quartz grains in rounded format, with 75-86% of 0.5 mm grain fraction and 11-25% of 1 mm coarse sand (Da Silva et al. 2016).

Figure 01. Localization of Vermelha beach (red spot) in Guanabara Bay, Rio de Janeiro, Brazil.



Source: The Author

Situated at the mouth of Guanabara Bay, Vermelha beach has full touristic flow and visibility. In 2006 was included as a buffer zone from a conservation unit called Natural Monument of Sugar Loaf and Urca hill, also known as MONA Pão de Açúcar (SMAC 2013). Tourists can reach the Sugar Loaf summit by cable cars where they will find an astonishing landscape view of Guanabara Bay, including Vermelha beach. Guanabara Bay is the second-largest bay on the coast of Brazil, and much of its contamination comes from clandestine sewage disposal on surroundings (Fistarol et al. 2015). For example, the western and northwestern parts of this Bay present poor water quality because it receives most of metropolitan Rio de Janeiro drainage without previous treatment (Ribeiro and Kjerfve 2002; Fistarol et al. 2015). Therefore, as the polluted waters flow from the inner bay towards the ocean, Guanabara bay proximity may impair Vermelha beach quality during low tide. The Vermelha beach sampling was performed during low tide to evaluate the Guanabara bay contribution to local water and sand pollution. The sampling was detailed in item 2.2.

Beach Sampling

A total of five collections of water and sand were conducted from March to May 2016 at Vermelha beach. The beach was split into three stations, one central and two outermost. Each station represented a replica of Vermelha beach (n=3). Three samples of seawater (100 mL), wet (50 g of intertidal sediment), and dry sand (50 g of supralittoral sediment) were collected using sterile materials. Seawater was collected at 01 m and wet sand at 20 cm depth of the intertidal sediment. Dry sand was collected at 15 cm depth below the supralittoral sand column. At this depth, the microorganisms are protected from solar radiation and higher surface temperature (Pereira et al. 2013). Samples were labeled, packed into a thermo-isolated box containing ice, and immediately transported to the Laboratory of Water Microbiology, Federal University of the State of Rio de Janeiro for total (TC) and thermotolerant coliforms (TEC) quantification. Given the proximity of the beach to the laboratory, the manipulation of the samples started after 01h of collection, reducing effects that may mask the results (APHA 2005).

Collections occurred during low tide, observing the interval from three to four hours after the high tide peak. A sampling at low tide is recommended by APHA (2005) for providing a more significant contribution of local contamination and lower effluent dilution. Tide prediction was consulted at the Brazilian Navigation and Water Resources Directory website (*Tábuas de marés da Marinha do Brasil 2016*). Abiotic variables (pH, salinity, and temperature in °C) were measured by

HANNA Hi 98282.2 multiparameter. Precipitation volume (mm) in Urca station (Rio de Janeiro, Brazil) 96 hours before sampling was obtained at *Dados pluviométricos (2016)*.

Fecal Bacteria Quantification

Total (TC) and thermotolerant coliforms (TEC) in marine water, wet and dry sand from the supralittoral zone were quantified by multiple tubes fermentation technique as described in detail in APHA (1992, 2005). This technique is a standard, reliable, and affordable method applied all over the world for water quality assessment (APHA 1992; IOS 1998, 2000; APHA 2005).

Mass standardization for fecal bacteria quantification in water and sand ensured an appropriate comparison between samples. Thus, before inoculation, wet and dry sand were weighted (50 g) and transferred into 100 ml of sterile dilution water (APHA 2005). The samples were agitated for 01 minute to transfer microorganisms from the sand into the liquid phase (Pinto et al. 2012; Wang et al. 2010). Three dilutions (10^0, 10^{-1} and 10^{-2}) of each sample were inoculated in five replicas of test tubes containing lactose broth. The methodology is detailed in APHA guidelines (1992). Bacterial growth (turbidity) with gas formation was considered a positive result, and the most probable number was determined following CETESB (2007). As sediment samples were dissolved in dilution water before analysis, all results were expressed in the most probable number in 100 ml of samples (MPN.100 mL^{-1}), whether liquid or solid.

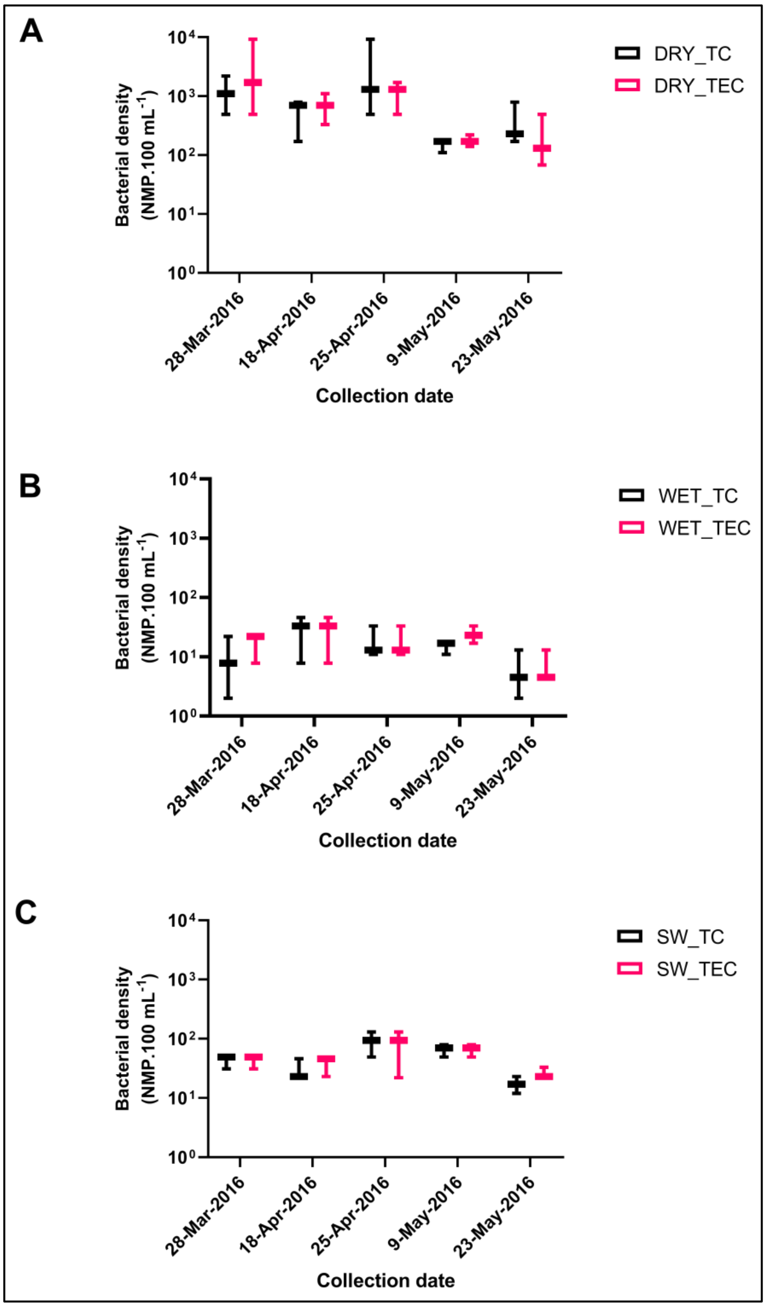
Statistical Analysis of Data

Only statistically significant results with $p>0.05$ were considered for discussion. All analyses and graphics were performed using GraphPad Prism version 8.3.0 for Windows, GraphPad Software, San Diego, California USA.

Shapiro Wilk and Kolmogorov-Smirnov test evaluated the parametric distribution of data. The differences among fecal coliforms densities (TC and TEC) within each beach compartments (seawater, wet, and dry sand) were tested by two-way ANOVA test. The statistical analysis showed weak evidence to accept the parametric assumption. Therefore, a correlation matrix between fecal bacteria (TC and TEC in seawater and wet sand) and abiotic variables (seawater temperature, salinity, and pH) were tested by Spearman correlation with one-tailed p-value and 95% of a confidence interval. Fecal bacteria in dry sand were excluded from correlation matrix analysis to avoid data inconsistency. Temperature, pH, and salinity are specific seawater parameters that could influence only wet, not dry sand. However, accumulated precipitation could affect fecal bacteria density in dry sand, and this analysis was included in the Spearman correlation matrix test.

Results

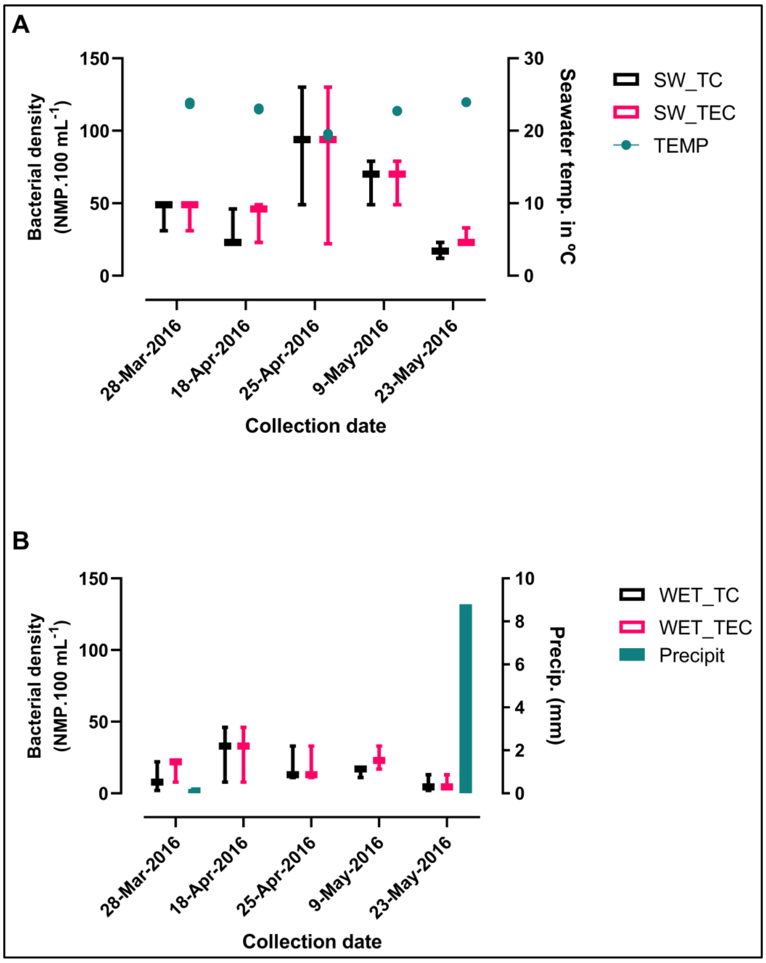
Figure 02. Box plot of fecal bacteria densities (total coliforms – TC; thermotolerant coliforms - TEC) in MPN.100 mL-1 at Vermelha beach, Rio de Janeiro, Brazil.



A) dry sand (DRY); B) wet sand (WET); and C) seawater (SW).
Source: The Author

Figure 02 shows fecal bacteria densities (TC and TEC) in seawater, dry, and wet sand by collection date. Dry sand presented total coliforms (TC) densities between 110.0 and 9,200 MPN.100 mL$^{-1,}$ while TEC densities varied from 68 to 9,200 MPN.100 mL^{-1}. TC densities in dry sand were significantly higher (two-way ANOVA test, $F_{2,6} = 5.510$, $p = 0.044$) than seawater ($12.0 \leq TC \geq 130.0$ MPN.100 mL^{-1}) and wet sand ($2.0 \leq TC \geq 46.0$ MPN.100 mL^{-1}). However, for TEC densities, there was no significative statistical difference among dry, wet sand and seawater. TEC densities for wet sand and seawater ranged, respectively, from 22.0 to 130.0 MPN.100 mL^{-1} and 4.5 to 46.0 MPN.100 mL^{-1}.

Figure 03. Box plot of fecal bacteria densities (total coliforms – TC; thermotolerant coliforms - TEC) in MPN.100 mL-1 at Vermelha beach, Rio de Janeiro, Brazil.



A) seawater (SW) and B) wet sand (WET) – left axis. The right axis shows A) seawater temperature in °C; and B) the accumulated precipitation 96 hours before collection (mm).
Source: The Author

The analysis of fecal bacteria density means by collection date revealed the highest densities in seawater (one-sample *t*-test; $t = 3.78$, $p = 0.195$ for TC and $t = 5.113$, $p = 0.0069$ for TEC) and wet sand (one-sample *t*-test; $t = 4.156$, $p = 0.0142$ for TC and $t = 5.348$, $p = 0.0059$ for TEC) in April

(Figure 02). On May 23, there was the lowest contamination observed for seawater (TC = 12.0; TEC = 22.0 MPN.100 mL^{-1}) and wet sand (TC = 2.0; TEC = 4.5 MPN.100 mL^{-1}). There was no significant difference within bacteria density in dry sand by the collection date.

From March to May 2016, there was no salinity (≈33) and pH (≈8.3) variation. However, seawater temperatures ranged from 19.3°C to 24.0°C on April 25 and May 23, respectively (Figure 03 A). The Spearman correlation matrix revealed an inverse correlation ($r = -0.90$; $p = 0.042$) between temperature and fecal bacteria (TC and TEC) in seawater (Figure 03 A).

Accumulated precipitation (8.8 mm) at Urca station was reported 96h before May 23 collection (*Dados pluviométricos 2016*). Spearman correlation matrix revealed an inverse correlation ($r = -0.89$; $p = 0.05$) between precipitation and fecal bacteria (TC and TEC) in wet sand (Figure 03 B). Dry sand presented higher fecal bacteria densities in the absence of rain (Figures 02 A and 03 B). However, there was no correlation between dry sand contamination and accumulated precipitation 96 hours before collection.

Following the microbial standards for sand beach quality listed in Table 01, Vermelha beach was classified as "excellent" under Rio de Janeiro city and Portugal guidelines (TC and TEC in dry and wet sand lower than 10,000 NMP/100 g of sand).

Discussion

Vermelha beach compartments presented different fecal bacteria densities by collection time (Figure 02). Dry sand presented the higher fecal bacteria densities (two-way ANOVA test, $F_{2,6} = 5.510$, $p = 0.044$), followed by seawater and wet sand. The bacterial density in dry sand was up to 4,600 times higher than wet sand (Figure 02 A and B). These data are in consonance with other studies that classifies dry sand as the highest contaminated beach compartment (Mendes, Nascimento, and Oliveira 1993; Abdelzaher et al. 2010; Pinto et al. 2012; Pereira et al. 2013; Sabino et al. 2014; Abreu et al. 2016; Testolin et al. 2017).

Bacteria can adhere to sand particles. Therefore, sand can present microbiological contamination higher than adjacent water Yamahara (Yamahara et al. 2007; Nevers and Boehm 2011; Boehm and Sassoubre 2014; Sabino et al. 2014). In this direction, some studies described higher concentrations of fecal bacteria (including *Escherichia coli* and *Enterococcus*) in wet sand compared to nearby water (Halliday and Gast 2011). In reverse, Testolin et al. (2017) suggested that microbiological contamination profiles for seawater and wet sand are similar. Wet sand appears as the principal reservoir for microbes. However, seawater can contaminate or dilute the wet sand depending on the degree of water contamination, including the presence of suspended organic matter undergoing decomposition (Pereira et al. 2013; Solo-Gabriele et al. 2016; Testolin et al. 2017).

Hence it should be noted that sand quality does not reside solely on water quality. Some authors believe that beach sand behaves as a passive element of cumulative pollution (Khiyama and Makemson 1973; Mendes, Nascimento, and Oliveira 1993; Mancini et al. 2005). According to Mancini et al. (2005), the level of contamination of beach sand is generally higher than that observed in water due to the phenomenon of bioaccumulation, intensified by the introduction of organic matter into this environment. Beachgoer adds organic matter to dry sand through body parts such as hair, nail, and skin scaling and food consumed at the local. Thus, the uses of the sandy beach zones by many visitors can lead to the proliferation of microorganisms nourished by food waste, which can be aggravated by the lack of good personal hygiene habits and sometimes by the presence of domestic animals or birds (Mancini et al. 2005; Testolin et al. 2017). All this organic matter introduced into the sand environment ends up favoring microbial survival (Abreu et al. 2016). However, several other factors also affect the bacteria's survival in beach sand, including temperature, humidity, organic matter, sunlight, pH, mineralogy, water retention capacity, and soil microorganisms (Solo-Gabriele et al. 2016).

Many studies assessed the relation between precipitation and fecal bacteria in seawater, as will be discussed later (Ackerman and Weisberg 2003; Noble et al. 2003; Sampson et al. 2006; Phillips et al. 2011; Zhang et al. 2013). Nevertheless, few studies addressed the effect of the precipitation on bacteria density in dry sand (Phillips et al. 2011; Pinto et al. 2012; Abreu et al. 2016; Testolin et al. 2017). Still, the results are contradictory. Testolin et al. (2017) suggested that dry sand is not under the influence of the tides. Therefore, illegal sewage discharge or residence runoff located near the beach can be a contamination source. Complementary, Abreu et al. (2016) reported that an atypical extreme precipitation event was responsible for higher values of biological and mycological contaminants in Portuguese sand beaches (Abreu et al. 2016). Contrastingly, in Vermelha beach, there was no correlation between dry sand contamination and accumulated precipitation 96 hours before collection. Phillips et al. (2011) and Pinto et al. (2012) otherwise described that after rain events, fecal bacteria densities decreased on dry sand, and increased in the water and wet sand. In this direction, considering the significant fecal bacteria decrease in wet sand associated with a precipitation event ($r = -0.89$; $p = 0.05$), Vermelha data agree with Phillips et al. (2011). Heavy rain could wash away fecal bacteria from sand, causing low densities in this zone (Phillips et al. 2011).

Similarly to dry sand, data association of precipitation and fecal bacteria densities in seawater are conflicting (Ackerman and Weisberg 2003; Noble et al. 2003; Sampson et al. 2006; Phillips et al. 2011; Zhang et al. 2013). Sampson et al. (2006) reported no relationship between beach contamination and rainfall after events lower than 6 mm. For storms, lower than 2.5 mm Ackerman & Weisberg (2003) did not detect a rainfall effect. However, these authors described a clear relationship between rainfall incidence and the decrease of water quality when rainfall indexes were higher than 2.5 mm. Contrastingly, at Vermelha beach, the lowest seawater densities of fecal bacteria were observed 96 h after 8.8 mm accumulated precipitation (Figure 03 B). Considering the divergences on this theme, Sampson et al. (2006) recommend studying beaches individually. Each beach with its specific characteristics that may influence the bacterial contamination densities.

Thereby, Vermelha geomorphology and hydrodynamics may explain higher densities of fecal bacteria in seawater than in wet sand. Vermelha beach is exposed to the ocean and continuously subjected to waves that stir up microorganisms from wet sand to water. The sand size between 0.5 mm and 01 mm (Da Silva et al. 2016) may also impact bacterial adherence. Coarse sand has a higher volume and thus possesses less surface area contact in comparison to finely-grained sediment. Tidal, waves, or precipitation events may directly or indirectly contribute to fluctuations in bacterial sand concentrations. Halliday and Gast (2011) suggest that erosional flow conditions generated by storms or tides may flush bacteria out of sediments or sands, resulting in some level of contamination of the water column. Solo-Gabriele et al. (2016) observed that *Escherichia coli* densities increased in storm events and after these events returned to baseline levels, shifting in a cyclic pattern.

Some researchers studied the relationship between abiotic variables, including pH, temperature, and salinity with fecal bacteria (Rozen and Belkin 2001; Schulz and Childers 2011; Byappanahalli et al. 2012; Zhang et al. 2013). According to Rozen and Belkin (2001), when released at seawater, enteric bacteria are challenged by stress factors, including pH, temperature, salinity, nutrient availability, light radiation, and oxidative stress. In this direction, Schulz and Childers (2011) noticed a decay rate increase of fecal *Bacteroidales* at higher temperatures. Briefly, high temperatures can decrease microorganism populations by desiccation (Byappanahalli et al. 2012). Indeed, in Vermelha fecal bacteria density decreased at higher temperatures and vice-versa ($r = -0.90$, $p = 0.042$; Figure 03 A).

Sand Quality Standards

Beach sand microbial communities are heterogeneous, including bacteria, viruses, protozoa, helminths, and fungi (WHO 2003; Solo-Gabriele et al. 2016). For decades, the scientific literature describes the isolation of fecal bacteria and other pathogens from beach sand (Mendes et al. 1996; WHO 2003; Phillips et al. 2011; Lescreck et al. 2016; Solo-Gabriele et al. 2016; Testolin et al. 2017). However, the capacity of these pathogens to infect beachgoers remains unproven (Mendes et al. 1996; WHO 2003; Canada 2012; Solo-Gabriele et al. 2016; Praveena et al. 2016). Although it recommended, systematic beach surveillance as part of pollution control is relatively limited worldwide. A routine sand beach microbial monitoring index is generally not justified (WHO 2003). Nevertheless, epidemiological studies investigating cause-effect, or a possible dose-response relationship linking beach sand microbial quality with skin, eye, ear, and gastrointestinal symptoms would improve this subject (WHO 2003; Dwight et al. 2005; Praveena et al. 2016; Solo-Gabriele et al. 2016).

Currently, only Rio de Janeiro (Brazil) and Portugal recommend bacteria standards for dry sand quality worldwide, as listed in Table 01. However, there are no standardized values, indicators, or methodologies for sand monitoring. While SMAC (2010) express results as MPN in 100 g of sand, Portuguese data can be expressed as MPN, CFU or pfc in g of sand (Mendes et al. 1996; Brandão et al. 2011). Therefore, in Table 1, all microbial values were converted into the same unit for better comparison.

Similarly, Rio de Janeiro city (Brazil), and Portugal recommend total coliforms and *Escherichia coli* monitoring. However, Portuguese guidelines also include the monitoring of thermotolerant coliforms*, Enterococcus faecalis,* and yeasts, such as *Candida spp.* in the sand (Mendes et al. 1996; SMAC 2010; Brandão et al. 2011). Similarly, fecal bacteria standards for recreational sand are conflicting in both countries (Table 01). For example, in Mendes, Nascimento, and Oliveira et al. (1993), the proposed limit for total coliforms is 10^2 times higher than that established in SMAC (2010). Even though primary sand contact is “not recommended” when total coliforms densities are up to 30,000 MPN.100 g^{-1} (SMAC 2010) (Table 01), Mendes, Nascimento, and Oliveira; Mendes et al. (1993; 1996) considers as “not recommended” sand values up to 1,000,000 MPN.100 g^{-1}. Contrastingly, in Brandão et al. (2011) the maximum admissible values were 100 pfc.g^{-1} for total coliforms and 20 pfc.g^{-1} for *E. coli* and *Enterococci*.

Although there are no international guidelines for determining the contamination of beach sand, reference values should be interpreted and modified according to local or regional factors such as exposure pattern, population behavior, severity, and nature of locally endemic diseases and economic, socio-cultural and environmental aspects (WHO 2003). In this context, Brazilian legislation (Brasil 2000) only recommends the evaluation of the microbiological and parasitological conditions of the sand for future standardization. However, the Brazilian Environmental Council has so far not taken any initiative to make beach sand analysis mandatory and establish reference values for its classification. Testolin et al. (2017) believe that constant microbiological parameters monitoring is essential for both water and sand because this measure can support actions to recover the coastal environment and reduce risks to public health. Unfortunately, despite the Brazilian coast extension, still few scientific studies deal with beach sand quality analysis in Brazil (Vieira et al. 2002; Pinto et al. 2012; Lescreck et al. 2016; Testolin et al. 2017).

Addressing the methodological approach for collecting sand samples, microorganism's identification and quantification require specific procedures and moderate laboratory infrastructure (WHO 2003; Solo-Gabriele et al. 2016; Testolin et al. 2017). However, the methods currently available for monitoring the microbiological quality of beach sand can be laborious and time-consuming (multiple tube methods) or very expensive (quantitative polymerase chain reaction - qPCR) (Testolin et al. 2017). Therefore, coastal managers and environmental agencies in developing countries must deal with institutional and economic limitations (Wagener 2005; Solo-Gabriele et al. 2016). In this direction, SMAC (2010) resolution should be reviewed to optimize quantification and avoid crossed contamination and false-positive results. Thus, the implementation of aseptically tools, including a sterile PVC tube as a grab for collecting sediments, is crucial for microbiological monitoring and accurate results. Furthermore, establishing an international standard unit for bacterial densities would also improve data comparison between studies.

Despite all the controversy between the cause-effect of contaminated sand and illness, microbiological quality of beach sand has recently become a public health issue (Brandão et al. 2011; Solo-Gabriele et al. 2016). Praveena et al. (2016) demonstrated that skin problems were the most predominant health symptoms indicated by beachgoers at Malaysian tropical beaches. The authors also showed that sand exposure duration was significantly correlated with the perceived health symptoms, indicating its possible relationship with the microbiological quality of beach sand. However, many investigators agree that more research is needed before guideline values for sand could be established (WHO 2003; Canada 2012; Praveena et al. 2016; Solo-Gabriele et al. 2016). In this direction, Health Canada Recreational Water Quality Guidelines indicate that testing of sand may be warranted in circumstances such as support for sanitary surveys or disease outbreak investigations (Canada 2012).

Portugal was the first country to recommend fecal indicators standards for sand. However, Rio de Janeiro, Brazil, was the first city worldwide to embrace a classification for recreational sands with the primary contact (Pinto et al. 2012). The contamination report bulletins are published biweekly at the municipal administration website (*Areia* carioca) since 2010. Noteworthy, this municipal stopped in November 2015, restarting in June 2017. Furthermore, another initiative exclusive from the city of Rio de Janeiro was the decree-law that prohibits animals in beaches' sand and obligates dog owners to collect their feces from public space (PRJ 2001). This act goes in consonance with Europe Union directive 66/2006 (The Council of the European Union 2006) and European Blue Flag Association criteria for blue flag accreditation (Brandão et al. 2011). For long is known that animals can contribute to sand fecal pollution (WHO 2003; Mancini et al. 2005; Sampson et al. 2006; Pinto et al. 2012; Canada 2012; Solo-Gabriele et al. 2016; Testolin et al. 2017). Animal excreta, including dogs and birds, are the principal microbial risk to human health encountered on beaches and in similar areas. Regulations, often local, may restrict access on a seasonal basis to frequently used beaches or place an obligation upon the owner to remove animal excreta. However, the animal prohibition in beaches from the municipality of Rio de Janeiro is under review.

CONCLUSIONS

The scientific contribution of this study was to establish a baseline of fecal bacteria (TC and TEC) in the dry sand of Vermelha beach, strengthening the coastal beach management. As a buffer zone from the conservation unit MONA Pão Açúcar, and subjected to intense touristic pressure, Vermelha should be considered as a reference beach for sand monitoring in Brazil. Fecal bacteria densities quantified in Vermelha beach were within the standards of "excellent" quality sand and water according to current legislation/guidelines. However, dry sand was the most contaminated compartment, followed by seawater and wet sand. The bacterial density in dry sand was up to 4,600 times higher than wet sand. High beach hydrodynamics may contribute to Vermelha quality. Vermelha seawater salinity and pH were within the expected range for the marine environment. No significant change in these variables was registered even after 8.8 mm accumulated precipitation. Higher seawater temperatures reduced significantly fecal bacteria densities in seawater, and vice-versa. The accumulated precipitation 96 h before collection decreased fecal bacteria in wet sediment.

Despite the vast Brazilian coast extension, there is still no Brazilian federative guideline for beach sand quality. Except for Rio de Janeiro city, recreational guidelines for beach quality is exclusive for seawater contamination. There are no established parameters for recreational beach sand classification, only recommendations, including Portugal. Additionally, the establishment of an international standard unit and methods for fecal index bacteria are required for the advance on this subject and data comparison between studies. Monitoring recreational beach sand is critical to reducing the risk of beachgoer exposure to pathogens.

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Qualidade da Areia Seca: Estudo de Caso de uma Praia Turística do Rio de Janeiro, Brasil

RESUMO

A contaminação costeira tornou-se uma crescente preocupação de saúde pública. Surtos de doenças entéricas e a ocorrência de dermatites e micoses durante o verão nas áreas de lazer estão geralmente associados à poluição da água do mar. Os microorganismos patogênicos podem atingir áreas costeiras através de descargas de esgoto, comprometendo a água do mar e a qualidade da praia. Embora a transmissão de doenças entéricas pela areia ainda não esteja clara, existe uma preocupação de que a areia possa atuar como reservatório ou vetor da infecção para seres humanos. Nesse contexto, as principais hipóteses deste estudo foram: (i) alterações na densidade dos coliformes fecais nos compartimentos da praia; (ii) areia seca é o compartimento de praia mais contaminado; (ii) as densidades de coliformes fecais estão dentro dos limites do padrão internacional para areia. Portanto, este estudo de caso quantificou e comparou por três meses as densidades de colformes totais (CT) e termotolerantes (TEC) na água do mar, areia molhada e seca de uma praia turística escolhida do Rio de Janeiro, Brasil. Além disso, para contribuir para o gerenciamento das praias costeiras, os dados de contaminação da areia pesquisados foram comparados com os limites padrão vigentes em todo o mundo até abril de 2018 para verificar a adequação. A praia Vermelha deve ser considerada uma praia de referência para o monitoramento de areia no Brasil. Apesar de ser uma zona tampão da unidade de conservação MONA Pão Açúcar, está sujeita a intensa pressão turística. Mesmo assim, as densidades de bactérias fecais quantificadas na praia Vermelha estavam dentro dos padrões de "excelente" areia e água de qualidade, de acordo com a legislação / diretrizes vigentes. No entanto, a areia seca foi o compartimento mais contaminado, seguido pela água do mar e areia úmida. A densidade bacteriana na areia seca foi até 4.600 vezes maior que a areia úmida. Exceto na cidade do Rio de Janeiro, as diretrizes recreativas para a qualidade das praias são exclusivas para a contaminação da água do mar. Não existem parâmetros estabelecidos para a classificação de areia recreativa na praia, apenas recomendações, incluindo Portugal. O monitoramento da areia seca na praia é fundamental para reduzir o risco de exposição dos banhistas a estes patógenos.

Palavras-Chave: Chuva; Coliformes Termotolerantes; Bactérias Fecais; Legislação.

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