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Original article

OCCURRENCE AND COMPARATIVE ANALYSIS OF BACTERIA IN SOURCES OF WATER IN LAPAI, NIGER STATE, NIGERIA

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ABSTRACT

This study was carried out to comparatively analyse the bacteria occurrences in Lapai Metropolis sources of water supply. Bacteriological quality of water samples was determined by enumeration through Total Viable Count (TVC) and Coliform count using the Most Probable Number (MPN) method. Maximum TVC and Coliform count - were recorded during the rainy season (2.46 x 10^8cfu/ml and > 1100 MPN index/100ml) in the water samples from streams, while minimum TVC and Coliform count were recorded during the dry season (9.0 x 10^6cfu/ml and 0MPN index/100ml) in the water samples from Boreholes. Total bacteria count from all the water sources in Lapai metropolis during the dry and rainy season indicated that *Escherichia coli* and *Klebsiella* sp occurred most. There are more occurrences of different bacteria species in the month of February than March. The other variations in the occurrence of bacteria in the rainy season are the presence of *Serratia* sp in the month of June only, and *Chromobacterium violaceum* in the month of July. There exists a significant difference ($P<0.05$) with the P-Value of 0.026 and 0.009 in the Total Viable Count during the dry and rainy season respectively, while there was no significant difference ($P>0.05$) with P-Value of 0.211 and 0.274 in the coliform count during the dry and rainy season. There was also significant difference ($P<0.05$) with P-Value of 0.002 in the percentage occurrence of bacteria during the dry season, while there was high significance difference ($P\leq0.001$) with P-Value of 0.001 in the percentage occurrence of bacteria during the rainy season. This study has indicated that most water samples examined did not meet up with the WHO standard for portable water, and the presence of these bacteria are potential pathogens that can significantly affect microbiology water quality, resulting to great health risk.

Key words: Comparative, analysis, Bacteria, Water, Sources

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INTRODUCTION

Water is one of the most essential natural resources on earth needed to sustain life. It is important and useful to all forms of life [1]. The availability of water and accessibility of portable water plays a vital role in economic advancement and social welfare of a nation as well as it is an important element in health and food production [2]. Despite the relative abundance of water in supply, the quality and accessibility of clean fresh water remains a global challenge. Ground water accounts for the ultimate and commonest supply system for most water use for consumption by humans to both urban and rural areas in Nigeria and other developing countries. Nearly all the states and communities in Nigeria depend directly or indirectly on ground water for drinking and other purposes [3]. Poor water quality continues to cause major threats to human health. Presently, contaminated water has been reported to kill quite a number of people compared to other deadly diseases such as cancer and AIDS [1].

The quality of drinking water has drastically deteriorated over the time due to insufficient and inadequate management of the piped water distribution system. This is as a result of direct discharge of untreated or improperly treated sewage or industrial effluent into the water bodies. The deterioration in water quality is also due to location of water sources such as wells and boreholes in close proximity to latrines. Contamination of natural fresh water with faecal materials, industrial wastes and other wastes materials including Agricultural wastes may result in to a noticeable increase in the risk of transmission of water borne diseases to the inhabitants or populace that uses such water [4]. Today, contaminated water has been reported to kill more people than HIV/AIDS, cancer, war or even accident especially children [5]. Diarrheal diseases caused by poor sanitation and consumption of contaminated water account for an estimate of 4.1% of total daily global burden of disease and are responsible for about 1.8 million death rates every year. About 88% of the total global burden is attributed to unsafe water supply, poor sanitation and poor hygiene [6]. Microbial contamination of most drinking water supplies especially contamination from human faeces is known to be the major contributor to diarrheal disease that kills millions of people especially children under the age of five years every year [7][1].

Adherence to the microbial quality of drinking water can offer a high degree or level of protection to consumers against any form of water borne disease and help to curtail the spread and outbreak of any water borne disease [8]. The Enterobacteriaceae family which includes gram-negative, non-spore forming bacteria are of major importance [9][10]. Four groups of the bacterial belonging to the family; Enterobacteriaceae such as *Escherichia coli, Enterobacter*, *Klebsiella* and *Citrobacter* are the most common known important indicator organisms of faecal coliform [11]. *Escherichia coli*s one of the specific indicators of faecal contamination in tropical and temperate regions. Investigation of the bacterial

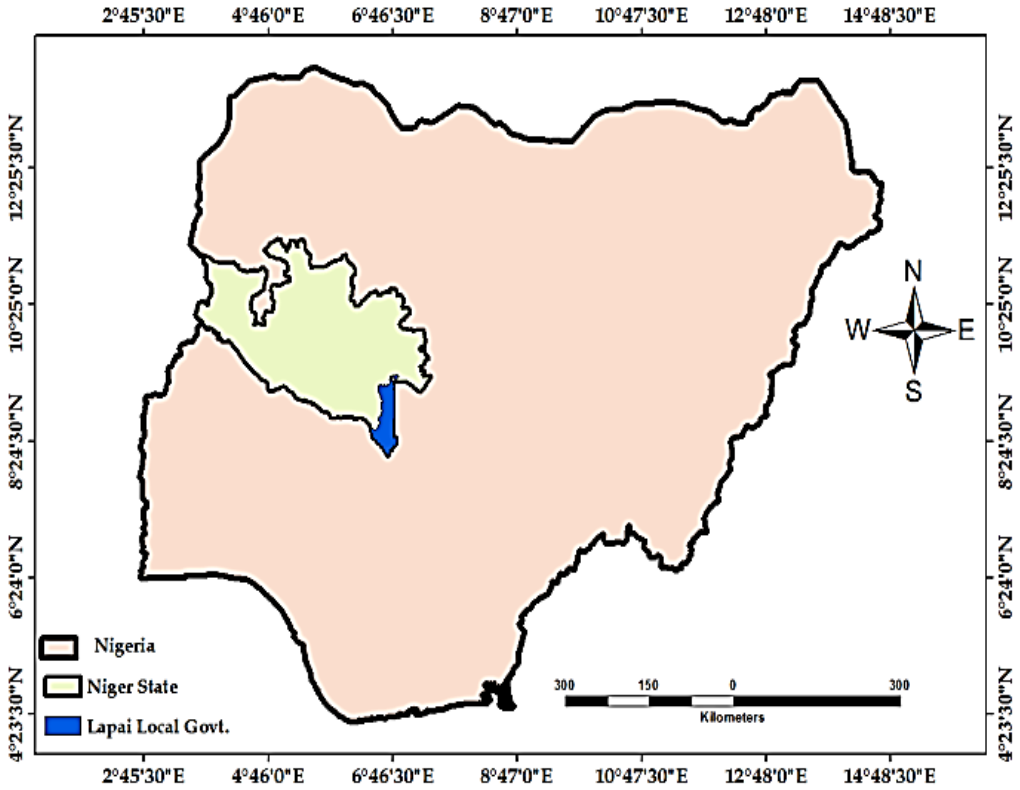
density of water could provide an approach to assess the reliability of monitoring data [12]. Enterohaemorrhagic *E. coli* have emerged as a serious gastrointestinal pathogen in many countries. Although the mode of transmission is mainly through the consumption of contaminated meat, outbreaks associated with water-borne enterohaemorrhagic *Escherichia coli* have also been described. In the Danube river basin, total coliforms, faecal coliforms and *Escherichia coli* indicate persistent contamination, with lower values of total coliforms in July and the highest value in August. Variations in these parameters could be spatio temporarily linked to the number of visitors in this ecosystem [13]. Indicator organisms are widely distributed in different proportions in different environment and habitats such as contaminated or untreated water, sewage, vegetables and even in food [14][15].

The reasons for seasonal variation in species and level of the faecal pollution most especially in rivers and in streams is due to land drainage occurring majorly during the raining seasons. That is, the level of faecal indicator bacteria, other bacteria groups and pathogenic enteric bacteria are mainly influenced by discharge of effluents from waste water treatment plants, industrial, agricultural and domestic wastes, particularly those containing human wastes as well as the nature of water shed [16]. Soil leaching and surface water run-off also contribute significantly to the faecal pollution or contamination of the aquatic or water environment [17]. In rivers and streams, the occurrence and survival of these faecal coliform is majorly as a result of their association with particles and their ability to remain resistant to environmental factors such as temperature, pH, Pressure, salinity and organic matter and nutrients present in water environment [18]. This study is aimed at analysing and comparing bacteria occurrence in Lapai water sources.

METHODOLOGY

Study Area

The research study was conducted in Lapai metropolis. Lapai is a Local Government Area in Niger State, Nigeria adjoining the Federal Capital Territory. Its headquarters is in the town of Lapai. Lapai is located between longitudes 4°27'30" to 13°60’95” North and 2°60'60" to 14°89'44". The region is bounded to the north by Minna, Paiko and Agaie local government areas, to the south by River Niger and Kogi state, to the west by Lavun local government and to the east by Gurara local government and the Federal Capital Territory of Nigeria. According to Nigerian National Population Commission (NNPC) 2006 figures, Lapai has a total population of over 184,000 spread across the area.



MAP OF NIGERIA SHOWING NIGER STATE AND LAPAI LOCAL GOVERNMENT

Sample Collection

After several preliminary visits and study to various cardinal points (North, South, east, and West) and areas in Lapai metropolis, twelve [12] sampling sites comprising of three [3] different water sources that includes; well water, surface water from streams and rivers and borehole water samples were collected from the four cardinal points (North, South, east, and West), one sample from well water, one sample from streams and rivers and one sample from borehole water in the four cardinal points making up three [3] samples from each cardinal point and twelve [12] samples in all.

Samples were collected once every month in each of the months of February and March, 2018 (representing the dry season) the months of June and July, 2018 (representing the rainy season). Each of the cardinal point selected had at least a borehole, well and a river or stream as a principal source of water for the inhabitant either for consumption or other domestic uses. The sampling points were selected such that the samples taken served as representative of the different sources from which each cardinal point in the community obtains water.

Total of forty-eight [48] water samples were collected from the four cardinal points for bacteriological assessment in the months of February and March (dry season) and the months of June and July (rainy season) of the year 2018. The sample collection represents two major seasons in Nigeria i.e. dry seasons and the rainy seasons. All water sampling, transportation and preservation

procedures were carried out according to standard methods for examination of water (World Health Organization guidelines for drinking water quality) [6]. Samplings especially for bacteriological analysis were done aseptically to ensure no external contamination of samples. All water samples collected were transported to the laboratory in an Ice Park within the time frame of two hours. Exactly 200mls capacity glass bottle was used for sampling. The bottles were fitted with ground glass stopper or screw caps. The cap and neck of the bottles were protected from contamination by covering with thin aluminium foil. The glass bottles were sterilised at 121^{o}C for 15 minutes. All samples collected were packaged in an ice pack and were transported to the laboratory. The samples were processed and analysed in the laboratory within the period of 2 hours.

Sample Collection from Streams and Rivers

The cap of the sterile sample bottle was aseptically removed and the mouth of the bottle was faced towards the flow way of the water. Where inconvenient and to avoid entering the water, the neck of the bottle was clamped to end of a stick by fixing the bottle neck in a retort stand and then was clamped and mounted on a stick. The neck was plunged downwards to about 30cm below the water surface [19] and the neck tilted slightly upwards to fill the bottle before the cover was replaced carefully and aseptically, and the bottle was labelled.

Sample Collection from Open Wells.

A sterile sample bottle was tied to a heavy length of rope and a stone was used as a weight in which the bottle was attached above the stone. The cap was removed aseptically from the bottle and the bottle was lowered into the well to the depth of about 1 metre from the surface of well water. When no more air bubbles were observed to rise to the surface, the bottle was raised out of the well and the cap was carefully replaced and the bottle was then labelled.

Sample Collection from Borehole

The hand pump was continuously operated for at least 5 minutes, and several litres of water was pumped to waste [19]. The sample of water was collected aseptically by allowing the water from the pump to flow directly into the sterile sample bottle and the cap of the bottle was carefully replaced. The bottle was labelled.

Total Viable/Heterotrophic Count

In the determination of total viable/heterotrophic count, a tenfold (10^{-1} to 10^{-10}) serial dilution was set up. 1ml of sample from the water (100ml) to be tested was transferred into the first test tube and this was done to the tenth test tube (Serial dilution). 1ml of the diluent from dilution factor 10^{-6} was inoculated into prepared plate of nutrient agar (20ml) by pour plate method. Wire loop was used to spread the sample onto the surface of the nutrient agar and the plate was incubated at 37^{o}C for 48 hours. Pure cultures of the isolate were obtained by sub-culturing on the surface of freshly prepared nutrient agar plates. The colonies were counted using colony counter, gram stained and subjected to Biochemical tests for identification of each bacteria species.

Determination of the Most Probable Number (MPN)

The most probable number method is also referred to as the multiple tube method. The method is based on an indirect assessment of microbial density in the water sample by reference to statistical table to determine the most probable number of microorganisms present in the original water sample.

The most probable number (MPN) method was used for the enumeration of total coliform isolates. 10mls of water from each of the sample source; well water, borehole water and stream was dispensed into each of three set of test tubes containing 10mls of double strength Lactose broth containing inverted Durham's tube, 1.0ml of each of the test water sample into three set of test tubes containing 10mls of single strength lactose broth and 0.1ml of each of the test water sample into each of three set of test tube containing 10mls of single strength lactose broth using sterile syringe. Durham's tubes were observed at the end of each incubation period for gas production, colour change and turbidity [19].

The most probable number (MPN) was carried out in three steps:

1 Presumptive test
2 Confirmatory test
3 Completed test.

1. *Presumptive Test:* Coliform count was obtained using the three tube method of the most probable number (MPN) method. Double strength and single strength

Lactose broth was prepared and using a sterile pipette, 10mls of double strength Lactose broth was dispensed into three test tubes containing inverted Durham tubes and the tubes were labelled LB2X. Also, 10mls of single strength Lactose broth was dispensed into 6 test tubes containing inverted Durham tubes and were labelled LB1X. The nine [9] set of test tubes were autoclaved at 121^0C for fifteen [15] minutes and was allowed to cool to room temperature. The water sample to be tested was mixed thoroughly by shaking. Using 10mls sterile pipette, 10mls of water sample to be tested was dispensed into 3 test tubes containing double strength Lactose broth, 1ml of water sample was also dispensed into 3 single strength Lactose broth tubes. Exactly 0.1ml of water sample was dispensed into the other 3 single strength Lactose broth tubes. All the nine [9] set of test tubes were shaken gently to ensure even mixture and were incubated aerobically at 37^0Cfor 24 to 48 hours for estimation of total coliform and at 44^0C for faecal coliform. All the tubes were examined for production of acid (yellow colour) and gas production after 24 to 48 hours of incubation. Positive presumptive tubes were retained and the most probable number was then estimated from the MPN (most probable number) statistical table.

2. *Confirmatory Test:* This was carried out by dispensing 0.1ml from Lactose broth positive presumptive tubes into a freshly prepared sterile Brilliant Green Lactose Bile tubes containing inverted Durham tubes. The tubes were shaken gently for even mixture and were all incubated at 44^0C for 48 hours. The tubes were all observed for gas production after 48 hours of incubation. The record of number of tubes showing positive confirmed test was taken. The Most Probable Number (MPN) was determined using the MPN statistical table.

3. *Completed Test:* Completed test was performed by streaking a loopful of broth from positive tube onto freshly prepared Eosin Methylene Blue (EMB) agar plate for pure colonies. The plates were incubated. Brilliant Green Lactose broth and Nutrient agar slant were inoculated with coliform culture from the Eosine Methylene Blue (EMB) agar plates. The broth tubes and the agar slant were incubated at 35^0C for 24 hours. The Lactose broth fermentation tubes were observed for gas production and the organisms found on the slant were Gram stained. The slides were observed for positive or negative Gram reaction and cell morphology was observed.

Characterization and Identification of Bacteria Isolates

Stock cultures of the pure isolates with different cultural characteristics were made on nutrient agar slants. Gram staining procedure was used to observe cell morphology and biochemical tests were performed to identify the isolates to species level. Various tests performed and used for identification of the isolates included; Oxidase test, Catalase test, Coagulase test, Urease test, Indole test and Citrate utilization test [20].

Statistical Analysis

SPSS was used to carry out statistical analysis of variance (ANOVA) on the Total viable count, coliform count and the percentage occurrence of bacteria. It was also used to test for statistical significance in the results and where significant differences were detected. Duncan's Multiple Range Test (DMRT) was further used to locate the significantly different means.

RESULTS

Total Viable Count of Bacteria during the dry season

Higher total viable counts (TVC) in the water samples were recorded during the dry season in Lapai West Stream (LWS) with 190 colonies (1.9×10^7), closely followed by Lapai West Well (LWW), while lower total viable counts were recorded in Lapai East Borehole (LEB) with 52 colonies (5.2×10^7) and 90 colonies (9×10^7) in the month of February and March respectively.

Table 1: Total Viable Count of bacteria from water sources in Lapai Metropolis during the dry season

		FEBRUARY			MARCH		
S/NO	LOCATION	Dilution factor	Number of colonies	Population (cfu/ml)	Dilution factor	Number of colonies	Population (cfu/ml)
1	LNW	10^{-6}	118	1.18×10^{8}	10^{-6}	111	1.11×10^{8}
2	LNB	10^{-6}	95	9.5×10^{7}	10^{-6}	140	1.4×10^{8}
3	LNS	10^{-6}	165	1.65×10^{8}	10^{-6}	152	1.52×10^{8}
4	LEW	10^{-6}	113	1.13×10^{8}	10^{-6}	130	1.3×10^{8}
5	LEB	10^{-6}	52	5.2×10^{7}	10^{-6}	9	9×10^{6}
6	LES	10^{-6}	171	1.71×10^{8}	10^{-6}	150	1.5×10^{8}
7	LWW	10^{-6}	180	1.80×10^{8}	10^{-6}	172	1.72×10^{8}
8	LWB	10^{-6}	100	1.00×10^{8}	10^{-6}	148	1.48×10^{8}
9	LWS	10^{-6}	130	1.30×10^{8}	10^{-6}	190	1.9×10^{8}
10	LSW	10^{-6}	111	1.11×10^{8}	10^{-6}	140	1.40×10^{8}
11	LSB	10^{-6}	170	1.7×10^{8}	10^{-6}	156	1.56×10^{8}
12	LSS	10^{-6}	122	1.22×10^{8}	10^{-6}	166	1.66×10^{8}

KEY: LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Total Viable Count of Bacteria during rainy season.

Higher total viable counts (TVC) in the water samples were also recorded during the rainy season in Lapai East Stream (LES) with 246 colonies (2.46×10^{8}), closely followed by Lapai West Well (LNS), while lower total viable counts were recorded in Lapai East Borehole (LEB) with 94 colonies (9.4×10^{7}), followed by 105 colonies (1.05×10^{8}) in Lapai North Borehole (LNB). The overall microbial count in different water samples and in both dry and rainy seasons indicated that total viable counts (TVC) in the water samples were highest during the rainy season in Lapai East Stream (LES) (2.46 X 10^{8}cfu/ml), Lapai West Stream (LWS) (2.43 X 10^{8}cfu/ml), Lapai North Stream (LNS) (2.4 X 10^{8}cfu/ml) , while the TVC were lowest during the dry season in Lapai East Borehole (5.2×10^{7}cfu/ml) and Lapai East Borehole (9×10^{7}cfu/ml) of the year. The stream water in the four cardinal points has the highest Total viable count in LES (2.46 X 10^{8}cfu/ml) followed by the well water from LSW (2.08 X 10^{8}cfu/ml) and the borehole water from LWB (1.82×10^{8}cfu/ml) appeared to have the least Total Viable count among them in the dry and rainy season. The borehole water from LEB (5.2×10^{7}cfu/ml) has the best Microbiological water quality throughout the period of analysis in both the dry and rainy seasons. Also, from the results in Tables 1 and 2, Water samples from Lapai West generally as a sampling location is seen to have the highest value of Total viable counts, especially during the rainy season and in all the different water sources analysed.

Table 2: Total Viable Count of bacteria from the water sources in Lapai Metropolis during the rainy season

		JUNE			JULY		
S/NO	LOCATION	Dilution factor	Number of colonies	Population (cfu/ml)	Dilution factor	Number of colonies	Population (cfu/ml)
1	LNW	10^{-6}	196	1.96×10^{8}	10^{-6}	165	1.65×10^{8}
2	LNB	10^{-6}	164	1.64×10^{8}	10^{-6}	105	1.05×10^{8}
3	LNS	10^{-6}	240	2.4×10^{8}	10^{-6}	190	1.90×10^{8}
4	LEW	10^{-6}	206	2.06×10^{8}	10^{-6}	172	1.72×10^{8}
5	LEB	10^{-6}	106	1.06×10^{8}	10^{-6}	94	9.4×10^{7}
6	LES	10^{-6}	246	2.46×10^{8}	10^{-6}	201	2.01×10^{8}
7	LWW	10^{-6}	183	1.83×10^{8}	10^{-6}	213	2.13×10^{8}
8	LWB	10^{-6}	170	1.7×10^{8}	10^{-6}	182	1.82×10^{8}
9	LWS	10^{-6}	192	1.92×10^{8}	10^{-6}	243	2.43×10^{8}
10	LSW	10^{-6}	185	1.85×10^{8}	10^{-6}	208	2.08×10^{8}
11	LSB	10^{-6}	162	1.62×10^{8}	10^{-6}	164	1.64×10^{8}
12	LSS	10^{-6}	226	2.26×10^{8}	10^{-6}	168	1.68×10^{8}

KEY: LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream

Most Probable Number of Coliform bacteria during the dry season

The most probable number of coliform bacteria from water sources in Lapai metropolis during the dry season indicated that Bore- hole water sources (LSB, LEB, LWB) have the least total coliform count in the month of March than in the month of February, except the water source LNB, where there is a record of much higher coliform count. Samples from other water sources had a relatively higher total coliform count of 1100 and above.

Table 3: Most Probable Number of coliform bacteria from the water sources in Lapai Metropolis during the dry season

		FEBRUARY	MARCH
S/NO	LOCATION	MPN Index/100ml (cfu/ml)	MPN index/100ml (cfu/ml)
1	LNW	1100	1100+
2	LNB	15	1100+
3	LNS	1100+	1100
4	LEW	210	1100+
5	LEB	04	00
6	LES	1100+	1100
7	LWW	1100+	1100
8	LWB	15	00
9	LWS	1100+	1100+
10	LSW	1100+	1100+
11	LSB	1100	15
12	LSS	1100	1100+

KEY:

LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Most Probable Number of Coliform bacteria during the rainy season

In table 4, Most Probable Number of coliform bacteria is lowest from Bore-hole water sources (LNB, LEB, LWB) in the month of June, except from LSB, where the MPN value is higher in the month of July. In all other water sources, the MPN values are higher, ranging from 1100 to 1100+.

Table 4: Most Probable Number of coliform bacteria from the water sources in Lapai Metropolis during the rainy season

		JUNE	JULY
S/NO	LOCATION	MPN Index/100ml (cfu/ml)	MPN index/100ml (cfu/ml)
1	LNW	1100	1100+
2	LNB	210	1100
3	LNS	1100	1100+
4	LEW	1100+	1100+
5	LEB	00	460
6	LES	1100+	1100+
7	LWW	1100+	1100
8	LWB	09	1100+
9	LWS	1100+	1100+
10	LSW	1100	1100+
11	LSB	210	23
12	LSS	1100+	1100

KEY: LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Bacteria population in the dry season

Total bacteria count from all the water sources in Lapai metropolis during the dry season indicated that *Escherichia coli* and *Klebsiella* sp occurred most in February and March. However, there are more occurrences of different bacteria specie in the month of February than March as shown in table 5 below, which also reflected in percentage occurrence.

Table 5: Occurrence of Bacteria in the water sources of Lapai Metropolis during the dry season.

S/NO	BACTERIA	FEBRUARY % of occurrence	MARCH % of occurrence
1	*Escherichia coli*	25.00	28.57
2	*Klebsiella* sp	25.00	21.43
3	*Enterobacter* sp	12.50	7.14
4	*Citrobacter* sp	0.00	7.14
5	*Proteus* sp	12.50	7.14
6	*Staphylococcus aureus*	12.50	7.14
7	*Serratia* sp	0.00	14.29
8	*Salmonella* sp	6.25	7.14
9	*Shigella* sp	6.25	0.00
Total		100	100

KEY: LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Bacteria population in the rainy season

Escherichia coli and *Klebsiella* sp are the most occurred bacteria specie in the months of June and July in the rainy season from the water sources of Lapai metropolis. The other variations in the occurrence of bacteria in the rainy season are the presence of *Serratia* sp in the month of June only, and *Chromobacterium violaceum* in the month of July.

Table 6: Occurrence of Bacteria in the water sources of Lapai Metropolis during the rainy season

S/NO	BACTERIA+	JUNE % of occurrence	JULY % of occurrence
1	*Escherichia coli*	29.41	27.78
2	*Klebsiella* sp	23.53	22.22
3	*Enterobacter* sp	11.76	11.11
4	*Citrobacter* sp	11.76	16.67
5	*Proteus* sp	11.76	5.56
6	*Serratia* sp	11.76	0.00
7	*Salmonella* sp	0.00	11.11
8	*Chromobacterium violaceum*	0.00	5.56
Total		100	100

KEY: LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Statistical Analysis of Total Viable Count of Bacteria.

The result of the Determination of significant difference in the Total Viable Count is represented in table 7. The analysis of variance (ANOVA)showed that there is significant difference in total viable count during the dry season. The lowest viable count was found in LEB (6.83) while LNW (7.06), LNB (7.06), LNS (7.20), LWS (7.20), LEW (7.08), LES (7.20), LWW (7.25), LWB (7.09), LSW (7.10), LSB (7.21) and LSS (7.15) had the highest viable count and there is no significant difference among them. Also, the analysis of variance (ANOVA) shows that there is significant difference in the Total Viable Count during the rainy season. The lowest viable count was found in LEB (7.0) while the highest viable count was found in LNS (7.35) which was not statistically different from LES (7.35), LWW (7.30), LWS (7.34), LSW (7.29) and LSS (7.29).

Table 7: Determination of significant difference in the Total Viable Count of bacteria

S/N	LOCATIONS	TVC-DRY	TVC-RAINY
1	LNW	7.06 ± 0.01^{b}	7.26 ± 0.04^{bc}
2	LNB	7.06 ± 0.08^{b}	7.12 ± 0.10^{ab}
3	LNS	7.20 ± 0.02^{b}	7.35 ± 0.07^{c}
4	LEW	7.08 ± 0.03^{b}	7.28 ± 0.04^{bc}
5	LEB	6.83 ± 0.12^{a}	7.00 ± 0.03^{a}
6	LES	7.20 ± 0.03^{b}	7.35 ± 0.04^{c}
7	LWW	7.25 ± 0.01^{b}	7.30 ± 0.03^{c}
8	LWB	7.09 ± 0.09^{b}	7.25 ± 0.01^{bc}
9	LWS	7.20 ± 0.08^{b}	7.34 ± 0.05^{c}
10	LSW	7.10 ± 0.05^{b}	7.29 ± 0.03^{c}
11	LSB	7.21 ± 0.02^{b}	7.21 ± 0.00^{bc}
12	LSS	7.15 ± 0.07^{b}	7.29 ± 0.06^{c}

Means followed by same superscript(s) along a column are not significant difference ($P > 0.05$).

KEYS: TVC-DRY= Total viable count during the dry season
TVC-RAINY= Total viable count during the rainy season
LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Statistical analysis of the Most Probable Number of Coliform bacteria

The result of the Determination of significant difference in the coliform Count is represented in table 8. The analysis of variance (ANOVA) reveals that there is significant difference in coliform count during the dry season. The lowest coliform count was found in LEB (0.30) while the highest coliform count was found in LWS (7.04) which is not statistically different from LSW (7.04). During the rainy season, the statistical analysis of variance (ANOVA) shows that the lowest coliform count was found in LEB (1.33) while the highest coliform count was found in LWS (7.04) which is not statistically different from LES (7.04) and LEW (7.04).

Table 8: Determination of significant difference in the coliform count of bacteria

S/N	LOCATIONS	MPN-DRY	MPN-RAINY
1	LNW	5.04 ± 2.00^{ab}	5.04 ± 2.00^{ab}
2	LNB	4.11 ± 2.93^{ab}	2.68 ± 0.36^{ab}
3	LNS	5.04 ± 2.00^{ab}	5.04 ± 2.00^{ab}
4	LEW	4.68 ± 2.36^{ab}	7.04 ± 0.00^{b}
5	LEB	0.30 ± 0.30^{a}	1.33 ± 1.33^{a}
6	LES	5.04 ± 2.00^{ab}	7.04 ± 0.00^{b}
7	LWW	5.04 ± 2.00^{ab}	5.04 ± 2.00^{ab}
8	LWB	0.59 ± 0.59^{a}	4.00 ± 3.04^{ab}
9	LWS	7.04 ± 0.00^{b}	7.04 ± 0.00^{b}
10	LSW	7.04 ± 0.00^{b}	5.04 ± 2.00^{ab}
11	LSB	2.11 ± 0.93^{a}	1.84 ± 0.48^{ab}
12	LSS	5.04 ± 2.00^{ab}	5.04 ± 2.00^{ab}

KEY:
Similar alphabet showed there is no significant difference (P>0.05) across the rows.
MPN-DRY=Most Probable Number during the dry season
MPN-RAINY==Most Probable Number during the rainy season
LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

Statistical analysis of the percentage occurrence of bacteria

The result of the Determination of significant difference in the percentage occurrence of bacteria is represented in table 9.The Statistical analysis of variance (ANOVA) shows that the lowest occurring organism during the dry season was *Chromobacterium violaceum* (0.00) which is not statistically different from *Shigella* sp (3.13), *Citrobacter*spp (3.57), *Salmonella* sp (6.70), *Serratia* sp (7.15), *Staphylococcus aureus* (9.82) and *Enterobacter* sp (9.82). The highest occurring organism during the dry season was *Escherichia coli* (26.79) which is not statistically different from *Klebsiella* sp (23.22). The organism with the highest occurrence during the rainy season is *Escherichia coli* (28.60)

Table 9: Determination of significant difference in the percentage occurrences of Bacteria

S/N	ORGANISMS	DRY-%	RAINY-T%
1	*Escherichia coli*	26.79±1.79^{b}	28.60±0.82^{e}
2	*Klebsiella* sp	23.22±1.79^{b}	22.88±0.66^{de}
3	*Entorobacter* sp	9.82±2.68^{a}	11.44±0.33^{bc}
4	*Citrobacter* sp	3.57±3.57^{a}	14.22±2.46^{cd}
5	*Proteus* sp	9.82±2.68^{a}	8.66±3.10^{abc}
6	*Staphylococcus aureus*	9.82±2.68^{a}	0.00±0.00^{a}
7	*Serratia* sp	7.15±7.15^{a}	5.88±5.88^{abc}
8	*Salmonella* sp	6.70±0.45^{a}	5.56±5.56^{abc}
9	*Shigella* sp	3.13±3.13^{a}	0.00±0.00^{a}
10	*Cromobacterium violation*	0.00±0.00^{a}	2.78±2.78^{ab}

KEYS:
Similar alphabet showed there is no significant difference($P>0.05$) across the rows.
DRY - %= percentage occurrences of bacteria during the dry season
RAINY - %=percentage occurrences of bacteria during the rainy season
LNW: Lapai North Well, **LNB:** Lapai North Borehole, **LNS:** Lapai North Stream, **LEW:** Lapai East Well, **LEB:** Lapai East Borehole, **LES:** Lapai East Stream, **LWW:** Lapai West Well, **LWB:** Lapai West Borehole, **LWS:** Lapai West Stream, **LSW:** Lapai South Well, **LSB:** Lapai South Borehole, **LSS:** Lapai South Stream.

DISCUSSION

The findings from this research study revealed that all the water samples examined from the four locations and from the three main water sources (well, borehole and stream) had high total viable counts and high coliform counts. The World Health Organisation (WHO) standard for viable count in portable water supply indicates that the Total Viable Count (TVC) should not exceed 100cfu/ml [6]. Based on the WHO standard, the result from this study shows that the well water with a Total Viable Count of between 1.11 x 10^{8} - 1.80 x 10^{8}cfu/ml during the dry season, a TVC of 1.6 x 10^{8}- 2.13 x 10^{8}cfu/ml during the rainy season and water from the streams with a TVC of between 1.65 x 10^{8} - 1.90 x 10^{8} during the dry season, a TVC of between 1.68 x 10^{8} _ 2.46 x 10^{8}cfu/ml during the rainy season are all of unacceptable quality for human consumption [6]. Borehole water from the four locations (LNB, LEB, LWB and LSB) are less contaminated with a TVC of between 5.2 x 10^{7} - 1.70 x 10^{8} during the dry season and TVC of between 9.4 x 10^{7} - 1.82 x 10^{8}cfu/ml during the rainy season are also of unacceptable Microbiological

water quality because of their high microbial loads.

Vulnerable water sources such as streams and wells from this study showed the highest level of faecal contamination of above 1100 (MPN) index/100ml and hence have the lowest quality and this is in agreement with the research carried out by [21]. The Most Probable Number (MPN) results revealed that surface water samples from streams were observed to be most contaminated with coliform count of between 1100 MPN index/100ml to >1100 MPN index/100ml in the four locations. The coliform count was >1100 MPN index/100ml most especially during the rainy season in Lapai East Stream (LES). The well water has lesser coliform count of 210 MPN index/100ml during the dry season to > 1100 MPN index/100ml during the rainy season. The well water from Lapai North and Lapai East were observed to be more contaminated compared to those of Lapai West and Lapai South. The result also revealed that only water samples from Lapai East Borehole and Lapai West Borehole of the four locations investigated recorded counts of 0 MPN index/100ml and later shifted to 460 MPN index/100ml in Lapai East Borehole during the rainy season. The well water, stream water especially are unacceptable for drinking and this could be due to proximity of some water sources such as wells, boreholes and streams to waste dump sites and animal droppings littered around [22]. The high faecal contamination level of > 1100 MPN index/100ml of water could also be as a result of the location of water sources in close proximity to potential contamination sources such as closeness to latrines and location of ground water in close proximity sites of open human defecation.

The research result indicated variations in the microbial loads and the quality of the twelve water samples been examined from the first location (LNW) down to the last location (LSS). This variation depended on seasonal changes [23], surfaces through which the water flows [24] [25] and human activities including the discharge of domestic wastes, urban and other waste water, Agricultural waste and washing of farm product directly into fresh water sources. The result also indicated that the rainy season of the year has the highest total viable counts of 2.46 x 10^8cfu/ml in Lapai East Stream (LES) and coliform count of > 1100 MPN index/100ml in other locations (LNW, LNS, LEW, LES, LWS and LSW). This is simply because most bacteria and coliform bacteria live on the surface of the earth where they are easily discharged by humans and other worm-blooded animals into the streams or rivers and wells in the form of surface run-off during the rainy season [26]. More significant of this highest number of bacteria occurrences is seen shortly after few days or weeks of warm rainy weather or at the beginning of the rainy seasons of the year and fewest population is seen during the dry season [26]. Also, samples taken after few hours of heavy rainfall has highest microbial load of 2.46 x 10^8cfu/ml in Lapai East Stream specifically in surface waters, and this is due to the discharge of surface run-off into the water body or land drainage occurring after the rainfall. The slope, topography and weather condition were also observed in the research to contribute to increase in the number of microorganisms in water [27].

Escherichia coli, Klebsiella sp*, Enterobacter* sp *and Proteus* sp were isolated in both rainy season and dry season and from the four locations with *Escherichia coli* having the highest number of occurrences of 28.57% in dry season and 29.41% in rainy season. *Escherichia coli, Salmonella* sp and *Citrobacter* sp all occurred during the rainy season and this is in agreement with the research conducted by [27]. The occurrence of these bacteria serves the reason for the increase in the cases of outbreak of waterborne diseases experienced most especially at the beginning of the rainy seasons.

The analysis of variance showed that there is significant difference ($P < 0.05$) with the P-Value of 0.026 and 0.009 in the Total Viable Count during the dry and rainy season respectively while there was no significant difference ($P > 0.05$) with P-Value of 0.211 and 0.274 in the coliform count during the dry and rainy season. There was also significant difference ($P < 0.05$) with P-Value of 0.002 in the percentage occurrence of bacteria during the dry season while there was high significance difference ($P \leq 0.001$) with P-Value of 0.001 in the percentage occurrence of bacteria during the rainy season.

Some researchers pose that higher faecal contamination is expected during the dry season in which they attribute this point to excessive evaporation of water and increase in temperature that favours the establishment of coliform in the water [28]. However, [27] disagrees with this and argued that highest number of coliform and viable bacteria count occur during the dry season in which they attributed discharge of potential contaminants such as domestic wastes, Agricultural waste, wash water from car wash, microbial seepage and other nutrient rich wastes through surface run-off or erosion into water sources. The result also revealed that there are variations in the Microbiological quality of the raw water in the four locations and from the three water sources (Well, Borehole and Stream) as well as of the water quality even at the point of consumption.

CONCLUSION

Most water sources in Lapai metropolis portray their unsuitability for drinking without any form of proper treatment. The major cause of water quality deterioration in the locations examined is simply due to lack of proper sanitation, poor personal hygiene, location of water source in close proximity to dump sites, latrines, population pressure on the water sources and lack of adequate protection of most water sources.

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