Detection of Coliform from Public Water Supply in Birnin Kebbi Metropolis, Kebbi State, North-Western Nigeria

By

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#### ABSTRACT

The study aimed to detect the presence of coliform bacteria from public water supply in Birnin Kebbi metropolis, using standard microbiological water quality analysis (MPN), The total viable bacterial count and coliform bacterial population in a total of 10 water samples collected over a period of two months at ten strategic points along Dukku water treatment plant and within Birnin Kebbi metropolis demonstrated that the public water supply in in the area has been heavily contaminated with Salmonella typhi, Escherichia coli, Citrobacter specie, Enterobacter specie and Serratia marcescens, and did not meet any of the WHO guidelines for water supply fit for any public purposes. As such, indiscriminate treatment, handling, transportation (through pipes) of public water supply could impact on the microbiological quality and promote increase of incidence of coliform bacteria to any public water supply, hence, the demands for necessary actions.

Keywords: Coliform bacteria, water supply, WHO guidelines, microbiological quality.

#### INTRODUCTION

Water is colourless, odourless and tasteless liquid and are the most abundant substances estimating to 70% or more of the weight of the human body by mass. It is a crucial component of metabolic process and serves as a solvent for many solutes (Environmental Protection Agency, 2005). Water is vital to the existence of all living organisms, but this valued resource is increasingly being threatened as human populations grow and demand more water of high quality for domestic purposes and economic activities (Kolawole, Ajavi, Olayemi and Okoh, 2011). Water contamination due to pathogenic agents, pesticides chemicals. heavy metals, water disinfectants, and thereby product as a consequence of industrial and agricultural activities leaching from soil, rocks, and atmospheric deposition and other

human activities has become a hazard to human health in several regions of world (Ritter *et al.,* 2002).

Water- borne diseases continue to be one of the major health problems especially in developing nations. The high prevalence of diseases such as diarrhea, typhoid fever, cholera and bacillary dysentery among the populace has been traced to the consumption of unsafe water and unhygienic drinking water production practices (Mead, Helm, Callan, and Atlas., 1999).

Access to safe drinking water has long been a central aim of public health and international development policy (Bain *et al.*, 2012). The Millennium Development Goals (MDGs) included target 7c to 'halve by 2015 the proportion of the population without sustainable access to safe drinking water (United Nations, 2013). The World Health Organization (WHO) and United Nations International Children Emergency Fund (UNICEF) through their Joint Monitoring Programme (JMP) were tasked with monitoring progress against the MDG target and adopted an indicator, 'use of an improved source' (WHO/UNICEF, 2013). The WHO Guidelines for drinking water quality recommend that faecal indicator bacteria (FIB), preferably *Escherichia coli* or alternatively thermotolerant coliform (TTC), should not be detectable in any 100ml drinking water sample (WHO, 2011). Yet numerous reports document faecal contamination of drinking water sources especially in low-income countries, including four of five nationally representative surveys commissioned by WHO and UNICEF (Bain *et al.*, 2012).

There is substantial evidence to demonstrate that improved sources of drinking water can contain faecal contamination. In a systematic review of microbial drinking water quality, many improved sources including piped water were found to be contaminated with *Escherichia coli* or thermotolerant coliform (Bain *et al.*, 2012). Earlier studies estimate that approximately 1.8 billion people are exposed to faecal contamination through drinking water (Onda *et al.*, 2012; Wolf *et al.*, 2013).

# Microbiology of water supply

The purpose of examining water microbiologically is to help in the determination of its sanitary quality and its suitability for general use (Ako *et al.*, 2011). The sanitary quality of water may be defined as the relative extent of the absence of suspended matter, color, taste, unwanted dissolved chemical, bacteria indicative of fecal presence, and other "aesthetically offensive" objects or properties. In short, the sanitary quality of water depends on its acceptability for internal consumption and other uses in which water comes directly or indirectly in contact with man (Okafor, 2011).

Microbiologically safe drinking water is defined by WHO (2005) as that water having acceptable quality in terms of its physical, chemical and bacteriological parameters. Bacteriological parameters, especially *Escherichia coli* and total coliform have been used to determine the general quality of drinking water worldwide (WHO, 2005). The supply of safe water is important to protect the health of the public, scarcity and pollution of water-both microbial and chemical- are a major problems faced by a rural population in several part of the countries (Anstiss and Ahmad, 2006). In developing countries, 2.6 billion people lack access to basic sanitation and 1.1 billion do not have access to improved water sources. This combination leads to 1.6 million deaths each year from preventable diarrheal diseases, 90 % of which are among children less than 5 years of age (WHO, 2004). Public and environmental health protection requires safe drinking water, which means that it must be free of pathogenic bacteria.

The microbiological quality of drinking water in municipal water distribution systems (WDS) depends on several factors. Free residual chlorine and/or chloramines are typically used to minimize bacterial recontamination and/or regrowth in WDS. Despite such preventive measures, regrowth of heterotrophic bacterial count (HBC) and opportunistic bacteria in bulk water and biofilms has yet to be controlled completely (Chowdhury, 2012). Biofilms can provide shelter for pathogenic bacteria and protect these bacteria from disinfectants. Some heterotrophic bacterial count may be associated with aesthetic and non-life threatening diseases. Research to date has achieved important success in understanding occurrence and regrowth of bacteria in bulk water and biofilms in Water Distribution System.

# Coliform bacteria

Coliform bacteria (*Escherichia, Klebsiella, Enterobacter*, and *Citrobacter*) are defined as Gramnegative, rod-shaped, non-spore-forming, oxidasenegative, aerobic or facultative anaerobic bacteria capable of fermenting lactose with production of acid and gas at 35°C in less than 48 hours (Anonymous, 2004). They are group of intestinal bacteria used as indicators to determine if treated water is acceptable for human consumption. Coliforms will not likely cause illnesses. However, the presence of coliforms in drinking indicates the presence of disease-causing

## Examination of water for coliform

Detection of coliform is one of the best ways to evaluate the effectiveness of water disinfection methods. The most important indicator bacteria, in terms of their importance, include *Escherichia coli*, coliforms and other thermotolerant coliforms. The presence of these bacteria in the water is an indicator of insufficient disinfection process and also recent and frequent contamination of water with human and animal feces (Rodríguez, 2012). The examination of water for general coliforms may be carried out by two methods: the multiple-tube and the membrane filtration methods.

### Multiple-tube method

Here, the procedure is carried out as the presumptive test, the confirmed test, and the completed test. Thus, the presumptive test alone is used for polluted water, whose consideration for direct portability without further treatment is not in question. The confirmed test alone is applied to potable water in the process of purification, chlorinated sewage effluents and bathing waters, and the finished water samples, while the completed test is used for drinking water.

In computing the MPN numbers for presumptive, confirmed, or completed test, in the multiple-tube method of determining numbers, the combination of sample sizes to be used should be obtained by knowledge of the quality of the water. Furthermore, the appropriate combination of water sample sizes to be used should be chosen by examining the MPN (Anonymous, 2006).

### • Membrane filtration method

This consists of filtering a measured volume of the sample through a membrane composed of cellulose esters and other materials. All the bacteria present are retained on the surface of the membrane and, by incubating the membrane face upward on a suitable medium and temperature; colonies develop on the membrane, which can then be counted. The volume of liquid chosen will depend on the expected bacterial density of the water and should be such that colonies developing on the membrane lie between 10 and 100. The great advantage of the filtration method over the multiple-tube method is its rapidity. Thus, it is possible to obtain direct counts of coliforms and *E coli* in 18 hours without the use of probability tables (Anonymous, 2006).

# MATERIALS AND METHODS

The areas of study were Dukku water treatment plant and within Birnin Kebbi metropolis, the capital city of Kebbi State, Nigeria. Kebbi State was created on 27<sup>th</sup> August in 1991 from the old Sokoto State. located in the North Western part of Nigeria between the latitude 11.6781<sup>o</sup>N and longitude 4.0695<sup>o</sup> E, with annual rainfall of about 800mn, and average temperature of about 26<sup>o</sup>C, ranging from 21<sup>o</sup>C in winter to 40<sup>o</sup>C between April and June, with a total population of 3,802,500 people according to National Population Census (NPC) estimate (Traditional State of Nigeria, 2010).

The study sites are Dukku water treatment plant located at the North West, along Makera village-Birnin Kebbi road by the left and Tarasa village-Birnin Kebbi road by the right, and within Birnin Kebbi metropolis as follows:

- Zabarmawa area (Sample B1).
- Takalau area (Sample B2).
- Badariya area (Sample B3).
- Bayan kara area (Sample B4).
- Gesse phase 1 area (Sample B5).

A total of ten (10) water samples were collected from ten different taps along Dukku water processing plant (A1 up to A5) and within Birnin Kebbi metropolis (B1 up to B5). All the water samples were collected at the points representative of the sampling sites, using sterile 250-ml capacity sampling bottles; the samples were transported to the laboratory in an ice jacket box and subsequently processed within 4 hours of sampling.

Quantitative bacteriological analysis of the water samples were carried out by using standard plate count (SPC) to enumerate total heterotrophic bacteria (THB) and using the multiple tube fermentation method to enumerate total coliforms bacteria (TCB). The total coliforms and total thermotolerant coliforms bacteria (TTCB) were detected and quantified with the use of Eosin methylene blue (EMB) agar and their incubation at 37 °C and 44.5 °C respectively. Their counts were expressed in cfu/100ml of the water. The IMViC and other biochemical tests including production of greenish metallic sheen on the EMB agar plates were used to confirm and identify *Escherichia coli* (Kolawole *et al., 2*011). A total of 3 replicate was use in this analysis.

All the media used in this study were weighted, prepared, sterilized according to manufacturer's instructions, while the inoculations using a pour plate technique in form of 3 replicates (while inoculating the samples) was used during the course of the study. For the coliform presumptive test, the combinations of sample sizes used in this study were 3 of 10ml each, 3 of 1.0ml each, and 3 of 0.1ml each.

### RESULTS

The pH values of the water samples obtained (figure1 and 2) was highest (7.50) in sample A5, and lowest (7.11) in sample B1 and B3 respectively. Table 1 indicated the highest (2.60±1.90 x 105) viable bacterial count for sample B4, with lowest (1.72±1.23 x 105) for sample A4. The biochemical characterization of isolates (Table 2) demonstrated the presence of: Salmonella typhi, Escherichia coli, Citrobacter specie, Enterobacter specie and Serratia marcescens. Figure 3 was the result showing Escherichia coli exhibiting the highest percentage of 80%, with Enterobacter specie having the least percentage of 40%. The most probable number of coliform bacteria of the water samples (Table 3) revealed the highest 150 MPN index/100ml for sample B4, with the lowest (3) observed for sample A1 A2 and A3 respectively.

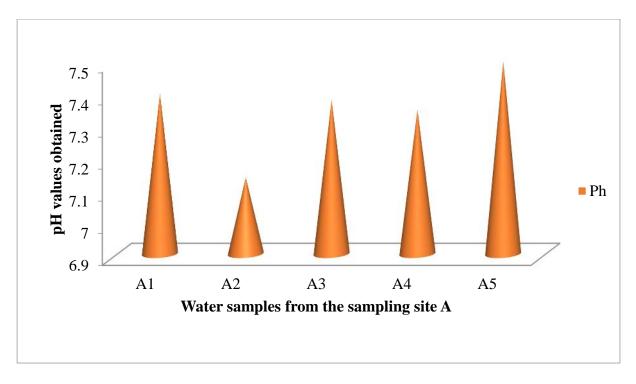


Figure 1: pH of the water samples collected from Dukku water processing plant (A1 up to A5)

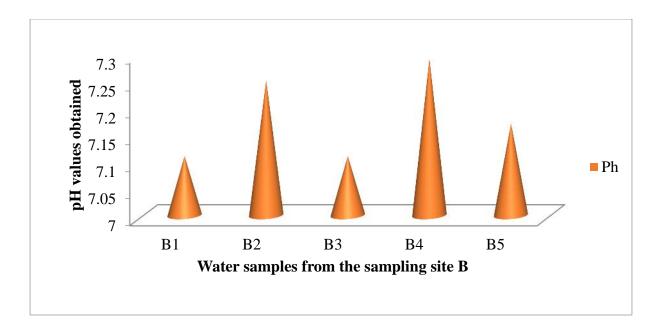


Figure 2: pH of the water sample collected within Birnin Kebbi metropolis (B1 up to B5)

Sample codes	TVBPC (cfu/0.1ml)	Sample codes	TVBPC (cfu/0.1ml)	
A1	2.28±1.60 x 10 <sup>5</sup>	B1	2.36±1.79 x 10⁵	
A2	2.04±1.45 x 10 <sup>5</sup>	B2	2.48±1. 87 x 10⁵	
A3	2.20±1.57 x 10⁵	B3	2.40±1,77 x 10 <sup>5</sup>	
A4	1.72±1.23 x 10⁵	B4	2.60±1.90 x 10⁵	
A5	1.92±1. 36 x 10 <sup>5</sup>	B5	2. 56±1.79 x 10⁵	

Table 1: Total viable bacterial plate count of the water sample A and
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Key: TVBPC = Total viable bacterial plate count, cfu/0.1ml = Colony forming unit/0.1ml

Isolate codes	Gr	Ur	Ind.	Citr.	TSI	Mr	Vp	Shape	Identified organisms
1	-	-	-	-	+	+	-	Rod	S. typhi
2	-	-	+	-	+	+	-	Rod	E coli
3	-	-	-	+	+	+		Rod	C. specie
4	-	-	-	+	+	-	+	Rod	E specie
5	-	+	-	+	+	-	+	Rod	S. marcescens

#### **Table 2:** Biochemical characterization of isolates from the water sample A and B

**Key:** Gr = Gram reaction, Ur = Urease, Ind. = indole, Citr. = Citrate, TSI = Triple sugar iron, Mr = Methyl red, Vp = Vogues Proskauer.

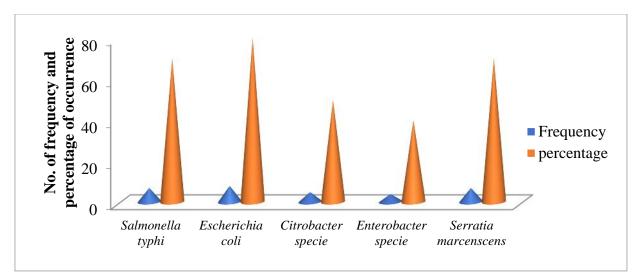


Figure 3: Frequency and percentage occurrence of identified organisms from the water samples

Sample codes	3 of 10 ml	3 of 1.0ml	3 of 0.1ml	MPN index/100ml	Lower limit of 90% confidence	Upper limit of 90% confidence
A1	0	0	1	3	1	9
A2	0	1	0	3	1	13
A3	0	1	0	3	1	13
A4	1	1	0	7	1	23
A5	2	0	0	9	1	36
B1	2	2	0	21	4	47
B2	2	2	1	28	10	149
B3	2	1	1	20	7	49
<b>B</b> 4	3	2	1	150	30	440
B5	2	2	0	21	4	47

Table 3: Most probable number (MF	PNI of coliform bacte	ria of water sample A and B
		ia u wale sample A and D

#### DISCUSSION

The pH values obtained ranges from 7.11 to 7.50, indicated a clear neutral pH, this is suitable for the growth of many species of bacteria, as the optimum growth for many bacteria is usually 7±1 or 2 (Rainy and Harding, 2005). The total viable bacterial count was highest  $(2.60\pm1.90 \times 10^5)$  for sample B4, while it was lowest  $(1.72\pm1.23 \times 10^5)$  for sample A4. This is transparent, because sample B4 was collected from a public tap in the town, which must have acquired a level of contamination from the water, as it moves through the pipes from the treatment plant to the public tap supply, while A4 sample was obtained from the treatment plant where the water has just

undergone treatment and therefore expected to contain less number of bacterial load. WHO standard on public water supply stipulates that the pipelines for public water supply be changed in every 10 years, but in Nigeria, this is not the case and therefore it is not uncommon to see public water pipelines bursting and gushing out water (WHO, 2011). For the biochemical identification of isolates, the heavy contamination of bacterial organisms (*Salmonella typhi, Escherichia coli, Enterobacter specie, Citrobacter specie* and *Serratia marcescens*) was observed, however some of these organisms are enteric, and may likely cause gastroenteritis, diarrhea, cholera, typhoid etc. The frequency and percentage of occurrence of the isolates, showed the highest (80%) percentage for *E* coli, while the lowest (40%) was observed for *Enterobacter specie*. This was contradicted with the statement made by IRC Center for Community water supply and sanitation, which stipulated that, the level of coliforms which should be presence in any giving water body should be less than 10/100ml of a sample, and the number of *E coli* should be less than 2.5/100ml of a sample. The most probable bacterial number observed was highest (150 MPN/100ml) for sample B4, while it was lowest (3 MPN/100) for sample A1, A2 and A3 respectively.

### CONCLUSION

The high count of total viable bacterial number and the presence of coliform bacteria attributed in this study on detection of coliform from public water supply in Birnin Kebbi metropolis, obviously confirmed the negative impact associated with the water supply, which may eventually cause diseases like cholera, bacteremia, gastroenteritis, Typhoid fever, etc., hence, the need for routine microbiological analysis of the water.

### RECOMMENDATIONS

- There should be proper and reliable treatment of water; this should be maintained during processing, transportation and domestic uses.
- The water meant for public supply should regularly be microbiologically treated
- Government should form and reinforce a water supply team (e.g. environmental health team) that will monitor the water supply system at all levels, just to maintain the sterility of water to the public supply and usage
- Sanitary waste disposal system should be provided, as the waste, particularly sewage contains some of the coliform bacteria, like *Citrobacter specie*, *Escherichia coli* etc., and can directly get into the water body and bring about fecal contamination

- Regular and effective environmental sanitation should be provided.
- Health education; public should be awared of the dangers associated with fecally polluted water.

## Acknowledgement

The authors' acknowledged the Kebbi State Water Board Management, especially Dukku water processing plant unit, Birnin Kebbi, for their transparency, accountability, immense contributions and supports for the conduct of this study.

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