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Antimicrobial Resistance Among Common Bacterial Pathogens in South Western Nigeria

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Abstract: The phenomenal increase in antibiotic resistant bacterial pathogens calls for regular review of antimicrobial sensitivity pattern among bacteria of clinical significance in an environment. This study was conducted to determine the common clinically significant bacterial pathogens in this environment and their antimicrobial susceptibility pattern. Bacterial pathogens isolated from various clinical specimens brought to the diagnostic laboratory of the University College Hospital between May and October 2005 were subjected to antimicrobial sensitivity testing using the disc diffusion and minimum inhibitory concentration tests. The most prevalent bacterial pathogen was found to be *Staphylococcus aureus* (47.5%), followed by *Pseudomonas aeruginosa* (24.6%), Klebsiella species (23%), Proteus species (3.3%) and *Escherichia coli* (1.6%). *P. aeruginosa* (24.6%) rated highest in frequency among the Gram-negative organisms while *S. aureus* was the main Gram-positive pathogen isolated. Generally, resistance rates to most of the antibiotics tested were high among both Gram-positive and Gram-negative isolates. However, both groups showed good susceptibilities to gentamycin and ciprofloxacin. Resistance to the old generation antibiotics such as streptomycin and chloramphenicol, nevertheless, remained persistently high. This study has further revealed that continued surveillance of changes in resistance patterns of bacterial pathogens to antibiotics is of utmost importance if effective management of infectious diseases is to be ensured.

Key words: Antimicrobial resistance · Bacteria · Nigeria

INTRODUCTION

Bacterial infections continue to be important causes of morbidity and mortality in developing countries [1]. However, there is a phenomenal increase in antibiotic resistant bacterial pathogens which is one of the major problems facing medicine and science today. This calls for regular review of the antimicrobial sensitivity pattern among bacteria of clinical significance in our environment [2]. There are many reasons for this alarming phenomenon and one of them is widespread and indiscriminate use of antibiotics which have been implicated in the development of serious problems of resistance to the older and less expensive antimicrobial agents. These include penicillin, ampicillin, co-trimoxazole, tetracycline and chloramphenicol [3]. Studies in Lagos and Ibadan have also shown that between 70% and 90% of strains of Enterobacteriaceae including *E. coli*, *Klebsiella* and *Proteus species* are resistant to many of the commonly available antibiotics and this, in many cases, has led to the use of newer and more expensive agents [2].

This resistance is associated with greater hospital mortality and longer duration of hospital stay [4, 5]. Moreover, infection with antibiotic resistant bacteria will make the therapeutic options for treatment rather difficult or virtually impossible [6]. The knowledge of prevailing susceptibility patterns is therefore vital to the selection and use of antimicrobial agents and to the development of appropriate prescribing policies [7]. This study was therefore conducted to determine the common and clinically significant bacteria isolates in this environment and their antimicrobial susceptibility pattern. This is to guide in antibiotic choice as well as in formulation of policy for the rational and effective use of antimicrobial agents.

MATERIALS AND METHODS

This study was conducted in the diagnostic laboratory of University College Hospital, Ibadan, Nigeria between May and October 2005. Specimens were collected from patients who presented in the hospital

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with various clinical diagnosis such as urinary tract infections, otitis media, pneumonia, bacterial conjunctivitis, wound sepsis etc. Information regarding patient's name, occupation, age, sex, ward or clinic, type of specimen taken were also recorded. Six hundred and ten strains of bacterial pathogens were isolated and identified from these various clinical specimens by conventional methods. Susceptibility testing was performed using both disc diffusion sensitivity and minimum inhibitory concentration (MIC) tests.

Susceptibility Testing: Antimicrobial disc susceptibility tests of the isolates were performed according to the recommendations of the National Committee for Clinical Laboratory Standards (NCCLS) [8] using the following antibiotic discs: amoxicillin/clavulanate (30ug), cefotaxime (30ug), chloramphenicol (10ug), ciprofloxacin (5ug), gentamycin (10ug), penicillin (2IU), streptomycin (10ug) and cefuroxime (30ug). Control organisms used as standards were *Staphylococcus aureus* NCTC 6571, *Escherichia coli* NCTC 10418 and a local sensitive strain of *Pseudomonas aeruginosa.*

Minimum inhibitory concentration estimations were determined by the microtube broth dilution technique as described in the NCCLS [8] guidelines with the same antibiotics mentioned above. The MIC was taken as the lowest antibiotic concentration that prevented the growth of the organism. The plates from the disc diffusion test were examined after overnight incubation at 37°C and the inhibition zone diameters were measured and compared with standards.

RESULTS

Six hundred and ten bacterial pathogens were recovered and identified from various clinical specimens. These included wound swabs 223(37.7%), blood 60(9.8%), sputum 40(6.6%), ear swab 100(16.4%) conjunctival swab 120(19.7%), urine 47(6.6%) and wound aspirate 20(3.3%).

Gram-negative bacilli accounted for 320(52.5%) of the isolates while 290(47.5%) were Gram-positive cocci. *S. aureus* 290(47.5%) was the most frequently isolated pathogen, followed by *P. aeruginosa* 150(24.6%), Klebsiella species 140(23%), *P. mirabilis* 20(3.3%) and *E. coli* 10(1.6%) (Table 1). Pseudomonas and Klebsiella species accounted for 290(47.6%)of the Gram-negative bacilli while *P. mirabilis* and *E. coli* make up the remaining 30(4.9%). Gram-positive cocci were predominantly *S. aureus* 290(47.5%).

Table 1: Frequency of Isolation Of Bacterial Pathogens From Different Specimens

Specimens								
Isolates	Ear Swab No (%)	Wound Swab No (%)	Conjunctival Swab No (%)	Urine No (%)	Sputum No (%)	Blood No (%)	Wound Aspirate No (%)	Among All Specimens No (%)
Klebsiella species	20 (20.0)	40 (17.9)	-	30 (63.8)	30 (75.0)	10 (16.7)	10 (50.0)	140 (23.0)
Pseudomonas aeruginosa	50 (50.0)	60 (26.9)	20 (16.7)	10 (21.3)	10 (25.0)	-	-	150 (24.6)
Escherichia coli	-	5 (2.4)	-	5 (10.6)	-	-	-	10 (1.6)
Proteus species	-	18 (8.1)	-	2 (4.3)	-	-	-	20 (3.3)
Staphylococcus aureus	30(30.0)	100 (44.8)	100 (83.3)	-	-	50 (83.3)	10 (50.0)	290 (47.5)
Total	100(100.0)	223 (100.0)	120 (100.0)	47 (100.0)	40 (100.0)	60 (100.0)	20 (100.0)	610 (100.0)

Table 2: Antibiotic Sensitivity Pattern of Bacterial Pathogens

Antimicrobial Agents								
Isolates	Amoxicillinclavulanate No (%)	Cefotaxime No (%)	Chloramphenicol No (%)	Ciprofloxacin No (%)	Gentamycin No (%)	Penicillin No (%)	Streptomycin No (%)	Cefuroxime No (%)
Klebsiella species(140)	40 (28.6)	60 (42.9)	30 (21.4)	80 (57.1)	100 (71.4)	0(0.0)	5 (3.6)	4 (2.9)
Pseudomonas aeruginosa (150)	0(0.0)	40 (26.7)	0 (0.0)	70 (46.7)	100 (66.7)	0(0.0)	0(0.0)	70 (46.7)
Escherichia coli (10)	2(20.0)	4 (40.0)	3 (30.0)	10 (100.0)	8 (80.0)	0(0.0)	0(0.0)	5 (50.0)
Proteus species (20)	5 (25.0)	10 (50.0)	0(0.0)	10 (50.0)	10 (50.0)	0(0.0)	5 (25.0)	15 (75.0)
Staphylococcus aureus (290)	120(41.4)	170 (58.6)	70 (24.1)	210 (72.4)	270 (93.1)	0(0.0)	40 (13.8)	140 (48.3)

Table 3: Minimum Inhibitory Concentration (MIC) of a Cumulative Percentage of Isolates

Antimicrobial Agents (μ/ml)								
	Amoxicillinclavulanate	Cefotaxime	Chloramphenicol	Ciprofloxacin	Gentamycin	Penicillin	Streptomycin	Cefuroxime
	MIC 50	MIC 50	MIC 50	MIC50	MIC50	MIC50	MIC50	MIC50
	MIC range	MIC range	MIC range	MIC range	MIC range	MIC range	MIC range	MIC range
Isolates	MIC 90	MIC 90	MIC 90	MIC 90	MIC 90	MIC 90	MIC 90	MIC 90
Klebsiella	32	0.5	8	2	1	64	4	4
species (140)	0.125-64	0.125-64	0.125-64	0.25-32	0.125-128	64-256	05-256	0.5-128
	64	64	64	32	32	256	256	8
Pseudomonas	64	8	16	16	0.5	64	64	0.5
aeruginosa (150)	16-256	0.125-64	16-256	0.125-64	0.125-16	64-256	16-128	0.25-64
	256	64	64	64	8	256	128	64
Escherichia	16	8	16	1.0	2	64	64	2
coli (10)	8-64	4-64	16-128	1.0-64	2-16	64-256	64-128	1.0-32
	64	64	128	8	16	256	128	16
Proteus	32	16	64	4	16	64	32	2
species (20)	16-64	16-64	64-256	4-32	8-64	64-256	32-64	0.5-64
	64	64	256	32	64	256	64	64
Staphylococcus	16	0.25	16	0.25	0.125	64	8	0.125
aureus (290)	0.125-128	0.125-64	0.5-128	0.125-64	0.125-64	32-256	0.5-128	0.25-64
	128	64	128	16	1.0	128	64	8

In vitro activities of eight different antibiotics against the bacterial isolates are illuminated in Tables 2 and 3. Resistance rates were high among both Gram-positive cocci and Gram-negative bacilli isolates. The number of strains resistant to amoxicillin/clavulanate was 100(71.4%) in Klebsiella species, 8(80%) in *E. coli* and 15(75%) in *P. mirabilis*. A high resistance rate to cefotaxime was also recorded in *P. aeruginosa 110*(73.3%), as well as in *E. coli* 6(60%), Proteus 10(50%) and Klebsiella species 80(57.1%). *P. aeruginosa* 80(53.3%) and *E. coli* 5(50%) also showed high resistance to cefuroxime. However, resistance to cefuroxime by Proteus was 5(25%). Majority of these Gram-negative organisms showed good susceptibilities to gentamycin. These are klebsiella species 100(71.4%), *P. aeruginosa* 100(66.7%), *E. coli* 8(80%) but that of Proteus species was 10(50%).

The susceptibilities displayed by these organisms to ciprofloxacin were intermediate as shown in Table 2. *S. aureus*, however, showed high susceptibility to ciprofloxacin 210(72.4%), gentamycin 220(93.1%) and a fair one to cefotaxime 170(58.6%). However, it showed a high resistance to streptomycin 250(86.2%) and chloramphenicol 220(75.9%) (Table2). Resistance to cefuroxime among Gram-positive and negative organisms were about the same except in Klebsiella where it is much higher 136(97.1%) and Proteus, where it is much lower 5(25%).

DISCUSSION

The most frequently isolated pathogen in this study was *Staphylococcus aureus* (47.5%) followed by *P. aeruginosa* (24.6%) and Klebsiella species (23%). This contradicts reports from other countries such as China [9], Egypt [10] and Israel [11] where *E. coli* was reported to be the most frequently isolated pathogen, followed by *S. aureus* and *P. aeruginosa.*

There are also high rates of antimicrobial resistance reported among these bacterial pathogens, which are in keeping with the results of studies conducted by Goosens in Belgium [12] El-Astal in Palestine [7] and Bakare in Nigeria [13]. Some strains of Klebsiella, Pseudomonas and Proteus species were resistant to ciprofloxacin, a quinolone, with MICs > 8ug/ml. This result is quite different from the report of Ogunsola et al who documented a 100% sensitivity of *P. aeruginosa* to ciprofloxacin in 1994 in Lagos, Nigeria [14]. However, the report is similar to those documented in other populations [15]. The emergence of resistance to quinolones by *P. aeruginosa* has also been reported in patients with complicated urinary infection [16]. High resistance to cefuroxime and cefotaxime was reported among all the Gram-negative bacteria in this study except Proteus. This resistance was also observed to be marked in *P. aeruginosa* and *E. coli.* In *S. aureus*, there is high susceptibility to ciprofloxacin and gentamycin and this picture is similar to the findings of Okesola *et al.* in 1999 [17]. Susceptibility to gentamycin is also high among the Gram-negative organisms though it is rather low in Proteus species. There was a generally high resistance pattern exhibited towards amoxicillin/clavulanate by all the Gram-negative organisms and *S. aureus.*

CONCLUSIONS

The high level of resistance exhibited by many of the pathogens in this study has demonstrated that the call for judicious use of antimicrobial agents cannot be overemphasized. The study has further confirmed the intrinsic resistance of Klebsiella and Pseudomonas species to most antibiotics, a situation which favours their continued existence in hospital environment. This high resistance rate may not be unconnected with widespread, indiscriminate and inappropriate use of antibiotics which is rampant in this environment. This calls for the education of both medical and paramedical staff on the rational use of antibiotics. Furthermore, the community at large must also be enlightened through regular health education programmes on the dangers inherent in self-medication.

It is also pertinent to say here that continued surveillance of changes in resistance patterns of these pathogens to antimicrobial agents is of utmost importance if effective management of infectious diseases is to be ensured.

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