

Variation of total coliforms and bacteria during dry and wet seasons in Rivers of Sigor Division, West Pokot County, Kenya

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Abstract

Clean potable water is a challenge in most developing nations. 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. The four rivers in this region are Weiwei, Chesogon, Lomut and Muruny. This study aimed at characterizing the strains and quantities of bacteria and coliforms in the four rivers during the wet and dry seasons. 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 characterized. Further biochemical tests were conducted to confirm the exact strains present. Total viable counts for the bacteria and coliforms were then enumerated. 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. 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.

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 weather varies from hot and warm days to cool and cold nights. Temperature ranges between a minimum of 10°C and a maximum of 35°C in different parts of the county. Rainfall varies from 400 mm (lowlands) to 1500 mm (highlands) per annum with evaporation rates of between 58 mm – 338 mm per month¹. 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 regions².

Escherichia coli is one of the most common bacteria resulting from consumption of untreated river³. There are other common bacteria such as those of *Fecal streptococcus* and *enterococcus* which have also been traced to river water⁴. 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 rods⁵. They have for a long time been used as indicators of potable water. They survive in the range of $37^{\circ}C^{6}$. The source of these microbes is the gastrointestinal 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 International Research Journal of Environmental Sciences ______ Vol. 10(1), 15-23, January (2021)

 $45^{\circ}C^{7}$. *Escherichia*, *Shigella* and *Salmonella* families are some of the members of fecal coliforms which are responsible for water-borne diseases⁸.

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.

Materials and methods

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). Figure-1 illustrates the four rivers and regions that were sampled.

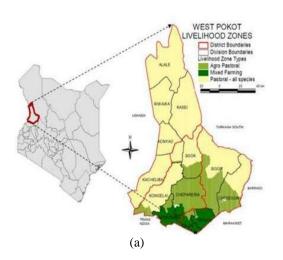




Figure-1: Map of West Pokot county in Kenya $(a)^9$ Sigor division showing the locations of the rivers $(b)^{10}$

A sample was collected from each site packed in sterile 500ml polypropylene bottles. They were then transported to Kenyatta University Microbiology Laboratory for analysis.

Sample Collection: Samples were collected from 4 locations located immediately upstream and at least 100m downstream. This was done four times over a period of eight months as from January to September 2013 which spanned through the dry season (January 2013 – March 2013) and the rainy season (April 2013 – June 2013) to cater for seasonal variations. Samples at the different courses of a particular river were used to make a composite sample. A total of 32 samples were collected during the sampling period with each point sampled four times. The sample size for the study was determined using Fisher formula with a confidence level of 95% with a margin error of 5%¹¹.

$$n = \frac{Z^2 P q D}{d^2} \tag{1}$$

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

Methods used: Inoculation and Morphological characterization of bacteria species.

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

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Test for *Escherichia coli:* A loopful of the positive broth from the presumptive test was streaked onto Eosin Methylene Blue agar (EMB) and incubated at a temperature of 37°C for 24 hours. The test was completed by making a thin smear for Gram staining from the metallic sheen colonies and confirmed by biochemical tests¹². The populations of the viable coliforms were obtained and a number of counted colonies multiplied by the dilution reciprocal.

Test for *Vibrio cholerae:* Water samples were streaked on TCBS (thiosulfate-citrate bile salts sucrose) agar warmed to room temperature and inoculated aerobically at a temperature of 35° C 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 temperature¹³.

Test for *Salmonella species:* One (1) ml of each sample from different sites was mixed well with 10 ml of selenite F broth and the mixture was incubated for 24 hours at 35°C. Streaking was carried out from the same enriched samples on Deoxycholate Citrate Agar (DCA), *Salmonella-Shigella* (SS) agar and MacConkey agar¹⁴. Enumeration of typical colonies was carried out using colony counter and Gram staining was done. Typical colonies were confirmed by biochemical tests such as Triple sugar iron and urease tests.

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 37°C for 18 hours.

Biochemical analysis of the isolates: Seven biochemical tests were conducted as outlined below.

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.

Indole Test: An isolated colony of the test isolates was emulsified in tryptophan broth and incubated at 37°C for 48 hours. 0.5 ml of Kovac's reagent was then added to the broth culture. A pink color appearing as a layer on the media after one minute indicated a positive test while a negative result was shown by no color change.

Methyl red test: A pure colony from the isolated bacteria was inoculated into a 0.5ml of sterile glucose-phosphate broth.

Overnight incubation was done at a temperature of 37° C, with a drop of methyl red solution added. The appearance of bright red color was an indication of a positive methyl red test showing the presence of *E. coli*.

Voges-Proskauer (VP) test: MR/VP broth was inoculated with a pure culture of the test organism and incubated for 24 hours at 35°C. After incubation, 1 ml of this broth was moved to clean test tubes, 0.6ml of 5% alpha naphthol was added than a further 0.2ml of 40% KOH. A gentle shake was given to the tubes them allowed to settle for 10 to 15 minutes. A positive test was shown by the development of a red color 15 minutes or more after the addition of the reagents indicating the presence of diacetyl, the oxidation product of acetoin. The yellow-brown color indicated a negative result.

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 24 hours at 35°C 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.

Triple Sugar Iron (TSI) test: A sterilized inoculation needle was streaked at the middle of a well-isolated colony of microbes. TSI agar was then inoculated, first by stabbing through the middle of the media to its bottom. The surface of the agar slant was thereafter streaked. The cap of the tube used was then loosened and left to incubate at 35°C in ambient air for 18 to 24 hours. Color changes of both the Slant/Butt as well as CO_2 and H_2S yielding metabolic processes were all recorded.

Citrate test: A needle tip was used to pick some bacterial cells from the cultured colonies. This was inoculated in the Simmons citrate agar (EMB agar containing: sodium citrate, an ammonium salt, and bromothymol blue indicator) on the slant and incubated for 24 hours at 37°C. Color changes in the media were observed. When it grew to medium and changed color from green to blue due to alkaline reactions, it confirmed a positive test.

Total viable counts/Microbial loads: From the samples collected from the four different rivers, the spread plate technique was done using a serial dilution of up to 10^4 to allow the growth of distinguishable colonies. From the 10^4 dilution, one milliliter of the sampled water was pipetted into sterile petri-dish and sterile melted EMB agar was added and mixed well by gently swirling the plate. The plate was covered and incubated at 35 - 37°C for 24- 48 hours. Resulting colonies were counted using a colony counter. The sample volume used, the size of dilution and the colonies formed were used to calculate the CFU/ml in the water sample.

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

$$N x D x V = CFU/ml \tag{2}$$

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

Presumptive Coliform Counts: Probable number technique was used to determine the presence of coliforms. A series of lauryl tryptose broth (LTB) fermentation tubes were inoculated with 10 ml, 1 ml and 0.1 ml of the sample¹². The formation of gas at a temperature of 37°C within 48 hours constituted a positive presumptive test. To confirm the test, inoculum from positive tubes were transferred to tubes containing 2% brilliant green bile lactose broth (Oxoid), dispensed in fermentation tubes fitted with inverted Durham tubes and incubated at a temperature of 37°C for 24 hours. The number of coliform units detected were then counted. The MPN results were graded according to water suitability for consumption. The water samples were graded according to the number of total coliform counts. The water samples that had a presumptive total coliform count between 1-3 was rated as satisfactory and the water samples that had counts above 10 were rated as unsatisfactory.

Data analysis: Numerical data of the microbes was given as mean \pm standard deviation. 95% confidence level (P > 0.05) was used for the statistical interpretations. Ms Excel (version 2016) and OriginLab (version 6.5) were used for statistical analysis.

Results and discussions

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.

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 broth¹⁶. 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 diseases¹⁷. 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 change¹⁸. It was however noted that Fecal streptococci species were the least dominant compared to other bacteria species analyzed.

Table-1: Bacteria species occurrence in the rivers of the sam	nple area during wet and dry seasons.
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	Rivers assessed								
Bacteria tests	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	+	-	+	-	+	-	-	-	

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 water¹⁹. 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 *E. coli, Shigella, 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.

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

		Biochemical Tests								
Isolates	Samples	Gram	Indole	MR	VP	Urease	TSI			Citrate
		Stain	Indole				S/B	CO_2	H ₂ S	Citrate
	R.Weiwei Dry	-rod	+	-	-	-	+/+	+	-	-
	Wet	-rod	+	+	-	-	+/-	+	-	-
	R.Chesogon Dry	-rod	-	-	-	-	+/+	+	-	-
E. coli	Wet	-rod	+	+	-	-	+/+	+	-	-
E. con	R.Lomut Dry	-rod	-	-	-	-	+/+	+	-	-
	Wet	-rod	+	-	-	-	+/+	+	-	-
	R.Muruny Dry	-rod	-	-	-	-	+/+	+	-	-
	Wet	-rod	+	+	-	-	-/+	+	-	-
	R.Weiwei Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	-	+	+	-/+	-	-	+
	R.Chesogon Dry	-rod	-	+	-	-	-/+	-	-	+
V. Cholerae	Wet	-rod	-	-	+	-	-/+	+	-	+
v. Cholerde	R.Lomut Dry	-rod	-	-	-	-	-/+	-	-	+
	Wet	-rod	-	+	-	+	-/+	-	-	+
	R.Muruny Dry	-rod	-	-	-	-	-/+	-	-	+
	Wet	-rod	-	+	-	-	-/+	+	-	+
	R.Weiwei Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	+	-	-	-/+	+	-	+
	R.Chesogon Dry	-rod	-	+	-	-	-/+	-	-	+
Salmonella &	Wet	-rod	-	+	+	-	-/+	-	-	+
Shigella	R.Lomut Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	+	+	-	-/+	-	-	+
	R.Muruny Dry	-rod	-	+	-	-	-/+	-	-	+
	Wet	-rod	-	-	+	-	-/+	-	-	+
	R.Weiwei Dry	+cocci	-	-	+	-	-/-	-	-	-
	Wet	+cocci	-	-	-	-	-/-	-	-	+
	R.Chesogon Dry	+cocci	-	-	+	-	-/-	-	-	-
Fecal	Wet	+cocci	-	-	+	-	-/-	-	-	-
Streptococci	R.Lomut Dry	+cocci	-	-	-	-	-/-	-	-	-
	Wet	+cocci	-	-	+	-	-/-	-	+	+
	R.Muruny Dry	+cocci	-	-	+	-	-/-	-	-	-
	Wet	+cocci	-	-	+	-	-/-	-	+	-

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 rivers³ 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 tracts²¹. 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 (CO₂). A few strains form hydrogen sulphide (H₂S) 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 gases. 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 seasons²². Their source is fecal discharge from infected people and cattle. Negative citrate test

confirmed strains of *Escherichia*, *Shigella*, *Morganella* and *Yersinia* species²³.

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.

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 number of bacteria colonies detected in the 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-3: Total number of bacteria colonies in the rivers of Sigor division.

		Total bacteria colony forming units (cfu/100ml sample)							
Sa	ample	LowestMaximumDetectable ValueDetectable ValueAverageStd. DeviationP value							
	R. Weiwei	86.00	95.00	90.67	3.22	0.05			
Wet	R. Chesogon	34.50	49.00	40.08	5.78	0.05			
Season	R. Lomut	55.00	89.00	74.60	13.75	0.05			
	R. Muruny	160.00	220.00	183.13	19.07	0.05			
	R. Weiwei	65.00	275.00	136.85	82.70	0.05			
Dry Season	R. Chesogon	27.00	121.00	57.07	33.57	0.05			
	R. Lomut	45.00	260.00	127.44	78.77	0.05			
	R. Muruny	40.00	290.00	165.81	83.09	0.05			

The number 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/100mls²⁶. 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.

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 sample²⁵. Authorities in this region should therefore move with speed to restore the hygiene of its river to avoid human, animal and capital losses.

Conclusion

E. coli, *V. cholerae, Salmonella* and *Shigella* species were found to be the most prevalent microbes in the four rivers of Sigor division, West Pokot county. *F. streptococci* was only observed during the wet seasons. Biochemical analyses of isolates conducted proved that the pathogens had several strains co-existing together in the water samples. Muruny river had the highest bacteria colony forming units with 183.13cfu/100ml during the dry season and 165.81/100ml during the wet season. A large disparity in colony forming units was observed in the rivers during the dry season. There were too many coliform units in the rivers during the wet season. The water in Sigor division was concluded to be unsatisfactory for animal and human consumption during both dry and wet seasons. Chesogon river, had the most coliform units detected.

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

Sampling site	Combination of		95 % confi						
	positives	MPN index/100 ml	Lower	Upper	Grade				
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

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References

- Report (2017). Meteorological department, West Pokot County, Kenya (2019). https://www.meteo.go.ke/index .php?q=today. Accessed on 20th October, 2019.
- Logan, L. K., & Weinstein, R. A. (2017). The Epidemiology of Carbapenem-Resistant *Entero bacteria ceae*: The Impact and Evolution of a Global Menace. *The Journal of infectious diseases*, 215(1), S28–S36. https://doi.org/10.1093/infdis/jiw282
- **3.** Cabral, J. P. (2010). Water microbiology. Bacterial pathogens and water. *International journal of environmental research and public health*, 7(10), 3657–3703. https://doi.org/10.3390/ijerph7103657
- Lin, J. & Ganesh, A. (2013). Water quality indicators: bacteria, coliphages, enteric viruses. *International Journal* of Environmental Health Research, 23, 6, 484-506, doi:10.1080/09603123.2013.769201
- Martin, N. H., Trmčić, A., Hsieh, T. H., Boor, K. J., & Wiedmann, M. (2016). The Evolving Role of Coliforms as Indicators of Unhygienic Processing Conditions in Dairy Foods. *Frontiers in microbiology*, 7, 1549. https://doi.org/10.3389/fmicb.2016.01549
- 6. Guyot, S., Pottier, L., Hartmann, A., Ragon, M., Hauck Tiburski, J., Molin, P., Ferret, E., & Gervais, P. (2014). Extremely rapid acclimation of *Escherichia coli* to high temperature over a few generations of a fed-batch culture during slow warming. *Microbiology Open*, 3(1), 52–63. https://doi.org/10.1002/mbo3.146
- 7. Dockins, W. S., & McFeters, G. A. (1978). Fecal coliform elevated-temperature test: a physiological basis. *Applied and environmental microbiology*, 36(2), 341–348.
- Dekker, J. P., & Frank, K. M. (2015). Salmonella, Shigella, and yersinia. *Clinics in laboratory medicine*, 35(2), 225– 246. https://doi.org/10.1016/j.cll.2015.02.002
- Report, NDMA (2014). National Drought Management Authority (NDMA). https://www.researchgate.net/ publication/285131644_Influence_of_Enclosure_ Management_Systems_on_Rangeland_Rehabilitation_in_C hepareria_West_Pokot_County_Kenya/figures?lo=1. Accessed on 26th June, 2020.
- **10.** Maps (2020). Downloaded and edited from Google maps. https://www.google.com/maps/place/West+ Pokot +County/@1.8935329,34.7209293,9z/data=!3m1!4b1!4m5! 3m4!1s0x178247e414a72f33:0x63bd5be869cf4ce0!8m2!3d 1.6210076!4d35.3905046 Accessed on 26th June 2020

- **11.** Fisher, L. D. (1998). Self designing clinical trials. *Statistics in medicine*, 17(14), 1551-1562.
- **12.** Sacchetti, R., De Luca, G., and Zanetti, F. (2009). Control of *Pseudomonas aeruginosa* and *Stenotrophomonas maltophilia* contamination of microfiltered water dispensers with peracetic acid and hydrogen peroxide. *International journal of food microbiology*, 132(2-3), 162-166.
- Furniss, A. L., Lee, J. V., and Donovan, T. J. (1978). The vibrios. HM Stationery Office London. U.K. ISBN-9054103264, 9789054103264.
- 14. Andrews, W. H., and Hammack, T. S. (2003). Food sampling and preparation of sample homogenate. *Bacteriological Analytical Manual*, 1-10.
- 15. Kranz, R., Weston-Hafer, K., and Richards, E. (2006). Identifying unknown bacteria using biochemical and molecular methods. Washington University in Saint Louis. (Lab Report) Retrieved from http://www.nslc.wustl.edu/ elgin/genomics/Bio3055/IdUnknBacteria06.pdf Accessed on 26th June, 2020.
- **16.** McDonough, P. L., Shin, S. J., & Lein, D. H. (2000). Diagnostic and public health dilemma of lactose-fermenting *Salmonella enterica* serotype *Typhimurium* in cattle in the Northeastern United States. *Journal of clinical microbiology*, 38(3), 1221–1226.
- 17. Humphries, R. M., & Linscott, A. J. (2015). Laboratory diagnosis of bacterial gastroenteritis. *Clinical microbiology reviews*, 28(1), 3–31. https://doi.org/ 10.1128/CMR.00073-14
- **18.** Reid, R. L., Porter, R. C., Ball, H. J. (1993). The isolation of sucrose-fermenting *Salmonella mbandaka.Vet Microbiol*. 37(1-2), 181-185. doi:10.1016/0378-1135(93) 90192-a
- 19. Kramer, A., Schwebke, I., & Kampf, G. (2006). How long do nosocomial pathogens persist on inanimate surfaces? A systematic review. *BMC infectious diseases*, 6, 130. https://doi.org/10.1186/1471-2334-6-130
- **20.** Lin, Y. C., & Peterson, M. L. (2010). New insights into the prevention of *staphylococcal* infections and toxic shock syndrome. *Expert review of clinical pharmacology*, 3(6), 753–767. https://doi.org/10.1586/ecp.10.121
- **21.** Testerman, T. L., & Morris, J. (2014). Beyond the stomach: an updated view of *Helicobacter pylori* pathogenesis, diagnosis, and treatment. *World journal of gastroenterology*, 20(36), 12781–12808. https://doi.org/10. 3748/wjg.v20.i36.12781
- 22. Manyi-Loh, C. E., Mamphweli, S. N., Meyer, E. L., Makaka, G., Simon, M., & Okoh, A. I. (2016). An Overview of the Control of Bacterial Pathogens in Cattle Manure. *International journal of environmental research* and public health, 13(9), 843. <u>https://doi.org/</u> 10.3390/ijerph13090843

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- 23. Morka, K., Bystroń, J., Bania, J. (2018). Identification of *Yersinia enterocolitica* isolates from humans, pigs and wild boars by MALDI TOF MS. *BMC Microbiol*, 18, 86. https://doi.org/10.1186/s12866-018-1228-2
- 24. Gwimbi P. (2011). The microbial quality of drinking water in Manonyane community: Maseru District (Lesotho). *African health sciences*, 11(3), 474–480.
- **25.** WHO. (1997). Guidelines for Drinking Water Quality. 2nd Edition. Vol. III. Geneva: Surveillance and Control of Community Supplies; 1997
- **26.** World Science, (2014). Bacteria total oliform. Retrieved from https://worldwidescience.org/topicpages/b/ bacteria+ total+coliform.html.Accessed on 10th June, 2020.