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Research Article

Physicochemical and Microbiological Analysis of some selected Borehole water in Dange-Shuni Local Government Area.

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The present study aimed to evaluate the microbiological and physicochemical analysis of borehole water in Dange/Shuni Local Government Area, Sokoto. A total of six (6) water samples were collected from Dange/Shuni Local Government from six different borehole water sources in Tuntube-Tsefe, Shuni, Rikina, Dange, Wababe and Fajaldu borehole Dange/Shuni LGA. The physicochemical parameters of the borehole water, including colour, odour, taste, pH, electrical conductivity, total dissolved solids, total hardness, chloride, nitrate, calcium, and magnesium contents, were analysed using standard laboratory procedures. The bacteriological analysis was carried out using the multiple-tube (most probable number) technique for the enumeration of both total coliform count and faecal coliform count. The sample was also isolated and identified using biochemical tests. Fungal isolation and identification were also carried out using standard techniques. The results of this study have shown that the physicochemical parameters analysed were within the standard limits, except for temperature, magnesium, and calcium contents. The total bacterial count ranged from 1.2×10^5 CFU/mL to 3.4×10^5 CFU/mL, which exceeds the WHO limit. The microbiological analysis also revealed the presence of bacterial and fungal species, which include: *Staphylococcus aureus* (16.7%), *Escherichia coli* (16.7%), and *Pseudomonas aeruginosa* (33.3%), *Bacillus subtilis* (33.3%), while fungal species include *Aspergillus flavus* (33.3%), *Endomyces* (16.66%), *Aspergillus niger* (16.66%) and *Rhizopus* (16.66%). In conclusion, the pathogenic microorganisms identified in Wababeborehole water, along with the high microbial load observed, make it unsuitable for drinking purposes. Thus, there is a need to create awareness about the current situation of the wash boreholes and the necessity for further treatment by consumers, before they can be used for both drinking and domestic purposes..

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INTRODUCTION

Sonawane, Vilas. (2020) stated that the water is a vital resource for human survival. The availability of good quality water is an indispensable feature for prevents diseases and improving quality of life. It is necessary to know details about different physical parameters like color, temperature, Total hardness, pH, sulphate, chloride, DO, BOD, COD, and alkalinity used to test water quality. This research work aims to analyze water quality using the Biological and Physicochemical parameters of water samples collected from six (6) wards in Dange/Shuni local government area of Sokoto State.

Ideally, drinking water should be safe and acceptable to all. Guidelines on the quality of drinking water by the World Health Organization (WHO) provided standards in terms of microbial, chemical and radionuclide content, as well as physical properties for safe drinking water. More so, in a bid to transform our world and ensure better lives for humans, the United Nations, of which Nigeria is a member, is guided by 17 sustainable development goals (SDGs). The SDGs came into effect in January 2016 with a Global agenda till 2030. Goal number six of the SDGs is targeted towards clean water and sanitation (UN, 2020). Four years into the implementation of the SDGs globally, it is important to examine how Nigeria is faring towards achieving the goal of access to clean potable drinking water with respect to quality and quantity.

The quality of drinking water has now become a very important topic in all countries of the world, especially in developing countries like Nigeria. Though, water plays an important role that can never be overemphasized in human life, it is known to have great potentials for transmitting a variety of diseases and illness when it is contaminated or polluted (Aleksander, & Ciszewski, 2016) Also water is one of the most essential natural resources needed by every living organism (Duru *et al.*, 2017; Onyango *et al.*, 2018). Whether it is used for drinking, bathing, food production or recreational purposes; portable and accessible water supplies are crucial for public health (Yusuf, 2007; WHO, 2018; WaterLife, 2018).

The chemical composition of ground water is a measure of its suitability as a source of water for

human consumption or for agricultural and industrial purposes. Ground water can be rich in dissolved solids, especially carbonates and sulfates of calcium and magnesium as well as chloride and bicarbonates, depending on the strata through which ground water flow, thus, additional treatment may be required in order to provide pleasant water for drinking and household use (Cadinuet *et al.*, 2023).

METHODOLOGY

Study Area

The study was carried out in the Laboratory of Microbiology Department, Sokoto State University, Sokoto, North-Western Nigeria. Water samples was obtained from Dange/Shuni local government with latitude 12.7956 'N and 5.4359 'E longitudes.

Sample Collection

A total of six (6) water samples were collected from Dange/Shuni Local Government in Six different borehole water sources in Tuntube-Tsefe, Shuni, Rikina, Dange, Wababe and Fajaldu borehole Dange/Shuni LGA. The borehole water samples was aseptically collected from the sources using sterile glass bottles after pumping water sample to waste for one or two minutes. The mouth of the tap was disinfected with cotton wool soaked in 70% ethanol. The water samples were packed in an ice box which was transported to the Microbiology Laboratory, Department of Microbiology Sokoto State University, Sokoto. All samples were stored at 4°C before the commencement of the practicals.

Media Preparation

All media preparation were carried out according to the manufacturers instruction thus, media used in the study includes the followings:

Nutrient Agar

Twenty-eight gram (28 g) of nutrient agar (NA) powder was dissolved in 1 litre of distilled water in a conical flask covered with cotton wool and aluminium foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes to be cooled to 45-50°C; about 20ml was dispensed aseptically into sterile Petri dishes.

MacConkey Agar

The medium was prepared by dissolving 7.8g of MacConkey agar powder in 300ml of distilled water. The conical flask's mouth was covered using sterile cotton wool and aluminium foil,

and then the mixture was shaken and subjected to heat on the hot plate for complete dissolution. The dissolved medium was sterilized using an autoclave at 121°C for 15 minutes (Cheesbrough, 2006).

Eosine Methylene Blue (EMB) Agar

The medium was prepared by dissolving 9.72g of EMB agar into 270ml of distilled water in a conical flask; this was covered and hot plated for good suspension. The medium was autoclaved at 121°C for 15 minutes (Cheesbrough, 2006).

Potato Dextrose Agar (PDA)

Thirty-nine grams (39 g) of potato dextrose agar (PDA) powder was dissolved in 1 litre of distilled water in a conical flask covered with cotton wool and aluminium foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile Petri dishes (Cheesbrough, 2006).

Physicochemical Analysis

The water samples collected was analysed for pH, temperature, odour, colour, taste, turbidity, electrical conductivity, total dissolved solids, total alkalinity, total hardness, chloride, nitrate and selected trace metals. The analysis was carried out at Agric chemical laboratory in UsmanuDanfodio University, Sokoto.

Determination of pH

The pH of the water samples was determined using the Wagtech pH meter. 10 ml of each of the samples was poured into a sterile beaker and the anode of the pH meter was dipped into it and readings were obtained when it was stable (Ademorati, 2000).

Measurement of Temperature

The temperature of each sample was determined with mercury-bulb thermometer by immersing the bulb vertically into the water samples and allowed to stand till the temperature reading is steady (Ademorati, 2000).

Determination of Colour, Odour and Taste

A 20 mL volume of each water sample was poured into a clean beaker. The sample was then shaken vigorously to check for any frothing and allowed to settle. Colour was determined through visual examination and then odours were determined through the sensation of smell. Taste tests for consumer acceptability were performed on the water samples. Small volumes

of each sample was tasted with the tongue and then immediately rinsed with distilled water (taste free) after each sample, and the results was recorded accordingly (Ademorati, 2000).

Determination of Turbidity

A two part calibrated turbidity tube was used, with calibrations from 5-25 turbidity units. The joined tubes were held over a white paper, while slowly pouring the water sample into the tube until the black cross at the bottom is no longer visible. At this point the reading was taken from the side of the tube as the turbidity value of the water sample (Ademorati, 2000).

Determination of Electrical Conductivity

A 20 mL volume of each water sample was poured into a clean beaker at room temperature. The Wagtech electrode was placed in the sample, and following stirring the electrical conductivity value was determined after the meter became stable (Ademorati, 2000).

Determination of Total Alkalinity

About 10 ml of water samples were put in curvet; alkaphot tablet was added, crushed and mixed until all particles have dissolved. Optical density reading was taken at 570 nm wavelength on a Wagtech photometer (Ademorati, 2000).

Determination of Total Dissolved Solids

The total dissolved solids were determined by submersing the Wagtech probe into 100 ml of each water sample, and the readings reading was recorded (Ademorati, 2000).

Determination of Chloride

To 50ml of the sample, 5 drops of a phenolphthalein indicator solution was added and neutralized with 0.1N sulphuric acid to the colorless side of phenolphthalein. One ml of potassium chromate indicator solution was added before titration with standard silver nitrate solution to the pinkish-yellow endpoint. A reagent blank titration was carried out in parallel to the sample titration. Chloride concentration was calculated as follows (APHA, 2005): Chloride, mg/l = [(A-B) (N) (35.45)/V] x 100, where A = Silver nitrate solution, in ml for sample titration; B = Silver nitrate solution, used for blank titration (in ml); N = Normality of the silver nitrate solution; n; and V = Sample volume in (ml) (APHA, 2005).

Determination of Nitrate

Test tube was filled with sample to 20 ml mark and one level spoonful (~ 15 ml) of nitrate test

powder (containing zinc dust 60%, and barium sulphate 40%) and one nitrates tablet (ammonium chloride) was added and was shaken for a minute. The tube was allowed to stand for a minute and then inverted 3-4 times to aid flocculation which was allowed to stand for two minutes to ensure complete settlement. The clear solution was dispersed into tube to 10ml mark and one nitricol tablet (sulfanilic acid, acting as the aromatic amine), was added, crushed, and mixed to dissolve, then it was allowed to stand for 10 minutes for colour development and readings were taken on the photometer at 570 nm wavelength (APHA, 2005).

Determination of Total Hardness

Total hardness was analyzed by titration of 50 ml water sample with standard EDTA. The EDTA was added in drops at pH 10 using Erichrome black T indicator until the colour changed into purple and the hardness was calculated by multiplying the average number of drops of EDTA used for the sample by the calibration factor of 20 (APHA, 2005).

Determination of Calcium

A 1000 ppm of calcium was prepared by dissolving 0.92g of calcium chloride (CaCl₂) in about 15ml of distilled deionized water and then later made it up to 250ml. (Ademoroti, 1996).

Determination of Magnesium

A 1000 ppm of magnesium was prepared by dissolving 2.59g of magnesium sulphate (MgSO₄) in about 15ml of distilled deionized water and then later made it up to 250ml. (Ademoroti, 1996).

Microbiological Analysis

Isolation and Identification of Bacteria

Serial Dilution

For each sample collected, 1ml of the samples was aseptically transferred into sterile test tube containing 9ml of distilled water to obtain 1 in 10 (10^{-1}) dilution factor; another 1ml was then transferred from the first test tube (10^{-1}) to the second test tube also containing 9ml of distilled water to obtain a dilution factor of 1 in 100 (10^{-2}). The procedure was repeated for 10^{-3} , 10^{-4} and 10^{-5} dilution factor (Cheesbrough, 2006).

Inoculation and Incubation

Thereafter, an aliquot of 1 ml of the 10^{-4} dilutions from each dilution was aseptically plated on Nutrient agar plates in duplicates, then

spread aseptically using a sterile spreader. Plates were incubated at 37°C for 24 hours before counting the colonies (Cheesbrough, 2006).

Colony Counting

The bacterial viable count observed on the plate was determined by comparing the morphological features or sizes of the colony. Less number of colonies was counted manually; while the larger of colonies was counted using any available colony counter machine and the mean count was recorded based on colony forming units.

Subculture

To obtain pure bacterial isolates, a distinct colony from mixed culture was picked using a sterile wire loop and placed on a fresh nutrient agar medium. After streaking, the petri dish was incubated at 37°C for 24 hours. All isolates from this pure culture was maintained in an agar slant for further analyses (John, 2003; Fox, 2011).

Gram Staining

A thin smear of bacterial isolates was prepared on a glass slide, air dried and then heat fixed by passing over a burner flame at least three times. The smear was covered with crystal violet stain for 1 minute and then washed with distilled water, it was then covered with Lugol's iodine and washed with distilled water after 1 minute, which was decolorized with acetone and rinsed with distilled water immediately for few seconds. The smear was covered with safranin and washed after 1 minute. The back of each of the slides was wiped out with cotton wool and allowed to air dried. Gram positive cell was incorporated or no counterstains was remain blue-violet in appearance, Gram negative bacteria, however, take on a pick color and are easily distinguishable from the Gram positives. Again, it was rinse for 5 seconds to remove any excess dyes (Cheesbrough, 2006).

Microscopy

The dried smear was examined microscopically with oil immersion and viewed with a microscope using x100 objective lens (Cheesbrough, 2006).

Biochemical Test

Catalase test

The test is used to differentiate those bacteria that could utilize hydrogen peroxide into oxygen and water. A Small portion of colony was picked using a sterile glass wire loop and transferred onto a drop of hydrogen peroxide on the glass

slide. The immediate bubbling was observed and indicated catalase positive while negative test was indicated by absence of bubbles (Cheesbrough, 2006).

Oxidase Test

The test is used to differentiate those bacteria that could utilize hydrogen peroxide into oxygen and water. A Small portion of colony was picked using a sterile glass wire loop and transferred onto a drop of hydrogen peroxide on the glass slide. The immediate bubbling was observed and indicated catalase positive while negative test was indicated by absence of bubbles (Cheesbrough, 2006).

Coagulase Test

Coagulases are enzymes that clot blood plasma and they produced by *Staphylococcus aureus*. The enzyme protease converts fibrinogen to fibrin resulting to blood clotting. The Slide method was used. Here, clean slide was divided into two sections, to one section the test organism was smeared on it using a sterile wire loop while a drop of distilled water was added to the other section serving as control. Then human plasma was added to both sections and the slide was rocked gently for some minutes. A clumping/agglutination of the plasma indicate the presence of coagulase (Cheesbrough, 2006).

Urease Test

A little portion of the organism was picked using a sterile wire loop and inoculated into prepared urease medium. This was then incubated at 37°C for 24 hours. A change in the colour of the medium from yellow to pink indicated urease positive test (Cheesbrough, 2006).

Indole Test

This test was used to determine which of the isolates has the ability to split indole from tryptophan present in buffered peptone water. The test is used to differentiate Gram-negative Bacilli especially those of the enterobacteriaceae. Peptone water was prepared and about 3 ml of it was dispensed in bijou tubes using a sterile pipette. Then, fresh sterile loops was used to pick a well-isolated colony of bacteria and inoculated into bijou tubes, thereafter, the tubes was incubated at 37°C for 48 hours. After incubation period, 0.5 ml of Kovac's Indole Reagent was added to the inoculated bijou tubes and gentle shaken. A red ring will be examined in the surface layer within

10 minutes which indicated indole positive reaction (Cheesbrough, 2006).

Citrate Utilization Test

This test is based on the ability of some organisms to utilize citrate as a sole source of carbon. It was carried out by inoculating the test organism in test tube containing Simon's citrate medium and incubated at 37°C for 24-48 hours. A deep blue colour indicates a positive result (Cheesbrough, 2006).

Sugar Fermentation Test

Each of the isolates was tested for its ability to ferment a given sugar with the production of acid and gas or acid only. Since most bacteria especially Gram negative bacteria utilize different sugars as source of carbon and energy with the production of both acid and gas or acid only, the test is used as an aid in their differentiation. The growth medium used was peptone water and the peptone water was prepared in a conical flask and the indicators; phenol red was added. The mixture was dispensed into test tubes containing Durham tubes and sterilized by autoclaving at 121°C for 15 minutes. 1% solution of the sugar was prepared and sterilized separately at 115°C for 10 minutes. This was then aseptically dispensed in 5ml volume into the tubes containing the peptone water and indicator. The tubes was inoculated with young culture of the isolates and incubated at 37°C. Acid and gas production or acid only was observed after about 24 hours of incubation. Acid production was indicated by the change of the medium from light green to yellow colour while gas production was indicated by the presence of gas in the Durham's tubes (Cheesbrough, 2006).

Most Probable Number (MPN)

The coliform counts were determined by the most probable number (MPN) techniques. Samples were incubated in Lactose broth tubes at 37°C for 48 hrs. Measured amounts of double and single strength MacConkey broth (purple colour) were sterilized in bottles containing inverted Durham tube to indicate the gas production. The bottles was arranged in three sets 50 ml, (10 ml and 1 ml and each had 5 bottles), and incubated at 37°C. Fermentation tubes were incubated with 50 ml, 10 ml and 1 ml of aliquot of the samples in accordance with standard methods (APHA, 2005). The tubes

were incubated at 37°C for 48 hrs. Positive tubes producing acid and gas were used to estimate the presumptive Most Probable Number (MPN).

The confirmed test for total coliform was achieved by plating a loopful of positive MacConkey broth on Eosine Methylene Blue (EMB) agar and incubated at 37°C for 24hrs, while the faecal coliform was achieved by transferring a loopful of broth from a positive tube to EC broth and incubated at 44.5°C for 24-48 hours and the tubes was observed for gas formation. Completed test for faecal coliform was carried out by plating a loopful of broth from a positive EC tube unto an Eosine methylene blue agar plate. The plates were incubated at 44.5°C for 48-hours and observed for a dark red colour with metallic green sheen (APHA, 2005).

Isolation and Identification of Fungi

Serial Dilution

Serial dilutions of the water sample was made by aseptically transferring 1 ml of the water sample into 9 ml of sterile distilled water; to make dilutions that range from 10^{-1} - 10^{-4} dilution factor.

Inoculation and Incubation

Aliquot of 1 ml of 10^{-3} dilution of the sample was inoculated into potato dextrose agar (PDA) in triplicate and incubated at 28°C for 72 hrs. Thereafter the developing colonies were counted and the mean values were thus determined (Ellis *et al.*, 20007).

Subculture

Isolates obtained was sub cultured on freshly prepared PDA media to obtain pure isolates.

Sterilized inoculating needle was used to cut a small portion of the isolates from the edges of each petri dish and separately inoculated into fresh media plates. The inoculating needles were sterilized by flaming to red hot after each cut to avoid mixed culture. The plates were then incubated for another seven (7) days at 37°C (Watanabe, 2010).

Macroscopy and Microscopic Examination

The macroscopic examination was based on colonial appearance, pigment production and micro morphology of the spore produced (Watanabe; Ellis *et al.*, 2007).

The pure culture of the isolate obtained was subjected to microscopic examination with the view to identify the organism present in the groundnut cake samples. Clean glass slides were used for the identification. A drop of water was placed in the center of the slide; a small portion of the fungal culture was cut out with a sterile inoculating needle. The piece was placed directly in the water and a smear is formed, a cover slip was used to cover the teased portion. The slides were mounted on the microscope stage, damped with chips, using lower and higher magnification (X10 and X40) objectives.

RESULTS

The result of the study is presented in Table 1 to 6. Table 1 shows the physicochemical components of the different borehole water samples obtained from Tuntube-Tsefe, Shuni, Rikina, Dange, Wababe and Fajaldu.

Table 1: Physicochemical Properties of Water Samples collected from six (6) Borehole Water.

| Samples | pH | EC µs/cm | K mg/l | Na mg/l | DO mg/l | BOD mg/l | Ca mg/l | Mg mg/l | CO ₃ mg/l | HCO ₃ mg/l | CL mg/l | NH ₃ mg/l | NO ₃ mg/l | PO ₄ mg/l |
|---------|------|-------------|-----------|------------|------------|-------------|------------|------------|-------------------------|--------------------------|------------|-------------------------|-------------------------|-------------------------|
| Shuni | 6.94 | 1149 | 40 | 32 | 3.8 | 18.2 | 116 | 88.8 | Nil | 24 | 8.5 | 1.0 | 0.8 | 0.64 |
| Tuntube | 6.09 | 217 | 4 | 6 | 2.6 | 16.8 | 880 | 54 | Nil | 40 | 4.0 | 1.6 | 2.0 | 0.60 |
| Wababe | 6.72 | 3.10 | 36 | 66 | 3.5 | 19.0 | 240 | 66.24 | Nil | 32 | 4.5 | 0.8 | 1.2 | 0.55 |
| Dange | 6.13 | 115.7 | 1 | 1 | 2.0 | 15.0 | 40 | 24 | Nil | 36 | 4.5 | 1.2 | 1.0 | 0.65 |
| Fajaldu | 6.99 | 100.9 | 1 | 1 | 2.3 | 16.5 | 30 | 33.6 | Nil | 40 | 5.5 | 1.6 | 1.8 | 0.80 |
| Rikina | 6.79 | 303 | 3 | 1 | 2.0 | 17.0 | 46 | 38.4 | Nil | 24 | 3.8 | 0.6 | 1.2 | 0.75 |

The results of the physicochemical analysis of borehole water shows that the PH value of all the water samples obtained from various locations/site (designated as SHN, TT, WBB, DNG, FJD, RKN) ranges from 6.09 to 6.94, Which is neutral, complying with the WHO standard water quality requirements (6.5 to 8.5). The pH values recorded in this study were slightly different from the results of Samuel *et al.* (2014) which had an average pH of 6.14. The slight difference might be because of different study site. Even though pH has no direct effect on human health it's indirect action on physiological process cannot be over

emphasized (Adenkuleet *al.*,2004; NSDWQ, 2007).

The conductivity of the water samples ranges from 1149 to 3.0 $\mu\text{S}/\text{cm}$. The higher conductivity was detected at Shuni Borehole water, while the lowest was detected at Rikina borehole site. Also the result showed that DO ranged from 3.8 to 2.0mg/L, while BOD levels of borehole water sample ranged from 18.2 to 15.0mg/L and the total sodium of water sample ranged from 32 to 1mg/L the higher sodium was detected at Shuni borehole water.

Table 2: Profile of Total bacterial count and Total coliform count obtained from borehole water

| Sample site | Total bacterial count (cfu/ml) | Combination of positive | coliform count (MPN/ml) |
|------------------|-------------------------------------|-------------------------|-------------------------|
| FJD | 2.9×10^5 | 2.2.0 | 21 |
| WBB | 1.5×10^5 | 2.3.0 | 29 |
| SHN | 2.5×10^5 | 3.2.0 | 93 |
| RKN | 1.2×10^5 | 2.2.1 | 28 |
| TT | 3.4×10^5 | 3.0.0 | 23 |
| DNG | 1.9×10^5 | 3.2.1 | 150 |
| WHO limit | 1.0×10^2 | | |

Key: TT=Tuntube-Tsefe, SHN=Shuni, RKN=Rikina, DNG=Dange, WBB=Wababe, FJD= Fajaldu

Table 3: Biochemical Characterization of the Isolates

| Sample | G.rxn | Shape | Ca | Co | Gl | Su | La | Ga | H2 | Ur | Ci | M | V | Mo | In | organism |
|--------|-------|-------|----|----|----|----|----|----|----|----|----|---|---|----|----|--------------------------|
| FJD | + | cocci | + | + | + | + | + | - | - | + | + | + | + | - | - | <i>S. aureus</i> |
| WBB | - | Rod | + | - | + | + | + | + | - | - | - | + | - | + | + | <i>E. coli</i> |
| SHN | - | Rod | + | - | - | - | - | + | - | - | + | - | - | + | - | <i>Pseudomonas</i> |
| RKN | + | Rod | + | - | + | + | - | - | - | - | + | - | + | + | - | <i>Bacillus subtilis</i> |
| TT | - | Rod | + | - | - | - | - | + | - | - | + | - | - | + | - | <i>Pseudomonas</i> |
| DNG | + | Rod | + | - | + | + | - | - | - | - | + | - | + | + | - | <i>Bacillus subtilis</i> |

KEY: FJD= Fajaldu, WBB= Wababe, SHN= Shuni, RKN=Rikina, TT= TuntubeTsefe, DNG= Dange, G.rxn= Gram Reaction, Cat= Catalase, Coa= Coagulase, Glu=Glucose, Suc= Sucrose, Lac= Lactose, H2s= Hydrogen Sulphide, Ure= Urease, Cit= Citrate, Mr= Methylred, Vp= Voges's Proskauer, Mot= Motility, Ind= Indole

TABLE 4: Percentage Frequency Occurrence Of The Bacterial Isolates

| Bacterial Isolates | FJD | WBB | SHN | RKN | TT | DNG | Percentage occurrence (%) |
|-------------------------------|-----|-----|-----|-----|----|-----|---------------------------|
| <i>Staphylococcus aureus</i> | 1 | 0 | 0 | 0 | 0 | 0 | 1(16.7) |
| <i>Escherichia coli</i> | 0 | 1 | 0 | 0 | 0 | 0 | 1(16.7) |
| <i>Pseudomonas aeruginosa</i> | 0 | 0 | 1 | 0 | 1 | 0 | 2(33.3) |

| | | | | | | | |
|--------------------------|----------|----------|----------|----------|----------|----------|---------------|
| <i>Bacillus subtilis</i> | 0 | 0 | 0 | 1 | 0 | 1 | 2(33.3) |
| Total | 1 | 1 | 1 | 1 | 1 | 1 | 6(100) |

Key:TT=Tuntube-Tsefe,SHN=Shuni,RKN=Rikina,DNG=Dange,WBB=Wababe,FJD=Fajaldu

Table 5: Macroscopic and Microscopic Characteristics of the Fungal Isolates

| Sample | Macroscopic characteristics | Microscopic characteristics | Fungi Identified |
|--------|---|--|--------------------------|
| FJD | Pin like green growth | Non-branched conidiophore with bulb end carries conidia | <i>Aspergillusflavus</i> |
| WBB | Large white colonies | A specie of yeast like fungus that is the perfect state of geotrichum, it form a real mycelium, cocci or ovoid ascopore. | <i>Endomyces</i> |
| SHN | Pin like black growth | Non-branched conidiophore with bulb end carries conidia like sun rays | <i>Aspergillusniger</i> |
| RKN | Cotton like white growth spotted with black color. | Sporangia contain spores | <i>Rhizopussp</i> |
| TT | Pin like green growth | Non-branched conidiophore with bulb end carries conidia | <i>Aspergillusflavus</i> |
| DNG | The texture is velvety to powdery from the front,the colour is initially white and then become rosy belge, pinkish, yellow or orange from the reverse it is tannish | Septatehypae, conidiophores,aleuriconida,arth roconidia and chlamydospores are observed,hyphae contain clamp connection (bridge)at the septa, conidiophores may be short or long, simple or branched | <i>Sporotrichum</i> |

Key: : FJD=Fajaldu, WBB= Wababe, SHN=Shuni,RKN=Rikina,TT=Tuntubetsefe,DNG=DangeShuni

Table 6: Frequency Distribution of the Fungal Isolates

| Isolates | Percentage Frequency (%) |
|--------------------------|--------------------------|
| <i>Aspergillusflavus</i> | 2(33.33) |
| <i>Endomyces</i> | 1(16.66) |
| <i>Aspergillusniger</i> | 2(33.33) |
| <i>Sporotrichum</i> | 1(16.66) |
| Total | 6(100) |

DISCUSSION

The study on physicochemical and microbiological quality of borehole water in

Dange/Shuni Local Government Area, Sokoto was performed. Water, including borehole water can be described as potable if it complies with

and meets certain physical, chemical and microbiological standards designed to ensure that a given water source is drinkable Watanabe,(2010). Potable water is essential for good health and the socioeconomic development of man (Nwachukwuet *al.*, 2013). Borehole water serves as the major source of drinking water in Nigeria, since only few Nigerians can actually afford treated bottled water for their daily water consumption (Nwachukwuet *al.*, 2013). Since borehole water sources could serve as sources via which waterborne pathogens could spread and cause waterborne infections in any given community, the physicochemical and bacteriological analysis of borehole water sources are vital and necessary to prevent the spread of disease-causing agents through them.

The results of the physicochemical analysis of borehole water shows that the PH value of all the water samples obtained from various locations/site (designated as SHN, TT, WBB, DNG, FJD, RKN) ranges from 6.79 to 6.94, Which is neutral, complying with the WHO standard water quality requirements (6.5 to 8.5). The PH values recorded in this study were slightly different from the results of Samuel *et al.* (2014) which had an average PH of 6.14. The slight difference might be because of different study site. Even though PH has no direct effect on human health it's indirect action on physiological process cannot be over emphasized (Adenkuleet *al.*,2004; NSDWQ, 2007).

The conductivity of the water samples ranges from 1149 to 303 μ S/cm. The higher conductivity was detected at Shuni Borehole water, while the lowest was detected at Rikinaborehole site. Although there is no disease or disorder associated with conductivity of drinking water (NSDWQ, 2007). The result showed that DO ranged from 3.8 to 2.0mg/L, while BOD levels of borehole water sample ranged from 18.2 to 17.0mg/L and the total sodium of water sample ranged from 32 to 1mg/L the higher sodium was detected at Shuni borehole water.

The total bacterial count of the isolates indicated that the sample of water from Tuntubetsefe had the highest bacterial count of 3.4×10^5 cfu/ml. while the lowest count was observed in the sample from Rikina borehole water site.

Coliform bacteria such as were observed in the water sample from Tuntube-Tsefe, Shuni, Rikina, Dange, Wababe and Fajaldu borehole water sources. The faecal count was also observed only in the sample obtained from Wababe borehole. The bacteria load for all the water samples analyzed in this study were generally high, and they exceeded the acceptable limit; which is the standard limit of coliform bacteria including heterotrophic bacteria in water bodies including borehole water (WHO, 2011; APHA, 2012). Conformation with microbiological standard is of special interest because of the capacity of water to spread waterborne diseases within a large human population (Quick, 2008). The highest bacteria count was observed from water samples from point A, while the least bacteria count which was observed in water samples from point D. This result agrees with the findings of Nwachukwuet *al.* (2013) that there are high counts of bacteria pathogens in most borehole water samples in some parts of Nigeria. In a related development, bacteria counts that were unacceptably high from several boreholes in Nigeria and elsewhere have been previously reported; and these studies showed similar results of high bacteria counts as obtained in this study (Obi and George, 2011; Palamuleni and Akoth, 2015). The major reasons that could be responsible for the high bacteria counts obtained from the various borehole water samples analyzed in this study could be as a result of human or animal faecal matter contamination resulting from surface water runoffs, and seepages from broken septic tanks. Effluents from abattoirs and seepage from sewage or septic tanks which are in close proximity to boreholes could also contribute to such contamination.

Bacterial species identified including *Pseudomonas aeruginosa*, *Bacillus subtilis* had the highest percentage frequency of 50% while the rest including *Staphylococcus aureus*, *Bacillus subtilis* and *Escherichia coli* had the lowest percentage distribution of 16.7%. The fungal species identified include *Aspergillus flavus*, *Aspergillus niger*, *Endomyces*, *Rhizopus* sp were *Aspergillus flavus* had the highest frequency occurrence (33.3%) while *Aspergillus niger*, *Endomyces* and *Rhizopus* had the lowest

percentage occurrence of 16.6%. The WHO guidelines for drinking water quality states that for all water directly intended for drinking, *E. coli* and thermo-tolerant coliform bacteria must not be detectable in any 100 ml of sample. The presence of *E. coli* in some of the water samples is an indication of faecal contamination, rendering the water unsafe for drinking. Water intended for human consumption should contain no faecal indicator organisms and the presence of *E. coli* in a drinking water sample always indicates recent microbial activity (Nwachukwuet *al.*, 2013). This study is in agreement with the findings of Adeiza *et al.* (2018) who identified *entrobacter* species including *Escherichia coli*. It's also in conformity with the finding of Onajita *et al.* (2018) who identified fungal species in borehole including *Rhizopus* sp and *Aspergillus niger*.

CONCLUSION

The results obtained from this study showed that the various water sources considered are contaminated with microorganisms including *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Bacillus subtilis*, *E.coli*, *Rhizopus* spp, *Endomyces*, *Aspergillus niger* and *sporotrichum*. The microbial loads observed in the borehole water were also beyond the prescribed limit for water quality, thus the water is unsuitable for drinking purposes.

Recommendations

- i. The borehole water should be effectively treated by disinfection or distillation so that bacteria and other harmful disease causing microorganism can be reduced or even completely eliminated
- ii. The construction of pit latrines near water sources should be avoided.
- iii. Public awareness on the dangers associated with the consumption of contaminated water should be increased
- iv. The communities should be educated on the need to keep their surroundings clean most especially around the boreholes.

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