Full Length Research Paper

Prevalence of community-associated multi-resistant *Staphylococcus aureus* among healthy women in Abuja, Nigeria

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Antimicrobial resistance has become a great public health problem worldwide and multi-drug resistance *Staphylococcus aureus* has been widely reported. This study determined the pattern of resistance to ten commonly used antibiotics. Urine samples collected from healthy women volunteers in the Abuja were cultured and screened for *S. aureus* using standard microbiological procedures. The antibiotic susceptibility of the isolates was investigated using disc diffusion technique. A total of 60 (40%) *S. aureus* isolates were isolated from 150 urine samples collected. Of these, 19 (32%), 22 (36%) and 19 (32%) were from married but not pregnant, pregnant and single women, respectively. The isolates were highly resistant to ampicillin (91.7%), clindamycin (78.3%), cephalexin (75%), methicillin (71.7%) and vancomycin (68.3%) but had very low resistance to gentamicin (3.3%), ciprofloxacin (3.3%), ofloxacin (3.3%), sparfloxacin (3.3%) and pefloxacin (10.0%). A total of 43 (71.7%) of the isolates showed multi-drug resistance and only 3 (5%) were susceptible to all the antibiotics tested. Multi-drug resistant *S. aureus* is highly prevalent in the urine of healthy women investigated in Federal Capital Territory. This calls for effective measures against irrational use of antibiotics.

Key words: Prevalence, community-associated, antibiotics, multi-resistant, *Staphylococcus aureus,* healthy women.
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INTRODUCTION

The widespread use of antibiotics has been responsible for the development of numerous problems including the emergence of multidrug resistant bacteria, increased number of nosocomial (hospital) and community-acquired infections; and increased health care costs (Snyder et al., 2000). *Staphylococcus aureus* is a pathogen of greater concern because of its virulence (Chambers, 2005), its ability to cause a diverse array of life threatening infections, and its capacity to adapt to different environmental conditions (Lowy, 1998; Lowy, 2003).

The development of resistance to penicillin in *S. aureus* is due to the production of β-lactamases; enzymes that destroy the antibiotic by hydrolysing the β-lactam ring and this quickly decreased the usefulness of penicillin for serious staphylococcal infections especially among hospitalised patients in whom resistant strains were frequently found before they spread to the community. These community-acquired strains are now uniformly resistant to all β-lactam antibiotics and show cross-resistance to other antimicrobial agents (Olayinka et al., 2004; Paul et al., 1982).

The level of antibiotic resistance in a given community or hospital can be predicted by these important measures; the proportion of resistant organisms introduced from outside the population, the extensive use

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Table 1. Frequency of Isolation of *Staphylococcus aureus* from three groups of women in Abuja, Nigeria.

Source	Number Sampled	S. aureus	
		No	**(%)**
Married	50	19	(838)
Pregnant	50	22	(44)
Single	50	19	(38)
Total	**150**	**60**	**(40)**

of antimicrobial agents and the proportion that is spread from person to person (Wenzel and Edmond, 2000). This study was carried out to determine the prevalence and multiple antibiotic resistance profiles of *S. aureus* isolated from urine samples of women resident in Abuja, Nigeria.

MATERIALS AND METHODS

Media

Mannitol salt Agar, Mueller Hinton Agar 11, nutrient agar and nutrient broth media all from LAB M (International Diagnostics Groups Plc, United Kingdom) were used.

Sample collected

First 'clean catch" urine sample were collected randomly from 150 healthy women who were classified into 3 groups (single, married but not pregnant, and pregnant women of ages between 20 to 40 years) over a period of 2 months from Abuja community after informed consent had been obtained from each woman. All the Volunteers were not on any antibiotic at the point of sampling. Samples (50 from each group) were collected into labelled sterile bottles, kept in an iced-bag and transported to the laboratory.

Bacteriology

On arrival at the laboratory, each urine sample was immediately inoculated (in duplicates) into mannitol salt agar plates and inoculated aerobically at 37°C for 24 h. The characteristic isolates were asceptically isolated and characterised using established microbiological methods that include colonial morphology, Gram stain characteristics, and catalase and coagulase tests (Cheesbrough, 2002;). Isolates that were Gram-positive cocci, catalase positive and coagulated human plasma were considered as *S. aureus* in this study.

Antibiotic susceptibility testing

The antibiotic susceptibility pattern of all isolated *S. aureus* to 10 µg ampicillin (Medreich, India), 30 µg cephalexin (Fidson, India), 5 µg ciprofloxacin (Fidson, India), 2 µg clindamycin (Pharmacia, Belgium), 10 µg gentamicin (Wuham, China), 10 µg methicillin (Oxoid, UK), 5 µg ofloxacin (Pathoteq Lab, India), 5 µg pefloxacin (Fidson, India), 5 µg sparfloxacin (Pathoteq Lab. India) and 30 µg vancomycin (Dumex-Alphama, S. Demark) were determined by the modified Kirby-Bauer diffusion technique (Cheesbrough, 2002). Standardised overnight culture of each isolate (containing 10^8 cfu/ml) was used to flood the surface of Mueller Hinton agar (MHA)

plates and excess drained off and dried while the petri dish lid was in place. The standard antibiotic discs were then asceptically placed at reasonable equidistance on the inoculated MHA plates and allowed to stand for 1 h. The plates (prepared in duplicates for each isolate) were then incubated at 37°C for 18 h (Ehinmidu, 2003). The diameter of the zone of inhibition produced by each antibiotic disc was measured, recorded and isolates were classified as "resistant", "Intermediate sensitive" or "sensitive" based on the standard interpretative chart updated according to the current NCCLS standard (Cheesbrough, 2002; NCCLS, 2002) and Fluka zone interpretative chart in accordance with WHO requirements.

RESULTS

For the purpose of this study, community-associated isolates were defined as isolates from the samples of the healthy women who were not on any antibiotic at the time of sampling and had not been admitted in hospital in the last one year. A total of 60 *S. aureus* were isolated from 150 urine samples screened. A total prevalence rate of 40% (60/150) was obtained and details of single point prevalence in each group are shown in Table 1.

The results of antibiotic susceptibility test showed that the isolates were highly resistance to ampicillin (91.7%), clindamycin (78.3%), cephalexin (75%), methicillin (71.7%) and vancomycin (68.3%) but had very low resistance to gentamicin (3.3%), ciprofloxacin (3.3%), ofloxacin (3.3%), sparfloxacin (3.3%) and pefloxacin (10.0%) as shown in Table 2. The prevalence of multi-drug resistance in the *S. aureus* isolates is shown in Table 3. Multi-drug resistance was defined in this study as resistance to four or more of the antibiotics tested. Thus, 43 (71.7%) of the isolates showed multi-drug resistance to the antibiotics and only 3 (5%) were susceptible to all the tested antibiotics.

DISCUSSION

S. aureus, a worldwide pathogen whose natural reservoir is human causes severe community-associated infections of skin and soft-tissue (Noble et al., 1967; Nordmann and Nass, 2005), and it is increasingly developing resistant to many antibiotics (Lowy, 2003). An overall prevalence of 40% of *S. aureus* was obtained from urine samples of healthy women in this study. This is in agreement with

Table 2. Antibiotic resistance pattern of *S. aureus* isolates from urine sample of women in Abuja, Nigeria.

Antibiotics	Number/60	(% Resistant)
Ampicillin (10 µg)	55	(91.7)
Cephalexin (30 µg)	45	(75.0)
Ciprofloxacin (5 µg)	2	(3.3)
Clindamycin (2 µg)	47	(78.3)
Gentamicin (10 µg)	2	(3.3)
Methicillin (10 µg)	43	(71.7)
Ofloxacin (5 µg)	2	(3.3)
Pefloxacin (5 µg)	6	(10.0)
Sparfloxacin (5 µg)	2	(3.3)
Vancomycin (30 µg)	41	(68.3)

Table 3. Prevalence of multi-drug resistance among 60 *S. aureus* isolates.

	Number	(%)
Fully Sensitive	3	(5.0)
Resistant to 1 agent	5	(8.3)
Resistant to 2 agents	5	(8.3)
Resistant to 3 agents	4	(6.7)
Resistant to 4 agents	7	(11.7)
Resistant to 5 agents	31	(51.7)
Resistant to 6 agents	3	(5.0)
Resistant to 7agents	1	(1.7)
Resistant to 8 agents	0	(0)
Resistant to 9 agents	0	(0)
Resistant to 10 agents	1	(1.7)

the report of Ehinmidu (2003) who reported 43.3%. In this study, the highest level of resistance is observed in ampicillin (91.7%), which is in agreement with the reports of Umolu et al. (2002) and Ehinmidu (2003). The resistance to cephalexin and clindamycin is in conformity with previous observations that most isolates of *S. aureus* are resistant to a large number of commonly prescribed antibiotics (Olukoya et al., 1995). The percentage resistance to methicillin and vancomycin is alarming and has been widely reported internationally (Fridkin, 2001; Hiramatsu et al., 1997) and even in our communities (Ikeh, 2003; Olayinka et al., 2005). The resistance may be due to the acquisition of resistance determining genes, like *mecA* (methicillin resistance) and *vanA, B, C* responsible for vancomycin resistance (Hiramatsu et al., 1997; Kim et al., 2000) or as a result of the thickening of the cell wall as reported by some authors (Kim et al., 2000; Denis et al., 2002). These were however not determined in this work.

Most of the isolates were highly susceptible to gentamicin, ofloxacin, ciprofloxacin, pefloxacin and sparfloxacin, which is in agreement with previous reports (Umolu et al., 2002; Ehinmidu, 2003; Olayinka et al., 2004). High susceptibility to gentamicin though very cheap, may be due to the complexity of the aminoglycoside and the route of administration. The fluoroquinolones are newer drugs with mode of action central on inhibition of the DNA replication which stops the multiplication of the bacteria cells and are relatively expensive therefore they are more likely less available for abuse.

The level of multi-drug resistance shown by the *S. aureus* isolates from healthy volunteers in this study is of great concern. About 72% of the isolates were resistant to at least four antibiotics, 95% were resistant to at least one antibiotic and only 5% were susceptible to all the antibiotics tested. These observations confirm the postulation that healthy members of the community are the highest reservoir of antimicrobial resistant bacteria (Lester et al., 1990; Lamikanra et al., 1996). The society is presently characterised with inappropriate prescription, unethical dispensing and indiscriminate use of antibiotics. The rate at which most antibiotics are losing the battle against resistant organisms should be of immerse

concern to the health professionals and calls for effective measures (including trainings) to promote rational use of antibiotics and thereby prolong their 'life expectancy'.

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