International Journal of Biotechnology and Microbiology
www.biotechnologyjournals.com
Online ISSN: 2664-7680, Print ISSN: 2664-7672
Received: 15-11-2023, Accepted: 01-12-2023, Published: 16-12-2023
Volume 5, Issue 4, 2023, Page No. 20-28

Assessment of the bacteriological quality of boreholes water in Bwarak, Pankshin and it environs

Adeniji A*, Toscanini M M, Bakrim B A
Department of Biology, Federal College of Education, Pankshin, Plateau State, Nigeria

Abstract
The purpose of this study is to investigate the bacteriological quality and public health Contamination of drinking water is an issue of serious environmental and health concern. This work investigated the bacteriological quality of boreholes drinking water in Pankshin Local Government Area, Plateau State. The study area comprised four sampling zones that cut across ten sampling points in dry and wet seasons. Microbiological analysis of water samples followed standard procedures of cultural characterization, microscopic observation, total viable bacterial and coliform counts. Biochemical identification of isolates involved gram staining, oxidase, citrate, urease, catalase, coagulase and hydrogen sulphide tests. Data analysis was done on the SPSS software (version 27) using descriptive and inferential statistics (at 5% level of significance). Seven (7) distinct species of bacteria were found in all water samples: *Salmonella* spp, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumonia*, *Enterobacter* spp, *Escherichia coli* and *Proteus mirabilis were identified*. In dry season, three species (*K. pneumonia P. aeruginosa* and *Enterobacter* spp) had the highest percentage occurrence of 20% each followed by *Proteus mirabilis* and *Staphylococcus aureus* (13.3% each) where total viable bacterial count ranged from 0.5 $x10^2$ cfu/ml to 1.8 $x10^4$ while coliform count was <50 cfu/ml. Wet season analysis revealed the presence of *K. pneumonia* as the most dominant (52.5%) contaminant found in water samples followed by *P. aeruginosa* (22.5%) and *Salmonella* spp (20%). Total viable count significantly ($P<0.05$) varied from 3.04±0.68 x 10^3 cfu/ml to 7.25±0.63 $x10^3$ cfu/ml whereas the mean coliform count was 4.39±0.49 $x10^3$ cfu/ml with insignificant differences among locations ($P>0.05$). Total *S. aureus* count had the highest overall mean value (1.99±1.05 $x10^3$ cfu/ml) while total *K. pneumonia* count had the lowest mean value (0.41±0.40 $x10^3$ cfu/ml). In conclusion, study revealed that borehole water of Bwarak, Pankshin and its environs is vulnerable to bacteriological pollution. Hence, proper bore-hole location and construction, control of human activities to prevent sewage from entering bore-hole environment is the keys to the avoiding bacterial contamination of drinking water.

Keywords: Bacteriology, water, health, environment, Pankshin

Introduction

Water is a basic necessity for life, needed by all forms of life including plants and animals and as well vital to many life processes, water can also serves as route via which pathogen that infect diseases can be transmitted to human body. It can contain heavy metals and other chemical substances that may adversely affect human health; as such a good supply of drinking water must be available to all consumers (WHO, 1996). Portability of drinking water is compounded in is odorless, colorless, practically tasteless and free from physical, chemical and biological contaminants (Engwa A.G. *et al*, 2015) [22].

In many developing countries with Nigeria inclusive, availability of portable water has become a critical and urgent problem and it is a matter of great concern to families and communities depending on non-public water supply system (Okonko *et al*., 2008). This result from increase in human population coupled with increased in industrialization, livestock farming and urbanization which have led to frequent contamination of surface sources of water such as: rivers, ponds, streams, and open skies which are major primary sources of water for the people living in these countries (Umeh *et al*., 2005). The issue of little or no supply of water led most resident resort to ground water sources such as boreholes as an alternative water sources. Thus, humans can abstract groundwater through a borehole, which is drilled into the aquifer for industrial, agricultural and domestic use. However, groundwater resources are commonly vulnerable to pollution, which may degrade their quality.

Boreholes is a ground water sources in which at least a depth of 150 feet (45.72m) is drilled to reach ground water and sources for drinking water (Olowo S.I., 2005 [35]; Onuh J.O. *et al*, 2009). It is drilled into the aquifer or ground water zone, some distance below the water table (Ukpong and Okon, 2013) [47]. The borehole is naturally clean and safe for consumption; quality of ground water is a function of natural processes as well as anthropogenic activities because the overlying soil acts as a filter, ground water is usually free of disease causing microorganisms. However, evidence has proved that underground water can be contaminated at any stage, especially in the era of industrialization where excess chemicals find their way into the water. The chemical composition of ground water is a measure of its suitability as a source of water for human consumption or for agricultural or industrial uses. Ground water can be rich in dissolved solids, especially carbonates and sulfates of calcium and magnesium as well as chlorides and bicarbonates, depending on the strata through which it flows. Thus, additional treatment may be required in order to provide pleasant water for drinking and household use (Adindu and Igboek, 2012). Experts further suggest that underground water can be contaminated with septic tanks, bused underground sewer pipes, toxic minerals and other varying contaminants. In Nigeria, groundwater contamination is one of the least recognize environmental problems, this is due to lack of proper awareness because groundwater problem are not readily detected and pathways for contamination are not as noticeable as those affecting surface water (Wright, J *et al.* 2004).

Borehole water has become the most used source of water dating back to ancient China (202BC- 220AD). The Han dynasty used deep boreholes, reaching as deep as 600m (2000ft) (Loewe, 1968). This borehole water fills the spaces between the rocks and soils making an aquifer (Driscoll, 1986). Ground water (borehole) depth and quality varies from place to place and this affects the quality of water obtained. The various kinds of rocks and soils which it moves through affect it. Pesticides and fertilizers applied to lawns and crops can accumulate and migrate to the water tables thus affecting both the physical, chemical and microbial quality of water. Water moving through underground rocks and soils may pick up natural contaminants, even with no human activity or pollution in the area (Beltaos *et al*., 2006). In some cases natural water may contain elevated concentrations of several potentially toxic elements or microbiological contaminants that may lead to diverse effects on human health (WHO 2004). In addition to nature's influence, water is also polluted by human activities, such as defecation, dumping garbage, poor agricultural practices and chemical spills at industrial sites (Coe, 2001). According to (Ukpong E.C. and Okon B.B. 2013) [47], water pollution is the modification of the physical, chemical and biological properties of water. Even though water may be clear, it does not necessarily mean that it is safe for drinking. It is very important to judge the safety of water with respect to its physical, chemical and bacteriological properties. Certain requirements must be met for water to be fit for human consumption. These requirements are freedom from organisms and chemical substances which might be injurious to health. Drinking water should be of such composition that consumers do not question the safety of the water. This implies that turbidity, color and taste should be unobjectionable, and macro organisms (e.g. worms, aquatic and fly nymphs) should be absent.

Enormous pressure has been exerted on the provision of safe drinking water especially in rural areas where less effective attention is paid on WHO standard and guidelines in water quality. Unsafe water has increasingly become an issue of serious environmental concern. It is a public health threat, placing persons at risk for diarrheal and other diseases as well as chemical intoxication. Unsanitary water particularly has devastating effects on young children in the developing world. Each year, more than 2 million persons, mostly children less than 5 years of age, die of diarrheal Disease. For children in this age group, diarrheal disease accounted for 17% of all death from 2000 to 2003 ranking third among causes of death, after neonatal causes and acute respiratory infections (WHO, 2005). Nearly 90% of diarrheal-related deaths have been attributed to unsafe or inadequate water supplies and sanitation conditions affecting a large part of the world's population.

The increase in the prevalence of waterborne disease across the world is alarming, and Nigeria is not left out since some outbreaks of waterborne diseases have also been reported in this part of the world (Obi, C.N and George, P. 2011; Palamuleni, L and Akoth, M. 2015). Since ground water (borehole water) is an important alternative sources of potable water to most people in the rural and urban areas across the world, it is vital to evaluate their physicochemical and microbiological quality since these sources of water are usually at risk of pollution from human and other environmental activities.

Inhabitants and users of borehole water within the study area have little or no knowledge about the bacterial quality of the borehole water used. Also there is no proper awareness of WHO standard and guideline that are meant to be observe when sinking a borehole in a particular location. This is because information on the bacteriological quality of water from borehole sources used for drinking and domestic purposes is limited. This has result to increase in enteropathogenic diseases in both children and adult in the community and also compromising of WHO standard and guideline for sinking of borehole.

Considering the boreholes, the necessity to assess the chemical and microbiology of borehole water in these regions is paramount, considering the rise of water borne diseases which is as a result of the modification of bacterial quality of the water.

Justification of the study

According to the (Ishaku *et al,* 2010), various sources of water supply in Plateau State include boreholes (46.6%), dug wells (14.6%), streams (1.80%) and water vending (37%) which gives ground water sources a total of 61.2% and surface water with 38.80%. The level of adequacy of the water was rated 28.38% as against 71.64% inadequacy. Inhabitants rely mostly on boreholes, vended water and hand-dug wells as sources for drinking water and for other domestic activities and because of this inadequacy, households are necessitated to collect waters in plastic containers or metal tanks for storage so they can use when the need arises. The population in Pankshin LGA cannot be sustained without reliable access to safe water and adequate quantity. The high birth rate has led to increased reliance on borehole water.

This research will help in determine water quality parameters and recommending for suitable action or creating awareness about water quality and water borne diseases. This research will also identify areas of water stress where less water is available for use, affecting the per capita consumption. The information from this research will be used to guide government agencies, researchers and other development organizations like NGO's to develop strategies, policies and institutional infrastructures to provide quality and accessible water resources to communities.

The study's objectives

The following are the precise goals:

1. Determine the level of contamination of borehole water with respect to location
2. Isolate and identify microorganisms contaminating borehole water use as sources of drinking water in the study area.
3. To determine the biochemical characteristics of the isolates.

Materials and methods

Study Area

The research was carried out in Bwarak, Pankshin and its environs, Pankshin town in Pankshin Local Government Area of the Jos Plateau (see Fig 3.1). The Jos Plateau has been described by PADP (2000), WWF (2001) and Plateau State Government Organization (2004). Pankshin LGA has 12 Wards with a population of 191,685 as at 2006 census, it covers an area of 1,524 km². The local government is

located within latitude 9^0 20' 0"N and longitude 9^0 27' 0"E having an elevation of about 1,217m (3,933) above the sea level, bounded to the north by Bauchi State, to the east by Kanke and Langtang North local government areas, to the south by Mikang and Shendam local government areas, and to the west by Mangu local government area. The major economic activities of the people of Pankshin are Animal husbandry, and farming predisposes them to water contamination (Blessing O.J, 2017). The state enjoys an abundant rainy season which usually starts from April and ends in October with an average precipitation of 1, 500mm. The daily mean temperature during the rainy season is 23°C. The dry season normally characterized by harmathan winds for the most part is from November to March.

Apparatus

The apparatus used for the experiment are: microscope; incubator, refrigerator, universal bottles, bayou bottles, syringe, needle, were loop, micropipette, Petridishes, masking tape, conical flask.

Reagents

The reagents used are Deoxycholate Citrate Agar (DCA), MacConkey Agar (MCA), Centrimide Agar (CMB), Selenite-F broth (SF), crystal violet, lugols lodine, Acetorie or acid-alcohol, safranin, methyl red, kovac reagent, Hydrogen peroxide and pepton water.

Sample Collection

Sampling was done between October and December 2013. It was during the period when the table was low. Samples were collected at 10 different point (boreholes) across the ten political wards that made up Pankshin metropolis. Four 2L plastic bottles were used to collect water from each of the 10 points making a total of 40 water samples The plastic container were prewashed with detergent, then with Nitric acid solutions, rinsed with distilled water and finally with the water source. The samples collected were preserved at 4^OC before the analysis. Those for heavy metal determination were preserved by adding three drops of Nitric acid at the field (Okiemen, Duru and Olorunfemi, 2002).

Table 1: Sampling Zone and Specific Sampled Areas

S/N	Sampling Zone	Specific Sampled Areas
1	Federal College of Education (FCE) Pankshin	BHA: First Boys Hostel Borehole BHB: Second Boys Hostel Borehole GHA: First Girls Hostel Borehole GHB: Second Girls Hostel Borehole
2	Bwarak Community	DOB: Double Across Street Borehole GH: Garikawa House Borehole
3	Top site Community	LUC1: First Locus street Borehole LUC2: Second Locus street Borehole
4	Staff Quarters	SQA: Staff Quarters first Borehole SQB: Staff Quarters second Borehole

Sources: Survey, 2023

Sterilization of Materials

The glassware and the ware loops were properly washed, air dried, wrapped with Kraft paper and sterilized in hot air oven at 180^0c for 2 hours.

Preparation of Media Used

Each medium was prepared as at when needed according to the manufacturer's instruction on the labels of the media and autoclaved at 121^0c for 40minutes. Different media such as Macconkey Agar (MCA), Sabouraud Dextrose agar (SDA), Nutrient Agar (NA) were prepared separately.

Preparation of Macconkey Agar (MCA)

25g of macconkey agar powder was weighed on a balance machine, suspended in 500m1 of distilled water. It was heated to boil with gentle swirling to dissolve completely. The medium for the isolation was sterilized by autoclave at 121^{0c} for 40minutes. The medium was cooled to 40-50^{0c} and poured in the sterile petri-dishes.

Preparation of Sabouraud Dextrose Agar

32.5g of sabouraud distilled agar powder was weighed on a balance machine, suspended in 500m1 of distilled water. This medium was heated to dissolve completely and was sterilized in autoclave at 121^{0c} for 40 minutes. The medium was cooled to 40-50^{0c} and poured into sterile petri-dishes.

Preparation of Nutrient agar

10g of nutrient Agar powder was weighed on a balance machine, and was suspended in 500ml of distilled water, boiled to solve completely, the media sterilized in autoclave at 121^0c for 40minute. The medium was cooled to 40-50^0c and poured into sterile petri-dishes.

Preparation of Normal Saline

3.4g of normal saline was suspended in 400m1 of distilled water, 10m1 syringe was used to transfer the dilute normal saline into the bijou bottles ready for dilution.

Culture of sample

1g of each sample was weighing on a weighing balance, dissolved properly in 10m1 of pepton water which was used to prepare ten folds serial dilution. Using pour plate method. The medium was allowed to solidify before drying in an incubator for 1 hour at 37^0c. The petri-dish of the medium was labeled according to the labeled samples. After the drying, micropipette was used to inoculate the dilute. Sample into the prepared medium and was spread with rubber wire loop properly to ensure even distribution. The pour plates were incubated in an inverted manner at 37^0c for 24 hours. The plates were observed and the single colonies were picked for sub-culture in order to obtain pure culture.

Microscopic examination

Microbial count

After incubation, the number of colonies on the petri-dish were counted using bacterial colony counting chambers. The total bacterial count, total Coliform count and total fungal count were taken.

Gram stain technique

Smear of each of the isolates were prepared by picking a small portion of discrete colony form the plates with the help of sterilized wire loop, into a drop of normal saline on glass slide, it was spread properly and allowed to dry. After making the smear, the slide were heat-fixed by carefully passing them over a Bunsen burner flame, about 3 times. The heat-fixed smear was then covered with crystal violet stain for 60 seconds. The stain was quickly washed off with clean water. The water was tipped off and the smear was covered with lugols lodine for 60 second. The lodine was washed with clean water. The smear was decolorized rapidly for about 20 seconds with 95% ethanol. The smear was quickly washed with clean water and then covered with dilute carbolfuschin for 30 second. The stain was washed off with clean water and the slide was allowed to dry at room temperature. The gram stained slide was examined first with x40 objective lens to check for the staining and distribution of the gram stained bacteria, then with oil immersion objective lens (x100) to look for the bacteria. Gram positive bacteria appeared purple while gram negative appear red or pink.

Biochemical test

Indole test

The sterile wire loop was used to inoculate organisms in a test tube containing 5m1 of peptone water (medium) and incubated for 48hours at 37^0c. After incubation, 0.5m1 of kovac's reagent was added into the tube and allowed to stand for 15minute a rose spank colour indicated positive reaction. While the negative reaction result in the indole reagent retaining it's yellow color.

Catalase test

The test demonstrates the presence of catalase which is an enzyme that catalys the release of oxygen form hydrogen peroxide (H_2 0_2). A colony of 24 hours old culture was picked using a sterile loop and then emulsified in a few drops of hydrogen peroxide on a clean slide presence of effervescence indicated catalase positive reaction whereas negative reaction showed no effervescence.

Motility test

This test was carried out to determine the presence of or absence of flagella as organelle of movement in the bacteria isolated. Discrete colonies of overnight culture was placed on microscopic slide containing a drop of peptone water and covered with a cover slip after a minute. Then, it was viewed microspically with high power objectives. Motile organisms were seen swimming around indicating a positive reaction while non-motile organisms indicated negative reaction.

Methyl Red Test

In carrying out this test, a test organism was inoculated in a test tube containing 5m1 of prepared peptone water and was incubated for 48hours at 37^0c after incubation, 0.5m1 of methyl red was added into the test tubes and allowed to stand for 15 minutes. A reddish color on the addition of indicators signified a positive result while a yellowish color denoted negative result.

Coagulase test

In carrying out this test, a test organisms was inoculate in a test tube containing 1ml of rabbit plasma which was labeled negative control and positive control, and was incubated at 37^0c and the suspensions was observed at half an hour intervals for a period of four hours. Positive result was indicated by gelling of the plasma which remains in place even after inverting the rube, while negative result remains until four hours at 37^0c, the tube is kept at room temperature for overnight incubation.

Oxidase test

A piece of filter paper was soaked with 1% of the substrate tetraethyl-p phenylenediamine dihydrochloride, the paper was moisten with sterile distilled water. A bit of the isolate were obtained with sterile wire loop and smeared on the wetter portion of the filter paper. The development of an intense purple color within 30 second, was observed.

Statistical analysis

Statistical analysis was carried out using One-way Analysis of Variance (ANOVA). Data were analysed using SPSS Version 27 computer software. Data were expressed as the mean $\pm$ standard error of mean and values of $P< 0.05$ were considered significant.

Results and discussions

Results

The Table 4.1 display the bacteria isolated from the borehole water sample point considered in the study, it was noted from the table that a total of six (6) bacteria species were isolated from the borehole water samples after undergoing bacteriological assessment. It was also observed from the table that *Klebisella pneumonia* was isolated in most of the sample point in comparison with other bacteria species isolated from all the sample points.

Morphological and cultural characters of the six species of bacteria isolated from the borehole were examine via microscope, table 2 present their gram stain, morphology, and the cultural form. It was observed that most gram stain of the bacteria isolated were red/pink which is has a result of their cell having less peptidoglycan and being more complex with various proteins, polysaccharides and lipids, also blue/purple stain was also observe which implies gram positive bacteria. The colonies of the bacteria isolated were found to be yellowish, greenish, whitish, pinkish, and black which were observed via microscope. All the bacteria isolated are gram negative bacteria expect *Staphylococcus aureus* that is gram positive bacteria.

The Table 3 below presents the biochemical characteristics of bacteria isolated from the borehole water in each sample point under consideration, from the table it was deduce that ten (10) bacterial were isolated from the borehole water, with the biochemical test revealing six species of the bacteria which include *Staphylococcus aureus Pseudomonas aeruginosa, Salmonella sp, Enterobacter sp, Proteus mirabilis, and Klebsiella pneumonia.* The isolated bacteria were subjected to various tests such as Oxidase test, Coagulase test, Urease test, citrate oxidation catalase, and motility test. From the Table 4.3 *E. coli* was H_2S, motility, and gas positive while citrate, urease, coagulase, and oxidase were negative for the organism, the organism also produces acid. The result for *Salmonells sp* produces positive for oxidase, citrate, and urease as well as double positive for hydrogen sulphide while coagulase test and motility test display negative, the organism display the production of both alkali and acid. *Pseudomonas*

aeruginosa was positive to oxidase test, urease test, acidic test, and gas test while negative for citrate, coagulase, and motility test. Acid producing *Klebsiella pneumonia* was positive for citrate and urease but negative for oxidase, coagulase and motility test. *Proteus mirabilis* was positive for oxidase, citrate, coagulase and urease test but negative for only motility test, the organism produce both acid and gas. Finally, *E. sp* was positive to production of gas only but negative to other biochemical tests.

The Table 4 below presents the percentage of occurrence of the bacteria found in the borehole water of the sample point, it was observe from the table that Total viable count (TVC) and Total Coliform count of the sample point BHA, SQA, and SQB are relative high (10%, 10%,and 10%) in comparison with their other counter parts, Total *Staphylococcus aureus* count TSaC) is relatively high only at sample point AG with (10%) of the sample, while Total *Escherichia coli* count (TEcC), Total *Pseudomonas aeruginosa* count(TPaC), Total *Salmonella sp* count (TSalC), Total *Proteus mirabilis* count (TPmC) and Total *Klebsiella pneumonia* count (TKpC) has a small percentage below 10% in their various sample point where they been Isolated. TVC has the highest total percentage of 52.0% for all the sample point followed by TCC with 47.0% and TPac with 22.5% of the isolated bacteria of the borehole water of the sample point while the rest of the bacteria species count were below 20%. This implies that the waters are not free from total coliforms which are probably from the environmental sources.

Table 1: Bacteria Isolated From Borehole Water

Sample Point	*Bacteria*
BH1	*Salmonelis sp. Pseudomonas aeruginosa*
BH2	*Enterocater sp.*
GH1	*Proteus mirabilis, Pseudomonas aeruginosa*
GH2	*Klebisella pneumonia, Pseudomonas aeruginosa*
LUC1	*Klebisella pneumonia*
LUC2	*Klebisella pneumonia*
GH	*Enterobacter sp*
SQA	*Escherichia coli*
SQB	*Proteus mirabilis*

Note: BHA= Boys Hostel A, BHB= Boys Hostel B, GHA= Girls Hostel A, GHB= Girls Hostel B, DOB= Double Across, GH= Gwarika House, LUC1= Locus 1, LUC2 = Locus 2, SQA = Staff Quaterz A, SQB = Staff Quarterz B,

Table 2: Morphological and Cultural Characteristics of Bacteria from Borehole Water in Bwarak and Its Environs

Bacteria	Gram Stain	Morphology	Cultural Characteristics
Staphylococcus aureus	Purple stain	Gram-positive cocci in gape-like cluster	Yellow colonies with yellow zones
Pseudomonas aeruginosa	Red stain	Gram-negative rods	Greenish colonies
Salmonella sp	Red stain	Gram-negative rods	Black colonies
Enterobacter sp	Pink stain	Gram-negative rods	Whitish colonies
Proteus mirabilis	Red stain	Gram-negative rods	Pinkish colonies
Klebsiella pneumonia	Pink stain	Gram-negative rods	Yellowish colonies

Sources: Laboratory Test, 2023

Table 3: Biochemical Test of Bacteria Isolated from Borehole Water in FCE and Environs

Sample name	OX	CT	Mot	UR	CO	Slope	Butt	H_2S	Gas	Species
BH1	+	+	-	+	-	Alkali	Acid	++	-	*Salmonells sp., Pseudomonas aeruginosa*
BH2	-	-	-	-	-	Acid	Acid	++	+	*Enterobacter sp.*
GH1	+	-	-	+	-	Acid	Acid	++	+	*Proteus mirabilis, Pseudomonas aeruginosa*
GH2	-	+	-	+	-	Acid	Acid	+++	-	*Klebsiella pneumonia*
LUC1	-	-	-	+	-	Acid	Acid	-	-	*Klebsiella pneumonia*
GH	-	-	-	-	-	Acid	Acid	-	+	*Enterobacter sp.*
SQA	-	-	+	-	-	Acid	Acid	+	+	*Escherichia coli*
SQB	+	+	-	+	+	Acid	Acid	+	+	*Proteus mirabilis*

Note: += present, -= absent, OX= Oxidase test, UR= Urease test, CT= Citrate test, Mot= Motility test, CO= Coagulase test, H_2S= Hydrogen Sulphide production.

Table 4: Percentage Occurrence of Bacteria Found in Borehole Water

Points	No of Samp.	TVC	TCC	TSalC	TEcC	TPac	TSalC	TEnC	TPmC	TKpC
BHA	4	4(10)	4(10)	0(0)	2(4.5)	2(5)	1(2.5)	2(4.5)	1(2.5)	4(10)
BHB	4	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
GHA	4	2(4.5)	2(4.5)	0(0)	1(2.5)	2(5)	0(0)	0(0)	1(2.5)	2(4.5)
GHB	4	2(5)	1(2.5)	1(2.5)	0(0)	0(0)	0(0)	0(0)	1(2.5)	2(5)
DOB	4	1(2.5)	1(2.5)	1(2.5)	0(0)	0(0)	0(0)	0(0)	0(0)	1(2.5)
GH	4	2(5)	1(2.5)	4(10)	0(0)	2(5)	0(0)	0(0)	0(0)	2(5)
LUC1	4	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
LUC2	4	4(10)	4(10)	0(0)	2(4.5)	2(5)	1(2.5)	0(0)	2(4.5)	4(10)
SQA	4	4(10)	4(10)	0(0)	1(2.5)	1(2.5)	2(5)	1(2.5)	0(0)	4(10)
SQB	4	2(5)	2(5)	2(5)	1(2.5)	0(0)	0(0)	1(2.5)	0(0)	2(5)
Total	40	21(52.5)	19(47.0)	8(20)	7(16.5)	9(22.5)	4(10)	4(9.5)	5(12)	21(52.0)

Note: *Tvc=total viable count, Tcc=total coliform count*, TSalC= *Total Salmonella sp* count, TEeC= Total *Escherichia coli* count, TPaC = Total *Pseudomonas aeruginosa* count, TPmC = Total *Proteus mirabilis* count, TKpC = Total *Klebsiella pneumonia* count, and TSaC = Total *Staphylococcus aureus* count

Discussions

Six species of bacteria namely; *Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella sp, Enterobacter sp, Proteus mirabilis, and Klebsiella pneumonia* were isolated from the borehole water with respect to their sampling points with *Klebsiella pneumonia* isolated mostly in the sampling point in comparison to other species the result us contrary to Azuonwu *et al*, (2017) where *Bacillus spp* was the most isolated with an occurrence of 7 (23.3%) and other organisms isolated included *Staphylococcus aureus, Shigella spp, Salmonella spp, Enterobacter spp, Streptococcus spp, Proteus spp* and *Escherichia coli*. Also Olajubu, F.A Ogunnika F. 2015 isolate Seven bacteria species, *Bacillus cereus, Bacillus subtilis, Pseudomonas aeruginosa, Klebsiella pneumoniae, Staphylococcus aureus, Salmonella paratyphi and Proteus vulgaris* with *Staphylococcus aureus* been the most frequently isolated among the bacteria, having been isolated in three of out of the seven samples examined and in Bello O.O. *et al*, 2013 [8] study eight (8) genera of bacteria which include *Escherichia coli, Klebsiella sp, Salmonella sp, Shigella sp, Enterococcus sp, Proteus sp, Pseudomonas aeruginosa and Staphylococcus aureus* were isolated from the water samples. The presence of these bacteria in drinking water system is quite alarming.

From the morphological and cultural characteristic table, most of the bacteria species isolated from the borehole in the sampling point were gram negative bacteria with rods except for *Staphylococcus aureus that is gram positive bacteria with* cocci in grape like cluster, the report are in consonance with Azuonwu *et al.* (2017) where *Staphylococcus aureus, Shigella spp, Salmonella spp, Enterobacter spp, Streptococcus spp, Proteus spp* and *Escherichia coli* were isolated and it found that most of these organisms are gram negative microorganisms and are usually associated with gastrointestinal illness.

In this present study, from the six bacteria species isolated in the sampling point *E. coli* has H_2S, motility, and gas to be positive while citrate, urease, coagulase, and oxidase were negative for the organism, the organism also produces acid, this support the finding of Aluko *et al*, 2001 having *E.coil* species with H_2S, motility positive and urease negative also the study of Angulo *et al*., 2007 found H_2S, motility and gas positive for most of the bacteria species. And the result for *Salmonells sp* produces positive for oxidase, citrate, and urease as well as double positive for hydrogen sulphide while coagulase test and motility test display negative, the organism display the production of both alkali and acid which is contrary to the study of Tassadaq *et al.* (2013) with *Salmonells sp* producing negative for oxidase and urease while citrate and lactose are positive. *Pseudomonas aeruginosa* was positive to oxidase test, urease test, acidic test, and gas test while negative for citrate, coagulase, and motility test. Acid producing *Klebsiella pneumonia* that has positive for citrate and urease but negative for oxidase, coagulase and motility test was in line with the report of Tassadaq *et al. (*2013) has positive was obtain for both citrate and lactose, and negative was obtain for oxidase. *Proteus mirabilis* was positive for oxidase, citrate, coagulase and urease test but negative for only motility test, the organism produce both acid and gas. Finally *E. sp* was positive to production of gas only but negative to other biochemical tests.

Result from the bacteriological assessment of the borehole water across the sampling point of the study area revealed that the total viable count (TVC) in borehole water samples collected in the study ranged from 0 to 4 cfu/100ml, constituting 52.5% of the total samples of the borehole water under consideration, similarly, the total coliform count (TCC) of the borehole water analyzed ranged from 0 to 4 cfu/100ml, with 47.0% of the total sample of the borehole water. The bacteria generally isolated in the course of this study at different percentages include *Enterobacter sp, Staphylococcus sp*., *Escherichia coil, Pseudomonas sp*., *Proteus sp*., and *Klebsiella sp*. (Table 4.4). These are known to be able to cause gastroenteritis in individuals whom consume the contaminated borehole water. *Klebsiella pneumonia* and *Pseudomonas aeruginosa* is the most predominant organism isolated from the borehole water across the sampling point with their total bacteria count percentage of 52.2% and 22.5% respectively this work is contrary to the report of Iroha C. *et al.,* 2016 that the results of the microbiological study revealed that the bacterial pathogens isolated in this study include *E. coli* (40%), *Staphylococcus aureus* (32%), *Pseudomonas aeruginosa* (16%) and *Klebsiella spp* (12%) with *Klebsiella spp* having the lowest percentage occurrence. The predominance of the two organism in the borehole water of the study area is slightly risky to the people in the environs as this organism are the common cause of urinary tract and wound infections where they cause acute or chronic infection, this prove was similarly to the study of (Bello *et al*., 2013) [8].

In this present study it also noted that the number and percentage of positive samples from the boreholes sampled. *Staphylococcus aureus* was isolated in BHA, DOB, GH and SQB having *2.5%, 2.5%, 10%* and *5%* percentage growth respectively. *Escherichia coli* was isolated from 1 of the total borehole water sampled having 2.5%. 5% of the bacteria *Pseudomonas aeruginosa* was isolated from 2 borehole water samples. *Salmonella sp* was isolated from 1 of the total borehole water sampled with *2.5%;* this result is in agreement with Nwandkor & obeagu, (2015) [33]., *Proteus mirabilis* and *Klebsiella pneumoniae were* isolated from 2 of the total boreholes sampled which is in agreement with Oshatogbe *et al.,* (2016) [37] and Nwandkor and Obeagu (2015) [33]. BHA and SQB borehole water sampling unit experience high prevalence of bacteria, with most of the bacterial species isolated from the borehole water analyzed. The high predominance of bacterial occurrence in BHA and SQB borehole water in the sampling unit is an indication of poor hygiene and sanitation, and a general collapse in the provision of safe drinking water.

A large portion of the water samples complies with EPA standard for coliform in water. According to EPA standard, every water sample that has coliform must be analyzed for either fecal coliforms or *E. coli* (EPA, 2003) with a view to ascertaining contamination with human or animal waste and possibly pathogenic bacteria or organism, such as Gardia and Cryptosporidium may be present (EPA, 2003). 2015This may be the result of aquifer contamination, which is a particular problem where fissured geological strata are combined with thin topsoil, and on the increase, notably in urban and periurban areas (WHO, 1997).

Bacteriological tests are extremely sensitive and specifically designed to reveal the evidence of water pollution (Ajayi & Adejumo, 2011) [1]. *E. coli* in borehole water is an indication of poor hygiene and sanitation, and a general collapse in the

provision of safe drinking water.Total coliform in all types of water sources exceeded WHO (2004) maximum permissible load (0/100 ml) for drinking water. Total coliform organisms may not always be directly related to the presence of faecal contamination or pathogens in the drinking water. It is noteworthy that *Salmonella sp.*, a waterborne pathogen (like a few other bacteria) is difficult to culture from water due to its ability to enter into a viable, but not culturable state after exposure to oligotrophic, aquatic environment (EPA/625/R-92/004, 1992). Yet, this investigation revealed the presence of *Salmonella sp* in 1 (2%) of the entire boreholes sampled, this result is in agreement with Nwandkor and Obeagu *2015* [33]. This gives a warning signal as to the rate of incessant typhoid fever that is regularly being reported in our clinics (Nwandkor and Obeagu 2015) [33]. Boy's hostel A water sample was not obtained directly from the borehole as the water is pumped into a tank and collected through a tap. This is an indicator that the water is not suitable for drinking and bathing. Since the water once pumped from the ground goes into the tank from where it is assessed, it can be surmised that the contamination of the water sample is from the tank or the outlet through which the water is gotten Water from deep boreholes is normally free from microbiological contamination and may be used by small communities without further treatments. This may depend on geographical locations and the location of the aquifer, (Nwandkor and Obeagu 2015) [33].

The *Staphylococcus* species is known to produce enterotoxin (Ajayi and Adejumo, 2011) [1] which targets the intestines. Proteus species is an intestinal flora, but also widely distributed in soils and water. *Pseudomonas aeruginosa* is an example of non-faecal coliforms, while E. coli are a fecal coliform Ajayi and Adejumo, (2011) [1] and Ehiowemwenguan, *et al*, (2014) [21]. The result obtained from this investigation can be compared to that obtained by Ajayi and Adejumo (2011) [1] and Nwandkor and Obeagu, (2015) [33].

Other potential risk factors which can cause water contamination includes indiscriminate disposal of refuse, poor personal hygiene, construction of septic tanks without proper adherence to guidelines, and the use of tanks without taking proper precautions to avoid the accumulation of bacteria. The distance from potential sources of contamination such as latrines, cattle pen, refuse pits, and hollows in the ground all may serve as a potential source of pollution especially if the distance is not up to 30 meter or more, this corroborates the work of Oshatogbe *et al, 2016.* [37]

Water and sanitation are the primary drivers of public health which means that once we can secure access to clean water and to adequate sanitation facilities for all people, irrespective of differences in their living conditions, a huge battle against all kinds of disease would have been won. In most countries the principal risks to human health associated with the consumption of contaminated water are microbiological in nature (although the importance of chemical contamination should not be under estimated).An estimated 80% of all diseases and over one third of deaths in developing countries are caused by water related diseases. In drawing up standards for drinking water quality, it will be necessary to take into account various local, geographical, socioeconomic and cultural factors (Nwandkor and Obeagu, 2015) [33], as some of these factors are responsible for contaminating the water..

It is recommended by the WHO that no microbial organisms should be found in 100ml of drinking water. The greatest risk from microbes in water is associated with consumption of potable water that is contaminated with human excreta, although other sources of exposure is significant (WHO, 2008).

Conclusion

The study has revealed that borehole water of Bwarak, Pankshin and its environs is vulnerable to bacteriological pollution. Hence, proper bore-hole location and construction, control of human activities to prevent sewage from entering bore-hole environment is the keys to the avoiding bacterial contamination of drinking water.

Recommendations

From the findings of this research, the researcher made the following recommendations

1. Proper sanitation goes a long way in reducing the contamination of water from the borehole. This calls for a conscious effort on the part of the users and the community as a whole. In disposing refuse, it should be done farther away from these water source and septic tanks should be built far away from borehole water sites.
2. The need for personal hygiene cannot be over emphasized as community health specialist from the ministry of health should ensure routine checks, by going round these communities inspecting these boreholes and create awareness on the need to always ensure proper hygiene habits.
3. Hospitals and laboratories should be visited by individuals suffering from stomach aches for proper check-up and test to determine the actual cause of the ache or pain, instead of treating oneself based on self-prescription.

Acknowledgement

Special appreciation to Tertiary Trust Fund (TETFUND) for sponsoring the research. Worthy of acknowledgement are individuals and institutions without whose assistance this research would not have been possible, the Provost FCE Pankshin, the Area Director Central Zone, Plateau State Ministry of Education, Research Assistance our families who provided various forms of assistance. To God be the glory.

References

1. Ajayi AO, Adejumo TO. Microbiological Assessment and Some Physico-Chemical Properties of Water Sources in Akungba-Akoko, Nigeria. Journal of Toxicology and Environmental Health Sciences, 2011, 342-346.
2. APHA, (2005). Standard Methods for the Examination of Water and Wastewater, 18th ed. American Public Health Association American Water Works Association and Water Pollution Control Federation, Washington, DC.
3. APHA, (2002). Standard Method for the Examination of Water and Waste Water. 20 ed. America Public Health Association, Washington, D.C.

4. APHA, (1998). Standard Methods for the Examination of Water and Wastewater, 14th ed. American Public Health Association American Water Works Association and Water Pollution Control Federation, Washington, DC.
5. Ashbolt NJ, Grabow WO, Snozzi M. Indicators of Microbial Water Quality, 2001, 289 – 316 In: Fewtrell, L. & Bartrom, J. (eds), Water quality: Guidelines, standards and health. World Health Organization and IWA Publishing, London UK.
6. Avery D. *The Third World Water Forum*. World Health Bulletin,2003:81(5):383.
7. Bartram J, Cortruro J, Exner M, Fricker C. Classmacher a. (2003). *Heterotrophic plate counts and drinking water safety. The significance of HPCs for water quality and human health*. IWA Publishing, London UK, 2003, 121-126.
8. Bello OO, Osho A, Bankole SA, Bello TK. Bacteriological and physicochemical analyses of borehole and well water sources in Ijebu-Ode, Southwestern Nigeria. *Int. J. Pharm. Biol. Sci*,2013:8:18–25.
9. Brooks GF, Jawetz E, Melnick JL, Adelberg EA. Jawetz, Melnick & Adelberg medical microbiology. New York, McGraw Hill Medical, 2010.
10. Chave J. Bacteriological Analysis of waterhttp://www.Watertest.Co.Uk/bacteriological analysis, html, 1999, 1.
11. Cheesbrough M. District Laboratory Practice in Tropical Countries PART 2. Cambridge University Press. The Edinburgh building, Cambridge CB22RU, United Kingdom, 2000, 234-236.
12. Chukwurah EI. Aquatic Microbiology. Otoba press limited, Onitsha, Nigeria, 2001.
13. Clark B, Koning N, Roos JC. The Continued Influence of Organic Pollution on the Water Quality of the Turbid Modder River. Water South Africa,1993:25(3):285-292.
14. Control of community supplies. World Health Organization, Geneva, Available at: www.klima-warnsignale.uni-hamburg.de Retrieved on the 5th March 2017
15. Council of the European Union. (1998). Council Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption. Official Journal L 330, 0032 – 0054.
16. Daniel H, Warwick LG, Paul TM, Stuart A, Christophe PS. Enumeration of water-borne bacteria using viability assays and flow cytometry: a comparison to culture-based techniques. Journal of Microbiological Methods,2003:55:585-597.
17. Diersing N. Water Quality: Frequently Asked Questions. Florida Brooks National Marine Sanctuary, Key West, FL, 2009.
18. Doyle MP, Erickson MC. Closing the door on feacal coliform assay. Microbiol.,2006:1:162-163. ISSN 1558-746.
19. Eckburb, PB, Bik EM, Bernstein CN, Purdom E, Dethlefsen L. Diversity of the human intestinal microbial flora. Science,2005:308:1635-1638.
20. Edberg SC, Rice EW, Karlin RJ, Allen MJ. *Escherichia coli the best biological drinking water indicator for Public Health Protection*. Symp. Ser. Soc. Appl. Microbiol.,2000:29:(1605)-1165.
21. Ehiowemwenguan G, Iloma AO, Adetuwo JO. *Physico-Chemical and Bacteriological Quality of Borehole Water in Eyaen Community Area of Edo State, Nigeria*. International Journal of Basic and Applied Science,2014:03(2):60-68.
22. Engwa AG, Tagbon NR, Iyala CPJ, Unaegbu M. Physicochemical and Microbial Analysis of Potable Water Sources in Enugu Metropolis. J Pub Health Epid,2015:7(3):65-70.
23. EPA. US Environment Protection Agency, Safe Drinking Water Act Amendment, 2002.
24. EPA, (2003). US Environmental Protection Agency Safe Drinking Water Act.
25. EPA/625/R-92/004. Manual: Technical Issues in Planning Water Reuse Systems, 1992, 17-28.
26. Grabow WOK. the Safety of Water Disinfection Chemical and Microbial Risks. Radisson Denville Resort, Miami Beach, Florida. USA, 2000, 15-17.
27. Havelaar A, Blumenthal U, Strauss M, Kay D, Bartram J. Guidelines: the current position, Water Quality: Guidelines, Standards and Health. World Health Organization (WHO). Published by IWA Publishing, London, UK, 2001.
28. Health Canada. A guideline to well water treatment and maintenance, 2008. http://www.who.intawatersanitation.org
29. Howard G, Smith M. Rapid assessment of drinking water quality: a handbook for implementation joint monitoring for water supply and sanitation WEDC. Longborough University, 2003. ISBN 184380: 042.
30. Ibe SN, Olplenye JI. Bacteriological Assessment of Borehole Water In Uli, Nigeria. African Journ of Applied Zoology & Environmental Biology,2005:7:116-119.
31. Mims C, Dockrell H, Goering R, Roitt I, Wakelin D, Zuckerman M. Medical Microbiology (3rd Ed.). Mosby, 2004, 287. ISBN 978-0-7234-3259-3.
32. NIS. Nig. Standards for drinking water quality. Oxford Press, 2007, 5.
33. Nwandkor UU, Obeagu EI. Bacteriological Assessment of Different Borehole Drinking Water Sources in Umuahia Metropolis. Int. J. Curr. Microbiol. App. Sci.,2015:4(5):1139-1150.
34. Obi CN, George P. The Microbiological and Physico-chemical Analysis of Borehole Waters used by Off-campus Students of Michael Okpara University of Agriculture, Umudike (MOUAU), Abia State, Nigeria. Res J Bio Sci,2011:6(11):602-7.
35. Olowo SI. Standard operating procedure for water production. Food forum magazine. Nig. Ins. Food Sci. Tech,2005:4:30-31.
36. Osei P. Water Quality of Boreholes and Hand-Dug Wells at Pakyi No. 1 in the Amansie West District of Ashanti Region of Ghana. Kwame Nkrumah University Of Science And Technology, Kumasi, 2014.
37. Oshatogbe AJ, Ijah UJJ, Riskuwa MJ. Bacteriological and physicochemical quality of water sources located in some slums in Sokoto metropolis, Sokoto state, Nigeria. Int J Original Res,2016:2(4):203-217. www.enrichjournals.com
38. Potential Threats to Our Groundwater". The Groundwater Foundation. Retrieved 24 September, 2015.

39. Review on Microbiological standards for water and their relationship to health risk. (2000). Barrell, R.A.E., Hunter, P.R., & Nichols. G. Communicable disease and public health, 2000, 3(1).
40. Ryan KJ, Ray CG. Sherris medical microbiology: an introduction to infectious diseases (4th ed.), 2004. McGraw-Hill Professional Med/Tech. ISBN 978-0-8385-8529-0.
41. Shelton T, Scibila SL. interpreting Drinking Water Quality Analysis. !jt~hd i t ionR utgers Cooperative Extension. New Jersey Agricultural Experiment Station, New Brunswick, 1998. NJ. Available: http:l/www.nj.gov/deplpwtaaddress.htm. via the INTERNET. Accessed 2007 May 24
42. Shigellosis. State of the art of new vaccine research and development (PDF). Immunization, Vaccines and Biologicals. World Health Organization, 2006, 10–2.
43. Szewzyk U, Szewzyk R, Mauz W, Schleifer KH. Microbiological safety of drinking water. Annu. Rev. Microbiol.,2000:54:81-127.
44. Tamberkar DH, Hirlkar NB. Rapid and modified field test for detection of feacal contamination in Drinking water. Journ. Sci. Ind. Res,2007:66(4):667.
45. Todar K. Todars online textbook of bacteriology. Available at www.textbookofbacteriology.net.
46. Uhuo CA, Uneke BI, Okereke CN, Nwele DE, Ogbanshi ME. The Bacterialogical Survey of Borehole Waters In Peri-Urban Areas Of Abakaliki; Ebonyi State, Nigeria. Int. Journ of Bact. Res ISSN: 2392-5690,2014:2(2):028-031.
47. Ukpong EC, Okon BB. Comparative Analysis of Public and Private Borehole Water Supply Sources in Uruan Local Government Area of Akwa Ibom State. International Journal of Applied Science and Technology,2013:3(1):76.
48. WHO. Guidelines for Drinking Water Quality: Surveillance and Control of Community Supplies. (2nded.). Geneva,1997:3:6-210.
49. WHO/UNICEF (2010). *Joint Monitoring Programme for Water Supply and Sanitation. Progress on Sanitation and Drinking-water: 2010 Update. Publications of the World Health Organization can be obtained from WHO Press, World Health Organization,* 20 Avenue Appia, 1211 Geneva 27, Switzerland
50. Wolf J, Prüss-Ustün A, Cumming O, Bartram J, Bonjour S, Cairncross S, Clasen T, *et al*. Systematic review: Assessing the impact of drinking water and sanitation on diarrhoeal disease in low- and middle-income settings: systematic review and meta-regression". Tropical Medicine & International Health,2014:19(8):928–942. doi:10.1111/tmi.12331
51. Wolf L, Nick A, Cronin A. How to keep your groundwater drinkable: Safer siting of sanitation systems – Working Group 11 Publication. Sustainable Sanitation Alliance, 2015.
52. World Health Organisation, WHO (2004) Water Sanitation and Health. Water related diseases. www.who.int/water sanitation and health/diseases/
53. World Health Organization (2006). Guidelines for drinking water quality. Third Edition, WHO press, Geneva, Switzerland. 398.
54. www.sundaymail.co.za/how-safe-is-borehole-water/ Retrieved 13th March 2017.
55. www.sundaymail.co.za/how-safe-is-borehole-water/ retrieved 15th March 2017.
56. www.vanguardngr.com/2016/12/regular-test-borehole-water-fg-advises-nigerians Retrieved 19th December 2016
57. Yabuuchi E. Bacillus dysentericus (sic) 1897 was the first taxonomic rather than Bacillus dysenteriae 1898". International Journal of Systematic and Evolutionary Microbiology,s2002,52(Pt3):1041. doi:10.1099/00207713-52-3-1041. PMID 12054222.