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ANTIBIOTIC SUSCEPTIBILITY PROFILE AND CHARACTERIZATION OF BACTERIA CONTAMINANTS FROM WELL WATER IN SABON TASHA KADUNA METROPOLIS, KADUNA

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ABSTRACT

Background: About 1.2 billion people worldwide lack access to safe drinking water. Diseases related to contamination of drinking water constitute a major burden on human health. Well water which is a common source of water in many Nigerian homes used for domestic activities is prone to bacteria contamination.

Aim: This study was aimed at isolating pathogenic bacteria present in well water and determining the susceptibility profile of the isolates with the highest occurrence, to commonly prescribed antibiotics, in Kaduna, Nigeria.

Methods: A total of 120 samples of well water were collected, Gram staining, and biochemical tests were carried out to identify the isolates. Antimicrobial susceptibility testing was done using the Kirby-Bauer Agar disc diffusion method.

Results: A total of 36 pathogenic bacteria isolates were obtained with a total colony count range of 1.0 x 10^5-6.2 x 10^5 cfu/ml. The following bacteria were isolated: *Escherichia coli (30.6%), Pseudomonas spp (22.2%), Proteus spp (11.1%), Staphylococcus spp (11.1%), Salmonella spp (8.3%), Klebsiella spp (8.3%), Paracoccus spp (5.6%), and Citrobacter spp (2.8%).* The result of antibiotic susceptibility screening of the highest occurring isolate, *E.coli*, showed that the isolates were susceptible to Ceftriazone (100%), Vancomycin (60%), Gentamicin (50%), and resistant to Streptomycin (100%), Penicillin G (100%), Chloramphenicol (80%), Ofloxacin (80%), Ciprofloxacin (60%), Cefoxitin (60%), Erythromycin (60%).

Conclusion: This study showed that well water in Sabon Tasha, Kaduna, is highly contaminated with antibiotic resistant bacteria, this can be a source of infection to the users and a potential means of transmitting multidrug resistant bacteria strains in the community.

Keywords: Well water, bacteria contaminants, antibiotic resistance.

INTRODUCTION

In most developing countries (including Nigeria), access to portable water has become a mirage and exploitation of groundwater through the construction of hand-dug wells is a major source of drinking water for majority of the populace (1). Access to safe drinking water is essential to health, as it serve as a powerful biological and environmental determinant of health (2). A satisfactory (adequate, safe and accessible) supply must be available to all. Diseases related to contamination of drinking water constitute a major burden on human health, interventions to improve the quality of drinking water provides significant health benefits (2). Water is one of the most important felt needs in public health and the availability of safe

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water dictates the quality of life since water is a basic requirement of life. About 1.2 billion people worldwide lack access to safe drinking water (3).

Despite the considerable investments of Nigerian government in water supply programme, over 52% of its population have no access to portable water (4). Water covers 70.9% of the earth's surface and is vital for all known forms of life (5). The World Health Organization asserted that open or poorly covered well heads pose the commonest risk to well-water quality; the possibility of the water being contaminated is further increased by the use of inappropriate water-lifting devices by consumers (23). The commonest physical defects leading to faecal contamination of dug wells are associated with damage to, or lack of, a concrete plinth, and with breaks in the parapet wall and in the drainage channel (6). The most serious source of pollution of well water is contamination by human waste from latrines and septic tanks resulting in increased levels of microorganisms, including pathogens (7). The impact of good quality drinking and domestic use water cannot be overemphasized. According to World Health Organization guidelines, quality drinking water must not contain *Escherichia coli* or thermotolerant coliform bacteria, giardia worms, viruses, *Cryptosporidium spp*, *Legionella pneumophila*, *Entamoeba hystolitical* and other opportunistic pathogens such as *Clostridium sp., Klebsiella sp.* and *Pseudomonas* (8). The presence of theses microorganisms could cause different disease conditions and clinical signs such as *giardiasis*, *cryptosporidiosis*, gastroenteritis, diarrhea, typhoid fever, cholera, bacillary dysentery, hepatitis, shigellosis etc (9). Water borne diseases are reported to account for 80% of illnesses in developing world, killing a child every 8 seconds. This is a global public health threat (10). Finding solutions to the different disease conditions caused by bacteria present in well water have been challenged by the current waves of resistant strains, it's therefore, necessary to investigate the quality and antibiotic sensitivity patterns of bacteria isolates from well water. Only a few studies on bacterial contamination of Well Water in North-Western Nigeria has been published; especially the antibiotic susceptibility of such bacteria to antibiotics. This study was therefore aimed at isolating bacteria contaminants from Well Water in Sabon Tasha, Kaduna, and to determine the antibiotic susceptibility of the highest occurring bacteria contaminant to commonly prescribed antibiotics.

MATERIALS AND METHODS

Study Area

The study area is Sabon-Tasha in Kaduna town, the ancient capital of Northern Nigeria and capital of present Kaduna State, located in the northern part of Nigeria. Kaduna is about 162 km away from the Federal Capital Territory (FCT) Abuja and 234 km away from Kano, the capital of Kano state. Sabon-Tasha is located in the southern part of Kaduna Metropolis, on latitude 10.44565N and longitude 7.4565N.

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Sample Collection

Forty different source locations of well water that gives the entire representation of the well water in Sabon-Tasha, Kaduna were randomly selected for the purpose of sample collection. Samples of the well water were aseptically collected in triplicate making a total of one hundred and twenty samples using a sterile 250mls screw capped bottle. The bottle, with the cap removed, was lowered into the well to fetch the water using a clean robe tied to the bottle. A piece of stone was also tied to the bottle to aid immersion of the bottle under water. The process was such that the bottle was not allowed to touch the bottom of the Well as well as the walls of the Well to avoid contamination from these sources. After fetching the water, the mouth of the bottle was wiped with a sterile swab and capped. The samples were labelled appropriately, placed into separate plastic bags and transported in an ice packed cooler to the Pharmaceutical Microbiology laboratory, Faculty of Pharmaceutical Sciences, Kaduna State University, Kaduna. The forty locations (Wells) where samples were collected are shown in Table 1 below.

Table 1: Sample Collection

S/No	Wells	Well type	Sample Code	Number of Samples
1	No. 1 Ogbodo Street U/Maigero, Kaduna State	Un-Cov.	A	A1 A2 A3
2	No 4, Ogbodo Street U/Maigero Kaduna State.	Un-Cov.	B	B1 B2 B3
3	No 1 Patience Nwodo Street U/ Maigero.	Un-Cov.	C	C1 C2 C3
4	No 2, Patience Nwodo Street U/Maigero.	Covered	D	D1 D2 D3
5	No. 1 Sokoto Street U/Maigero, Kaduna State	Un-Cov.	E	E1 E2 E3
6	No. 3 Sokoto Street U/Maigero Kaduna State	Un-Cov.	F	F1 F2 F3
7	No. 5 Delta Street U/Maigero , Kaduna State.	Covered	G	G1 G2 G3
8	No.10 Christ the King Close U/Maigero	Covered	H	H1 H2 H3
9	No. 8 Engr. Richard Obukofe Street Kaduna State	Covered	I	I1 I2 I3
10	No.13 , Nicholas Mamman Street Kaduna State	Un-Cov.	J	J1 J2 J3
11	No. 5 Jacob Street U/Maigero, Kaduna State	Covered	K	K1 K2 K3
12	Dangi Son Street U/ Maigero, Kaduna State	Covered	L	L1 L2 L3
13	No. 7 Dattawa Street U/Maigero, Kaduna State.	Covered	M	M1 M2 M3
14	Joseph Mathew Street U/ Maigero Kaduna State	Un-Cov.	N	N1 N2 N3
15	Near NNPC Water Intake U/Maigero Kaduna State	Un-Cov.	O	O1 O2 O3
16	No 35, Delta Street U/Maigero Kaduna State	Un-Cov.	P	P1 P2 P3
17	No. 35, Delta Street U/Maigero Kaduna State	Covered	Q	Q1 Q2 Q3
18	No.10 Jacob Street, Kaduna State.	Covered	R	R1 R2 R3
19	No. 2 Sokoto Street U/Maigero, Kaduna State.	Un-Cov.	S	S1 S2 S3
20	No. 9 Joseph Matthew Street U/Maigero,	Covered	T	T1 T2 T3
21	JJ 13, Marwa Street, Sabon Tasha, Kaduna State	Un-Covered	U	U1 U2 U3

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22	No 49, Zittistreet,U/ Galadima,Sabon-Tasha	Covered	V	V1 V2 V3
23	No 18, U/ Kadara,Sabon-Tasha, Kaduna State.	Un-Covered	W	W1 W2 W3
24	No 30, Romi Road, U/ Barde, Sabon-Tasha	Covered	X	X1 X2 X3
25	No 21, U/ Matari,Sabon-Tasha, Kaduna State.	Covered	Y	Y1 Y2 Y3
26	No 11, U/ Dodo, Sabon-Tasha, Kaduna State.	Un-Covered	Z	Z1Z2Z3
27	No 7, U/ Gimbiya, Sabon-Tasha, Kaduna State.	Covered	a	a1a2a3
28	No 28, Sen. Aziz Street, G.R.A, Sabon-Tasha	Covered	b	b1b2b3
29	No 15 U/ Bulus, Sabon-Tasha, Kaduna State.	Un-Covered	c	c1c2c3
30	Near St. Mary's Catholic church, U/Boro, No 30, Sabon-Tasha, Kaduna State.	Covered	d	d1d2d3
31	No 35, Patrick's Street, U/ BoroSabon-Tasha	Un-Covered	e	e1e2e3
32	No 5, Alkali Street, U/ Pama, Sabon Tasha, Kaduna State.	Covered	f	f1f2f3
33	No 29, Doka street, Court Road, Sabon-Tasha, Kaduna State.	Covered	g	g1g2g3
34	No 51, Tsuani Kura, Sabon Tasha, Kaduna.	Covered	h	h1h2h3
35	No 12, U/ Biso, Sabon-Tasha, Kaduna State.	Un-Covered	i	i1i2i3
36	JJ 20 Marwa Street, Sabon Tasha, Kaduna State	Un-Covered	j	j1j2j3
37	No 35, U/ Biso, Sabon-Tasha, Kaduna State.	Un-Covered	k	k1k2k3
38	No 8, U/ Kadara, Sabon-Tasha, Kaduna State.	Covered	l	l1l2l3
39	No 20, Zittistreet,U/ Galadima, Sabon-Tasha	Covered	m	m1m2m3
40	JJ 26, Marwa Street, Sabon Tasha, Kaduna State.	Covered	n	n1n2n3

Determination of Aerobic Plate Count and Mean Colony Count

Standard Plate Count Method was used to determine the total aerobic colony count of the samples (11). A five-fold serial dilution of each sample was made and plated out on an agar plate using spread plate technique. The plates were incubated at 37°C for 24 h. The average bacteria loads of the well water samples obtained from the different locations were expressed as Colony Forming Units per millilitre (CFU/mL) of well water (Table 2).

$$\text{Mean colony count (CFU/ml)} = \frac{\boldsymbol{Mean\ plate\ count\ (n)}}{\boldsymbol{Dilution}(\mathbf{10^{-3}})\times\boldsymbol{sample\ size}(\mathbf{10^{-1}})}$$

Preparation of Samples

Serial dilutions of samples were made up to 10^{-5} in distilled water. This was done by dispensing 1ml of each sample into a test tube containing 9ml of distilled water; this will make the content 10ml and give a dilution of 10^{-1}. The process was repeated until a 10^{-5} was obtained. Then 1ml of each 10^{-3} and 10^{-4} dilution (from every sample) was dispensed using sterile syringe into appropriately labelled petri dishes. About 20ml of Nutrient agar was dispensed into the plates containing the diluted samples. The plates were gently stirred so as to mix the media and sample; this was then allowed to gel. The plates were inverted and incubated at 37°C for 24 hours.

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Purification, Gram Staining and Identification of Isolates

Each bacteria colony was isolated in a pure form by sub culturing in a fresh Nutrient agar plate and pure cultures were temporarily preserved on nutrient agar slants at 4°C for further work. An overnight activated culture was then used to determine colony morphology, gram staining and biochemical tests (Table 3). The following biochemical tests were carried according to Chesbrough (12) to identify the isolates: indole test, catalase test, citrate utilization test, methyl red, Voges Proskauer test and oxidase test.

Growth on Selective Media

The isolates were further identified by growing them on selective media, among which were: eosin methylene blue, MacConkey agar, mannitol agar, and salmonella-shigella agar (Table 4)

Antibiotic Susceptibility Test

The highest occurring isolate *E. coli* was screened for antibiotic susceptibility using the Kirby-Bauer disc diffusion method. A suspension of each isolate was prepared in sterile normal saline. The isolate was standardised by making a turbid suspension of each isolate in sterile normal saline, this was compared with 0.5 Mcfarland standard. A sterile swab was dipped into the standardized bacteria suspension (1.5 x 10^8cfu/ml), pressed on the side of the bottle to allow excess drip-off, and then used to evenly streak the entire surface of the Mueller-Hinton agar (Oxford Basingstoke, England) plate in three directions, rotating the plate in approximately 60^0 to ensure even distribution. With the petri dish lid in place, the surface of the agar was allowed to dry for about 3-5 minutes. Sterile forceps were then used to place the antibiotic discs in a circular pattern on the agar plate, the plate was allowed for 30 minutes pre-diffusion time and thereafter incubated at 37°C for 16-18 hours. This procedure was carried out for all the isolates. After incubation, the zone of inhibition in diameter for each antibiotic was measured and interpreted as sensitive, intermediate and resistant according to Clinical and Laboratory Standard Institute (13) guidelines. The following antibiotic discs were tested: Penicillin 10 µg, Ceftriazone 30µg, Vancomycin 30µg, Chloramphenicol (30 µg), Ciprofloxacin (5 µg), Gentamicin (10 µg), cefoxitin (30 µg), Ofloxacin (30 µg), Streptomycin (10 µg), and ceftriaxone (30 µg). The basis for the selection of these antibiotics is due to the fact that they form part of the commonly prescribed agents in this environment.

Determination of Multiple Antibiotics Resistant Index (MARI) and Multidrug Resistance (MDR)

The MARI for each isolate was determined by dividing the number of antibiotics to which the test isolate was resistant to, by the total number of antibiotics tested (14). The isolates that showed resistance to three or more classes of antibiotics were classified as multidrug resistant

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RESULTS

In this study, the mean colony counts per well water sampled in Sabon Tasha, Kaduna was 3.27 x 10^5 CFU/ml with a ranges between 1.0 x 10^5 to 6.2 x 10^5 CFU/ml (Table 2). The well water at No. 1 Ogbodo Street U/Maigero, Kaduna State had the highest bacteria count (6.2 x 10^5 CFU/ml) while JJ 13, Marwa Street, Sabon Tasha and No 8, U/ Kadara, Sabon-Tasha had the least (1 x 10^5).

Table 2: Mean Colony Count (MCC)

S/N	Samples	MCC (×10^5cfu/ml)	S/N	Samples	MCC (×10^5cfu/ml)
1	A	6.2	21	U	1.0
2	B	2.1	22	V	2.0
3	C	5.8	23	W	1.8
4	D	5.3	24	X	3.4
5	E	3.1	25	Y	2.1
6	F	2.9	26	Z	5.1
7	G	4.7	27	a	2.8
8	H	2.6	28	b	3.4
9	I	1.2	29	c	3.7
10	J	5.1	30	d	2.2
11	K	3.2	31	E	2.5
12	L	4.8	32	F	3.3
13	M	2.8	33	G	4.4
14	N	1.9	34	H	2.9
15	O	2.1	35	I	3.1
16	P	5.4	36	J	2.8
17	Q	5.8	37	K	2.0
18	R	2.7	38	L	1.0
19	S	2.8	39	M	3.3
20	T	4.4	40	N	3.1

In the process of transporting the samples from sites to the laboratory, 4 samples were contaminated and were discarded; remaining a total of 36 samples. Biochemical analysis and morphological examination of the colonies on various media showed that *E. coli*, *Pseudomonas* spp., *Staphylococcus aureus*, *Proteus* spp., *Salmonella* spp were possible contaminants of well water in the area sampled (Tables 3 & 4).

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Table 3: Biochemical Tests

S/N	Samples	TSI			Ct	Ca	Co	Ox	Mr	Vp	In	Suspected organism
		G.U	G.P	H_2S								
1	A_1	+	+	-	-	+	-	-	+	-	+	*Escherichia coli*
2	A_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
3	A_1	-	-	-	-	+	+	-	+	+	-	*Staphylococcus aureus*
4	A_3	+	+	-	+	+	-	-	+	-	-	*Klebsiella* spp.
5	C_1	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
6	D_1	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
7	F_3	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
8	G_1	-	-	-	-	+	+	-	+	+	-	*Staphylococcus aureus*
9	J_3	+	-	+	-	+	+	-	+	+	-	*Salmonella* spp.
10	K_2	+	+	+	+	+	-	-	+	-	-	*Proteus* spp.
11	L_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
12	L_3	-	-	-	-	+	+	-	+	+	-	*Escherichia coli*
13	K_1	+	+	+	+	+	-	-	+	-	-	*Citrobacter* spp.
14	N_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
15	O_1	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
16	Q_2	+	-	+	-	+	+	-	+	+	-	*Salmonella typhi.*
17	R_2	+	+	-	+	+	-	-	+	-	-	*Klebsiella* spp.
18	P_1	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
19	S_1	+	+	-	-	+	-	-	+	-	+	*Escherichia coli*
20	S_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
21	S_1	-	-	-	-	+	+	-	+	+	-	*Staphylococcus aureus*
22	S_3	+	+	-	+	+	-	-	+	-	-	*Klebsiella* spp.
23	T_1	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
24	U_1	+	+	-	+	+	-	+	-	-	-	*Pseudomonas* spp.
25	V_3	-	-	-	+	+	-	+	-	-	-	*Paracoccus.*
26	W_1	-	-	-	-	+	+	-	+	+	-	*Staphylococcus aureus*
27	X_3	+	-	+	-	+	+	-	+	+	-	*Salmonella* spp.
28	a_2	+	+	+	+	+	-	-	+	-	-	*Proteus* spp.
29	a_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
30	b_3	-	-	-	-	+	+	-	+	+	-	*Paracoccus*
31	c_1	+	+	+	+	+	-	-	+	-	-	*Citrobacter* spp.
32	d_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
33	g_1	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*
34	h_2	+	+	+	+	+	-	-	+	-	-	*Prosteus spp.*
35	j_1	+	+	+	+	+	-	-	+	-	-	*Prosteus spp*
36	k_2	+	+	-	+	+	-	-	+	-	+	*Escherichia coli*

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Key: += positive, - = negative, IN=indole, Ca= catalase, Ct=citrate, H2S= hydrogen sulphide, Mr= methyl red, Vp= Voges Proskauer, G=gas, S=sugar, Ox=oxidase, Co= coagulase, G. U= glucose utilisation, G. P=Gas production

Table 4: Cultural Characteristics of Isolates on Selective Media

S/No	Samples	EMB	MAC	MANN	SSA	Presumptive Organism
1	A_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
2	A_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
3	A_1	Less dark	Pale pink	Yellow	Pink	*Staphylococcus aureus*
4	A_3	Less dark	Pink	Pink	Pink	*Klebsiella* spp.
5	C_1	Less dark	Green	Yellow	Black	*Pseudomonas* spp,
6	D_1	Less dark	Green	Yellow	Black	*Pseudomonas* spp.
7	F_3	Less dark	Green	Yellow	Black	*Pseudomonas* spp.
8	G_1	Less dark	Pale pink	Yellow	Pink	*Staphylococcus aureus*
9	J_3	Less dark	Colourless	Yellow	Black	*Salmonella typhi.*
10	K_2	Less dark	Colourless	No growth	Black	*Proteus* spp.
11	L_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
12	L_3	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
13	K_1	Blue-black	Pink	Yellow	Black	*Citrobacter* spp.
14	N_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
15	O_1	Less dark	Green	Yellow	Black	*Pseudomonas* spp.
16	Q_2	Less dark	Colourless	Yellow	Black	*Salmonella typhi.*
17	R_2	Less dark	Pink	Pink	Pink	*Klebsiella* spp.
18	P_1	Less dark	Green	Yellow	Black	*Pseudomonas* spp.
19	S_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
20	S_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
21	S_1	Less dark	Pale pink	Yellow	Pink	*Staphylococcus aureus*
22	S_3	Less dark	Pink	Pink	Pink	*Klebsiellai* spp.
23	T_1	Less dark	Green	Yellow	Black	*Pseudomonas* spp,
24	U_1	Less dark	Green	Yellow	Black	*Pseudomonas* spp.
25	V_3	Less dark	Pale pink	Yellow	Pink	*Paracoccus* spp.
26	W_1	Less dark	Pale pink	Yellow	Pink	*Staphylococcus aureus*
27	X_3	Less dark	Colorless	Yellow	Black	*Salmonella typhi.*
28	a_2	Less dark	Colourless	No growth	Black	*Proteus* spp.
29	a_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
30	b_3	Less dark	Pale Pink	Yellow	Pink	*Paracoccus spp*
31	c_1	Blue-black	Pink	Yellow	Black	*Citrobacter* spp.

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32	d_1	Blue-black	Pink	Yellow	Pink	*Escherichia coli*
33	g_1	Blue-Black	Pink	Yellow	Pink	*Escherichia coli*.
34	h_2	Less dark	Colorless	No growth	Black	*Prosteus spp.*
35	j1	Less dark	Colorless	No growth	Black	*Prosteus spp*
36	k2	Blue Black	Pink	Yellow	Pink	*Escherichia coli*

Key: EMB. = Eosin methylene blue, MAC. = MacConkey agar, MANN. = Mannitol agar, SSA. = Salmonella-Shigella agar EMB: Blue-black= positive, Less dark = negative

In this study, contaminants isolated include *E. coli (11), Pseudomonas* spp. (7), *Proteus* spp. (4), *Staphylococcus* spp. (4), *Salmonella* spp. (3), *Klebsiella* spp. (3), *Paracoccus* spp. (2) and *Citrobacter* (2) as shown in Figure 1 below.

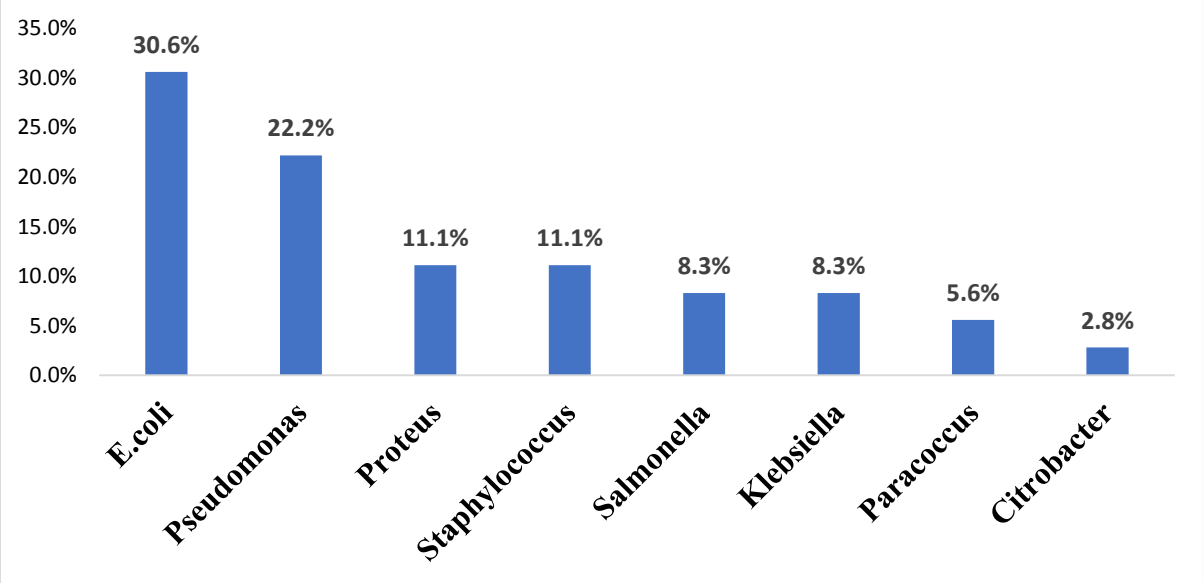

Figure 1: Percentage Occurrence of Bacteria Isolates in Well Water

The susceptibility test was carried out for *Escherichia coli* only as it's recommended as principal indicator of faecal water contaminant by World Health Organization and more so, it is the highest occurring contaminant while the other isolated organisms were ignored. The result from this study shows that different species of *Escherichia coli* showed different antimicrobial sensitivity to different antimicrobial agents (Figure 2). The isolates were susceptible to Ceftriazone (100%), Vancomycin (60%), Gentamicin (50%), and resistant to Streptomycin (100%), Penicillin G (100%), Chloramphenicol (80%), Ofloxacin (80%), Ciprofloxacin (60%), Cefoxitin (60%), Erythromycin (60%).

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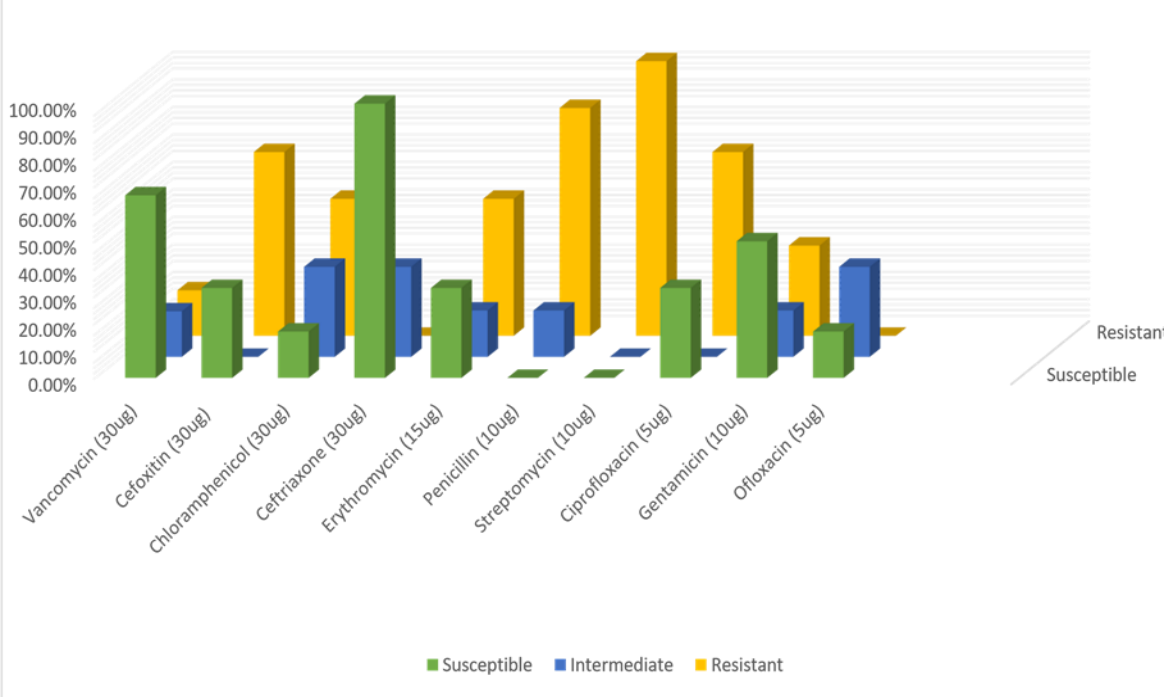

Figure 2: Resistant Pattern of *E. coli* to Commonly Prescribed Antibiotics

One hundred percent of the isolates had MARI ≥ 0.2, an indication that the isolates have been pre-exposed to the antibiotics tested (Table 5).

Table 5: Multiple Antibiotics Resistant Indices (MARI) Of *E. coli* Isolated From Different Well Water

S/N	Well	MARI	Antibiotic to which Isolates were Resistant to
1	B1	0.4	FOX, P, S, OFX
2	D3	0.7	V,FOX,C,E,CIP,S
3	E3	0.5	FOX,C,P,CIP,OFX
4	F1	0.5	FOX,S,CIP,CN,OFX
5	H1	0.4	E, P, S, CN
6	T2	0.5	C, E, P, S
7	K1	0.7	V,FOX,C,E,CIP,S
8	G1	0.5	FOX,C,P,CIP,OFX
9	I1	0.5	FOX,S,CIP,CN,OFX
10	J1	0.4	E, P, S, CN
11	Q2	0.5	FOX,S,CIP,CN,OFX

Keys: MARI=Multiple Antibiotics Resistant Indices; V= Vancomycin, FOX= Cefoxitin, C= Chloramphenicol, CIP= Ciprofloxacin, E= Erythromycin, OFX= Ofloxacin, CN= Gentamicin, P= Penicillin, S= Streptomycin.

DISCUSSIONS

The spread of disease through contamination of water source particularly in developing and under developed countries are common phenomenon and has been well reported (15). This study was conducted to determine the occurrence of bacteria contaminants present in well water samples collected within Sabon Tasha community of Kaduna State. The coliform bacteria are the main pathogen indicators in drinking water supplies. Among the coliform bacteria, presence of the fecal coliforms usually confirms fecal contamination of water supply since they usually exist in the intestine of animals as well as in humans. Diseases related to contamination of drinking water constitute a major burden on human health. Interventions to improve the quality of drinking water provides significant health benefits (2). In this study, the mean colony counts per sample ranges between 1.0×10^5 to 6.2×10^5 CFU/ml (Table 2). This exceeded the recommended limit for portable water (16), which states that the coliform count in drinking water should be zero cfu/100ml. Also, in this study, contaminants isolated include *E. coli* (11), *Pseudomonas* (8), *Proteus* (4), *Staphylococcus* (4), *Salmonella* (3), *Klebsiella* (3), *Paracoccus* (2) and *Citrobacter* (1) with *E. coli* having the highest occurrence (Fig 1). Similar microorganisms have been isolated from studies done in Delta state where well water used for drinking and cleaning purposes was contaminated with pathogenic microorganisms (17). The high prevalence or occurrence of *Escherichia coli* in well water samples in Sabon Tasha, Kaduna, also corresponds with the study by Ogu *et al* in Delta state (17). According to Ogu, the prevalence of *E. coli* in the well water was found to be 26.7%, which was lower compared to this study. The presence of these bacteria contaminants could be attributed to the fact that most of the wells in the study area were uncovered, and most were situated too close to septic tanks, and some constructed too low from the ground level. As a result of these conditions, these well water could be easily contaminated through wind, leakages from septic tanks, seepage from contaminated ground water and waste waters, run-off among others. Presence of these pathogens pose a special health risk for infants, young children and people with severely compromised immune system especially when consumed, (18). These could also cause serious disease conditions and clinical signs such as giardiasis, cryptosporidiosis, gastroenteritis, diarrhea, typhoid fever, cholera, bacillary dysentery, hepatitis, shigellosis etc (19). The presence of *E. coli* and its high occurrence in water gives an indication of fecal contamination (20). It is desirable for water used for drinking purposes not to contain bacteria of fecal origin. The higher level of prevalence of *E. coli* in well water may be due to the possibilities of relatively slow movement of water along the lateral plains in the soil substratum. The susceptibility test was carried out for *Escherichia coli* only as the study only focuses on the isolate with the

highest occurrence while the other isolated organisms were ignored as they were not of concern for this study.

The result from this study shows that different species of *Escherichia coli* showed different antimicrobial sensitivity to different antimicrobial agents (Fig. 2). The *E. coli* isolates obtained were from well number B_1, B_3, E_3, F_1, T_1 and T_2. The susceptibility profile of the E. *coli* isolates can be summarized thus: the isolates were sensitive to Ceftriaxone (100%), Vancomycin (60%), and Gentamicin (50%), but showed different degree of resistance to other antimicrobial agents thus: 100% resistance to Penicillin and Streptomycin, 80% resistance to Chloramphenicol and Ofloxacin, 60% resistance to Cefoxitin, Ciprofloxacin and Erythromycin (Figure 2). This is similar to a study by Adzitey *et al* (21)., which had similar susceptibility profile of *E. coli* isolate in drinking water in Ghana but his study excluded Vancomycin (21).

The multiple antibiotic resistance index of *E. coli* in this study is greater than 0.2 for all the isolates indicating high exposure of the *E. coli* isolates to the antibiotics (Table 5). This is similar to the work of Bello *et al.,* (22) who reported high level of multiple resistance among *E. coli* isolates from well water in Akure, Ondo State.

CONCLUSION

This study was able to establish the presence of faecal contaminants in Well water in Sabon Tasha area of Kaduna, with *E. coli* as the most prevalent. The presence of this bacteria contaminants could be attributable to the fact that most Wells in the area were uncovered and situated close to septic tanks. The *E. coli* species isolated showed sensitivity to fewer commonly prescribed antimicrobial agents (Ceftriaxone, Vancomycin and Gentamicin). The isolates were resistant to most of the commonly prescribed antimicrobial agents used in this study (Penicillin, Streptomycin, Chloramphenicol, Ofloxacin, Cefoxitin, Ciprofloxacin and Erythromycin. This poses a very serious health thread as Well Water is a common source of drinking water in this community, and indeed most developing countries. This can also be a source of infection to the users and a potential means of transmitting multidrug resistant bacteria strains in the community.

RECOMMENDATIONS

Government should employ the use of Environmental Health Officers, whose duties are to certify every Well fit for drinking or otherwise. Infections arising from water borne pathogens are most likely to respond to treatment with Ceftriaxone, Vancomycin and Gentamicin in this environment, while the use of Penicillin, Streptomycin, Chloramphenicol, Ofloxacin, Cefoxitin, Ciprofloxacin and Erythromycin may be of no medical value. The conduct of laboratory microbial culture and sensitivity test for every case of infection before treatment will be of great value in this environment.

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Authors' Contributions: This study design was carried out by PSK. BSG and ISG conducted the lab experiments, while CDJ and IJC made the first draft of this publication. All the authors reviewed the final version of this manuscript and it was approved by all.

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