Bacteriological Examination of Spring Water in Five Different Locations In Enugu Metropolis, Enugu State, Nigeria

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ABSTRACT: Investigation of bacteriological quality of five different spring water located in Enugu metropolis, Nigeria was analyzed for portability purpose using MPN technique and standard plate method. Samples were collected from each site weekly for a period of three month (between May and July, 2011). Mesophilic count from the samples were generally high exceeding the limit of 1.0 x 10^2 cfu/ml for water, with a range between1.8 – 3.9 x 10^2 cfu/ml and Ogui new layout having the highest range of 3.9 x 10^2 cfu/ml. The MPN count ranges from 7 to 1600 MPN per 100ml of water, with Abakpa and Ogui new layout showing MPN value less that acceptable range of less than 10 only within the first two weeks of sampling. The isolated organisms were identified to be *Klebsiella* spp, *Escherichia coli, Enterobacter aerogenes, Staphylococcus aureus* and *Pseudomonas aerogenosa*. This implies that the spring water were fecal contaminated and hence unsuitable to serve as source of drinking water.
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1. INTRODUCTION

Water is required for proper physiological functioning of all living things including man. An average adult consume up to one liter or more per day. Yet water on certain occasion can constitute hazard to human health when it contain pathogenic microorganisms. Idakwo and Abu (2004) reported that a wide variety of microorganisms pathogenic to human beings are transmitted through contaminated water. According to WHO (1982) 300,000 people die every day from water related disease. Water taken from rivers, streams and other different reservoir may look clean and have no undesirable odor or taste .unfortunately, however, pathogens found in water not only harmful but also are invisible to the naked eye and may be odorless and tasteless. These bacteria can cause more serious illnesses, such as severe diarrhea, hepatitis or typhoid fever (Anon, 2000). The transmission of disease through water is one of the primary concerns for safe water supply. Different diseases like cholera, typhoid, dysentery and hepatitis have been linked to drinking water contaminated by human waste (Eubank *et al*., 1995).

Ground water has traditionally been considered to be the water source least susceptible to contamination by indicator bacteria or human pathogens. This is certainly true of ground water from deep, confined aquifers. It was generally assumed that water passing through the soil was purified by active microbial processes and by filtration; therefore, there was little concern with ground- water contamination (USGS, 2007). In United States, ground water supplies at least 100 million people with drinking water (Prescott *et al.*, 2002).

Water from spring is equated to water from a deep well in the number of existing bacteria. Though spring water is consider as aesthetically acceptable for domestic use, presence of poorly designed pit latrines, poor waste water management as well as poor and inadequate spring protection, may lead to contamination of water from springs with pathogenic bacteria (Haruna *et al.,* 2005). It therefore implies that water supply must not be considered safe, just because it is a ground water, since ground water can be polluted from surface or fission in soil.

Several studies have demonstrated the contamination of groundwater with pathogens, in particular viruses. Nola *et al*, (1998) reported the isolation of *Pseudomonas aeruginosa, Aeromonas hydrophila* and indicators of fecal contamination in spring and well water. Potgieter, *et al.*, (2006) reported an average counts for total coliforms, faecal coliforms, faecal enterococci and *Clostridium perfringens* exceeding the South African recommended guideline limits in ground water source.

The contamination of ground water by pathogenic microorganisms is acknowledged as an important health risk worldwide. To protect against the transmission of waterborne disease, ideally, the occurrence and levels of all pathogen in drinking water should be monitored (Powell *et al*, 2001). The microbiological examination of water is used

worldwide to monitor and control the quality and safety of various types of water. These include potable water (water intended for drinking or use in food preparation), treated recreational water (swimming pools, Sap pools and hydrotherapy pools) and untreated waters used for recreational purposes such as sea, rivers, and lake water (Barrell *et al*, 2000).

In developing countries like Nigeria, limited numbers of people have access to safe drinking water. The State water Corporation, usually fall short in providing safe water to the citizens, hence people in Enugu metropolis usually resort to spring water across the city as a source of drinking water, and because of the general belief that the water is safe, no further treatment is taken prior to consumption or its use for domestic purposes.

The aim of this study was to evaluate the bacteriological quality of these spring water across the city to ascertain its suitability as source of drinking water.

2.0 Materials and Methods

2.1 Sample Collection

Spring water samples were collected from five locations; Abakpa, Coal camp, Emene, Ogui new layout, and Uwani within Enugu in eastern part of Nigeria. The samples were collected aseptically with a 100ml capacity bottle by holding the bottle slightly upward toward the water current. Samples were collected in duplicate and transported to the laboratory in ice box and analyzed within six hours.

2.2 Standard plate count

The total count was conducted by pour plate technique on plate count agar (PCA) and counting the colonies developed after the incubation at37^{O}C for 24hours according to APHA (1998).

2.3. Detection and Enumeration of Coliforms

Enumeration of coliform was carried out according to the method described by APHA, 1998; Each tube was inoculated into 5sets of tubes as follows; first, 10ml into tube containing 40ml of lactose broth, usually designated as double strength lactose broth (DSLB) with Durham tubes, then 1.0ml of the 20ml of lactose broth, usually designated as single strength lactose broth (SSLB) with Durham tubes and then 0.1ml inoculated into five tubes each containing 20ml of lactose broth usually designated as single strength lactose broth (SSLB) with Durham tubes. The tubes were incubated at 35^{O}C for 24- 48 hours. Following incubation, tubes showing gas production were counted and compared with MPN table adapted from APHA 1998, for determination of most probable number (MPN) of coliform per 100ml of water.

The media used for bacteriological analysis of water include plate count agar (PCA) nutrient agar (NA), Lactose broth (LB) and Eosin Methylene blue agar (EMB). All the media used were weighed out and prepare according to the manufacture's specification, with respect to the given instruction and directions. A serial dilution method was for viable count and the presumptive test for coliforms. The sterility of each batch of test medium was confirmed by inoculating one or two uninnoculated tube or plates along with the inoculated tests. The uninoculated tube or plates were examined to shown no evidence of bacterial growth was discarded. The pure cultures of bacterial isolates were subjected to various morphological and biochemical characterization tests to determine the identity of the bacteria isolates with reference to Bergey's Manual of Determinative Bacteriology.

3. Results

The bacteriological analysis of the water sample is shown in tables 1 – 3. The total viable count (TVC) indicates that Ogui New layout spring have the highest bacterial load after 24hrs of incubation having a value of 3.9x102 CFU/ml, which was higher than recommended value. The total viable counts for all the water samples were generally high exceeding the limit of 1.0x102 CFU/ml for water (Table1).

The Most Probable number (MPN) for the presumptive total coliform count of the water samples ranged from 7 – 1600 MPN per 100ml. Water samples from Abakpa and Uwani having the lowest total coliform count of 7 and 9 MPN per 100ml respectively in the month of May, which is less than acceptable range. The water samples from Coal camp and Ogui New layout had total coliform count of 1600 MPN/100ml in the last week of July. (Table 2)

The bacterial isolated from the water samples in this work include *Klebsiella* species, *Escherchia coli, Enterobacter aerogenes, Staphylococcus aureus, and Pseudomonas aerugionosa* (Table3), with *Staphylococcus aureus* and *Pseudomonas aerugionosa* not isolated from Abakpa and Uwani water sampled.

TABLE 1: Mean total bacterial count of spring water sample.

Source/ Sampling site	Mesophilic count CFU/ml x 10^2
Abakpa	2.0
Coal camp	3.5
Emene	2.2
Ogui New layout	3.9
Uwani	1.8

Table 2: Most Probable number (MPN) of coliforms presence in the water sample from various locations in Enugu.

Coliform count by source (MPN index per 100ml)						
Months	**Sample per week/ sources**	**Abakpa**	**Coal camp**	**Emene**	**Ogui new layout**	**Uwani**
May	1	7	17	17	17	9
	2	9	17	17	17	9
	3	11	17	17	21	11
	4	17	17	17	33	17
June	1	17	33	26	33	17
	2	17	33	27	34	17
	3	22	33	33	80	21
	4	33	34	33	180	26
July	1	33	180	34	280	33
	2	34	280	34	350	33
	3	140	350	280	380	140
	4	180	1600	350	1600	280

Table 3: Distribution of Bacterial isolates from water sample.

Isolates	**Abakpa**	**Coal camp**	**Emene**	**Ogui**	**Uwani**
Klebsiella spp	+	+	+	+	+
Escherichia coli	+	+	+	+	+
Enterobacter aerogenes	+	+	+	+	-
Staphylococcus aureus	-	+	+	+	+
Pseudomonas aerogenosa	-	+	-	+	-

4. Discussion

Standard plate count and coliform count have been used extensively as a basis for regulating, the bacteriological quality of drinking water. In this study, both regulatory parameters were exceeding above the WHO guideline value. Study results clearly indicate that most of the spring water sources are highly contaminated. The total bacterial count for all the water samples were generally high exceeding the limit of 1.0x102 CFU/ml which is the standard limit for heterotrophic count for drinking water (EPA, 2002). The high total heterotrophic counts are an indicative of high organic and dissolve salt in the water. The primary sources of these bacteria are animal and human wastes which include surface runoff, pasture, and other land area where animal waste are deposited.

The microbial load is high in water sample from coal camp and Ogui New layout, which is close to refuse dump, farm land and residential building as compare to other springs far away with count lower than the above. The result compare favorably with the result of Banwo (2006), which indicate the presence of bushes and shrubs makes likely possibility that smaller mammals may have been coming around this water bodies to drink and thereby passing out feces into the water.

Generally, underground water is believed to be the purest known, because of the purification of the soil, as pathogenic microorganisms and dissolved organic matter are removed from the water during subsurface passage through adsorption and trapping by fine sandy materials, clays and organic matter. Hence resulting in water with a lower microbial population (Gordon and John, 1996; Prescott et al, 2002,). However, ground water can also be contaminated. The high coliform obtain in the sample may be an indication that the water sources are fecal contaminated (EPA, 2003).

The result also indicate that the water sample show high level of contamination in July which is the peak of rainy season, compare to may indicating that the runoff from the farm land have gain access to the water body.

Klebsiella spp, *Escherichia coli, Enterobacter aerogenes, Staphylococcus aureus* and *Pseudomonas aerogenosa* which were isolated are well established pathogens or opportunistic pathogen of public health significance. *Staphylococcus aureus* is known to produce enterotoxin (Bennett and Lancette, 1998). *Enterobacter aerogenes* isolated from the water sample are example of non fecal coliform and can be found in vegetation and soil which serve as source by which the pathogen enters the water. *E coli* is a well established index of fecal contamination, the presence of these organisms is an indication that the water from the springs fell short of the standard required for untreated drinking water as stipulated by World Health organization (WHO, 1985). *Staphylococcus aureus* is usually harmless but are often able to cause infection once they gain entry into damage skin or deeper body.

5. Conclusion and Recommendation

In conclusion, people leaving in the study area are at risk since the work have shown that there is contamination of water used as source of drinking

water in that area by fecal coliform. This work has revealed hitherto unknown facts about the alternative source of drinking water in Enugu metropolis which calls for urgent action. Regulatory agencies should intensive their effort toward providing clean and portable water to the public and also engage in a campaign for cleanliness of the environment, good refuge and sewage disposal systems, since the awareness of the relationship between health, water, hygiene and sanitation is significant of availability of portable water.

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Reference

1. American Public Health Association (APHA). Standard Methods for Examination of ter waste. 20th ed., American Public Health Association, American Water Works Association, Water Environmental Federation, Washington, DC. 1998.
2. Anonymous. Bergey's Manual of Determinative Bacteriology ed 8th, Waverly Press, Inc. Mt. Royal and Guilford Aves. Baltimore, Md., U.S.A. 1974; 289-389.
3. Anonymous. Water treatment devices for disinfection of drinking water. *Health and Environment, Canada*, 2000; 312-315
4. Banwo, K. Nutrient Load and Pollution Study of Some Selected Stations along Ogunpa River in Ibadan, Nigeria. M.Sc. Dissertation. University of Ibadan, Ibadan, Nigeria, 2006; 107.
5. Bennett, R. W. and Lancette, G. A Bacteriological Analytical Manual, 8th Edition, Revision A . 1998; Chapter 12.
6. Barrelle, R.A.E, Hunter, P.R and Nichols G. Microbiological standards for water and their relationship to health risk. *Communicable Disease and Public Health*. 2000; 3 (1): 8 -13.
7. EPA, US Environment Protection Agency, Safe Drinking Water Act Amendment. 2002: http:// www. epa.gov/safe water /mcl. Html.
8. EPA, US Environmental Protection Agency Safe Drinking Water Act. 2003 EPA816 – F – 03 – 016.
9. Eubank, W. Carpenter, J.D., and Maltsberger B.A. Bacteria in drinking water Department of health, The Ohio State University 1995; Pp 441-449.
10. Gordan, M.; Fair, and John, Gever, G**.** Water supply and Waste Removal in: Waste supply and Waste Removal In: Waste Engineering Vol. John Wiley and Sons 1996; pp 220-236.
11. Haruna, R., Ejobi, F. and Kabagambe, E.K. The quality of water from protected springs in Katwe and Kisenyi parishes, Kampala city Uganda. *African Health Science* 2005; 5 (1):14 -20.
12. Idakwo, P.Y and Abu, G.O distribution and statistical analysis of bacteria in lake Alau in the arid Northern Nigeria. *Nigeria Journal of Microbiology* 2004; 19 (1-2):535 – 542.
13. Nola, M., Njine, T, Monkiedje, A., Foko, V.S., Djuikom, E. and Tailliez, R. Bacteriological quality of spring and well water in Yaounde (Cameroon). *Sante*. 1998; 8 (5) :330-6
14. Prescott, L.M; Harley, J.P. and Klein, D.A. Microorganisms in aquatic environments In Microbiology 5th edition, Mc Gram – Hill Companies Inc. New York. 2002; 632 – 663.
15. Potgieter, N., Mudau, L.S. and Maluleke, F.R. (2006) Microbiological quality of groundwater sources used by rural communities in Limpopo Province, South Africa. *Water Sci. Technol*. Vol. 54 (11-12):371-7.
16. Powell K. L. Cronin A.A., Pedley, S., and Barrett, M.H. Microbiological quality of groundwater in UK urban aquifer: Do we know enough? In Groundwater Quality: Natural and Enhanced Restoration of Groundwater pollution (Proceedings of the ground water Quality 2001 conference held at Sheffield UK, June 2001) IAHS publ. no 275, 2002. Thornton, S F. and Oswald S. E. (ed) P. 91- 97.
17. USGS (US Geological Survey) Bacteria and Their Effects on Ground-Water Quality. 2007; http://mi.water.usgs.gov/h2oqual/GWBactHOWeb.html (Accessed on 27/07/2012).
18. World Health Organization Guideline for drinking water quality Vol. EFP 1982; 82: 39.

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