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Antibiotic susceptibility and plasmid profile of *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* isolated from wound patients in Abakaliki metropolis, Ebonyi state

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Abstract: *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* has become increasingly recognized as an emerging opportunistic pathogen of clinical importance. A total of 18 isolates comprising of 10 (55.6%) *S. aureus*, 6 (33.3%) *P. aeruginosa* and 2 (11.1%) *E. coli* were recovered from 15 pus samples of wound patients attending Federal Teaching Hospital Abakaliki I (FETHA I) and Federal Teaching Hospital Abakaliki II (FETHA II). All the isolates were subjected to antibiotic susceptibility testing using Kirby-Bauer disk diffusion method on Mueller-Hinton agar. *S. aureus* showed the highest susceptibility percentage of 80.0% to ciprofloxacin, followed by 60.0% to augmentin and 50.0% to streptomycin, while the highest resistance percentage was obtained with lincocin (100.0%), followed by ampiclox (80.0%). *P. aeruginosa* showed the highest susceptibility percentage of 83.3% to ciprofloxacin, followed by 66.6% to streptomycin and gentamycin (66.6%), while the highest resistance percentage was obtained with streptomycin (33.3%) and gentamycin (33.3%). *E. coli* showed the highest susceptibility percentage to gentamycin and streptomycin with 100% activity. The antibiotics with reasonable resistant profile was observed in 10 isolates (5 *S. aureus*, 5 *P. aeruginosa* and 1 *E. coli*) with isolate code S_1, S_2, S_3, S_4, S_5, Ps_1, Ps_2, Ps_3, Ps_4, Ps_5 and Ec_1 which showed resistance to atleast 5 antibiotics, hence this isolates were subjected to plasmid profile analysis. Only three isolates (S_1, S_4 and Ec_1) showed the presence of plasmids within the range of 1.8 kbp to 10.4 kbp. Hence antibiotic resistance of an organism does not always confer the presence of plasmid.

Keywords: Antibiotics, Plasmid, *S. aureus*, *P. aeruginosa*, *E. coli,* Wound Infection, Abakaliki

1. Introduction

Wound is a breach in the skin, and exposure of subcutaneous tissue following loss of skin integrity providing a moist, warm and nutritive environment that is conducive for colonization and proliferation of opportunistic and pathogenic microorganisms (1). Wound infection is one of the health problems that are caused and aggravated by the invasion of pathogenic organisms in different parts of the body. In developing countries, large number of people die daily of preventable and curable diseases such as wound infections (2). The wound sometimes gets infection by either single or multiple organisms. Wound infections are mostly due to nosocomial pathogens that differ from country to country and from hospital to another within the same region, which remains the major source of post-operative morbidity (3).

Previous studies from different parts of the country showed that *Pseudomonas species, Staphylococcus aureus, Klebsiella* species*, Escherichia coli, Proteus* species*, Streptococcus* species*, Enterobacter* species and coagulase negative staphylococci are the most common pathogens isolated from wound (3). Despite technological advances in surgery and wound management, wound infection has been regarded as the most common nosocomial (hospital-acquired) infection, especially in patients

undergoing surgery (4). Antibiotic resistant bacterial nosocomial infections are a leading problem in intensive care units (ICU) (5). Antimicrobial resistance in nosocomial infections is increasing with both morbidity and mortality greater when infection is caused by drug resistant organisms (6).

Typing techniques useful for establishing clonal relationships between individual isolates in hospital settings are therefore important to recognize nosocomial transmission and guide infection control practice (7). Typing techniques such as PFGE, SDS-PAGE and RAPDPCR have been found to be useful for epidemiological study of *Pseudomonas aeruginosa* (7,8). Plasmid profile analysis examines the total bacterial plasmid content, or subjects plasmids to restriction endonucleases and separates the cleaved plasmid DNA by electrophoresis for analysis (9).

Owing to this, the analysis for this plasmid goes a long way to describe the susceptibility of the organism to antimicrobial agents (9). Hence, the present study determined the plasmid profiles of *Staphylococcus aureus, Pseudomonas aeruginosa* and *Escherichia coli* isolated from wound patients in Abakaliki metropolis, Ebonyi state in order to establish the relationship between antimicrobial resistance's and the presence or absence of plasmids.

2. Materials and Methods

2.1. Study Area

The study area was Abakaliki metropolis, Ebonyi state, Nigeria. Abakaliki is the capital of Ebonyi state. The study area is located between Latitude 06^0 4′N and longitude 08^0 5′E and rainfall pattern is bimodal (April-July), September-November with a short spell sometimes in August. The annual rainfall is between 1000mm-1500mm.The vegetation of the area is predominantly derived Savannah. The mean annual temperature is about 24°C and the relative humidity is between 60-80% (10). Abakaliki has various social amenities and infrastructures, which include hospitals, pharmacy and chemist shops, good roads as well as pipe borne water among others. From all indications, there is very high possibility of misuse of antibiotics by the populace through self-prescriptions as well as overuse and underuse of prescribed antibiotics. Also, many cars and motorcycles found in Abakaliki show high possibility of wound sustenance, whereas the presence of many media laboratories depicts great possibility of amateur laboratory scientists leading to inappropriate diagnosis of wound infections.

2.2. Collection of Samples

A total of 15 pus samples used in this study were collected using sterile swab sticks from 15 wound patients having different types of wounds in two major hospitals in Abakaliki. The hospitals used were Federal Teaching Hospital Abakaliki I (FETHA I) and Federal Teaching Hospital Abakaliki II (FETHA II) in which in which 10 and 5 samples were respectively collected and sent to the Applied Microbiology Laboratory, Ebonyi State University, Abakaliki for analysis after immediate labeling of the samples. All the isolates were identified using conventional techniques (11).

2.3. Antibiotic Susceptibility Testing

Antibiotic susceptibility testing was carried out using the procedure described by Cheesbrough (12) after the organisms had been standardized to 0.5 McFarland standard. Antibiotic susceptibility testing of the isolates was performed using Kirby – Bauer disk diffusion method on Mueller-Hinton agar against the antibiotics. The inhibition zone sizes were interpreted using standard recommendations of NCCLS (13).

2.4. Plasmid Profile Analysis

Plasmid profile analysis was carried out at the Department of Microbiology Laboratory, University of Nigeria Nsukka (UNN). Plasmids DNA were extracted from culture cells using the alkaline sodium dodecyl sulphate (SDS) method as described by Olukoya and Oni (14) and modified by Yah *et al*. (15). The DNAs were electrophoresed on 0.8% agarose gel stained with ethidium bromide and visualized by UV-transillumination. Plasmid sizes were estimated by comparing with previously characterized plasmid.

3. Results

The organisms were then characterized as shown in Table 1 and 2. Three bacteria were isolated in this study and are known to contaminate wound.

Table 1. *Identification of the bacterial isolates using cultural characteristics, motility and Gram reaction*

Isolates	Culture media			Shape/ Consistency	Elevation/Surface	Motility	Gram reaction
	Nutrient agar	Blood agar	MacConkey agar				
S. aureus(S_1-S_{10})	White	Golden yellow	White/pink	Round/moist	Raised/Smooth	-	+
P. aeruginosa(Ps_1-Ps_6)	Green	Dark Greenish-blue	Pale	Rhizoid/dry	Flat/Smooth	+	-
E. coli(Ec_1-Ec_2)	Creamy	Mucoid pink	Smooth pink	Round/dry	Flat/rough	+	-

Key: S = Staphylococcus aureus, Ps = Pseudomonas aeruginosa, Ec = Escherichia coli, + = Positive, - = Negative

Table 2. *Biochemical reaction of the bacterial isolates*

Sample code	Catalase	Coagulase	Indole	Comments
S_1-S_{10}	+	+	-	*S. aureus*
Ps_1-Ps_6	+	-	-	*P. aeruginosa*
Ec_1-Ec_2	+	-	+	*E. coli*

Key: + = Positive, - = Negative

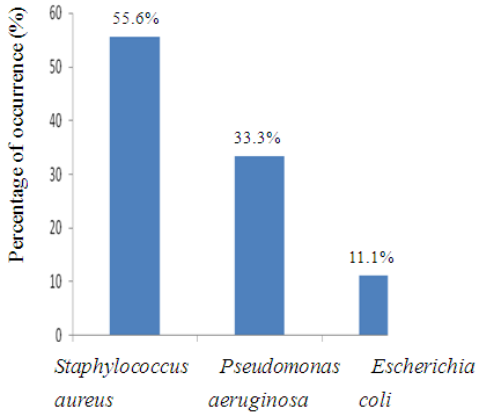

Figure 1. *Percentage occurrence of individual isolates*

The percentage occurrence of the three isolates, *S. aureus* (55.6%), *P. aeruginosa* (33.3%) and *E. coli* (11.1%) as shown in Figure 1.

The *S. aureus* isolates showed very high level of sensitivity to ciprofloxacin with inhibition zone diameter of 20 mm, 19 mm, 10 mm, 21 mm, 16 mm, 18 mm, 12 mm, 18 mm, 17 mm and 20 mm for S_1, S_2, S_2, S_3, S_4, S_5, S_6, S_7, S_8, S_9 and S_{10} respectively, followed by norfloxacin with inhibition zone diameter of 18 mm, 13 mm, 10 mm, 19 mm, 16 mm, 10 mm, 10mm and 16 mm for S_1, S_2, S_2, S_3, S_4, S_5, S_6, S_7, S_8, S_9 and S_{10} respectively, followed by erythromycin and gentamicin while high level of resistance was observed with floxapen which showed 5 mm, 6 mm, 6 mm and 4 mm zone of inhibition to *S. aureus* isolates (S_6, S_7 and S_{10}) and no zone of inhibition to other *S. aureus* isolates (S_1, S_2, S_2, S_3, S_4, S_5, S_8 and S_9), followed by lincocin with inhibition zone of 5 mm, 6 mm and 8 mm (S_6, S_7 and S_9) and showed no inhibition zone to *S. aureus* isolates (S_1, S_2, S_2, S_3, S_4, S_5, S_8 and S_{10}) (Table 3).

Table 3. *Inhibition zone diameter (IZD) of S. aureus against antibiotics*

Isolate code	CPX	NB	CN	LC	S	RD	E	CH	APX	FLX
S_1	20	18	-	-	16	-	-	15	16	-
S_2	19	13	10	-	-	18	18	-	-	-
S_3	10	10	16	-	18	-	20	-	-	-
S_4	21	19	10	-	15	17	19	14	18	-
S_5	16	16	-	-	11	-	-	16	-	-
S_6	18	10	18	5	-	-	10	10	13	5
S_7	12	11	13	6	11	16	12	12	10	6
S_8	18	18	17	-	15	9	11	-	9	-
S_9	17	9	12	8	16	-	18	11	-	-
S_{10}	20	16	11	-	-	11	-	15	-	4

Key: S = S. aureus, CPX = Ciprofloxacin (10 µg), NB = Norfloxacin (10 µg), CN = Gentamycin (10 µg), LC = Lincocin (20 µg), S = Streptomycin (30 µg), RD = Rifampin (20 µg), E = Erythromycin (30 µg), CH = Chloramphenicol (30 µg), APX = Ampiclox (20 µg), FLX = Floxapen (20 µg), - = No inhibition

P. aeruginosa isolates (Ps_1, Ps_2, Ps_3, Ps_4, Ps_5 and Ps_6) were highly sensitive to ciprofloxacin with inhibition zone diameter of 20 mm, 18 mm, 19 mm, 13 mm, 16 mm and 16 mm respectively, followed by streptomycin with inhibition zone diameter of 18 mm, 18 mm, 16 mm, 13 mm and 15 mm for *P. aeruginosa* isolates (Ps_1, Ps_2, Ps_3, Ps_5 and Ps_6). Highest level of resistance was observed with ceporex which showed inhibition zone diameter of 10 mm and 9 mm for *P. aeruginosa* isolates (Ps_1 and Ps_6) and showed no inhibition against the other isolates (Table 4).

They were highly sensitive to ciprofloxacin with inhibition zone diameter of 20 mm and 19 mm, followed by streptmomycin with inhibition zone of 18 mm and 18 mm for the *E. coli* isolates with isolate code Ec_1 and Ec_2 respectively. But showed highest resistance to ceporex with inhibition zone diameter of 6 mm to *E. coli* isolates (Ec_2) and showed no inhibition zone to Ec_1, followed by ampicillin as shown in Table 5.

Table 4. *Inhibition zone diameter (IZD) of Pseudomonas aeruginosa against antibiotics*

Isolate code	CPX	AUG	CN	S	CEP	NA	SXT	PN	PEF	OFX
Ps_1	20	16	15	18	10	11	18	10	16	18
Ps_2	18	15	16	18	-	16	-	15	18	17
Ps_3	19	17	18	16	-	10	-	18	19	13
Ps_4	13	-	-	-	-	-	-	-	-	-
Ps_5	16	12	-	13	-	-	-	11	-	12
Ps_6	16	10	18	15	9	-	9	10	15	14

Key: Ps = Pseudomonas aureus, CPX = Ciprofloxacin (10 µg), AUG = Augmentin (30 µg), CN = Gentamycin (10 µg), S = Streptomycin (30 µg), CEP = Ceporex (10 µg), NA = Nalidixic acid (30 µg), SXT = Septrin (30 µg), PN = Ampicillin (30 µg), PEF = Peflacin (10 µg) OFX = Tarivid (10 µg), - = No inhibition

Table 5. *Inhibition zone diameter (IZD) of Escherichia coli against antibiotics*

Isolate code	CPX	AUG	CN	S	CEP	NA	SXT	PN	PEF	OFX
Ec_1	20	18	19	18	-	12	-	12	16	20
Ec_2	19	12	17	18	6	10	16	-	15	-

Key: Ec = Escherichia coli, Ciprofloxacin (10 µg), AUG = Augmentin (30 µg), CN = Gentamycin (10 µg), S = Streptomycin (30 µg), CEP = Ceporex (10 µg), NA = Nalidixic acid (30 µg), SXT = Septrin (30 µg), PN = Ampicillin (30 µg), PEF = Peflacin (10 µg) OFX = Tarivid (10 µg), - = No inhibition

Table 6. *percentage susceptibility and resistance pattern of bacterial isolates*

Isolate code	CPX	AUG	CN	LC	S	RD	E	CH	APX
S. aureus									
Susceptible %	8(80.0)	6(60.0)	3(30.0)	0(0.0)	5(50.0)	3(30.0)	4(40.0)	4(40.0)	2(20.0)
Resistance %	2(20.0)	4(40.0)	7(70.0)	10(100.0)	5(50.0)	7(70.0)	6(60.0)	6(60.0)	8(80.0)
P. aeruginosa									
Susceptible %	5(83.3)	NT	4(66.7)	NT	4(66.7)	NT	NT	NT	NT
Resistance %	1(16.7)	NT	2(33.3)	NT	2(33.3)	NT	NT	NT	NT
E. coli									
Susceptible %	2(100.0)	NT	2(100.0)	NT	2(100.0)	NT	NT	NT	NT
Resistance %	(0.0)	NT	(0.0)	NT	(0.0)	NT	NT	NT	NT

Table 6. *continues*

Isolate code	FLX	SXT	AUG	CEP	NA	PN	PEF	OFX
S. aureus								
Susceptible %	0(0.0)	NT	NT	NT	NT	NT	NT	NT
Resistance %	10(100.0)	NT	NT	NT	NT	NT	NT	NT
P. aeruginosa								
Susceptible %	NT	1(16.7)	1(16.7)	0(0.0)	1(16.7)	2(33.3)	4(66.7)	2(33.3)
Resistance %	NT	5(83.3)	5(83.3)	6(100.0)	5(83.3)	4(66.7)	2(33.3)	4(66.7)
E. coli								
Susceptible %	NT	1(50.0)	1(50.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(50.0)
Resistance %	NT	1(50.0)	1(50.0)	2(100.0)	2(100.0)	2(100.0)	2(100.0)	1(50.0)

Key: NT = Not tested

The plasmid profile of the isolates *Escherichia coli* and *Staphylococcus aureus* with isolated code Ec_1, S_1 and S_4 are shown in Figure 2 below.

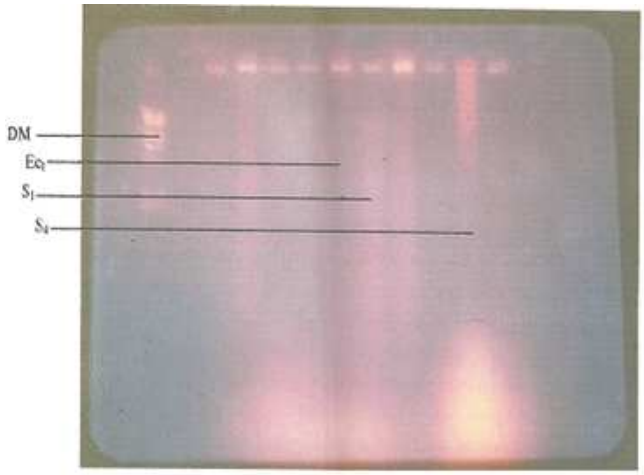

Figure 2. *Plasmid profile of the multiantibiotic resistance Escherichia coli (Ec_1) and Staphylococcus aureus (S_1 and S_4).*

Key: DM = DNA Marker

Table 7. *Number and plasmid DNA with corresponding molecular weight*

Isolate Code	Number of plasmids	Mobility (mm)	Molecular weight (kbp)
S_1	3	12,15 and21	6.0, 4.6 and 1.8
S_2	Nil	Nil	Nil
S_3	Nil	Nil	Nil
S_4	2	9 and 12	10.4 and 6.0
S_5	Nil	Nil	Nil
Ec_1	1	15	4.6
Ps_1	Nil	Nil	Nil
Ps_2	Nil	Nil	Nil
Ps_3	Nil	Nil	Nil
Ps_4	Nil	Nil	Nil
Ps_5	Nil	Nil	Nil

The number, electrophoretic mobility and corresponding molecular weight of plasmid DNA analyzed are presented in Table 7.Out of the ten isolates (S_1, S_2, S_3, S_4, S_5, Ec_1, Ps_1, Ps_2, Ps_3, Ps_4 and Ps_5) analyzed, only three isolates (S_1, S_4 and Ec_1) showed the presence of plasmids. S_1 revealed 3 plasmids with mobility of 12 mm, 15mm and 21 mm and the corresponding molecular weight of 6.0 kbp, 4.6 kbp and 1.8 kbp respectively; S_2 showed the presence of 2 plasmids with mobility of 9 mm and 12 mm and corresponding molecular weight of 10.4 kbp and 6.0 kbp respectively; While Ec_1 showed the presence of only 1 plasmid with mobility of 15 mm and 4.6 kbp.

4. Discussion

Wound infection is a major concern among healthcare practitioners, not only in terms of increased trauma to the patient, but also in view of its burden on financial resources and increasing requirement for cost-effective management within healthcare system (1). The result of this study reveals the presence of *Staphylococcus aureus, Pseudomonas aeruginosa* and *Escherichia coli* (Table 1 and 2) at the percent rate of 55.5%, 33.3% and 11.1% respectively (Figure 1). This could be as a result of the fact that healthcare workers carry these microorganisms in their wears and stand the chance of transmitting them to immunocompromized wound patients (15). Also, these organisms are opportunistic and can only cause infection in patients with breached immunity; this can also be seen in wounds caused by trauma or open wounds. For instance, *S. aureus* is a skin flora and can also enter and colonize open wounds, thereby causing infection therein (1). The ubiquity or versatility *Pseudomonas aeruginosa* around the hospital environment cannot be over emphasized.

This result of this study is similar to the report of Shittu *et al* (16) in Ile-Ife, who showed that *S. aureus* (25.3%), followed by *P. aeruginosa* (9.3%), *E. coli* (12.3%), *K. pneumonia* (1.9%) among others were isolated from wound infections. Alghalibi *et al*. (17) reported the presence of *S. aureus* (47.8%), *P. aeruginosa* (23.0%), *E. coli* (5.3%) as part of the organisms isolated from burnt wound infection in Teiba Center for Burns, Sana'a City, Yemen. Sani *et al*. (18) observed that *S. aureus* was more frequently isolated (62.0%) than *Streptococcus pyogenes* (38.0%) from wounds infections in Niger State. Subsequently, this result is also similar to the work of Tran *et al*. (19) in Thailand; Emele *et al.* (20) in Edo State-Nigeria; Mashita*et al*. (21) in Japan; Rasteger and Alaghehbundan (22), in Iran, as well as Yah *et al*., (15), in Benin City- Nigeria, Nwachukwu *et al.* (23) in Abia State-Nigeria, Ohalete *et al*. (24) in Imo State-Nigeria. This showed that *S. aureus* is the leading aetiologic agent of wound infections, followed by *P. aeruginosa.* This could be as a result of the varying decree of possibility of patients' encounter with the agents. For instance, *S. aureus* was most prevalent in view of its existence as a normal flora of the skin having greater possibility of entering and colonizing the wound once there is trauma (25). *P. aeruginosa* occurs in high prevalence rate in hospital equipments and environment including hospital beds. This could be the cause of its high prevalence against *E. coli*, which is strictly an enteric organism (26). But it is in conflict with the result of Etok *et al*., (27) in Awa-Ibom State, Nigeria, who had more *E. coli* following *Proteus* spp. and *Pseudomonas* spp. in percentage occurrence. The variation in the results could be as a result of the differences in the duration of stay of the patients in the hospital, as Yah *et al.* (15) demonstrated that high prevalence of *P. aeruginosa* is recorded in patients having prolonged stay in the hospital.

Furthermore, result of antibiotic susceptibility test (Table 3, 4 and 5) carried out on the three isolates revealed that out of 10 isolates of *S. aureus* used in this study, 8(80%) were susceptible to ciprofloxacin, 6(60%) to norfloxacin and 5(50%) to streptomycin; the least (20%) sensitivity was recorded with ampiclox. This is in agreement with the result of Udoh and Njirinze (28), in Uyo, Nigeria, Tillotson *et al*. (29) in USA, but contrary to the work conducted in Michael Okpala University of Agriculture, Umudike, Abia State by Chigbu and Ezeronye (30) and in Zaria, Nigeria (31). Also, 100% resistance was recorded with lincocin and floxapen, followed by ampiclox (80%), gentamicin and rifampincin (70% each), chloramphenicol and erythromycin (60% each); least resistance was obtained with Ciprofloxacin. This is similar to the study result of Uwaezuoke and Aririatu (32), in Imo State University, in which 66.7% and 10.4% sensitivities were recorded for streptomycin and ampiclox respectively. Also in line with work of Sani *et al*. (18) in Niger State, Nigeria, where reasonable resistance was recorded with Erythromycin (60%), ampiclox (54.0%) and gentamycin (39%).

More so, out of 6 isolates of *P. aeruginosa* used, 83.3% sensitivity was recorded with ciprofloxacin, followed by peflacin, gentamycin and streptomycin (66.7% each) and then augmentin (50%); least sensitivity was found with nalidixic acid and septrin (16.7% each). A 100% resistance of *P. aeruginosa* was obtained with ceporex, followed by 83.3% each of nalidixic acid septrin; least resistance was observed with ciprofloxacin (16.7%) (Table 6). This result is in agreement with the work of Udoh and Njirinze (28) in Uyo, Akwa Ibom State where ciprofloxacin and nalidixic acid gave 66.7% sensitivity and 100% resistance respectively. Subsequently, the result of this work is in consonance with the work reported by Akingbade *et al*. (2) in South West Nigeria, Masaadeh and Jaran (33) in Irbid, Jordan, and Anjum and Mir (34) in Islamabad, Pakistan.

Also observed in this study is 2 isolates of *Escherichia coli* tested against various antibiotics (Table 6). All showed 100% sensitivity against ciprofloxacin, gentamycin, peflacin, and streptomycin. 50% sensitivity was recorded against septrin, augmentin and tarivid. On the other hand, 100% resistance was recorded against ceprorex. nalidixicacid, and ampicillin.

Generally, the quinolones (ciprofloxacin and norfloxacin) followed by the aminoglycoside showed some degree of effectiveness on the isolates; this could be as a result of the fact that they have not been exposed to intensive abuse in our community. However lincocin, floxapen, ceprorex, nalidixic acid, septrin and ampicillin proved ineffective. This could be attributed to poor storage of antibiotics, abuse of antibiotics by patients as a result of self-prescription, misdiagnosis by clinicians as well as due to the emergence of resistance, which could be plasmid or chromosomes borne. A similar report about quinolones has been made by Yah *et al*. (15), in Benin City, Nigeria.

Interestingly, in the view of high resistance recorded in this work against most of the antibiotics, a plasmid analysis was carried out on 10 isolates out of 18 isolates used in this study. This was to check whether or not the resistance was plasmid borne. The result of the plasmid analysis revealed that only 3 isolates out of 10 isolates had plasmids (Table 7). The three isolates were 2 *Staphylococcus aureus* (S_1 and S_4) and 1 *Escherichia coli* (Ec_1). The *Escherichia coli* isolates had only one plasmid with molecular weight of 4.6kbp and mobility of 15 mm while the two isolates of *S. aureus*(S_1 and S_4) had 3 and 2 plasmids respectively with molecular weights of 6.0 kpb and 1.8 kpb and 10.4 kpb and 6.0 kpb respectively and mobility of 12 mm; 15 mm; 21 mm and 9 mm; 12 mm respectively. The study indicated that although S_1 (having 3 plasmids) showed reasonable degree of sensitivity. Also, the *E. coli* was sensitive to a good number of antibiotics. The result revealed that the presence of plasmids DNA does not always suggest resistance to antibiotics, as there are different types of plasmids performing different functions of bacteria (35). For instance the plasmid contained in the *E. coli* isolate could be a virulence or col plasmid and not R-plasmid. Also, Ps_4 (*Pseudomonas aeruginosa*) isolate showed little sensitivity to only one antibiotic (Table 4), yet had no plasmids on analysis. Also, S_5 and PS_5 were sensitive to only a few antibiotics, yet they did not have plasmids on analyses (Table 3 and 4). The result is suggestive to the fact that antibiotic resistances are not always plasmid borne (35,36). The result of plasmid analysis also showed that the increase in molecular weight had a retarding effect on the mobility of rate of the plasmid; an increase in molecular weight reduced the mobility of the plasmid. This is compared favorably with the work of Anyim *et al.* (37), which confirmed that the higher the molecular weight of a plasmid the lower the electrophoretic mobility. It was also suggested that the DNA molecule is negatively charge, migrating through an agarose gel towards the anode at a rate which is dependent upon the molecular weight (38).

The above results were also supported by a curing experiment carried out on the 10 isolates used for plasmid profiling. After curing, all the isolates were sensitive including S_1 (which was formally resistant and had 3 plasmids) suggesting that the resistance obtained in S_1 was plasmid-mediated rather than chromosome-mediated and that other resistance recorded was not associated with the presence of plasmids. This is compared favorably with the work of Yah *et al*. (15), who demonstrated that 48.9% of their antibiotics resistance was chromosome-mediated while 39.7% antibiotics resistance could not be ascertained.

5. Conclusion

In conclusion, there is an alarming increase of infections caused by antibiotic-resistant bacteria. This study has highlighted diverse plasmid profiles and wide spread antimicrobial resistance patterns of some clinical strains of *Staphylococcus aureus, Pseudomonas* a*eruginosa* and *E. coli* from Nigeria. Therefore the rational use of antimicrobials must be a priority. Public health policy on

appropriate prescribing and use of antibiotics must be instituted and affected. Subsequently, antibiotic resistance of an organism does not always confer the presence of plasmid.

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