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Research Article

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Antibiogram and Molecular Analysis of Clinical Bacteria Isolates from the Three Geographical Regions of Ondo State, Nigeria

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Abstract

This study shows diversified forms of multidrug resistant bacteria agents that were obtained from designated health facilities in Ondo State, Nigeria. One hundred and fifty (151) clinical bacteria isolates collected from designated hospitals in Ondo, Okitipupa, Owo and Akure were identified. From Ondo North (SSH, Ikare and FMC, Owo), Seventy (70) bacterial isolates were obtained and this includes 15 (21.4%) Gram +ve organisms consisting of Staphylococcus spp, Bacillus spp, Streptococcus spp, and Corynebacterium spp. While 55 (78.6%) of the isolates were Gram-ve of various species. In Ondo Central (SSH, Akure and Trauma Centre, Ondo), Sixty-five (65) bacterial isolates obtained comprises 16 (24.6%) Gram+ve species of Staphylococcus and Streptococcus only. While 49 (75.4%) were Gram-ve bacterial species. Similarly, Ondo South (SSH, Okitipupa), Thirty-three (33) bacterial isolates were obtained, 8 (24.2%) were Gram+ve of the species of Staphylococcus, Streptococcus and Enterococcus. While 25 (75.8%) were Gram-ve of diverse species. Thirty two (32) of the 151 isolates subjected to antibiotic susceptibility test were extremely resistant to both the convectional antibiotic discs and the E-tests strips. These resistant strains were further identified molecularly with their plasmid profile studied. This is of epidemiological significance and shows the necessity to sort alternative therapy for these multiple antibiotic resistant strains and improve our health management services.

Keywords: Antibiogram, Clinical bacteria isolates, Molecular analysis, Ondo State, Nigeria

Introduction

The trend of diseases spread and distribution within the environment is linked with some factors uniquely suited to the spread of microbial infections as it houses both susceptible patients and patients with difficult-to-treat infections [1]. There is a great risk that some patients may contact microbial infections which may be difficult to treat due to microbial drug resistant [2; 3]. Escalations in both community and hospital-acquired antimicrobial-resistant bacteria are threatening the ability to effectively treat patients, emphasizing the need for continued surveillance, more appropriate antimicrobial prescription, prudent infection control, and new treatment alternatives [4].

One of the major contributions to health care delivery in the 20th century was the discovery of potent antimicrobial agents [5]. However, the emergence of resistance of bacteria to the antimicrobials raises serious concern and poses a growing threat to health-care delivery and a danger to the public globally [5, 6]. Antibacterial drugs have been regarded for more than 60 years to cure infections, whether the infection was acquired in the community or in the hospital setting [7]. That is, the discovery of antimicrobial agents had a major impact on the rate of survival from infections but the changing patterns of antimicrobial resistance (AMR) caused a demand for new antibacterial agents [8].

As studies on antimicrobial resistance in different parts of Nigeria have been conducted, pathogenic bacteria isolation and antimicrobial susceptibility is variable and influenced by geographical location, variations in patient population, infection control practices, level of health facility, and regional antibiotic uses [9].

In most hospitals, the causative pathogens originate from the environment as well as the endogenous flora of the patient's skin, mucous membranes or hollow viscera [10]. The most commonly isolated bacterial pathogens are *Staphylococcus aureus*, *Escherichia coli*, *Enterobacteriaceae*, Coagulase Negative Staphylococci (CoNS), Enterococci and *Pseudomonas aeruginosa* [11, 12]. Although the microbial pathogens isolated varies across different hospitals, but literature reports have documented an increasing proportion of Gram-positive and Gram-negative organisms in recent times (10). Furthermore, there is an increase in incidence of clinical isolates attributed to antimicrobial resistant pathogenic bacteria like methicillin resistant *Staphylococcus aureus* (MRSA) and Vancomycin Resistant *Staphylococcus aureus* [13].

In the United States of America, a study was conducted from small community hospital among all patients who were admitted and *S. aureus* was reported as the commonest isolate (25.8%), followed by *Enterobacteriaceae* (12.4%), streptococci species (11.2%), CoNS (10.1%), *Enterococci* species (7.9%) and *Pseudomonas aeruginosa* (6.7%), but MRSA was isolated from only 4.5% of the patients [12]. Another similar study in USA among patients who underwent operation for hollow viscus injury documented Escherichia coli as the most commonly isolated microorganism (64.7%) followed by *Enterococci* species (41.2%) and *Bacteroides* (29.4%) (14). Findings from these two studies suggest that the etiological agents of pathogenic organisms depend on the hospitals and the kind of patients admitted.

Findings from a study carried out at a University hospital in Nigeria showed that the commonly isolated bacteria were *S. aureus* (25%), *Pseudomonas aeruginosa* (20%), *Escherichia coli* (15%), Klebsiella oxytoca (10%) and Proteus mirabilis (10%) [5]. Similarly, in a prospective survey done in Central African Republic among admitted patients, it was found that methicilin-susceptible S. aureus was the most frequent species isolated followed by Enterobacteriaceae and *P. aeruginosa*. A strain of *E. cloacae* harbouring extended spectrum beta lactamase (ESBLs) was also isolated [15]. Frequent isolation of *S. aureus* (28.8%) and *Escherichia coli* (27.1%) have also been reported among patients with abdominal surgical wounds in Ethiopia [16].

Studies have shown an increase in the trend of clinical isolates with attributable to antimicrobial resistant pathogens such as MRSA. Data collected between 2003 and 2007 by Weigelt *et al*., reported that the proportion of MRSA significantly increased (from 16.1% to 20.6%) among culture positive patients readmitted in 97 US hospitals [11]. Another study reported that MRSA was the most frequent pathogen recovered and the prevalence rate of MRSA was almost doubled during the study period increasing from 0.12 infections per 100 patients to 0.23 infections per 100 patients [17].

The evolution of disease-causing microorganisms has resulted in many antimicrobials losing their effectiveness after more than 50 years of widespread use. As microorganisms evolve, they adapt to their environment. The microorganisms evolve new mechanisms to resist the antimicrobials by changing their genetic structure from something that stops them from growing and spreading such as an antimicrobial [18]. The genetic structure changes ensure that the offspring of the resistant microbes are also resistant. It is harder to eliminate infections from the body due to antimicrobial resistance. Some infectious diseases are now more difficult to treat than a few decades ago [19].

Use of antibacterial drugs has become widespread over several decades to cure infections whether or not their use is appropriate [20]. Consequently, antibacterial drugs have become less effective resulting in an accelerating global health security emergency [21]. Over the past few years several studies in the world and African countries such as Nigeria had reported the presence of antimicrobial resistant strains from clinical specimens. Identification and recognition of resistance pattern of bacteria is necessary from time to time to give appropriate treatment and avoid adverse clinical outcomes [20].

Currently, many studies have shown that commonly isolated bacteria from different specimen such as *S. aureus*, *E. coli* and *P. aeruginosa* are resistant to many commonly used antimicrobials. Different studies conducted on antimicrobial resistance in Nigeria and Ethiopia indicated increasing resistance rates of these commonly isolated pathogens to commonly prescribed antibiotics, including ampicillin, amoxicillin, penicillin, tetracycline and cotrimoxazole [22; 23]. In another study conducted in Nepal on isolates of wound infections, *Staphylococcus aureus* showed high level of drug resistance to oxacillin and cotrimoxazole [24].

According to the World Health Organization (WHO), AMR is possibly the single biggest threat facing the world in the area of infectious diseases [7]. Systematic literature review study conducted in London from 2000-2012 in the world show that the cost of AMR is a vast range of figures, from £5 million to more than £20 million and reported additional costs per patient per episode for hospital costs [18].

Ondo State is a large State in the South Western part of Nigeria with a population of 3.7 million people. It spreads from the coastal area of the South to the grassland area of the Northern part with fishing and arable farming being the major occupation of the people. With this diversity, it is expected that the bacteria spread too will be greatly varied. The state has 15 General Hospitals, 4 Specialist Hospitals and 18 Comprehensive Health Centers. Not all have standard medical laboratory, no knowledge about infection rate in the State. This study helps to determine the susceptibility pattern and resistance genes of different clinical isolates from major hospitals in Ondo State, Nigeria and thereby provide baseline information for use in the treatment of infectious diseases. This influences empiric antimicrobial choices which should be based on local isolation and susceptibility studies. Thus, knowledge of local pathogenic bacteria isolates and susceptibility patterns is required to detect any changes on time through periodic investigation so that modification and recommendation for empiric therapy of bacterial infections could be made. There is a paucity of data regarding antimicrobial resistance of common clinical bacterial isolates in Ondo State hence this study was initiated to provide data regarding bacteriological identification and antibiogram of clinical isolates in the three geographical locations of Ondo State, Nigeria.

Materials and Methods

Sample Collection

Fifty (50) Agar slants in bijou bottles were submitted to each of the twelve (12) selected hospitals namely State specialist hospitals at Ikare, Ondo, Okitipupa and Akure; General hospitals at Iwaro, Ileoluji, Ore, and Igbokoda for the collection of clinical isolates. Others are the Federal Medical Center, Owo and the Mother and Child hospital Ondo. Collections of bacterial isolates were done periodically, from the above named Hospitals in the three Senatorial districts of Ondo State. The bacterial isolates were brought to Adekunle Ajasin University, Akoko in slants; they were subcultured, purified and stored in agar slants in the Refrigerator.

Purification/ Subculture of Isolates

All isolates collected were purified by sub-culturing them on freshly prepared Blood Agar and MacConkey agar plates. After incubation at 37°C for 24hrs, the isolates will now be stored at 4°C until further analysis.

Identification of Isolates

This was done through observation of isolate's morphology on

agar plates, staining procedures (Gram's stain, spore staining) biochemical tests (catalase, oxidase, sugar fermentation) and use of API -20E Test kit as described by Cheesbrough and Cowan and Steel [25, 26].

Susceptibility Test

Disc diffusion technique described by CLSI, [27] was employed. E-test strips were used to determine the MIC of the sensitive antibiotics..

Plasmid Curing

Plasmid curing of resistant isolates was done according to the method described by Tomoeda *et al*., [28]

Molecular Study of Multidrug Resistant Strains

Isolates that are multidrug resistant were further studied using molecular methods. DNA extraction and amplification was done using Polymerase Chain Reaction (PCR). The processing protocol include an initial preheat at 94°C for 2 minutes, followed by 35 cycler of denaturation at 94°C for 15s, this was followed by annealing and extension of the gene of interest.

Plasmid Extraction

A single bacterial colony was picked up and grown overnight in a test tube of Muller Hilton broth at 37°C. One (1 ml) of Muller Hilton broth was pipette in an eppendorf tube and centrifuged at 10,000 rpm for 2 min. The cell pellets obtained were re-suspended in 600µl Extraction buffer (200mM Tris –Hcl PH 8.0, 25Mm EDTA, 25Mm Nacl and 1% SDS), vortex then incubated at 65°C for 35 minutes. Deproteinization was done with Phenol/chloroform: Isoamyl alcohol (6:6:1). The tube was mixed vigorously, and centrifuged at maximum speed for 5 mins. The supernatant was transferred into a sterile 1.5 ml eppendorf tube and equal volume of chloroform: isoamyl alcohol (24.1) was added. The sample was left at room temperature for 5 minutes and centrifuged at maximum speed for 10 min and the supernatant was transferred into a sterile tube. Precipitation was done with 2 volumes iced ethanol (100%) and incubated at 4°C for 2hrs. centrifugation was done at maximum speed. The plasmid DNA pellet was washed with 550 µl of cold (4°C) 70% ethanol and centrifuged at 12,000 rpm for 15 min. The supernatant was discarded; the pellet was dried for 30 min and re-suspended in 30 µl of sterile deionized water [29].

Result

One hundred and fifty one (151) clinical microbial isolates collected from designated hospitals in Ondo, Okitipupa, Owo and Akure are presented in Tables 1. The sex distribution of the patients showed an almost equal ratio. This is shown in Table 2. Table 3 shows the age distribution of patients from whom sample were collected. Age range of 81-90 had the lowest percentage of participants (3.97%) while those in the age range of 31 – 40 had the highest percentage (23.84%). Patients within the ages of 21 -60 formed the bulk (67.56%) of participants. The distribution of sample types are shown in Table 4 and urine samples, 79 (52.32%) were the most collected sample.

In Table 5, the Gram negative organisms identified with use of microbact kits were shown. With the *E. coli* being the most frequently isolated. Thirty two (32) of 151 bacterial isolates subjected to antibiotic susceptibility test were extremely resistant to both the convectional antibiotic discs and the E-tests strips. The susceptibility pattern of the isolates shows that Nitrofurantoin, Ciprofloxacin and Augumentin were the most active antibiotics with the highest activity recorded by the Nitrofurantoin against *Salmonella pullorum* (24.0mm), *Yersinia aldora* (23.3mm), *Actinobacillus* spp. (22.3mm) and *S.aureus* (21.3mm). This is followed by Ciprofloxacin against *E.coli* (22.5mm) and Augumentin against *E. hermanni* (20.0mm) as shown in Table 6. The agarose gel electrophoresis plates for the identification of resistant genes on the plasmid of the isolates are shown in Fig 1.

Table 1: Distribution of Samples Collected from Hospitals

S/NO	Hospital	No of Samples	Percentage (%)
1	IK	32	21.19%
2	OW	31	20.53%
3	OK	27	17.88%
4	AK	29	19.21%
5	T	32	21.19%
Total	**5**	**151**	**100%**

Degree of Confidence 95%

Key: IK = State Specialist Hospital Ikare, OW = Federal Medical Center Owo, OK = State Specialist Hospital Okitipupa, AK = State Specialist Hospital Akure and T = Ondo State Trauma and Surgical Center Ondo.

Table 2: Sex of Patients from whom samples were collected

Sex	Frequency	Percentage
Male	76	50.33%
Female	75	49.67%
Total	**151**	**100%**

Degree of Confidence 95%

Table 3: Ages of patients from whom samples were collected

S/NO	Age Range	Frequency	Percentage (%)
1	1-10	7	4.64%
2	11-20	11	7.28%
3	21-30	24	15.89%
4	31-40	36	23.84%
5	41-50	22	14.58%
6	51-60	20	13.25%
7	61-70	15	9.93%
8	71-80	10	6.62%
9	81-90	6	3.97%
Total	**1 – 90**	**151**	**100%**

Degree of Confidence 95%

Table 4: Frequency distribution of samples

S/NO	Site of Isolate	NO. of Isolate	Sample (%)
1	Urine	79	52.32%
2	Wound Swab	19	12.58%
3	HVS	14	9.27%
4	Semen	11	7.28%
5	Sputum	10	6.62%
6	Ear Swab	7	4.64%
7	Stool	3	1.99%
8	Throat	2	1.33%
9	Urethral Swab	2	1.33%
10	Blood	1	0.66%
11	Eye Swab	1	0.66%
12	ECS	1	0.66%
13	Soft Tissues	1	0.66%
Total	13	151	100%

Degree of Confidence 95%

Table 5: Frequency of Gram Negative Isolates Identified using MicrobactTM 24E

S/NO	Isolate	Test Code	% Probability	Occurrence	Percentage
1	*Escherichia coli*	67604760	99.49	31	20.53%
2	*Escherichia hermannii*	67600260	62.26	1	0.66%
3	*Klebsiella ornithimolityca*	77763777	99.89	4	2.65%
4	*Klebsiella oxytoca*	47623776	99.95	8	5.30%
5	*Klebsiella pneumonia*	47533776	76.63	2	1.32%
6	*Klebsiella terrigena*	47462776	95.90	1	0.66%
7	*Seratia odorifera biogp-1*	77765772	99.74	2	1.32%
8	*Enterobacter gergoviae*	77526327	83.23	2	1.32%
9	*Enterobacter aerogenes*	77577777	99.64	2	1.32%
10	*Enterobacter agglomerans*	47712261	65.54	2	1.32%
11	*Enterobacter cloacae*	67567775	97.33	2	1.32%
12	*Aeromonas hydrophila*	457704662	99.91	6	3.97%
13	*Acinetobacter baumannii*	45422021	98.93	5	3.31%
14	*Acinetobacter haemolyticus*	00100000	42.26	1	0.66%
15	*Citrobacter amalonaticus*	67734761	83.88	1	0.66%
16	*Citrobacter youngae*	57700620	69.04	1	0.66%
17	*Citrobacter gillenii*	17462760	84.59	1	0.66%
18	*Budricia aquatic*	01000220	64.51	4	2.65%
19	*Pseudomonas aeruginosa*	447534221	96.62	10	6.62%
20	*Pseudomonas fluorescens-25*	405004220	97.15	5	3.31%
21	*Bergeyella zoohelcum*	400100000	80.04	2	1.32%
22	*Actinobacillus sp*	401500001	99.86	2	1.32%
23	*Burkhodreria cepacia*	457534020	94.76	2	1.32%
24	*Burkhodreria pseudomallei*	475673745	99.81	6	3.97%
25	*Chrysobacterium menigosepticum*	401600220	99.94	1	0.66%
26	*Yersinia pestis*	01400020	95.35	1	0.66%
27	*Yersinia aldovae*	01100020	78.18	1	0.66%
28	*Pasteurella multocida*	465540220	98.0	4	2.65%
29	*Moraxella sp*	460100000	99.97	4	2.65%
30	*Morganella morganii biogp-1*	65314020	77.97	1	0.66%
31	*Salmonella pullorum*	67000220	94.00	1	0.66%

Degree of Confidence 95%

Table 6: Antibiotics Susceptibility Pattern of the Bacterial Isolates

	CAZ (30µg)	CRX (30µg)	GEN (10µg)	CXM (5µg)	OFL (5µg)	AUG (30 µg)	NIT (30µg)	CPR (5µg)	CTR (30µg)	ERY (5µg)	CXC (5µg)
Escherichia coli	0	16.2±0.2	5.8±0.7	8.00±0.1	16.4±1.0	18.7±1.2	12.9±0.9	**22.5±3.1**	0.9±0.1	12.6±0.	0
Escherichia hermannii	2.5±0.3	9.5±0.5	3.0±0.2	7.2±0.7	10.8±2.5	**20.0±5.8**	8.7±2.1	20.5±4.5	0	3.7±0.2	0
Klebsiella ornithimolityca	1.5±0.7	5.8±2.2	0.4±0.1	10.0±3.2	12.5±1.5	9.5±0.7	13.1±0.9	17.9±4.6	0.8±0.3	0	0
Klebsiella oxytoca	0	2.7±0.2	9.4±0.6	11.5±0.9	16.3±3.0	9.6±1.5	10.1±2.9	8.4±0.6	0.7±0.1	9.7±0.3	0
Klebsiella pneumonia	0.7±0.3	6.0±1.2	4.4±0.5	8.1±0.3	10.7±2.5	0	9.4±0.6	8.4±0.6	8.8±2.3	9.7±0.3	0
Klebsiella terrigena	0.5±0.1	3.0±0.5	0	6.2±0.3	11.3±2.7	11.6±3.0	7.6±1.2	16.7±5.6	0	2.4±0.2	0
Seratia odorifera biogp-1	0	0.7±0.2	0	1.5±0.1	7.6±1.5	0	0.3±0.0	8.4±0.6	0	0	0
Enterobacter gergoviae	0	0.8±0.3	5.5±0.2	7.0±0.3	5.9±2.7	4.4±0.3	6.8±2.0	0	7.5±1.8	0	0
Enterobacter aerogenes	0	5.8±2.2	0.4±0.1	0	10.5±4.5	8.2±0.5	0	10.5±3.6	0	2.7±0.2	0
Pseudomonas aeruginosa	0	0	6.3±0.1	0	15.9±3.9	7.4±0.3	13.7±2.7	1.5±0.5	9.1±0.8	10.±5.1	0
Staphylococcus aureus	2.3±0.3	1.0±0.1	12.3±3.6	0	19.0±5.0	6.9±2.0	**21.3±7.0**	13.4±0.8	0	8.2±2.2	0
Corynebacterium sp	0	6.3±0.3	0	2.5±0.3	0	0	0	7.6±0.9	0	6.7±2.1	0

Table 6: Antibiotics Susceptibility pattern of the Bacteria Isolates											
Enterobacter gergoviae	0	0	7.5±2.3	5.3±1.2	0	0	6.8±2.0	0	7.5±1.8	0	0
Enterobacter cloacae	0	2.7±0.2	0	0	0	0	6.8±2.0	0	0	0	0
Aeromonas hydrophila	0	7.8±2.5	8.3±1.1	0	6.5±1.5	15.6±3.7	6.5±1.9	0	4.5±0.3	0	0
Acinetobacter baumannii	3.0±1.0	0	10.4±1.6	0	0	7.6±1.0	0	6.4±0.7	5.7±0.1	11.9±2.3	0
Acinetobacter haemolyticus	0	10.0±3.	0	8.1±0.3	10.7±2.5	0.0±0.0	13.7±0.6	8.4±1.5	0	9.7±0.3	0
Citrobacter amalonaticus	0.5±0.1	3.0±0.5	0.0±0.0	6.2±0.3	11.3±2.7	11.6±3.0	7.6±1.2	16.7±5.6	0	2.4±0.2	0
Citrobacter youngae	0	0	0	0	6.0±1.5	0	0	5.4±1.2	0	0	0
Citrobacter gillenii	2.2±0.0	0	7.7±2.5	8.9±2.3	0	9.6±1.0	10.8±2.0	0	7.5±1.8	8.2±1.5	0
Budricia aquatic	0	10.5±3.6	0.4±0.1	0	10.5±4.5	8.2±0.5	0	5.9±2.1	0	2.7±0.2	0
Pseudomonas fluorescens-25	0	0	7.8±3.0	0	12.9±2.0	7.4±0.3	13.7±2.7	3.7±1.5	0	10.2±5.1	0
Bergeyella zoohelcum	4.7±0.3	5.0±0.2	0	0	10.0±2.7	0	11.3±7.0	4.4±0.6	0	5.2±1.2	0
Actinobacillus sp	1.0±0.0	2.3±0.1	9.5±3.2	0	12.0±3.	0	**22.3±6.5**	14.5±2.9	0	13.7±4.2	0

Table 6: Antibiotics Susceptibility Test on Bacteria Isolates (Cont'd)											
Burkhodreria cepacia	0	0	9.3±3.0	0	6.1±2.6	0	10.8±2.0	0	7.5±1.8	0	0
Burkhodreria pseudomallei	0	2.7±0.2	5.7±1.3	0	0	0	6.8±2.0	0	5.0±1.2	0	0
Chrysobacterium menigosepticum	1.5±0.2	0	10.4±1.5	0	0	7.6±1.0	0	6.4±0.7	8.5±2.	1.9±0.4	0
Yersinia pestis	2.0±0.5	0	8.7±1.9	5.0±2.0	0	6.8±1.4	15.0±4.3	4.9±1.5	7.7±1.1	12.9±2.3	0
Yersinia aldovae	4.2±1.3	1.5±0.1	10.3±1.6	0	**20.0±5.6**	7.5±2.1	**23.3±6.5**	11.1±2.8	0.9±0.1	10.6±3.1	0
Pasteurella multocida	1.3±0.2	0	13.5±3.1	0	18.6±4.7	7.9±2.5	20.4±6.0	14.0±3.2	0	7.3±1.0	0
Moraxella sp	0	5.7±0.7	9.3±2.2	0	10.9±3.2	0	16.4±3.4	3.7±1.2	0	8.0±2.1	0
Morganella morganii biogp-1	5.1±0.8	0	6.8±2.9	11.4±1.8	0	9.6±1.0	10.8±2.0	0	7.5±1.8	8.2±1.5	0
Salmonella pullorum	1.5±0.1	1.9±0.2	2.4±0.5	1.9±0.1	13.7±5.5	9.7±2.5	**24.0±6.0**	18.2±3.0	9.0±1.5	14.4±3.2	0
Enterococcus sp	0	11.0±4.1	6.5±2.3	5.3±1.2	15.0±5.0	0	6.8±2.0	0	7.5±1.8	9.2±1.3	0
Streptococcus sp	11.2±1.3	5.5±0.1	12.3±1.6	0	11.0±3.6	10.5±2.1	14.3±6.5	11.1±2.8	10.9±0.1	12.±3.1	0

P<0.05 Keys: CAZ = Ceftazine (30µg), CRX = Cefuroxime(30µg), GEN = Gentamicine (10µg), CXM = Cefixime (5µg), OFL = Oflaxacin (5µg), AUG = Augmentin (30µg), NIT = Nitrofuratoin (30µg), CPR = Ciprofloxacin (5µg), CTR = Cenptraiaxome (30µg), ERY = Erythromycine (5µg) and CXC = Cloxacelline (5µg); SEM = Standard Error of Mean.

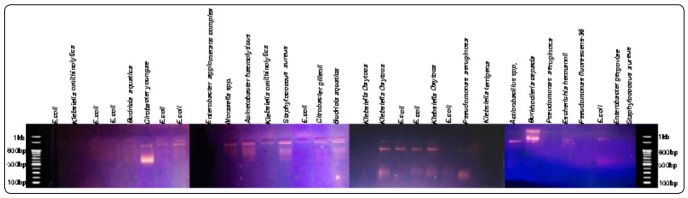

Figure 1: Molecular identification of clinical isolates

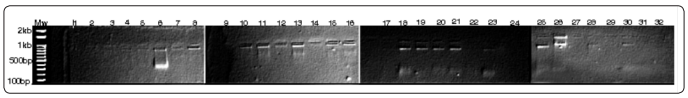

Figure 2: Plasmid profile of clinical isolates

Discussion

This study shows the distribution pattern and resistant nature of bacteria isolated from clinical samples obtained from hospitals in various geopolitical zones in Ondo State, Nigeria. A total of 151 isolates were obtained out of which *Staphylococcus aureus* (17.88%) and *Escherichia coli* (28.48%) apart from other coliforms organisms that constitute 21.19% of total isolates were predominant. The consequences of bacterial resistance to both individuals and the public are severe. Infections caused by resistant bacteria as observed in this study include failure to respond to standard treatment. This corroborates with WHO global report [7] and the study of Schnuriger *et al*. [21] which shows that resistant bacterial strains often leads to untreatable serious infections with high rate of mortality. The incidence of multiple antibiotic resistance in this study is alarming in that patients receiving organ transplants, cancer treatment and other advanced therapies are particularly vulnerable to infection which could be difficult to treat under this conditions. Hence, according to the study of Smith and Coast [18], when treatment of an infection fails in such patients, the infection is likely to become life-threatening and may be fatal.

Ciprofloxacin, Oflaxacin and Augmentin are relatively the most effective antibiotics against most test clinical isolates used for this study (Table 8). World Health Organization (WHO) study shows that 2-70 percent *E. coli* strains and 8-77 percent of *K. pneumonia* were resistance to third-generation cephalosporins and fluoroquinolones in correlation with this study. This has great impact on health and economic burden of infections caused by *E. coli* strain. This also correlates with previous investigations [7, 18], which shows that this may lead to unnecessarily high usage of broad-spectrum antibacterial drugs, which will exacerbate the resistance problem. Antibiotics will be ineffective against these resistant microorganisms which lead to persistence and spread of these infections in the community and thus expose the general population to the risk of contracting a resistant strain of infection [30]. Hence the observation made on potency and efficacy of selected antibiotics on test organisms in this study will be helpful for clinicians and health management system.

Baseline survey carried out by Drug Administration and Control Authority [9] at national level showed that almost all prescribers used clinical symptoms and signs to prescribe antibiotics, while culture and sensitivity tests were done for only 2.2% of patients. Moreover, most of prescribers did not know bacterial resistance patterns to commonly prescribed antibacterial in their health facilities [9]. Furthermore, lack of continuous education and updated information for prescribers and dispenser might have contributed to the development of antimicrobial resistance as reported by Getachew *et al*., [31].

Based on the study of O'Neill *et al*. [32], antibacterial drugs used to prevent post-operative surgical site infections have become less effective or ineffective. And this may be part of factors responsible for the frequently encountered multiple resistant strains in this study. Complimentary to this, the use of expensive and potentially toxic second or third line drugs in treatment of infections with resistant strains often leads to longer hospital stay due to side effects in correlation with the report of Mulu *et al*., [33].

This study helps to provide information on bacteria spread within the geographical region studied. The antibiotic susceptibility profile of these pathogens will be a general guide to the clinicians and a template for future studies as well as combating the menace of multiple antibiotic resistant clinical strains within the populace.

Declarations

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