

## Bacteriological Investigation of Well Water Samples from Selected Market Locations in Ibadan Nigeria

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**ABSTRACT:** This study was carried in Ibadan, to investigate the bacteriological and physicochemical quality of selected well water sampled from 6 locations for the presence of coliforms, an index of faecal contamination of drinking water. All samples investigated were found to be heavily laden with coliforms and other isolates; *Pseudomonas* and *Klebsiella* were obtained in varying percentage. The  $P^H$  of the 24 well water sampled were progressively acidic with the exception of 2 well water with  $P^H$  7.0 . Though the regressive analysis to determine the significant of the extrinsic and intrinsic values of the sampled water extends beyond the  $P$ -value  $\leq 0.5$ , the total viable count obtained from six well water elicited the water as grossly contaminated, therefore, it is unsafe for drinking to avoid water-borne infection.

**KEYWORDS:** Bacteriological, Well Water, Ibadan

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### I. INTRODUCTION

Water is everywhere, but little is available to drink. The total amount of water on earth is constant ; it neither increases nor decreases, but may change location due to climatic changes, human activities or both. Everything originated in the water, and everything is sustained by water. All life on earth depends on water. Many uses water for many purposes drinking, irrigation, fisheries, industrial processes, transportation and waste disposal. The origin of water on the earth is not clear so far. However, the current presumption is that the primordial earth had no oceans, and perhaps very little atmosphere. It is believed that the volatile constituents bound in the earth's crust, oozing to the surface through volcanoes, rock atmosphere. This way, perhaps this remarkable combination of hydrogen and oxygen- called water- came into being and eventually became an indispensable component of the earth's environment (Sobseyet.al.,2002).

Water is not only essential to life but it is the predominant inorganic constituent of living matter, forming, in general, nearly three quarters of the weight of the living cell. It makes up to some 5 percent of the body weight of an adult human and can form as much as 98 percent of the mass of certain jellyfish. Organisms which contain relatively small amounts of water are generally in dormant state or show very slow development; seeds and certain invertebrates that live in arid environments are examples. On the other hand, high rainfall over a land mass invariably means a large biomass per unit area( Annan,2003 ).

Water serves as the second natural medium for the growth of microorganisms and stands next to soil. The growth of microorganisms in water mainly depends on the amount of available mineral nutrients and the dissolved oxygen present in it. It has been observed that as the amount of organic matter increases in water, the number of microorganisms also increases but up to certain limit. The number of bacteria and other microbes will always be higher in river passing by thickly populated cities than of the villages because persons living in cities, are continuously disposing sewage water and other waste products in rivers which contain a very high amount of mineral nutrients- a medium for their growth. Moreover, the pH, temperature range and inorganic phosphate content as well as the situation of the lake and river also support the growth and cause a dense population of microorganisms. These organisms (bacteria, blue green algae, etc) form a heavy blooms under these conditions(Adeniyi,2003).

The possible zinc, copper, and the poor quantity of nitrate nitrogen etc. It has been noticed that the excess of calcium is harmful for the luxuriant growth of microorganisms, specially to algae in general. However, in spite of enormous quantity of the substance that exists, only a small proportion of it is actually usable by human beings. Humans use water in the home, in industry, in agriculture and for recreation. Water well is an excavation or structure located in the ground by digging, driving, boring or drilling to access groundwater in the underground aquifers mechanically or manually(Kirby, 1997).

Water borne diseases are the most important concern about the quality of water. Safe drinking water is defined by the W.H.O as treated surface or untreated but uncontaminated underground water such as bore-holes, springs and sanitary wells (W.H.O. 1996). Water borne disease can cause dysentery, typhoid fever, Salmonellosis and vibrio illness depending on the etiologic agent associated with each infections. And the socio economic impact is financial and manpower loss. On average a family spends about 10% of the monthly household income(Galbraith,1987).

Availability of facilities, financial constraints, illiteracy and bad governance are the major obstacles in the provision of water of good quality in developing countries. In Nigeria, treated pipe borne water is limited to urban areas for those that can afford it. The study is designed to analyse the bacteriological quality of well water in use, from selected local but popular markets in Ibadan, Nigeria.

## **II. MATERIALS AND METHODS**

### **Study Areas**

Four well water of varied depths was randomly collected from selected six markets locations; Oja-Oba market designated as OOM sample, Bodija market as BM sample, Oje market as OM sample, Gege market as GM sample, Sango market as SM sample and Molete-Bode market as MB sample in ancient city of Ibadan.

### **Collection of Water Sample**

Water samples were collected with sterile 200ml screwed capped glass bottles. The lid was aseptically removed and the bottles lowered into the well. The bottle was brought up to the surface and covered with a screw cap when no air bubbles. All the sampled bottles were immediately labelled and transported in ice-pack to the laboratory for bacteriological analysis within 6 hours of collections.

### **Determination of pH of the Water Sample**

The pH of each water sample collected was taken using a calibrated hand held pH meter and recorded for further analysis.

### **Bacteriological Analysis**

The water sample was shaken thoroughly and a one in one thousand dilution of each sample was carried out with sterile distilled water. 1ml of the dilution (1:100) was inoculated into 20ml of melted, cooled nutrient agar. The nutrient agar was mixed thoroughly and poured into a sterile petri-plate. This was allowed to solidify and then incubated at 37<sup>0</sup>c for 24 hours. The number of discrete colonies were counted and expressed as colony forming unit per ml (cfu/ml). 1ml of sterile water in 20ml agar was used as control.

### **Presumptive coliform test**

A total of 11 tubes with Durham tube suspended, divided into 3 each were used for each of the six water samples. 50ml of double strength MacConkey broth were added to 50 of water sample. 10 mls of double strength MacConkey broth were added to 10mls of water samples in 5 tubes and 1ml of water sample were added to 5ml of single strength MacConkey broth each. The bottles were incubated at 37<sup>0</sup>c for 48 hours and were examined for acid and gas production. The number of positive bottles (without acid and gas with yellowish colour profile) were recorded. The most possible number of coliforms per 100ml of sample was estimated from probability table.

### **Differential Coliform Test**

Subcultures were made from presumptive positive test into fresh tubes containing 5ml sterile peptone water. The tubes were incubated at 44<sup>0</sup>c and examined after 48 hrs. 3 drops of Kovac's reagent was added to each tube, pink rose ring color were regarded to be laden with index of faecal contamination of drinking water *Escherichia coli*.

### **Complete Confirmatory Test**

A loopful of presumptive positive and negative test was sub-cultured on MacConkey agar plate and incubated at 37<sup>0</sup>c for 24 hours. The significant bacterial isolates obtained were inoculated on nutrient agar slopes for biochemical identification.

### **Biochemical Identification**

Conventional microbiology tests; Gram stain, indole test, catalase test, oxidase test, citrate utilisation test, and triple iron sugar test were carried out on the isolates.

### **Statistical analysis**

Statistical analysis of the total viable bacteria count, the intrinsic and extrinsic of the examined well water were carried out by ANOVA method of interpretation.

### III. RESULTS

**Table 1.0**  
**INTRINSIC AND EXTRINSIC PROPERTIES OF THE EXAMINED WELL WATER (DEPTH OF THE WELL/PH)**

Location	Depth (M)	P <sup>H</sup>
Oja-Oba market	20.0	4.0
	11.7	3.2
	30.0	4.5
	35.0	5.2
Bodija market	25	4.0
	42	5.7
	9	4.3
	60	7.0
Oje market	30	4.3
	45	5.0
	12	3.0
	49	5.4
Gege market	10	3.2
	27	4.1
	15	3.7
	32	4.3
Sango market	30	4.2
	37	5.0
	53	5.9
	44	4.8
Molete Bode market	14	3.2
	25	4.0
	33	4.5
	65	7.0

**Table 2.0 Distribution of Bacterial Isolates in the Examined Well Water**

Location	Code	<i>Escherichia coli</i>	<i>Pseudomonas aeruginosa</i>	<i>Klebsiella spp</i>
Oja Oba market	OM <sub>1</sub>	28	-	24
	OM <sub>2</sub>	32	-	16
	OM <sub>3</sub>	22	-	18
	OM <sub>4</sub>	16	-	12
Bodija market	BM <sub>1</sub>	26	16	14
	BM <sub>2</sub>	24	14	8
	BM <sub>3</sub>	22	-	5
	BM <sub>4</sub>	28	-	15
Oje market	OM <sub>1</sub>	30	12	-
	OM <sub>2</sub>	18	10	-
	OM <sub>3</sub>	22	14	-
	OM <sub>4</sub>	24	-	-
Gbagi market	GM <sub>1</sub>	36	16	-
	GM <sub>2</sub>	24	14	8
	GM <sub>3</sub>	28	12	-
	GM <sub>4</sub>	26	-	9
Sango market	SM <sub>1</sub>	26	8	-
	SM <sub>2</sub>	28	14	-
	SM <sub>3</sub>	24	12	8
	SM <sub>4</sub>	30	16	10
Molete-Bode market	MB <sub>1</sub>	28	12	-
	MB <sub>2</sub>	20	8	-
	MB <sub>3</sub>	18	14	-
	MB <sub>4</sub>	24	-	14

**Table 3.0 Total Coliform Count of the Examined Well Water CfU/ml**

Location	W <sub>1</sub>	W <sub>2</sub>	W <sub>3</sub>	W <sub>4</sub>
Oja Oba market	32.5	12.5	25.0	7.0
Bodija market	31.5	24.5	16.0	31.5
Oje market	30.0	13.5	31.5	22.5
Gege market	35.0	31.5	7.5	17.5
Sango market	12.5	30.0	17.0	35.5
Molete Bode market	25.0	7.0	32.5	25.0

**Dilution factor 10<sup>-6</sup> inoculum size 0.2ml**

**Cfu/ml = Viable count × Reciprocal of Dilution factor × Reciprocal of inoculums size**

#### IV. DISCUSSION AND CONCLUSION

The distributions of the bacterial isolates in the water sampled varied according to the environment surrounding the well. *Escherichia coli* was predominant in all the well sampled. At Oja-Oba market, the well, a locally dugged well were surrounded by various raw foodstuff seller's in an unkempt open places and there are no provision for toilets and lavatory for their use, *Escherichia coli* and *Klebsiella spp* were mostly predominant and no *Pseudomonas aeruginosa* were recorded in this sample.

The situation in Bodija Market which is a feeder market to Oja-Oba market is similar, though there are variation in the distribution of samples, *Pseudomonas aeruginosa* were recorded this sample.

Oje-market predominantly colonized by local herb sellers, the well water at this location was covered with a wooden plank, though there were litters of herbs around the well, there was no grown-up vegetation around this location. There was no *Klebsiella* in the samples but *Escherichia coli* and *Pseudomonas aeruginosa* were recorded. Gbagi market and Sango market were predominantly known for locally dyed cloth production and every othe fabric works. The well though covered with a metallic lid was located at the end of major gutter while Sango well water was at the entrance of the major gate of the market. The distribution of the isolates in the two market slightly varied. The Molete-Bode market was colonized by cereal sellers and locally manufactured provisions, *Klebsiella spp* was only recorded on the fourth well water sampled. *Escherichia coli* and *Pseudomonas aeruginosa* were recorded in varied numbers. With the exception of the P<sup>H</sup> of the water obtained From Bodija and Molete, the P<sup>H</sup> of the remaining four examined water examined were acidic. This findings does not conforms with the acceptable recommended standardized pH range of 6.5 to 8.5(SEPA,2011). The pH values of water can be used to predict the parameter of microbial pathogens that can be obtained from such water. The values outside the recommended range can indirectly affect human health as secondary contaminants in water.

The distribution of bacterial isolates were examined, *Escherichia coli*, an index of faecal contamination of water were pre.0)dominant in all the 24 well water sampled (Table 2.0). *Pseudomonas aeruginosa*, a nutritionally non-exacting bacteria were also recorded in 5 of the 6 well water with the exception of Oja-oba market that has no pseudomonas growth and Bodija market where well number BM3 and BM4 elicited no growth. *Klebsiella spp* were also predominant in Oja-oba and Bodija market samples while the rate of growth in Gbagi and Sango market were twice per the four well sampled. Only sample MB4 had 14 growth noticed in the water sampled.

The total coliform count in the examined well water using inoculums size of 0,2ml from 10<sup>-6</sup> dilution as shown in Table 3.0. The presence of coliforms in the water sampled varied from well investigated. The outlook of the values obtained in the Table 3.0 elicited the significant contamination level of each well, which could be faecal oral source, hence an evidence of poor hygienicity in and around the well location.

Though the regressive analysis indicated no significant difference between the extrinsic and intrinsic values of the sample water(P-values > 0.05). The statistical significant could not be considered as more relevant than the clinical significant which is more important because of the bacterial involved that are potential pathogen. The possible contamination ways could be due to poor hygiene, lack of treatment scheme for the water and ever other dirty oriented marketing activities in and around those well. It is imperative that the concern public health outfits should create awareness to the marketers and sink a modern bore-hole water neede for their day to activities.

#### REFERENCES

- [1]. Abraham, M. (1984). Water that Goes to waste. World Health No. 6, 1984 p. 67.
- [2]. Adeniyi, J. (2003). Text of a Message delivered at the 31<sup>st</sup> Environment Day, Ibadan, Nigeria. The Guardian Newspaper 5<sup>th</sup> June, 2003 p. 37.
- [3]. Anita, K.H. & Sidney, A.C. (1992). Waterborne Diseases in Encyclopedia of Microbiology Academic Press Inc. p. 337-384
- [4]. Annan, K (2003). Text of message delivered at the 31<sup>st</sup> Environment day, Ibadan, Nigeria. The Guardian Newspaper 5<sup>th</sup> June, 2003 p. 37
- [5]. APHA (1992). Standard methods for the Examination of Water and Waste water. American public health association, America water works Association and Water Pollution Control Federation. Washington 17<sup>th</sup> Ed. Part 9000.

- [6]. Bickford, T.M. Lindsey, B.D. and Beaver, M.R. (1996). Bacteriological Quality of Ground Water used for Household Supply.
- [7]. *Ecological Water Resources Investigation. Report 96-4212.*
- [8]. C.D.C. (2002). Water-borne Disease on the Rise, Health News, Nov. 2002. [www.body1.com/news/index.cfm](http://www.body1.com/news/index.cfm)
- [9]. Cheesebrough, M. (1984). Water and Sanitation Decade in Medical Laboratory Manual for Tropical Countries. Vol. II. Butterworth Heinemann 206-224.
- [10]. Cheesebrough, M. (2002). Water related Diseases and Testing of Water supplies in District Laboratory Practice in Tropical Countries Part II. Cambridge University Press P. 143-157.
- [11]. Cowan, S.T. (1981). Manual for the Identification of Medical Bacteria. Cambridge University Press 2<sup>nd</sup> ed.
- [12]. Dadswell, J.V. (1990). Microbiological Aspects of Water Quality and Health. Journal of the Institutions of Water and Environmental Management. (J. IWEM) Vol. 4, No. 6, Dec. 1990 P. 515-519.
- [13]. Duran, C.J. (1984). Safe Water for Every Family. World Health. No. 6, Nov. 1984.
- [14]. Enteric Archives (2002). Surveillance of Water-borne Disease and Water Quality. July-Dec. 2001.
- [15]. Environmental Literacy Council (2002). Water are the most Dangerous Water-Borne Diseases? Ask an Expert, Nov. 19, 2002. [www.envliteration.org](http://www.envliteration.org)
- [16]. Galbraith, N.S. et al (1987). Water and Disease after Croydon: A Review of Water-borne and Water Associated Diseases in the U.K.J. IWEM, No. 1 Jan-Feb., 1987 P. 7-21.
- [17]. Green, J.H. (1984). An Introduction to Human Physiology. Oxford University Press, 4<sup>th</sup> S.I. Ed.
- [18]. Hawker, L.E. and Linton A.H. (1971). Microorganisms: Function Form and Environment. Edward Arnold Ltd. 1<sup>st</sup> Ed. P. 515-528, 583-585.
- [19]. Holden, W.S. (1970). Water Treatment and Examination. Churchill, London, P. 193-221.
- [20]. Hugo, W.B. and Russel, A.D. (ed.) (1984). Pharmaceutical Microbiology. Blackwell Scientific Publications. 3<sup>rd</sup> ed. P. 320
- [21]. Hutin, Y.; Luby S. and Paquet, C. (2003). A Large Cholera Outbreak in Kano City: The Importance of Hand Washing with Soap and The Danger of Street Vended Water. J. Water & Health, Vol. 1, 2003.
- [22]. Illinois Dept. of Public Health (1999). Well Sampling for Coliform and Nitrate Environmental Health Fact Sheet. April, 1999. [www.idph.state.il.us/envhealth](http://www.idph.state.il.us/envhealth)
- [23]. IPCS News New Drinking – Water Quality Guidelines. The Newsletter of the International Programme on Chemical Safety, Issue 4, Nov. 1993.
- [24]. Jawetz, E. Melnick, J.L. and Adelberg, E.A. (1978). Review of Medical Microbiology. Lange Medical Publications 13<sup>th</sup> ed. P. 88-90.
- [25]. Kirby, C. (1997). Shallow Aquifer Water Quality Risks. Water and Sanitation for All: Partnerships and Innovations. 23<sup>rd</sup> WEDC Conference, Durban, South Africa, 1997. P. 385-387.
- [26]. Krovacek, K. et al. Enterotoxigenicity and Drug Sensitivity of *Aeromonas hydrophila* isolated from Well Water in Sweden – A case study. Int.J Fd.Microb. 8 P. 149-154.
- [27]. Lloyd B. and Helmer, R. (1991). Surveillance of Drinking Water Quality in Rural Areas. Longman Grp. UK., P. 1-4.
- [28]. Manitoba Health (2001). How do I Disinfect my well. Well Water Fact Sheet. March, 2001.
- [29]. Manitoba Health (2001). How do I know if my well water is safe from Bacterial Contamination. Well water fact sheet. March, 2001
- [30]. Mendie, V.E. (2002). Cyclical growths of contaminants in drinking water packaged in polythene bag. Nig. J. Pharmacy, 33, April-June, 2002, pg. 27-32.
- [31]. Neira, M. (1997). Cholera: A challenge for the 21<sup>st</sup> century. World Health, 50<sup>th</sup> Year, No. 1, Jan.-Feb. 1997
- [32]. Nola, M. Njine T, Monkiedje A, Sikati foko V, Djuikom E, Tailliez R (1998). Bacteriological Quality of Spring and Well Water in Yaounde, Cameroun. Cahiers d'etudes et de recherches francophones/santé vol. 18, No. 5, Oct.-Nov. 1998. P. 330-336.
- [33]. Osho A and Fagade, O.E. (2000). Occurrence of Coliform and Faecal Streptococcus in Ago-Iwoye Well Water. Nig. J. Sci. Vol. 34, No. 4 P. 323-329
- [34]. Oyo State Government and Unicef B Zonal office profile of the Urban Local Governments of Ibadan: Planning Baseline Data, April 1997.
- [35]. Packam, R.F. (1990). Cryptosporidium and Water Supplies – The Badenoch Report. J. IWEM, 1990 4<sup>th</sup> Dec. P. 78-80.
- [36]. Pelczar M.J., Reid, R.D. and Chan, E.C.S. (1977). Microbiology of Domestic Water and Sewage in Microbiology. Mc Graw-Hill Bk. Co. NY P. 781-806.
- [37]. Petrie, A.S. Horan, N.J., Clapham D.B. Cram, A.G. (1994). Seasonal Variations in the Quality of Spring Waters Used as Water Supplies. J. IWEM, 1994, 8<sup>th</sup> June, P. 320-326.
- [38]. Reuters News Service: Cholera Kills 100 in Northern Nigeria. Nov. 23, 2001
- [39]. Shual ed. (1980). Legal aspects of water quality management in Israel under conditions of scarcity. [Atar.msc.huji.ac.il/ingeo/emppp/cv](http://Atar.msc.huji.ac.il/ingeo/emppp/cv).
- [40]. Sobsey, M.D. and Pfaender, F.K. (2002). Faecal Indicator Detection and Testing in Drinking Water – Context, Purposes, Needs and Criteria. Water and Sanitation WHO, Geneva.
- [41]. Strauss, B.; King, W., Ley, A., Hoey, J.R. (2001). A Prospective study of rural drinking water quality and acute gastrointestinal illness. Biomed Central Public Health, 29<sup>th</sup> Aug. 2001. [www.biomedcentral.com](http://www.biomedcentral.com)
- [42]. Water Resources (2002). Application of standards for bacteriological quality of public drinking water supplies and private wells: water quality standards. Dept. of Environment, Gov. of Newfoundland & Labrador. Reviewed April 9<sup>th</sup> 2002. [www.gov.nf.ca/env/Env/waterres](http://www.gov.nf.ca/env/Env/waterres)
- [43]. WHO (2002). Microbiological agents in drinking water – *Aeromonas* 2<sup>nd</sup> ed. [www.who.int](http://www.who.int)
- [44]. WHO (1993). New drinking water guidelines. IPCS news, issue 4, Nov. 1993.
- [45]. WHO (1996). Water and Sanitation Fact Sheet 112, Nov. 1996. [www.who.int/inf](http://www.who.int/inf)
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