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Variation of Total Coliforms and Bacteria during Dry and Wet Seasons in Rivers of Sigor Division, West Pokot County, Kenya

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Authors' contributions

This work was carried out in collaboration among all authors. Authors NAG, KS, MJ and KA designed the study. Authors MJ and KS performed the statistical analysis. Authors NAG and KA wrote the protocol. Author NAG wrote the first draft of the manuscript. Authors NAG and KS managed the analyses of the study. Author NAG managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aims: There has been an increase in gastro-intestinal and urinary infections in Sigor division, West Pokot, Kenya. These infections result from bacteria and coliforms which are majorly propagated in water systems. Residents of this area are pastoralists and small-scale farmers relying on river water for their consumption and economic needs. This study aimed at characterizing the strains and quantities of bacteria and coliforms in the four rivers during the wet and dry seasons.
Study Design: An independent measures design was used.
Place and Duration of Study: Samples were taken from four rivers (Weiwei, Chesogon, Lomut and Muruny) in Sigor division, West Pokot county, Kenya. The study was conducted between January and October 2013.
Methodology: Sampling was done at various points of the river in a stratified manner for characterization and analysis. Four main pathogens namely *E. coli, V. cholerae, Shigella* and *Salmonella* species as well as *F. streptococci* were isolated, cultured using different media and

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characterized. Further biochemical tests were conducted to confirm the exact strains present. Total viable counts for the bacteria and coliforms were then enumerated.

Results: The results found out that *E. coli, V. cholerae, Salmonella* and *Shigella* species were abundant in the rivers while *F. streptococci* were only observed during the wet season. Biochemical tests conducted on the isolates revealed that the strains co-existed in the water samples. Weiwei river had the greatest number of bacteria strains. Muruny river was found to have the largest population of bacteria colony forming units (cfu's). There was a large disparity in cfu's in the rivers during the dry seasons. Chesogon river had the highest population of coliform units.

Conclusion: The raw water in all the rivers were concluded to be unsafe for human consumption according to WHO standards.

Keywords: Clean river water; bacteria; coliforms; gastro-intestinal infections.

1. INTRODUCTION

There is a global demand for clean potable drinking water. Clean water is a scarce resource in most Arid and Semi-Arid Lands (ASAL); such as in West Pokot County, found in the north-west part of Kenya and bordering Uganda to the East. The region enjoys a mixed weather pattern with cold nights and warmer days. Different parts of Sigor division experience slightly different temperatures. This usually ranges between 10°C to 35°C. Precipitation in the lowland parts (around Chesogon) is quite little (400 mm, evaporation rate of 58mm per annum). The highland parts, especially around Lomut and Muruny region is quite high (1500 mm) per annum. This condition also affects the evaporation rates which averages 338 mm per month [1]. Majority of the residents of this county, especially in Sigor division are pastoralists while a few practice crop farming. These residents derive water for their animals and domestic use from the rivers. There are poor hygiene measures and most residents have no toilets thus dispose their urine and fecal wastes in bushes. This wastes gradually leach towards the rivers, especially during rainy seasons and contaminate the water.

Deposition of animal and human contaminants into the rivers introduce bacteria into the water bodies. Majority of these bacteria constitute the *Enterobactericeae* family that is responsible for many clinical cases in ASAL regions [2]. *Escherichia coli* is one of the most common bacteria resulting from consumption of untreated river [3]. There are other common bacteria such as those of *Fecal streptococcus* and *enterococcus* which have also been traced to river water [4]. All these microbes are harmful, especially to people with vulnerable immune systems such as the old and young children. They cause several gastro-intestinal diseases, most which are chronic such as cholera, typhoid and dysentery amongst others. Some of the common short-term effects of these diseases include diarrhea, cramps and pneumonia symptoms. At advanced stages, the diseases cause death. The public health system in West Pokot county, like other parts of the country is quite challenged to respond to outbreaks of the disease in time thus leading to fatalities.

Total coliforms are gram-negative, aerobic or facultative anaerobic, non-spore forming rods [5]. They have for a long time been used as indicators of potable water. They survive in the range of 37°C [6]. The source of these microbes is the gastro-intestinal walls of warm-blooded animals. They are passed out as fecal waste. For a long time, they were thought to be an indicator of the intensity of fecal contamination in water bodies. However, a more accurate indicator are the fecal coliforms. These ones survive at a relatively higher temperature of around 45°C [7]. *Escherichia, Shigella* and *Salmonella* families are some of the members of fecal coliforms which are responsible for water-borne diseases [8].

The major seasons in West Pokot county are the dry and wet seasons. During the dry season, there is less precipitation and temperature, sunlight intensity and wind strength are high. There is a lot of particulate discharge into rivers. The elevated temperature provides a warmer ground for several fecal coliforms to thrive. There is also a lot of fecal discharge from cattle as other water pans have dried up and rivers are their only source of water. In the process of drinking water, the animals discharge their waste into or near rivers. During wet seasons, there is more precipitation, less temperature wind strength and sunlight intensity. Due to poor vegetation in the division, most agricultural residues and human fecal discharge in bushes are leached into the rivers. Poor sewerage

systems in the small townships in the division also ensure more fecal waste is discharged into the rivers. Therefore, the fecal microbes are replenished during wet seasons and thrive in the dry ones. This cycle continues and, in the process, cases of water-borne diseases have been on the rise.

The main objective of this study was to enumerate the bacteria and coliform population in four rivers of Sigor division, West Pokot county during wet and dry seasons. The four rivers in question are Rivers Weiwei, Chesogon, Muruny and Lomut. Determination of the exact microbial pathogen population at specific times of the year in the rivers will guide policy makers on the appropriate measures to take.

2. MATERIALS AND METHODS

2.1 Study Design

A cross-sectional experimental design was followed for the study. Sampling was done at various stages of the rivers, based on their topography. Samples were collected at both the wet and dry seasons at the chosen sampling points in Sigor division (between latitude 1.1359°N and longitude 35.7121°E). Fig. 1 below illustrates the four rivers and regions that were sampled.

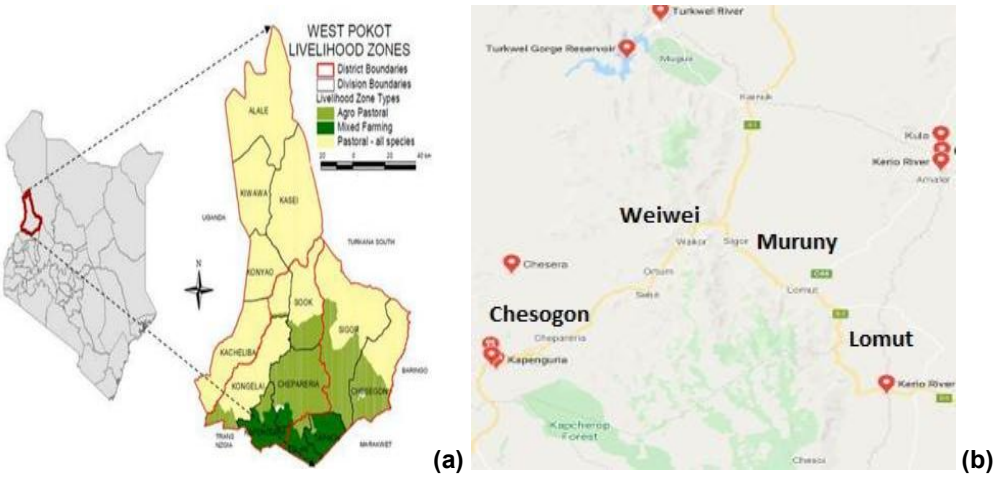
500ml polypropylene sample-bottles were used in the sampling process. The sample bottles were sterilized to avoid contamination of the samples. The samples were then taken to a Microbiology lab. (in Kenyatta university, Kenya) where analysis was scheduled to be done.

2.2 Sample Collection

Sampling was done between January and September of 2013. This was intended to cover both wet and dry seasons. The first 4 months (up to late March) enjoy a dry season in Sigor division. The next 4 months (up to July) experience a lot of rainfall thus forming the wet season. Water from different sections of the rivers was sampled between the upstream and downstream points. A 100m threshold deviation between any two points was maintained to avoid overriding samples. Each point was samples quadruple times and a total of 32 points were sampled. The samples of different river courses were pooled together to form a composite sample. The Fischer method was used to determine the sample size (confidence level being 95% i.e. 5% margin error) [11].

$$n = \frac{Z^2 PqD}{d^2} \quad (1)$$

Where; n = sample size, Z = is the appropriate value from a normal distribution for the desired confidence level which was 1.96 in this study, p = anticipated prevalence which was 3% (0.03) in this study, q = failure which was calculated as (100% - 3%) giving 97% (0.97), D = design effect which was given a value of 2 because replication was carried out based on 3% prevalence and Z value of 1.96 and d = allowable error (0.086), and. A final sample size of 30 samples was finally obtained.



(a) (b)

Fig. 1. Map of West Pokot county in Kenya (a) [9] Sigor division showing the locations of the rivers (b) [10]

Sampling of the water was done by 'grab method' using polypropylene sampling bottles. The researchers had personal protective gear to ensure they do not come into contact with the water samples. Transfer of the water samples was done while slightly slanting the sampling bottles very cautiously to ensure the samples do not come into contact with the researchers.

2.3 Methods Used

Inoculation and Morphological characterization of bacteria species.

Four bacteria species namely; *Vibrio cholera, Escherichia coli, Salmonella-Shigella* (SS) and *Fecal streptococci* species and were isolated, cultured using several media and characterized.

2.3.1 Test for *Escherichia coli*

A positive broth of bacteria was developed by conventional methods. A little amount of this was added onto Eosin Methylene Blue (EMB) agar for incubation at 98.6°F for one day. A lean smear was then made for gram staining onto the metallic sheen clusters. This was later to be confirmed by biochemical tests [12]. Enumeration of the viable coliforms was done before factoring with the dilution ratio.

2.3.2 Test for *Vibrio cholerae*

Water samples were streaked on TCBS (thiosulfate-citrate bile salts sucrose) agar warmed to rtp and inoculated aerobically at a temperature of 35ºC for 24 hours. Immediately after the incubation period, the colonies formed were morphologically examined to avoid the color for positive *V. cholera* test (yellow) reverting to green color. This usually happens at room temperature [13].

2.3.3 Test for *Salmonella species*

A composite mixture of the samples and selenite F broth (1:10) was made and incubated at 98.6°F for one day. A micro-spatula was used to leanly whizzle this impregnated samples onto MacConkey agar, SS agar and Deoxycholate Citrate Agar (DCA) [14]. The bacteria clusters were then counted using a colony counter before Gram staining. Urease and Triple Sugar Iron (TSI) biochemical tests were later used to confirm these bacteria colonies.

2.3.4 Test for *Fecal streptococci*

Dulcitol selenite broth was added onto (Xylose lysine deoxycholate) XLD media for primary enrichment. The mixture was modified by addition of sodium acid selenite. The constituents were then dissolved in sterile flasks covered with foil and heated to 88°C in a water bath to obtain a sterile clear medium without pH adjustments. The mixture was then incubated at body temperature for 18 hours.

2.3.5 Biochemical analysis of the isolates

To identify the exact strain of *E. coli, V. cholerae, Salmonella species, Shigella species* and *Fecal streptococci* in the samples, biochemical tests were conducted. This experiments also aimed at identifying any co-existence of the strains in the water samples. The strains of bacteria present in the Seven biochemical tests were conducted as outlined below.

2.3.5.1 Gram stain

From the pure colonies, a loopful of the bacterial cells was picked and smeared on a glass slide. Fixing was done by passing the slide over hot flame after which the smear was flooded with crystal violet stain. A gram iodine mordant was added after a minute, washed and flooded with a decolorizer for 10 seconds. After washing, the slide was flooded with safranin for 1 minute after which it was washed, blot dried and observed under a microscope. Gram-positive bacteria stained purple while the negative stained red/pink.

2.3.5.2 Methyl red test

A bacteria cluster not previously contaminated from the isolated broth was inoculated into a sterile glucose-phosphate broth ($0.5cm^3$). The mixture was then incubated at 98.6°F for 12 hours. 2ml of methyl red indicator was then added. Positive test of *E. coli* strains was marked by a bright red coloration.

2.3.5.3 Voges-Proskauer (VP) test

A broth constituting of methyl red and Voges-Proskauer reagents was cultured together with purified samples at 98.6°F for one day. The incubated mixture was added onto alpha naphtha solution (5%) (in a ratio of 3:1) followed by a droplet of potash soda (40%). The mixture was gently agitated in test tubes before allowing to settle for quarter an hour. A chemical reaction was to proceed behind the scene to yield diacetyl, if the bacteria strains were present. Presence of diacetyl was confirmed by red

coloration while its absence was marked by yellow-brown coloration.

2.3.5.4 Indole test

Tryptophan broth was emulsified with a cluster of bacteria test isolates and incubated at 98.6°F for 2 days. A drop of Kovacs reagents was added and the color of the product monitored. The confirmatory color was pink while no color change indicated negative results.

2.3.5.5 Urease test

A slant containing the urea Christensen's Urea Agar was streaked with a loop full of colonies of the test organisms. The slants with the loosened caps were incubated for one day at 90°F after which the color was observed. The color changed from light orange to bright pink in positive results and remained light orange in negative results.

2.3.5.6 Triple Sugar Iron (TSI) test

A purified and isolated cluster of bacteria was finely spread on a sterile petri dish. A sterile inoculation needle was then struck at the centre of the cluster. TSI agar was then inoculated by streaking at the middle, the agar slant surface and lower part of the media. The cap of the tube used was then loosened and left to incubate at 90°F in ambient air for about 21 hours. Color changes of both the Slant/Butt as well as CO_2 and H_2S yielding metabolic processes were all recorded.

2.3.5.7 Citrate test

A needle tip was used to pick some bacterial cells from the cultured colonies. The mixture was inoculated in Simmons citrate agar on the slant and incubated for a day at 98.6°F. Confirmatory of the bacteria strains was marked by green-blue coloration.

2.3.6 Total viable counts/Microbial loads

The spread plate technique was applied in enumeration of the bacteria microbes. A dilution factor of 10^4 was used in diluting the samples collected from the 4 rivers. This ratio ensured further growth of desired bacteria clusters. An aliquot of the diluted sample river water (1ml) was gently agitated with sterilized EMB agar on a plate. The mixture was incubated for about 36 hours between 90-98.5°F. A colony counter was used to enumerate the exact colonies available before re-calculating using the dilution factor used.

Total bacteria were enumerated as Colony Forming Units (CFU/ml) using the formula [15];

$$N\ x\ D\ x\ V = CFU/ml \quad (2)$$

Where; N = Number of colonies, D = Dilution factor, V = Volume factor

2.3.7 Presumptive coliform counts

Coliform enumeration followed the Most Probable Number (MPN) method due to its accuracy and reproducibility. Positive presumptive tests were monitored by gas production from anaerobic reaction of the river water samples with a series of lauryl tryptose broth (LTB) [12]. The optimal gas production temperature was 98.6°F after two days. Inoculum from the positive fermentation tubes was mixed with 2% brilliant green bile lactose broth (Oxoid). The mixture was then dispensed in sterile fermentation tubes fitted with inverted Durham tubes and incubated for a further day at 98.6°F. This was the confirmatory test after which the microbes cfu's were enumerated. MPN population were graded according to potable water suitability. MPN of between 1-3 was rated as satisfactory and the water samples that had counts above 10 were rated as unsatisfactory.

2.4 Data Analysis

Numerical data of the microbes was given as mean ± standard deviation. 95% confidence level (P = .05) was used for the statistical interpretations. Ms Excel (version 2016) and OriginLab (version 6.5) were used for statistical analysis.

3. RESULTS AND DISCUSSION

3.1 Presence of Bacteria Species in the Rivers

The colonies of *E. coli, V. cholera* and *Shigella-Salmonella* were found to be quite abundant in both wet and dry seasons of the rivers based on the growth in their media. *F. stroptococci* colonies were only present in mild amounts during the wet seasons of Rivers Weiwei, Lomut and Chesogon. Table 1 highlights the occurrence of these bacteria species in the rivers during the wet and dry seasons.

Table 1. Bacteria species occurrence in the rivers of the sample area during wet and dry seasons

Bacteria tests	Rivers assessed							
	R. Weiwei		R. Chesogon		R. Lomut		R. Muruny	
	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry
E. coli using EMB	++	+	+	+	++	+	+	+
V. cholerae using TCBS	+	+	++	+	+	-	+	+
Shigella and *Salmonella spp.* using SS	++	-	+	+	+	+	+	-
F. streptococci using XLD	+	-	+	-	+	-	-	-

Presence of the metallic green sheen on the plates containing EMB agar indicated positive presence of *E. coli* in the water. This coloration was more pronounced in the wet samples of rivers Weiwei and Lomut. This is attributed to their positioning with reference to human settlement in the division. Fecal discharge and other wastes are deposited from the sewerage system of the area. During dry seasons, there is less precipitation and therefore less water in the sewer system. The metallic green sheen on EMB agar results from a group of *E. coli* bacteria that have a fast rate of fermenting the lactose in the EMB agar broth [16]. The rapid fermentation of lactose by these bacteria cause a sharp drop in pH resulting to the color formed. Some of these bacteria include *Citrobacter* and *Enterobacter* species responsible for travelers' diarrhea and bacteremia diseases [17]. At advanced stages, patients are susceptible to chronic infections such as urinary tract infections (UTI) and neonatal meningitis. Majority of the water samples tested positive for *Vibrio cholerae* indicating golden yellow spots on the background of TCBS agar medium. This resulted due to fermentation of sucrose leading to these colorations. TCBS agar was chosen for its high selectivity and therefore it can be precisely confirmed that there were indeed traces of *V. cholerae* in the river samples except in River Lomut during its dry season. Chesogon river had more pronounced colonies of the species in its wet season. This are partly attributed to sewer discharge as well as fecal discharge from cattle. Chesogon river is located closer to Chesera plains, on the western part of Sigor where pastoral farming is more pronounced than the other areas of Sigor. Contrary to Chesogon, Lomut is on the eastern, greener side where most of her residents are farmers. There is very little effluent discharge during the dry season, justifying absence of *V. cholerae* species in its water.

Shigella and *Salmonella* species were present in most of the rivers except R. Weiwei and Muruny during the dry season. Weiwei river however indicated more traces of the pathogens, with pronounced pink-red spots on the media used. The spots had a larger surface area (diameter, about 13mm) and were more spread out. The SS media used is highly selective to *Salmonella* species but slightly inhibitory to some *Shigella* species. tests of these species indicate lack of fecal discharge into the rivers. Weiwei residents should however be wary of the pronounced amounts of the species during wet seasons as they are more likely to catch typhoid and gastroenteritis. *Fecal streptococci* species were the least in the water samples. Their traces were spotted by dark spots on the pink-red XLD agar media background. The colors result from fermentation of xylose, lactose and sucrose in the dulcitol broth and media to acids and thus color change [18]. It was however noted that *Fecal streptococci* species were the least dominant compared to other bacteria species analyzed.

3.2 Biochemical Analyses of the Isolates

There were mixed findings to isolates of the bacteria when analyzed with more specific tests. From the gram stain tests, all the samples tested negative for rod shaped bacilli bacteria. However, the water samples appeared to be contaminated with cocci bacteria, possible *Staphylococcus aureus* or *Staphylococcus pneumoniae*. These isolates have a long lifespan in many environments such as air, soil and water [19]. It is therefore not surprising to observe these microbes in both wet and dry seasons. These pathogens cause skin infections, toxic shock syndrome and pneumoniae (*S. pneumoniae*) [20]. Indole test confirmed presence of *E. coli* in water samples of R. Weiwei (both wet and dry seasons) and wet season samples of the other rivers. There might be presence of other microbes co-existing with *E.*

coli such as *P. vulgaris, M. morganii* and *providenica;* all confirmatory tests of Indole test. The flora of all these pathogens include both soil and water. Various water samples tested positive to MR test. Only the samples with *F. streptococci* couldn't test positive due to the nature of their isolates. Other bacteria such as *Shigella, E. coli, Salmonella, citrobacter, Proteus* and *Yersinia* species can all perform mixed acid fermentation when supplied with glucose. These microbes were present throughout all seasons in various rivers of Sigor division. Table 2 illustrates the biochemical tests of the isolates of the river water samples analyzed.

VP test seeks to identify presence of *Enterobacter* species in bacteria isolates. The most common species is *Enterobacter cloacae.* Negative test of VP is also confirmatory of *E. coli* strains. All the *E. coli* isolates tested negative for all the seasons. On contrary, most of the *F. streptococci* isolates tested positive for this test. Only isolates of Lomut and Weiwei rivers on their wet seasons formed red colonies to confirms positive for Urease test. This is attributed to urinary discharge into the rivers [3] during the wet season, probably from the sewerage systems. This implies there is a high likelihood of the residents of the two rivers to have *H. pyroli* infections that can affect their gastrointestinal and urinary tracts [21]. The triple sugar iron test targeted all bacteria that could ferment various forms of carbohydrates. The products of the tests determine the nature of bacteria present. Color change to either the slant or butt of the tube to yellow confirmed presence of *E. coli, Pseudomonas aeruginosa*, *Salmonella enterica* and *Shigella sonnei*. The *E. coli* isolates and some of the *V. cholerae* and *Salmonella* and *Shigella* isolates with samples of various rivers confirmed these microbes. *F. streptococci* isolates did not exhibit these strains. Another group of bacteria leads to formation of a gas in an anaerobic media (CO_2). A few strains form hydrogen sulphide (H_2S) gas with the isolates. The wet samples of Lomut and Muruny *F. streptococci* isolates were found to exhibit these compounds while all the others gave negative tests. *Salmonella enterica* strains have been associated with formation of these gas. The citrate test differentiates gram-negative bacteria of the family of *enterebacteriaceae*. Most tests gave a green color (negative) except for the *V. cholerae* and *Salmonella* and *Shigella* species isolates which gave a shade of blue on the slant side of the tube (positive test). The samples collected from wet seasons of Weiwei and Lomut rivers also tested positive for these strains of bacteria. *Salmonella, Citrobacter, klebsiella* and *Enterobacter* strains have been associated with this test. All these pathogens can survive well both in water and soil samples, thus present in both dry and wet seasons [22]. Their source is fecal discharge from infected people and cattle. Negative citrate test confirmed strains of *Escherichia, Shigella, Morganella* and *Yersinia* species [23].

From these tests above, it is rather clear that all the four rivers were heavily infested with numerous strains of pathogenic bacteria of different isolates. The several tests done confirmed this. The wet samples of the rivers, especially River Weiwei indicated that this water required a lot of treating before consumption. Otherwise, the water was not fit for consumption by both people and cattle.

3.3 Total Bacteria Viable Counts

There was a great difference in the number of colonies detected in the study area during the wet and dry seasons. The population of bacteria colonies detected in the study rivers during the wet season were closely harmonized together (standard deviation ranging between 3.22 to 19.07, $P > 0.05$). On contrary, during the dry season, there was a great dispersion in number of bacteria colonies detected (standard deviation ranging between 33.57 to 83.09, $P > 0.05$). This deviation can be attributed to the nature of the rivers during the two seasons. At the wet season, there is a lot of water and dispersion of microbes is quite evenly distributed. Therefore, the number of bacteria microbes at any two points of the same river is likely to be similar when all other factors are held constant. During the dry season, some spots of the river, especially close to sewer lines, urban settlements and water drinking points for cattle, have more microbes than other points of the river. This is because there is less water to distribute the microbes evenly in the water systems. River Muruny was the most inconsistent with this disparity in total bacteria microbes. All the rivers had bacteria colonies above the permissible World Health Organization (WHO) limits of 0.00 cfu/100ml [24,25]. Water from the rivers of Sigor division can thus be concluded to be unsuitable for human and animal consumption. While Weiwei river had numerous types of pathogens as shown above, Muruny river had the most quantities of the pathogens as illustrated in Table 3.

Table 2. Biochemical tests for various bacteria strain present in the rivers of Sigor division

Isolates	Samples	Biochemical tests								
		Gram stain	Indole	MR	VP	Urease	TSI			Citrate
							S/B	CO_2	H_2S	
E. coli	R.Weiwei Dry	-rod	+	-	-	-	+/+	+	-	-
	Wet	-rod	+	+	-	-	+/-	+	-	-
	R.Chesogon Dry	-rod	-	-	-	-	+/+	+	-	-
	Wet	-rod	+	+	-	-	+/+	+	-	-
	R.Lomut Dry	-rod	-	-	-	-	+/+	+	-	-
	Wet	-rod	+	-	-	-	+/+	+	-	-
	R.Muruny Dry	-rod	-	-	-	-	+/+	+	-	-
	Wet	-rod	+	+	-	-	-/+	+	-	-
V. Cholerae	R.Weiwei Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	-	+	+	-/+	-	-	+
	R.Chesogon Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	-	+	-	-/+	+	-	+
	R.Lomut Dry	-rod	-	-	-	-	-/+	-	-	+
	Wet	-rod	-	+	-	+	-/+	-	-	+
	R.Muruny Dry	-rod	-	-	-	-	-/+	-	-	+
	Wet	-rod	-	+	-	-	-/+	+	-	+
Salmonella & Shigella	R.Weiwei Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	+	-	-	-/+	+	-	+
	R.Chesogon Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	+	+	-	-/+	-	-	+
	R.Lomut Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	+	+	-	-/+	-	-	+
	R.Muruny Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	-	+	-	-/+	-	-	+
Fecal Streptococc	R.Weiwei Dry	+cocci	-	-	+	-	-/-	-	-	-
	Wet	+cocci	-	-	-	-	-/-	-	-	+
	R.Chesogon Dry	+cocci	-	-	+	-	-/-	-	-	-
	Wet	+cocci	-	-	+	-	-/-	-	-	-
	R.Lomut Dry	+cocci	-	-	-	-	-/-	-	-	-
	Wet	+cocci	-	-	+	-	-/-	-	+	+
	R.Muruny Dry	+cocci	-	-	+	-	-/-	-	-	-
	Wet	+cocci	-	-	+	-	-/-	-	+	-

Table 3. Total number of bacteria colonies in the rivers of Sigor division

Sample		Total bacteria colony forming units (cfu/100ml sample)				
		Lowest detectable value	Maximum detectable value	Average	Std. deviation	*P value*
Wet Season	R. Weiwei	86.00	95.00	90.67	3.22	.05
	R. Chesogon	34.50	49.00	40.08	5.78	.05
	R. Lomut	55.00	89.00	74.60	13.75	.05
	R. Muruny	160.00	220.00	183.13	19.07	.05
Dry Season	R. Weiwei	65.00	275.00	136.85	82.70	.05
	R. Chesogon	27.00	121.00	57.07	33.57	.05
	R. Lomut	45.00	260.00	127.44	78.77	.05
	R. Muruny	40.00	290.00	165.81	83.09	.05

Table 4. The number of observed coliform units in the rivers of Sigor division

Sampling site	Combination of positives	MPN index/100 ml	95 % confidence limit		Grade
			Lower	Upper	
Dry season					
RM	5-4-4	350	160	820	Unsatisfactory
RW	5-5-2	500	200	2000	Unsatisfactory
RL	5-5-4	1600	600	5300	Unsatisfactory
RC	5-5-5	≥2400	-	-	Unsatisfactory
Wet season					
RM	5-5-5	≥2400	-	-	Unsatisfactory
RW	5-5-4	1600	600	5300	Unsatisfactory
RL	5-5-5	≥2400	-	-	Unsatisfactory
RC	5-5-5	≥2400	-	-	Unsatisfactory

Key: RM=River Muruny, RW=River Weiwei, RL=River Lomut, RC= River Chesogon; MPN=Most Probable Number; P<0.05

The population of bacteria colonies in the rivers during the dry season were found to be significantly higher than those during the wet season. During the wet season, there is more precipitation and liquid discharge into the rivers. The concentration of bacteria colonies is thus limited by the high amount of water present. However, during the dry season there is less precipitation and the rivers have less water. This implies that the concentration of bacteria colonies present is quite higher. Additionally, during the dry season the temperature is high promoting more breeding of the pathogens. Both seasons however had bacteria colonies above permissible WHO standards for clean drinking water (0cfu/100ml water sample). The acceptable *E. coli* limits for 30 days in a water system not to be considered as bacteria-polluted is 126cfu/100mls [26]. Going by these standards, only Chesogon river during the dry season is bacteria-safe. Muruny river had bacteria colonies surpassing these levels during both wet and dry seasons. This is attributed to its nearness to Sigor township therefore experiencing more sewer contamination compared to the other rivers.

3.4 Total Coliform Units

The level of contamination amongst the four rivers varied amongst the seasons with the highest coliform counts recorded during the wet season and the lowest recorded during the dry season as illustrated in Table 4.

The number of coliforms observed during the wet season was higher than the limit of 2400 units for all rivers except Weiwei river which had 1600 coliform units. Most rivers had excessive coliform units that could not even be accurately counted. The high amounts of coliform units in the rivers during the wet season can be attributed to discharge of human excreta. This can either be in the form of sewerage or directly through leaching out of fecal matter disposed in the bush by residents lacking pit latrines. Chesogon river had the most coliform units during both dry and wet season due to its location where there is high pastoral activity. A good number of her residents also lack pit latrines and defecate in the bushes. Most coliforms, especially the fecal ones are responsible for a string of gastro-intestinal and urinary infections. The permissible WHO standards for maximum coliform units in clean potable water is 0MPN/100ml of water sample [25]. Authorities in this region should therefore move with speed to restore the hygiene of its river to avoid human, animal and capital losses.

4. CONCLUSIONS

Strains of *E. coli, V. cholerae, Salmonella* and *Shigella* species were confirmed to be present in the four rivers of Sigor division. *F. streptococci* was only found present on during the wet season. The bacteria were found to co-exist together. There was a large disparity in bacteria colony forming units between the dry and wet season, with the population of bacteria being more during the dry season. This would indicate more prevalence of gastro-intestinal and urinary tract infections during the dry season.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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