

ANTIBIOGRAM PATTERN OF BACTERIA ISOLATES FROM GROUND WATER (BOREHOLE WATER) RESOURCES IN PORT HARCOURT, SOUTHERN NIGERIA

ABSTRACT

The study considered antibiogram of bacteria isolates from selected ground water (borehole water) resources susceptibility pattern of the isolated microorganisms in Port Harcourt during the dry and wet season of 2020. Seven (7) sampling locations designated as Abuloma, Borikiri, Eagle Island, Fimie, Macoba-Isaka, Rumuekini and Rumuokoro in Port Harcourt Metropolis were established. Water samples were collected from boreholes in 10 ml sterile sampler and placed in an ice-packed container and send to the laboratory for the analysis of *Vibrio*, *Staphylococcus*, Total Heterotrophic bacteria, *Salmonella*, *Shigella*, Total coliform and Fecal coliforms). Bacteria isolates were examined for the susceptibility and resistance of Ofloxacin, Augmentin, Cefazidime, Gentamicin, Cefixime, Ciproflaxin, Cefturoxime and Nitrofurantoin. There were observed increased rate of resistance antibiotics in this study which could be attributed to the widespread misuse of various antibiotics without control. The *E. coli* percentage resistance isolates from the water sources discovered that Ofloxacin, Ciproflaxin, Nitrofurantoin had no value in percentage resistance of the *E. coli* isolated. Augmentin had 50% (Abuloma), 100% (Rumuokoro, Rumuekini, Makoba-Island and Eagle Island). Cefazidime had 100% (Borikiri, Fimie, Abuloma, Rumuokoro, Rumuekini, Makoba-Island and Eagle Island) correspondingly. Gentamicin had 50% (Abuloma), 60% (Rumuekini), 100% (Rumuokoro, Makoba-Island and Eagle Island). Cefixime had 100% (Borikiri, Fimie, Abuloma, Rumuokoro, Rumuekini, Makoba-Island and Eagle Island) while Cefuroxime had 100% for (Borikiri, Fimie, Abuloma, Rumuokoro, Rumuekini, Makoba-Island and Eagle Island). The susceptibility in percentage of the *E. coli* isolated revealed that Ofloxacin had percentage value of 100% (Fimie, Rumuokoro, Rumuekini, Abuloma, Makoba-Isaka, Eagle Island and Borikiri) respectively. Augmentin and Gentamicin had 50% in Abuloma. There was no percentage value for Cefazidime, Cefixime and Cefuroxime while Ciproflaxin appeared as 100% (Fimie and Borikiri) and 50% (Rumuokoro and Abuloma) respectively. Nitrofurantoin had percentage values as 100% in Abuloma, Borikiri, Eagle Island, Fimie, Makoba-Isaka, Rumuekini and Rumuokoro correspondingly. Findings holds that the microbial counts of groundwater resources are moderately lower, intense commercial activities increase the rate of occurrence of microbial activities in water bodies, the high percentage resistance is a factor of indiscriminate waste disposal pattern and drug abuse of the citizenry. Thus, it is recommended that groundwater source (boreholes) be sited away from septic tanks source(s) as possible interference under the influence of proximity could become inevitable. More so, public health education on the dangers of drug abuse and indiscriminate waste disposal be encouraged.

Keyword: Groundwater, Antibiogram, Susceptibility, Resistance, Borehole sources

INTRODUCTION

Water is generally known to occupy over 70% of the earth crust. It is a fundamental requirement for life and this becomes inevitable for humans (Gideon *et al.*, 2017). Water is by far the most abundant substance in animal and plant tissues as well as the world around us. It accounts for

about 70% of the human body and it is higher in many growing plants (Odesina, 2003). Water is not only used all over the world in large quantities for drinking purposes, but it is used even in greater quantities for cooking, solvent in industrial processes, bleaching, raising steam to drive engines or turbines to generate electricity as well as washing (Bibiye, 2013 & Odesina, 2003). However, due to increases in human population, industrial and recreational activities, and agricultural activities among others the access to quality water consumption has become a teething problem to mostly in developing countries. However, having access to good and potable water has been an age-long challenge super imposed by mostly anthropogenic activities and less of nature mostly in Sub-Sara Africa.

Bacteria from different sources of water (human, animal and environmental) are able to interact and resistance evolves as a consequence of promiscuous exchange and shuffling of genes, genetic platforms and genetic vectors (Baquero *et al.*, 2008). The most important felt needs in public health requirement is water. However, the availability of safe water dictates the quality of life since water is one of the basic necessities for life. About 1.2 billion people worldwide lack access to safe drinking water (Wilkes *et al.*, 2009). The sub-Sahara Africa accounts for over 1/3 of the number, and is lagging behind in progress towards the MDG target, with only about 60% of the population using improved sources of drinking water (WHO, 2010). Water intended for human consumption must be free from organism and from concentration of chemical substances that may be a hazard to health. Infectious diseases posses' major public health challenge to man when water sources are mostly contaminated by *E. coli* (Feachem, 2001).

In public and environmental health, antimicrobial resistance is a threat. It is a characteristic of pathogen causing different diseases but generally not a problem of disease pathology rather one of the limited therapy options. These antibiotic resistance bacteria have been found in a surprising diverse range of environments including clinics, animal pens, orchards, food, sewage as well as chlorinated and un-chlorinated water supplies (Sammie *et al.*, 2012; Chopra *et al.*, 2001). Bacteria are common contaminants worldwide and the release of human and animal wastes in to the environment exacerbates bacterial contamination especially in aquatic setting. Increase resistance to antibiotics may pose great challenge for the effective treatment of bacterial infection. However, Biograms are used by clinicians to access local susceptibility rates, as an aid in selecting empiric antibiotic therapy. Anantibiogram shows the aggregate number of bacteria tested against antimicrobial and incorporates the extent of bacterial isolates vulnerable to every antimicrobial operator tested (Fridkin *et al.*, 2001). Plasmid analysis helps in the differentiating isolated microorganisms and is it useful in the epidemiologic investigation of enteric disease outbreak (Soumik *et al.*, 2010). Whereas antibiotics are metabolites having preferential antimicrobial activity; hence they are widely used for curing of human ailments caused by microorganisms. These antibiotics compounds are used either in their natural form or as semi-synthetic derivatives. The latter are usually produced by isolating the antibiotic nucleus and subjecting it to chemical modification according to Kemper (2007) and Wright (2007). The overuse of antibiotic in human and animals for treatment leads to the release of antibiotics and antibiotic resistant strains into the environment as stated by Ghafur *et al.*, (2010) and Silbergeld *et al.*, (2008). Antibiotic resistance is a natural phenomenon and represents an evolutionary response to the strong selective pressure resulting from these compounds (Ogbonna & Azuonwu., 2019; Chaturvedi *et al.*, 2008). In this scenario, an antibiotic may kill virtually all the bacteria causing a disease in a patient but a few bacteria that are genetically may survive. These go on to reproduce or to transfer their resistance to others of their species through the processes of gene exchange of cases.

According to Prescott *et al.*, 2001, the wide spread of water borne diseases as a result of contamination of water sources, concerns have been raised that the disease failed to be cured due to resistance to commonly prescribed antibiotics by the contaminating microorganisms originating from livestock excreta and human sewage. (Matthew *et al.*, 2007) reported that for the past years antibiotic resistant bacteria species are very ubiquitous in the environment and their negative impact has greatly increased drastically. The improper use of antibiotic and lack of awareness are considered as the most important factor for the emergence, selection, and dissemination of antibiotic resistant bacteria species in the environment (Neu, 1992; Abera *et al.*, 2013).

Several literatures have shown that inadequately treated sewage and wastes are mainly the sources of antibiotic resistant bacteria in the environment (Inversen *et al.*, 2002; Hanwood *et al.*, 2001). This is said to be largely due to the failure or neglecting regulatory agencies in controlling drug use and as such these drugs are usually obtained over the counter without the supervision of the prescription by a licensed medical officer of health (Sanders, 2005).

MATERIAL AND METHOD

Study Area

The study areas are fast growing sub-urban in Port Harcourt with geographical coordinates of Latitude 4° 49' 27.0012" N and Longitude 7° 2' 0.9996" E. It has 9 meters above sea level with a tropical climate. It has a significant rainfall pattern in most months of the year and a short dry season with little effect (Okafor, 1973; Demographia, 2016; www.wikipedia.com, 2010). There is temperature variation throughout the year.

Sample Collection

Samples were collected in seven (7) different boreholes with the aid of a sterilized 10mls plastic containers haven rinsed these containers with the sample. The containers were placed under the running tap and were Stoppard while the tap was still running. Samples were thus placed in an ice-packed cooler and were transported to the microbiology laboratory of Rivers State University for microbial analysis.

Chart 1: Sample Location (Areas)

Materials

All glass wires were sterilized in a hot box oven at 160°C for 1hr. Samples were serially diluted with normal saline. Media for selection of microorganisms' isolates were SS agar, N agar, EM

S/N	Sample Location	GPS	Sample Water Source	Sample Codes
1	Abuloma	4.7432°N 7.0821°E	Ground water (Borehole)	AB
2	Borikiri	4.7463°N 7.0364°E	Ground water (Borehole)	BO
3	Eagle Island	4.8667°N 6.9833°E	Ground water (Borehole)	EI
4	Fimie	4.7829°N 6.9773°E	Ground water (Borehole)	FI
5	Macoba-Isaka	4.8873°N 6.9415°E	Ground water(Borehole)	MI
6	Rumuekini	4.7878°N 7.0415°E	Ground water (Borehole)	RK
7	Rumuokoro	4.4424°N 6.5915°E	Ground water (Borehole)	RO

agar, MacConkey agar, and Mannitol salt agar and TCBS agar. 0.1ml aliquot was used, Samples were inoculated. The inocula incubated at 37°C for 2hrs and the special at 45°C for 48hrs. Pour plate technique for culture medium and nutrient agar were used. The nutrient's agar was prepared according to the manufacturer's instruction and allowed to cool to 45°C.

Microbiology Test Analysis

Colonies were sub-cultured to obtain pure isolates which were then characterized by gram's staining for the identification and characterization of bacteria. Biochemical tests (Voges Proskauer, Methyl red, Citrate, Catalase, Coagulase, Oxidase, Motility and Sugar fermentation tests) were respectively carried out. Identity of the isolates was matched with Bergery's Manual of Determinative Bacteriology (BMD) for Confirmation (Holt *et al.*, 2002).

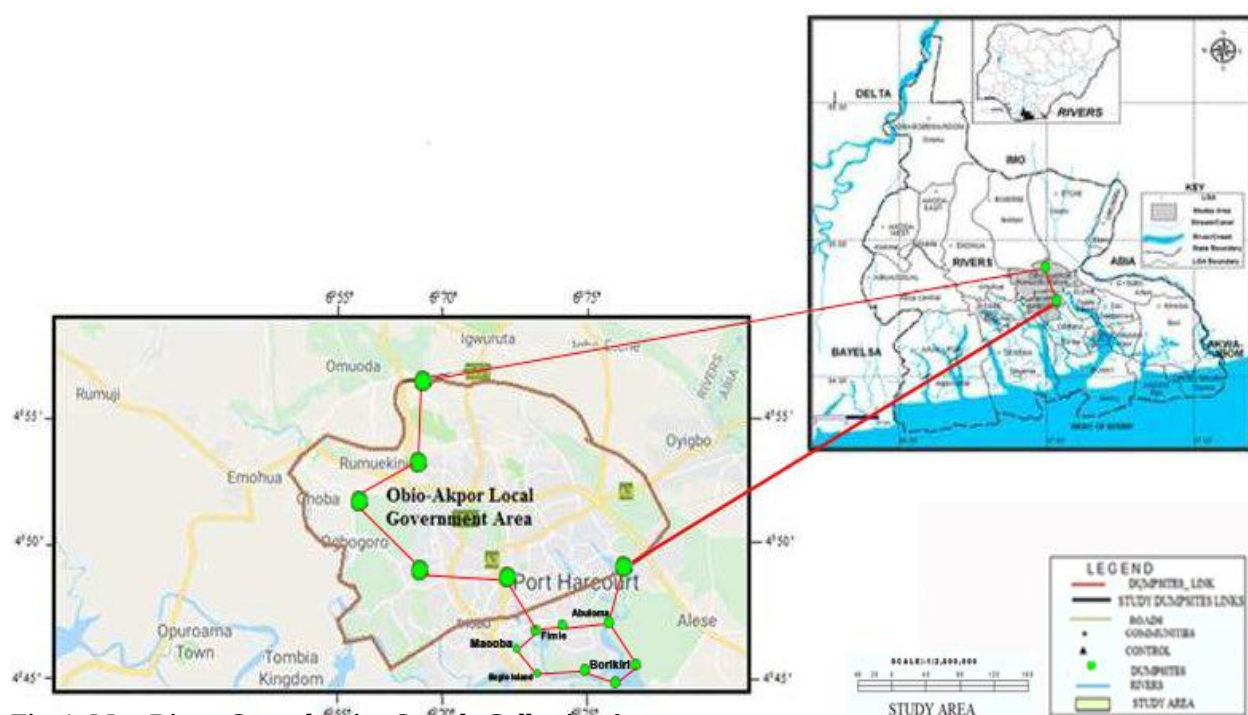


Fig. 1: Map Rivers State showing Sample Collection Area

Bacteriology Test Analysis

TBC, TCC and other microorganisms were enumerated, serial dilution and specific dilution into clean and sterilized Nutrient agar plate method was carried out using 0.1ml dilution of 10^{-1} to 10^{-2} of bacteria suspension, and inoculated plate were incubated at 37°C for 28-28hrs at home temperature. Bacteria colonies of the plate were counted randomly and purified by sub-culturing into fresh agar plate using the Streak technique.

Antibiotic Sensitivity Test Analysis

The antibiotic sensitivity test was performed by the Kirby-Bauer technique. Discs were commercially prepared for Gram-positive and Gram-negative organisms. The isolate from a fresh culture was cultured for about 18-24 hours in peptone water was prepared according to the manufacturers formula. Mueller-Hinton Agar was prepared in line with manufacturer's formula. 25 ml of the medium was inoculated in each plate and was allowed to set and solidify. A turbid suspension of the isolates was made in distilled using 0.5 McFarland Standard and prepared as a comparator. A sterile swab was dipped into the bacteria suspension, pressed on the side the sides of the bottle to allow excess drip off, then used to evenly Streak the entire surface of the Mueller-Hinton agar plates. The plates were incubated at 37°C for 24 hours. Zone of

inhibition was measured in millimeters, recorded after incubation period and interpreted was grouped using the criteria of the National Committee on Clinical Laboratory Standard (NCCLS) of the World Health Organizations.

RESULT

Microbial Count of Groundwater Sources

In Abuloma, Heterotrophic Bacteria Count, Total Coliform Count and Feacal Coliform Count during the dry season had mean 0.65×10^6 cfu/ml, 5.53×10^4 cfu/ml and 1.55×10^4 cfu/ml while their wet season values were 1.8×10^6 cfu/ml, 7.25×10^4 cfu/ml and 2.55×10^4 cfu/ml respectively. At Borikiri, Heterotrophic Bacteria Count, Total Coliform Count and Feacal Coliform Count during the dry season had mean values of 0.65×10^4 cfu/ml, 0 cfu/ml and 0 cfu/ml with a corresponding mean value of 1.55×10^4 cfu/ml, 0.34×10^4 cfu/ml and 0 cfu/ml during the wet season. Also at Eagle Island, Total Heterotrophic Count, Total Coliform Count and Feacal Coliform Count during the dry season had mean values of 10.25×10^6 cfu/ml, 8.25×10^4 cfu/ml and 3.4×10^4 cfu/ml and a wet season value of 11.75×10^4 cfu/ml, 0.9×10^4 cfu/ml and 5.2×10^4 cfu/ml respectively. The mean value for Fimie and Macoba-Isaka for Total Heterotrophic Count, Total Coliform Count and Feacal Coliform Count during the dry season was 0.2×10^6 cfu/ml, 0 cfu/ml, and 0 cfu/ml; 0.6×10^4 cfu/ml, 0.5×10^4 cfu/ml and 0 cfu/ml with their corresponding wet season mean values as 0.8×10^4 cfu/ml, 0.3×10^4 cfu/ml and 0 cfu/ml; 1.35×10^4 cfu/ml, 0.6×10^4 cfu/ml and 0.2×10^4 cfu/ml respectively. Rumuekini and Rumuokoro had a separately dry season mean value of 4.8×10^4 cfu/ml and 1.25×10^4 cfu/ml for Total Heterotrophic Bacteria Count dry, 4.25×10^6 cfu/ml and 0 cfu/ml for Total Coliform Count, 2.4×10^4 cfu/ml and 0 cfu/ml for Feacal Coliform Count their respective wet season mean values were 9.2×10^4 cfu/ml and 2.75×10^4 cfu/ml for Total Heterotrophic Bacteria Count, 4.9×10^4 cfu/ml and 0.2×10^4 cfu/ml for Total Coliform Count and 3.7×10^4 cfu/ml and 0 cfu/ml for Feacal Coliform Count (Table 1, Fig. 1). Total Coliform Count dry season and Feacal Coliform Count dry and wet seasons were significant at $P < 0.05$ for Borikiri, Fimie and Rumuokoro while at Macoba-Isaka, there was significance in FCC during the dry season at $P < 0.05$ (Table 1).

Table 1: Variation of Bacteriological Parameters of Borehole Water across sampling Locations

Sample Location	THB		TCC		FCC		t test (p-value)
	Dry Season	Wet Season	Dry Season	Wet Season	Dry Season	Wet Season	
ABULOMA	0.65×10^6	1.8×10^6	5.35×10^4	7.25×10^4	1.55×10^4	2.55×10^4	0.05
BORIKIRI	0.6×10^6	1.55×10^6	0	0.3×10^4	0	0	0.05
EAGLE ISLAND	10.25×10^6	11.75×10^6	8.25×10^4	0.9×10^4	3.4×10^4	5.2×10^4	0.05
FIMIE	0.2×10^6	0.8×10^6	0	0.3×10^4	0	0	-
MACOBA-ISAKA	0.6×10^6	1.35×10^6	0.5×10^4	0.6×10^4	0	0.2×10^4	-
RUMUEKINI	4.8×10^6	9.2×10^6	4.25×10^4	4.9×10^4	2.4×10^4	3.7×10^4	-
RUMUOKORO	1.25×10^6	2.75×10^6	0	0.2×10^4	0	0	-

TBH=Total heterotrophic bacteria, TCC=Total coliform bacteria, FCC=Feacal coliform count.

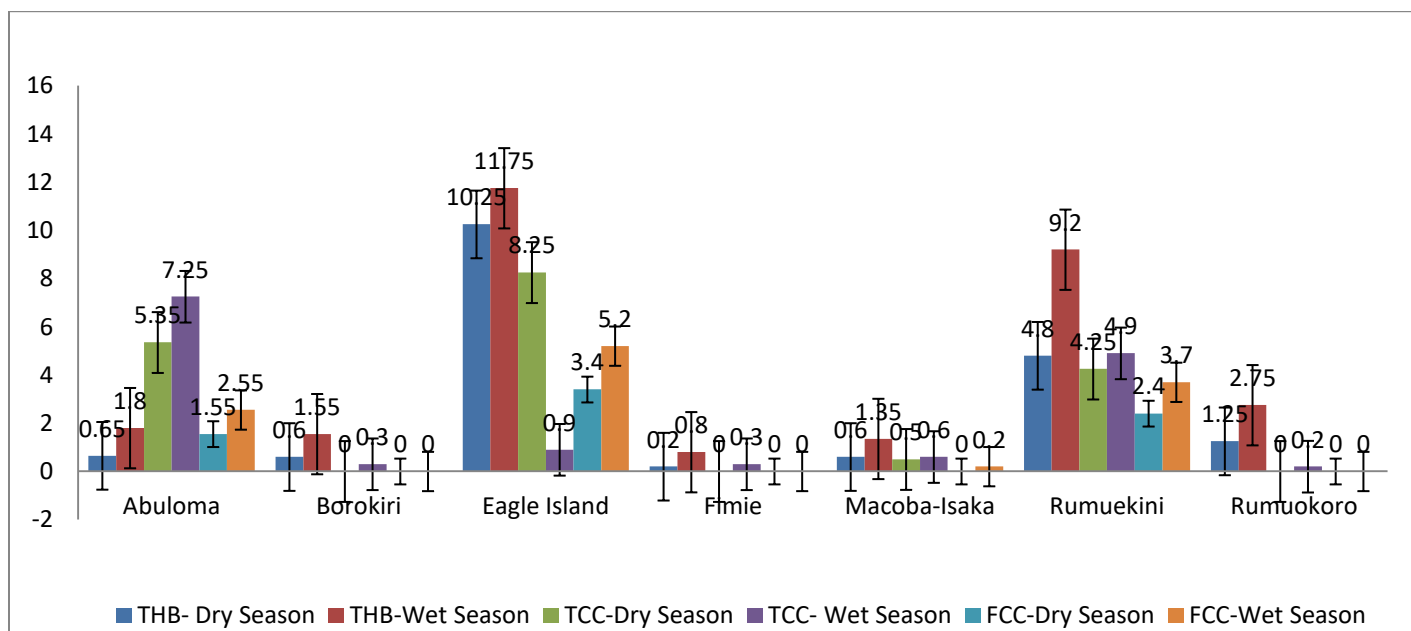


Fig. 2: Variation Plot of Bacteriological Parameters of Borehole Water across Sampling Locations

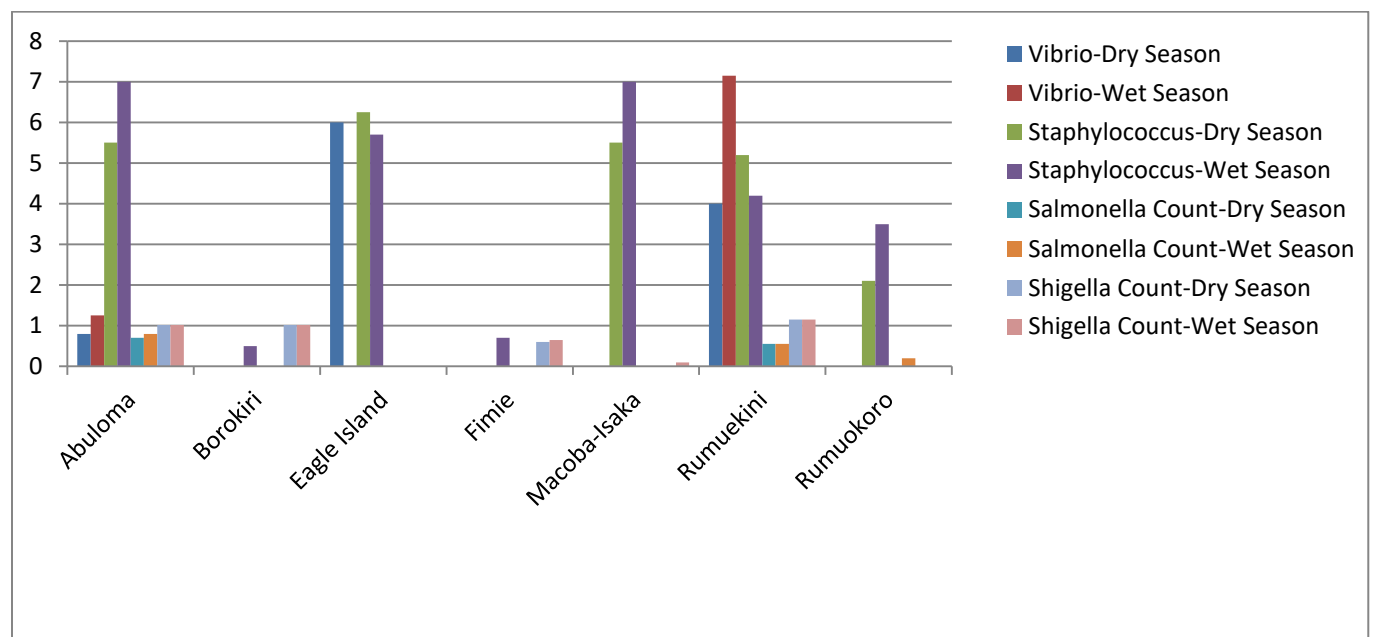
In Abuloma, Borikiri, Fimie and Eagle Island the mean *Vibrio* count during the dry season were (0.8×10^4) cfu/ml, (0)cfu/ml, (6×10^4) cfu/ml, and (0) cfu/ml. Their respective wet season count were 1.25×10^4 cfu/ml, 0cfu/ml, 0 cfu/ml and 0cfu/ml respectively. The dry season mean value of Macoba-Isaka, Rumuekini and Rumuokoro were 0cfu/ml, 4×10^4 cfu/ml and 0cfu/ml with a corresponding wet season values of 0cfu/ml, 0cfu/ml, 7.15×10^4 cfu/ml and 0cfu/ml respectively (Table 2). Abuloma, Borikiri and Eagle Island showed mean count during the dry season for *Staphylococcus* was 5.5×10^4 cfu/ml, 0cfu/ml and 6.25×10^4 respectively. Fimie, Macoba -Isaka, Rumuekini and Rumuokoro had mean values for *Staphylococcus* count varies as 0cfu/ml, 5.5×10^4 cfu/ml, 5.2×10^4 and 2.1×10^4 during the dry season. For the wet season, mean value for *Salmonella* Count in Abuloma, Borikiri and Eagle Island were 7×10^4 cfu/ml, 0.5×10^4 cfu/ml and 5.7×10^4 cfu/ml while Fimie, Macoba-Isaka, Rumuekini and Rumuokoro mean values for the wet season varies from 0.7×10^4 cfu/ml, 7×10^4 , 4.2×10^4 and 3.5×10^4 respectively (Table 2).

The mean count for *Salmonella* during the dry season for Abuloma, Borikiri and Eagle Island were 0.7×10^4 cfu/ml, 0cfu/ml and 0cfu/ml while the mean *Salmonella* counts for Fimie, Macoba-Isaka, Rumuekini and Rumuokoro mean during dry season were 0cfu/ml, 0cfu/ml, 0.55×10^4 and 0cfu/ml respectively. More so, for the wet season, Abuloma, Borikiri and Eagle Island had their mean values of 0.8×10^4 cfu/ml, 0cfu/ml and 0cfu/ml while for Fimie, Macoba-Isaka, Rumuekini and Rumuokoro, the mean values were from 0cfu/ml, 0cfu/ml, 0cfu/ml, 0.55×10^4 cfu/ml and 0.2×10^4 cfu/ml respectively. *Shigella* Count for Abuloma, Borikiri and Eagle Island mean value for dry season ranged from 1.02×10^4 cfu/ml, 1.02×10^4 cfu/ml and 0cfu/ml respectively for Fimie, Macoba-Isaka, Rumuekini and Rumuokoro mean value for these locations during the dry season varied from 0.6×10^4 cfu/ml, 0cfu/ml, 1.15×10^4 and 0cfu/ml respectively. For mean values for *Shigella* count during the wet season or Abuloma, Borikiri, Eagle Island were 1.02×10^4 cfu/ml, 1.02×10^4 cfu/ml, and 0cfu/ml, However, Fimie, Macoba-Isaka, Rumuekini and Rumuokoro location ranged from 0.65×10^4 cfu/ml, 0.1×10^4 cfu/ml, 1.15×10^4 and 0cfu/ml respectively (Table 2.; Fig. 2).

Table 2. Variation of Microbial Parameters of Borehole Water across Sampling Locations.

S L	Vibrio Count		Staphylococcus		Salmonella Count		Shigella Count		t test (p value)
	Dry Season	Wet Season	Dry Season	Wet Season	Dry Season	Wet Season	Dry Season	Wet Season	
AB	0.8×10^4	1.25×10^4	5.5×10^4	7×10^4	0.7×10^4	0.8×10^4	1.02×10^4	1.02×10^4	-
BO	0	0	0	0.5×10^4	0	0	1.02×10^4	1.02×10^4	-
EI	6×10^4	0	6.25×10^4	5.7×10^4	0	0	0	0	0.05
FI	0	0	0	0.7×10^4	0	0	0.6×10^4	0.65×10^4	0.05
MI	0	0	5.5×10^4	7×10^4	0	0	0	0.1×10^4	-
RU	4×10^4	7.15×10^4	5.2×10^4	4.2×10^4	0.55×10^4	0.55×10^4	1.15×10^4	1.15×10^4	0.05
RK	0	0	2.1×10^4	3.5×10^4	0	0.2×10^4	0	0	-

AB=Abuloma,BO=Borikiri, EI=Eagle Island, FI=Fimie, MI=Macoba-Isaka,RU=Rumuekini, RK=Rumuokoro,
SL=Sample Location.

**Figure 3: Variation Plot of Microbial Parameters of Borehole Water across Sampling Locations**

Distribution of Bacteria Isolates in Groundwater across Sampling Locations

The distribution and the percentage occurrence of bacterial isolates in groundwater showed wide variations. The present of *staphylococcus* sp. was observed with 65% in Eagle Island, 56% (Rumuokoro), 25% (Abuloma), 50% (Makoba-Isaka) and 30% (Rumuekini). It was absent in Fimie and Borikiri locations. *Micrococcus* sp. was present in Eagle Island with 15% occurrence, 15% (Rumuokoro) and 5% (Abuloma). No occurrence of this species in Fimie, Borikiri, Isaka and Rumuekini. *Shigella* sp. occurred in Fimie with 30%, 3% in Abuloma and 10% in Rumuekini while there was no occurrence in Eagle Island, Rumoukoro, Borikiri and Isaka. *Proteus* sp. was present in Eagle Island 20% occurrence, 30% (Rumuokoro), 10% (Fimie), 25% (Borikiri), 5% in Abuloma and 10% (Isaka). No occurrence in Rumuekini. *E. Coli* occurred only in Abuloma and Rumuekini with 30% and 16% respectively. It was absent in Eagle Island, Rumuokoro, Fimie, Borikiri and Isaka (Table 2). The presence of *Enterobacter* sp. occurred only in Abuloma with 7% occurrence. All other locations had no occurrences. *Citrobacter* sp. was

present in Abuloma and Rumuekini locations having 5% and 4% separately. It was absent in other locations. *Pseudomonas* sp was present in Abuloma only with 7% occurrence and was absent in other locations. However, *Klebsiella* sp, *Salmonella* sp., *Bacillus* sp., and *Vibrio* sp. were absent in Eaagle Island and Rumuokoro locations. *Klebsiella* sp.was still absent inFimie, Borikiri, Abuloma and Rumuekini but was present in Isaka with 30% occurrence. *Salmonella* sp. was further absent in Borikiri and Isaka but was present in Fimie (10%), Abuloma (5%) and Rumuekini (10%). *Bacillus* sp.was further absent in Isaka and was present in other locations with 50% (Fimie), 75% (Borikiri), 3% (Abuloma) and 10% Rumekini. Furthermore, *Vibrio* sp. was also absent in Fimie and Borikiri but was present in other locations with percentage occurrence as 5% (Abuloma), 10% (Macoba-Isaka) and 20% (Rumuekini) while *Serratia* sp.was present in Eagle Island and Rumuokoro 5% each but was absent in other locations. The column-wise summation of the percentage occurrence of the isolates for the seven sampling locations defined one hundred per cent (100%) signifying a perfect enumeration (Table 2.).

Table 3: Distribution and Percentage Occurrence of Bacterial Isolates in Groundwater across the Seven Sampling Locations

Isolates	Eagle Island		Rumuokoro		Fimie		Borikiri		Abuloma		Isaka		Rumuekini	
	O	%	O	%	O	%	O	%	O	%	O	%	O	%
<i>Staph. sp.</i>	+	65	+	56	-	0	-	0	+	25	+	50	+	30
<i>Micro. sp.</i>	+	15	+	15	-	0	-	0	+	5	-	0	-	0
<i>Shig. sp.</i>	-	0	-	0	+	30	-	0	+	3	-	0	+	10
<i>Prot. sp.</i>	+	20	+	30	+	10	+	25	+	5	+	10	-	0
<i>E. coli</i>	-	0	-	0	-	0	-	0	+	30	-	0	+	16
<i>Enter. sp.</i>	-	0	-	0	-	0	-	0	+	7	-	0	-	0
<i>Citro. sp.</i>	-	0	-	0	-	0	-	0	+	5	-	0	+	4
<i>Pseu. sp.</i>	-	0	-	0	-	0	-	0	+	7	-	0	-	0
<i>Klebs. sp.</i>	-	0	-	0	-	0	-	0	-	0	+	30	-	0
<i>Salm. sp.</i>	-	0	-	0	+	10	-	0	+	5	-	0	+	10
<i>Bacil. sp.</i>	-	0	-	0	+	50	+	75	+	3	-	0	+	10
<i>Vibrio sp.</i>	-	0	-	0	-	0	-	0	+	5	+	10	+	20
<i>Serr. sp.</i>	+	5	+	5	-	0	-	0	-	0	-	0	-	0
Total	100		100		100		100		100		100		100	

O = Occurrence, % = Percentage of Occurrence, - = Absent, + = Present, *Staph. sp.*=*Staphylococcus* sp., *Micro. sp.*= *Micrococcus* sp, *Shig. sp.*=*Shigella* sp., *Prot. sp.*=*Proteus* sp., *E. Coli*=*E.coli*, *Enter. sp.*=*Enterobacter* sp., *Citro. sp.*=*Citrobacter* sp.=*Pseu. sp.*= *Pseudomonas* sp., *Klebs. Sp.*=*Klebsiella* sp., *Salm. sp.*=*Salmonella* sp., *Bacil. Sp.*=*Bacillus* sp., *Vibrio sp.*, =*Vibrio* sp., *Serr. Sp.*=*Serratia* sp.

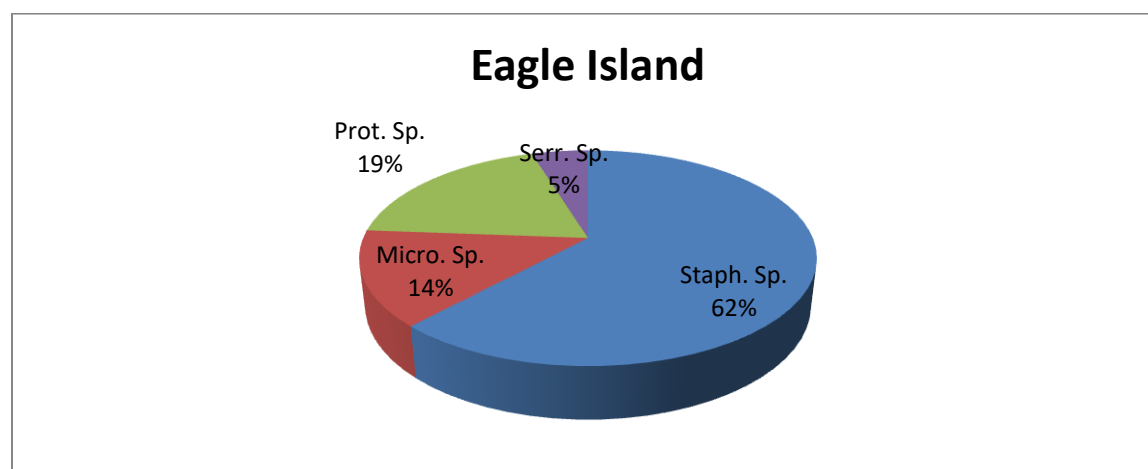


Image 1. Percentage (%) Resistance of *E. coli* isolated from water sources

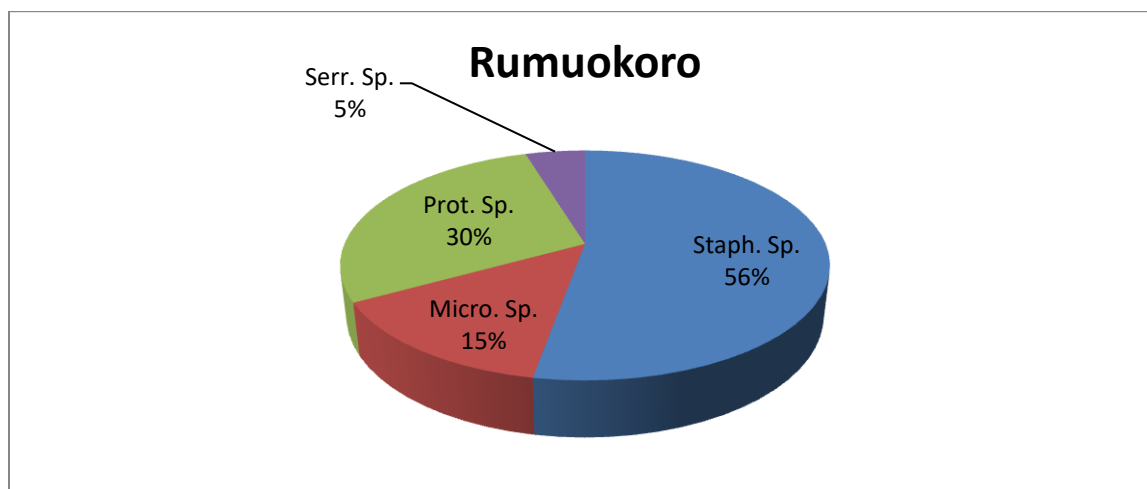


Image 2. Percentage (%) Resistance of *E. coli* isolated from water sources

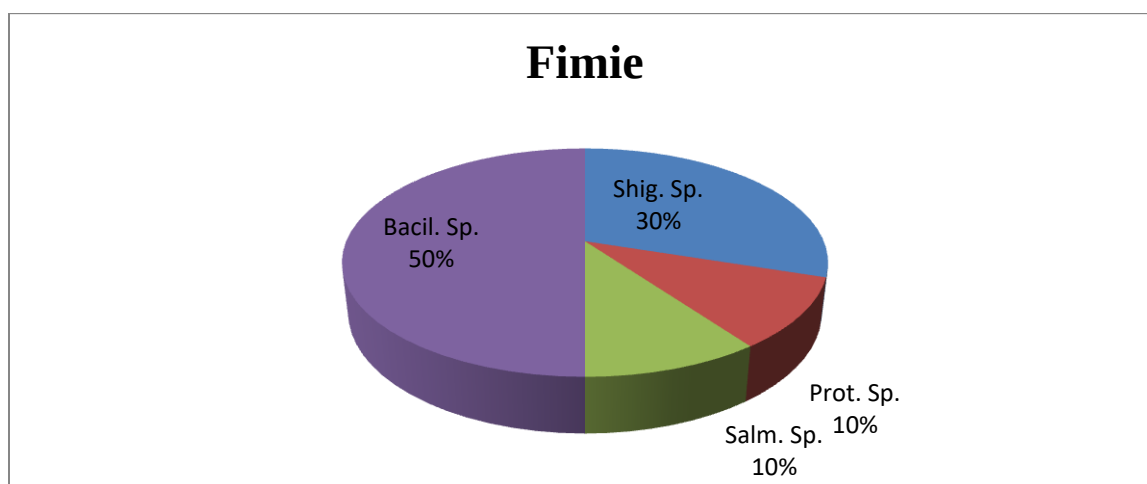


Image 3. Percentage (%) Resistance of *E. coli* isolated from water sources

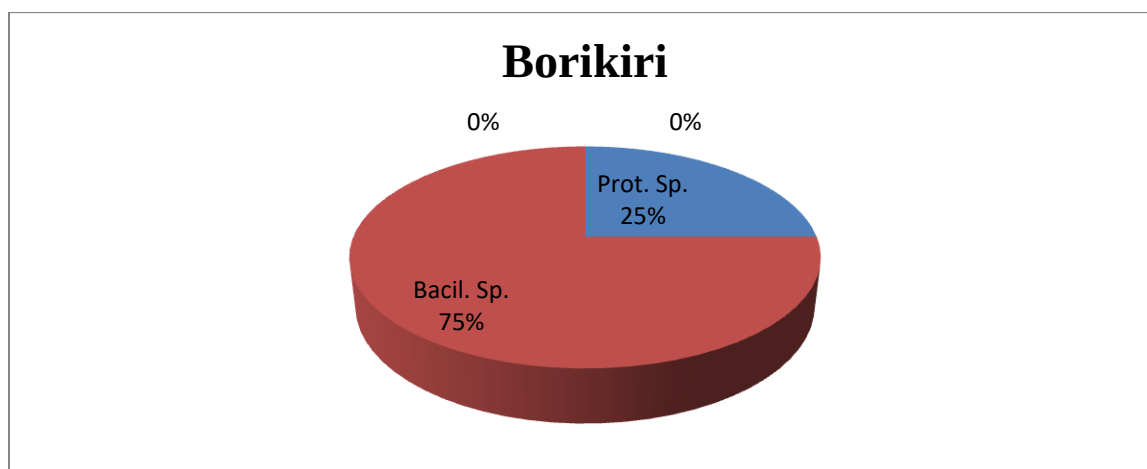


Image 4. Percentage (%) Resistance of *E. coli* isolated from water sources

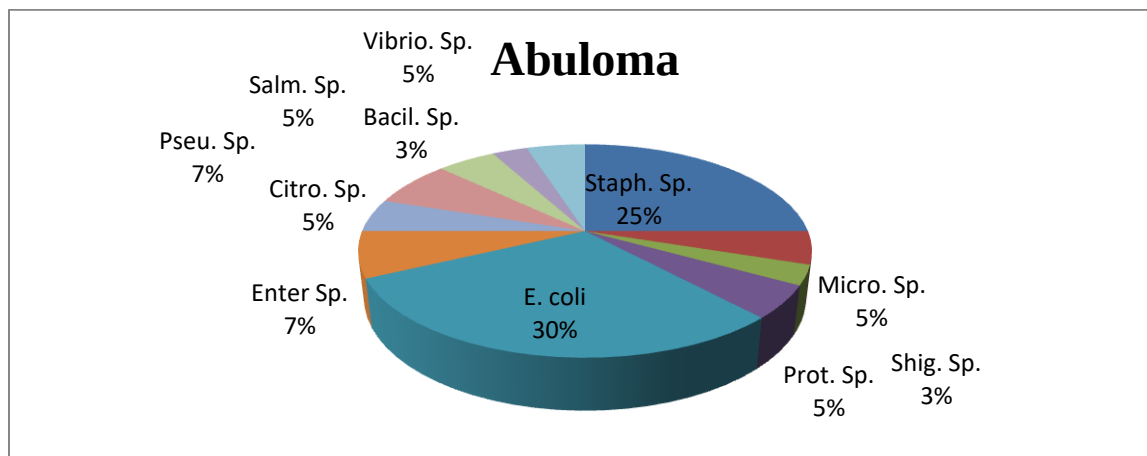


Image 5. Percentage (%) Resistance of *E. coli* isolated from water sources

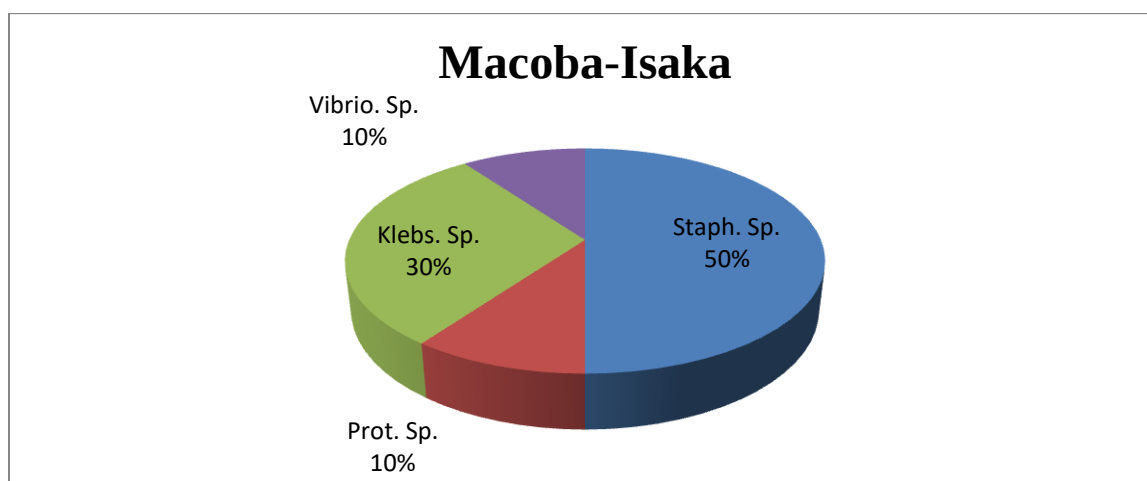


Image 6. Percentage (%) Resistance of *E. coli* isolated from water sources

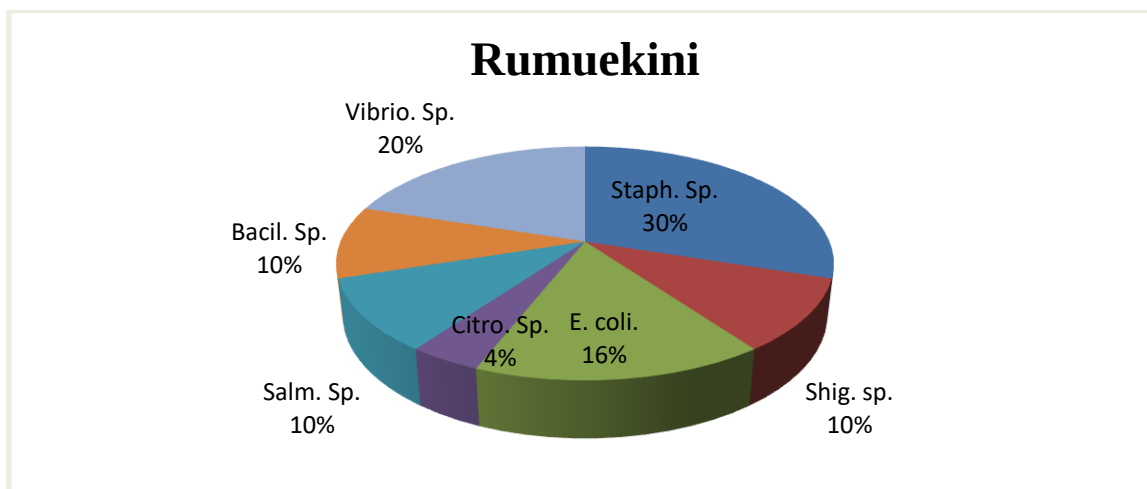


Image 7. Percentage (%) Resistance of *E. coli* isolated from water sources

The *E. coli* percentage resistance isolates from the water sources discovered that Ofloxacin (**OFL**), Ciproflaxin (**CPR**), Nitrofurantoin (**NIT**) had no value in percentage resistance of the *E. coli* isolated. Augmentin (**AUG**) had 50% (Abuloma), 100% (Rumuokoro, Rumuekini, Makoba-Island and Eagle Island). Ceftazidime (**CAZ**) had 100% (Borikiri, Fimie, Abuloma, Rumuokoro, Rumuekini, Makoba-Island and Eagle Island) correspondingly. Gentamicin (**GEN**) had 50% (Abuloma), 60% (Rumuekini), 100% (Rumuokoro, Makoba-Island and Eagle Island). Cefixime (**CXM**) had 100% (Borikiri, Fimie, Abuloma, Rumuokoro, Rumuekini, Makoba-Island and Eagle Island) while Cefuroxime (**CRM**) had 100% for (Borikiri, Fimie, Abuloma, Rumuokoro, Rumuekini, Makoba-Island and Eagle Island) respectively (Fig. 4).

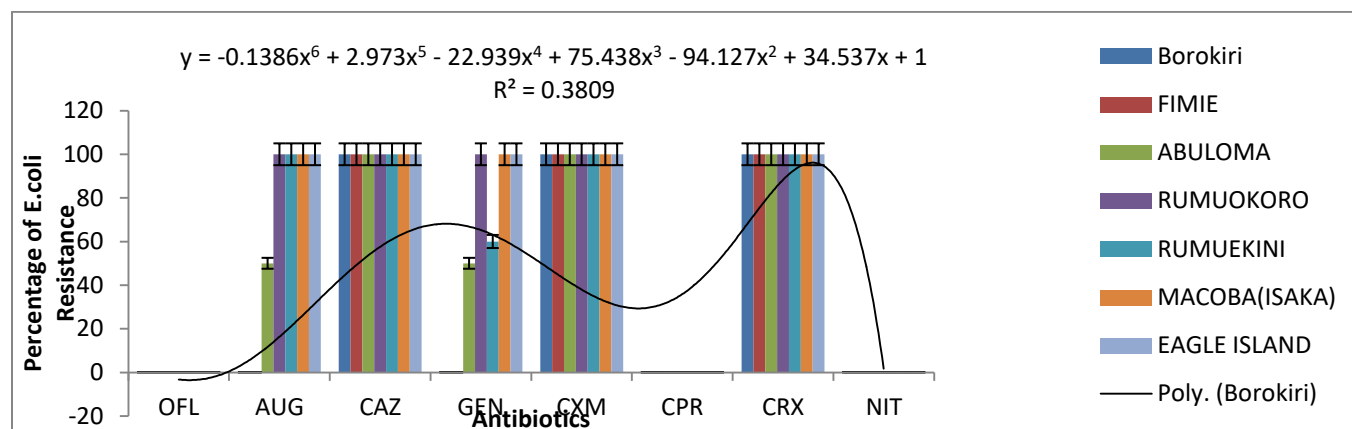


Fig.4. Percentage (%) of Resistance of *E. coli* isolated from seven locations to various commonly use Antibiotics

Percentage (%) of Susceptibility of *E. coli* isolated from water sources

The susceptibility in percentage of the *E. coli* isolated from the seven water sources under investigation revealed that Ofloxacin had percentage value as 100% (Fimie, Rumuokoro, Rumuoekini, Abuloma, Makoba-Isaka, Eagle Island and Borikiri respectively). Augmentin had 50% in Abuloma. There was no percentage value for Cetazidime, Cefixime and Cefuroxime. Gentamicin appeared 50% (Abuloma) and 40% (Rumuekini) while Ciproflaxin appeared as 100% (Fimie and Borikiri) and 50% (Rumuokoro and Abuloma) respectively. Nitrofurantoin had percentage values as 100% for Abuloma, Borikiri, Eagle Island, Fimie, Makoba-Isaka, Rumuekini and Rumuokoro correspondingly (Fig. 5).

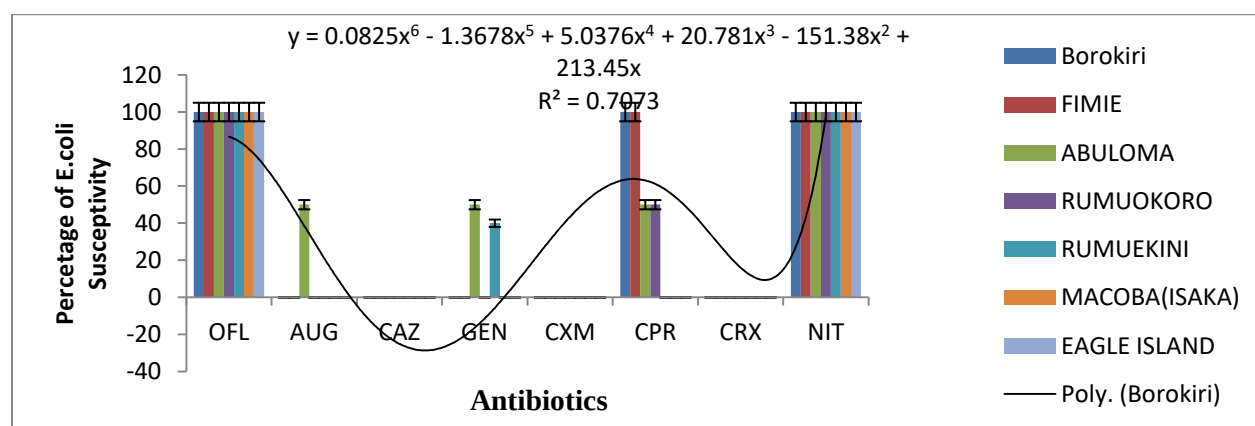


Figure 5 Susceptibility (%) of *E. coli* isolated from seven locations to various commonly use Antibiotics. (OFL)=Ofloxacin, (AUG) =Augmentin, (CAZ)=Cetazidime, (GEN)Gentamicin, (CXM)=Cefiximine, (CPR)=Ciproflaxin, (CRX)=Cefuroxime, (NIT)=Nitrofurantoin.

DISCUSSION

Chemotherapeutic from antimicrobial have been adjudged to exercise some health benefit to humans. The microbial counts of groundwater resources in this study are far less than the values obtained by Hao-chang *et al.*, 2014 in China and Nain *et al.*, 2015 in Delhi, India. Elsewhere in Abakili South-east Nigeria, values obtained by Iroha *et al.*, 2013 were also higher than the values obtain in this study. The observed low values in this study could be attributed to very low precipitation which inhibits dilution rate during the study period. The occurrences of *Escherichia coli* were observed in Abuloma and Rumuoekini while there were no observed occurrences in the other five sample stations. However, these occurrences in Abuuloma and Rumuoekini were far less than that obtained by Hao-chang *et al.*, 2014 in China; Onuoha (2015) and that of IWA, 2011 both in Nigeria. These observed differences could be attributed to intense commercial activity on going in China than the current study in Nigeria. The bacteriological properties such as THB, TCC and FCC values in the current study were moderate and comparable to that obtained by IWA (2011) in Nigeria except TCC and FCC in Rumuekini and Eagle–Island whose values were very much higher. The observed higher values could be linked with the different commercial activities, terrain factor, and waste disposal factors including transportation. The distribution and percentage occurrence of bacterial isolates in groundwater across the seven sampling locations revealed that *Staphylococcus* in Eagle-Island, Rumuokoro, Macoba-Isaka and Rumuekini were much higher than the other sampling stations. At Fimie, *Shigella* and *Bacillus* species were high while at Macoba-Isaka, *Klebsiella* value was also high. These high values were in line with the study carried out by Cooke (1976) and Falodun and Adekanmbi (2016).

The percentage resistance isolates of *E coli* revealed further that ofloxacin, ciproflaxin and nitrofurantoin had no percentage resistance. Augmentin and Gentamycin all in Abuloma had 50% resistance while at Rumuokoro, Macoba-Isaka and Eagle-Island it had 100% resistance. Cefuroxime also had 100% resistance in all the seven sample location. Values obtain in this study were far higher than those obtained by Patoli *et al.*, 2010 in Karachi, Pakistan. The reason for this high value resistance in this study as compared with Patoli *et al.*, 2010 could be attributed to heavily contamination with potentially pathogenic multidrug resistance strains of *E coli*. This is line with the research carried out by Bello *et al.*, 2013 in Akure, Ondo State, Nigeria. The percentage susceptibility of *E coli* was found to be 100% in Abuloma, Borikiri, Eagle Island, Fimie, Makoba-Isaka, Rumuoekini, and Rumuokoro for Ofloxacin whereas Augmentin and Gentamycin had 50% each in Abuloma. Ciproflaxin had 100% susceptibility in Fimie and Borikiri while Nitrofurantoin also had 100% susceptibility in all the sampling location. The result obtains in this study varied significantly with that obtained by Ogu *et al.*, 2017 in Delta State, Nigeria. While in Delta State, the susceptibility values were moderately low as against very high values in Port Harcourt. Very high anthropogenic activities (e.g. indiscriminate disposal of waste, drug abuse etc.) including weather condition may have contributed to this variability. This corroborate with the works of Ogu *et al.*, 2017; Nain *et al.*, 2015 and Hao-Chang *et al.*, 2014.

SUMMARY OF FINDINGS

1. The microbial counts of groundwater resources in this study are moderately lower than those obtain by Hao-Chang *et al.*, 2014.
2. Intense commercial activities increase the rate of occurrence of microbial activities in water bodies.
3. The high percentage resistance is a factor of indiscriminate waste disposal pattern and drug abuse of the citizenry.
4. Anthropogenic and weather factors underpin the high percentage susceptibility in this study.

CONCLUSION

Antibiogram shows the aggregate number of bacteria tested against antimicrobial and incorporates the extent of bacterial isolates vulnerable to every antimicrobial operator tested. In the current study, Ofloxacin, Nitrofurantoin and Cefixmine were most apparently susceptible antibiotics among others while Augmentin, Ceftazidime, Gentamycin, Ciproflaxin were observed to be most resistance antibiotic for the *Escherichia coli* from the groundwater sampled. Gentamycin appeared as intermediates in Abuloma and Rumuekinki which could also mean non susceptibility by the bacteria possibly at forthcoming time to this antibacterial agent. The increased rate of resistance antibiotics in this study could be attributed to widespread misuse of various antibiotics without control. Consequently, it could be recommended that groundwater source (boreholes) be treated before it should be acceptable for domestic use. Furthermore, boreholes should be sited away from septic tanks source as possible interference under the influence of proximity becomes inevitable.

RECOMMENDATION

1. All water for domestic and human consumption should be treated.
2. There should be public education and enlightenment campaign on the dangers associated with drug abuse and indiscriminate waste disposal.

REFERENCE

- Baquero, F. Martinez, J.L and Canton, R. (2008). Antibiotics and antibiotic resistance in water environments. *Curr Opin Biotechnol*, 19 (260-265).
- Bibiye, Alaye A.S. (2013). Introduction to environmental to pollution and control, Nybraide Publisher & Co., Second Edition, (8-13; 26-32).
- Chaturvedi, S., Chandra, R. and Rai, V. (2008). Multiple antibiotics resistance patterns of rhizospheric bacteria isolated from *Phragmites australis* growing in constructed wetland for distillery effluent treatment, *J Environmental Biology*, 29(1): pp (117-24).
- European Centre for Disease Prevention and Control (ECDC). Factsheet for experts, (2013).
- Feachem, R.G (2001). Drinking Water Quality, Environmental Health Engineering in Tropics 2nd Edition, University Press, London, PP 78-92.
- Gideon, I.O., Inamul H.M, Alexander, A.O., and Iftikhar. A.T., (2017). Antibiotic Susceptibility Profile of Bacteria Isolated from Drinking Water Resources in Amai Kingdom, Delta State, Nigeria. *Annual Research and Review in Biology* 14 (1); 1-9, 2017, Article no. ARRB. 34326 ISSN; 2347-565X, NLM ID;101632869.
- Kummer, R.N., Solanki, R. and Kumar, N. J. I. (2013) An Assessment of Seasonal Variation and water quality Index of Sabarmati River and Kharicut Canal at Ahmedabad, Gujarat. *Electronic Journal of Environmental, Agricultural and Food Chemistry*, 53 (3), 161-163.
- Matthew, A.G., Robin, C and Liamthong, S., (2007). Food-borne pathogens and diseases, *Am. J. Microbial, Clin. Pathol.*, 4(2): 115-133.
- McManus P.S., Stockwell V.O. (2001). Antibiotic use for plant disease management in the United States. *Plant Health Progress* doi 10.1094/PHP-2001-0327-01-RV

- Neu, H.C., (1992). The Crisis antibiotic resistance, *Am. J. Sci.*, 257:1064-1073.
- Prescott, M. Harley, P. and Klein, A. (2001) *Microbiology* 5th edition. McGraw- Hill Book Co. New York.
- Samie A., Makonto T. E., Odiyo J., Ouaboi-Egbenni P.O., Mojapelo P. and Bessong P.O., (2011). Microbial quality, diversity and antibiotic susceptibility profiles of bacterial isolates from borehole water used by schools in greater Giyani Municipality, Mopani District, South Africa. *Afr. J. Microbiol.Res.* 2011;5 (3);198-210.
- Soumik, B., Sohini, C., Goutam, C., Thandarararaya, R., Swapan, K.N., Ranajit, K., and Hemanta, K., (2010). Plasmid mediated streptomycin and sulfamethoxazole resistance in *Shigella flexneri* 3a. *International journal of Antimicrobial Agents*, 36, 348-351.
- Silbergeld E.K., Graham J., Price L.B. (2008). Industrial food animal production, antimicrobial resistance, and human health. *Annu Rev. Publ. Health* 29:151-169.
- Waters, A.E., Contente-Cuomo, t., Buchhagen, J., Liu, C.M., Watson, L., Pearce, K., Foster, J.T., Bowers, J., Driebe, E.M., Engelthaler, D.M., Keim, P.S. and Price, L.M. (2011). Multidrug-resistant *Staphylococcus aureus* in US meat and poultry. *Clinical Infectious Diseases*, 52(10), 1227-1230.
- World Health Organization (WHO). Progress on Sanitation and Drinking-Water: 2010 Update Report; World Health Organization (WHO): Geneva, Switzerland, 2010.
- Wright G.D (2007) The antibiotic resistance nexus of chemical and genetic diversity nature reviews Microbiology 5, 175-186.