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Antibiotic Susceptibility Pattern of *Staphylococcus aureus* Isolated from Clinical Samples in Specialist Hospital, Sokoto

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Authors' contributions

This work was carried out in collaboration among all authors. Authors NMB and AAI designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author MDH managed the analyses of the study. Author UIA managed the literature searches. All the authors read and approved the final manuscript.

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ABSTRACT

Aim: The study was to determine the antibiotic susceptibility pattern of *Staphylococcus aureus* isolates against some conventional antibiotics.
Study Design: Hospital based cross-sectional study.
Place and Duration of Study: The study was conducted in Specialist Hospital, Sokoto Metropolis, Sokoto State Nigeria, between June 2018 and September 2018.
Methodology: One hundred (100) *Staphylococcus aureus* isolates were used in this study. Gram's staining, catalase, coagulase and mannitol fermentation tests were used to identify and confirm the

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isolates. Antibiotic susceptibility testing was carried out by disc agar diffusion test.
Results: In the present study 63.0% of the *Staphylococcus aureus* isolates were from male subjects, while 37.0% were from female subjects. The age group with the highest number of isolates was 11-20 years (37%) and the least (9%) was seen in 41-50 years. Subjects with urine samples had the highest frequency of *Staphylococcus aureus* isolates with 32.0% and those with high vaginal swab had the lowest 6.0%. The antibiotics susceptibility testing showed that 40% of *Staphylococcus aureus* isolates were susceptible to Clindamycin, 64% to Ciprofloxacin, 57% to Erythromycin, 71% to Gentamicin, 34% to Cefoxitin, 46% to Quinupristin/Dalfopristin, 58% to Tetracycline and Sulphamethaxazole –Trimethoprim respectively. Screening for MRSA was carried out by antibiotic sensitivity testing using cefoxitin and a prevalence of 66% was obtained. This study showed that Gentamicin and Ciprofloxacin were the most active antibiotics against *Staphylococcus aureus*.
Conclusion: There is the need for consistent on-going antimicrobial resistance surveillance for important and commonly isolated clinically significant pathogens of staphylococcal species to form the basis for developing and implementing measures that can reduce the burden of antimicrobial resistance and prevent a probable impending public health problem.

Keywords: Antibiotics; Staphylococcus aureus; MRSA; clinical samples.

1. INTRODUCTION

Staphylococcus aureus is a Gram-positive coccus, catalase, and coagulase positive bacterium that emerged as one of the major human pathogens of public health concern, and has over the past decades, been a leading cause of nosocomial and community-acquired infections [1]. The bacterium is well characterized and known to have a wide range of virulence factors that cause a significant inflammatory response [2]. This pathogen affects both immunocompromised and immunocompetent individuals, often leading to high morbidity and with complications, which constitute problem to health care delivery system [3]. Variety of factors contribute to the ability of *S. aureus* to cause infection (virulence); enzymes, toxins, adhesion proteins, factors that help the bacteria to evade the innate immune defense, and antibiotic resistance mediate survival of the bacteria and tissue invasion at the site of infection [4].

The emergence of multidrug resistance Gram-positive bacteria (Staphylococci, Enterococci and Pneumococci) is an important and great health concern. Perhaps the pathogen of greatest concern is *S. aureus*, possibly because of its ability to adapt to various environmental conditions, its capacity to cause an array of life threatening conditions, and its intrinsic virulence [5]. *Staphylococcus aureus* is known to be notorious in the development of resistance to new antibiotics and continues to defy attempts at medical control [5]. The resistance of *S. aureus* isolates to commonly used antibiotics in Nigeria and other parts of the world has been widely reported [6]. This increase in emergence of resistance strains has being attributed to the indiscriminate use of antibiotics in both human and veterinary medicine especially in the developing countries. Many strains of *S. aureus* carry a wide variety of multi-drug resistant genes on plasmids, which aid the spread of resistance even among different species [7]. In Nigeria, most symptomatic patients engage in indiscriminate use of antibiotics prior to consulting the physicians when the situations cannot or would be difficult to control. The physicians on the other hand usually treat the patients with broad-spectrum antibiotics before microbiological investigations [8].

2. MATERIALS AND METHODS

2.1 Study Design

Cross-sectional study designed to determined the antibiotic susceptibility profile of one hundred (100) *Staphylococcus aureus* isolates obtained from medical microbiology laboratory of Specialist hospital Sokoto.

2.2 Bacterial Isolates

A total of 100 isolates of *Staphylococcus aureus* were collected from various clinical specimens (wound swab, nasal swab, ear swab, high vagina swab, pus and urine samples) obtained from the routine diagnostic tests in medical microbiology laboratory of Specialist Hospital and transported to the medical microbiology laboratory in the school of medical laboratory sciences, Usmanu Danfodiyo University Sokoto, Nigeria.

2.3 Identification of Bacteria

Diagnostic procedures consisted of Gram staining, biochemical tests like Catalase, Coagulase and Mannitol fermentation test.

2.3.1 Gram staining technique

Gram staining was carried out according to Chessbrough [9].

2.3.2 Biochemical tests

Isolates found to be gram positive cocci were subjected to biochemical tests like catalase and coagulase using technique described by Chessbrough [9] and also, sub cultured on Mannitol Salt Agar.

2.3.3 Catalase test

Two drops of 3% hydrogen peroxide solution was placed on a cleaned glass slide. A colony of the test organism was collected using a sterile glass rod and then emulsified into the drop of hydrogen peroxide. Bubbles of gas indicated a catalase positive test, while absence of bubbles indicated a catalase negative test [9].

2.3.4 Coagulase test

Slide Test to detect bound coagulase; A drop of normal saline was placed on two separate cleaned grease free glass slide. A colony of the organism was picked and emulsified in each of the drops to make a suspension. Using a wire loop a loopful of plasma was added onto one of the suspensions, mixed gently and observed for clumping of the plasma immediately. No plasma was added to the second suspension, it served as the negative control of the test. Clumping of the plasma indicates the organism is *S. aureus* while no clumping indicates other *Staphylococcus species* [9].

2.3.5 Mannitol fermentation test

Isolates were directly inoculated on Mannitol Salt Agar MSA (Oxoid, England), a selective and differential media of *S. aureus* and incubated at 37°C for 24 hours. Organisms that were able to grow on Mannitol Salt Agar (Oxoid, England) with fermentation of Mannitol and acid production to give yellow colour in the media around the colonies were characterized as *S. aureus* [9].

2.4 Antibiotic Susceptibility Testing

The antimicrobial susceptibility testing for *Staphylococcus aureus* was performed in accordance to Clinical and Laboratory Standards Institute (CLSI) [10]. Standard inoculum was prepared by making a direct saline suspension of isolates colonies by selecting from an 18h agar plate (nutrient agar). The suspension was adjusted to a turbidity of 0.5 McFarland standard equivalent which resulted in a suspension containing approximately 1 $x10^8$ colony forming unit (CFU)/ml. Antimicrobial susceptibility was performed on Mueller-Hinton Agar by the standard Kirby-Bauer disk diffusion method. This was done by dipping a sterile swab stick into the bacterial suspension and carefully swabbing the entire surface of Mueller Hinton agar plates. The antibiotic single discs (Oxoid) were then placed on the surface of the inoculated plates and gently pressed. The plates were incubated at 37°C for 18–24 h. The diameter of zone of inhibition was measured in millimeters and isolates were scored as sensitive, intermediate or resistant by comparing with values recommend in the CLSI M100 inhibition zone standard [10].

2.5 Screening for MRSA

Zones of inhibition ≥22 mm with 30 µg cefoxitin were recorded as Methicillin Susceptible *Staphylococcus aureus* (MSSA), while zones of inhibition ≤21 mm with 30 µg cefoxitin was recorded as Methicillin Resistant *Staphylococcus aureus* (MRSA) [10].

2.6 Statistical Analysis

The data collected was presented in tables, and analyse using Statistical Package for Social Sciences (SPSS) version 25 and the confidence level was set at 95% (P = .05). Comparative resistance rates of *S. aureus* strains from the different clinical specimens were statistically analysed by Chi square - test.

3. RESULTS AND DISCUSSION

In this study, a total of 100 *Staphylococcus aureus* isolates were collected from clinical samples of patients attending Specialist Hospital Sokoto from the medical microbiology laboratory. Analysis of the gender specific distribution of patients infected with *Staphylococcus aureus* in Specialist Hospital Sokoto shows that Males had higher infection rate (63.0%) than females

(37.0%) (Table 1). However, the age group with the highest frequency of *Staphylococcus aureus* infection was found to be individual aged 11-20 years while the least was in the 21-30 years (Table 2). Different clinical specimens from which *Staphylococcus aureus* was isolated were analysed, the highest number of isolates was from urine samples 32(32.0%) and the least was from high vaginal swab 6(6.0%) (Table 3).

Sensitivity and resistance pattern of *Staphylococcus aureus* to various antibiotics showed that the highest frequency of sensitivity was observed with Gentamicin (71%) followed by Ciprofloxacin (64%). The least was observed with cefoxitin (34%) (Table 4). Antibiotic resistance pattern of Methicillin resistant *Staphylococcus aureus* (MRSA) shows that 66(100%) of MRSA were resistant to Cefoxitin, 44(66.7%) to Clindamycin and 38(57.6%) to Quinupristin/Dalfopristin (Table 5).

In the present study, it has been observed that male subjects were more infected with *Staphylococcus aureus* (63%) than female subject (37%), which is in agreement with what was reported by Kumurya and Ado [11] at Aminu Kano Teaching Hospital that males had (61.8%) and females (38.2%). This is probably due to the nature of job men engage that females do not, especially farming in the Northern part of the country.

Also, in this study the highest frequency of isolates of *Staphylococcus aureus* (37%) was observed in the age group 11-20 years. This is in contrast to previous study by Nwankwo et al. [12] who reported the highest frequency (47.3%) among neonates and infants 0-10 years. This difference can be attributed to distribution of specimen collection as more were collected from age group 11-20 years than 0-10 years during the period of this study.

The prevalence of *Staphylococcus aureus* was found to be higher from urine samples 32.0% compared to other samples. This is in contrast to previous study by Kumurya and Ado [11] who reported the highest prevalence of 38.1% from blood cultures. This may be attributed to the issue of urine contamination with *S. aureus* from the surface during sample collection.

The highest frequency of susceptibility in this study occurred with Gentamicin and Ciprofloxacin having (71.0%) and (64.0%) respectively. The least was cefoxitin having (34.0%). A similar study depicted that the most potent of all the antibiotics tested was Rifampicin, with 54% sensitivity [13]. The level of resistance to Cefoxitin and Clindamycin could be associated with earlier exposure of these drugs to the isolates which may have enhanced development of resistance and probably due to high rate of antibiotics abuse in this area arising from self-medication which is often linked with failure to comply to treatment, inadequate dosage and availability of antibiotics to consumers across the counters with or without prescription [14].

Screening for methicillin resistant isolates in this study showed a prevalence rate of 66%. This is in line with a study in Zaria [15] where similar prevalence of 69% was obtained. In other studies elsewhere in Nigeria, a lower prevalence of 25.5% was reported from Kano by Nwankwo et al. [12] and 34.7% was reported a few years [16]. In addition, the prevalence of MRSA was also

Table 1. Prevalence of *Staphylococcus aureus* isolates based on gender

Gender	No. tested	Percentage	X^2	P-value
Male	63	63.0	20.885	0.002
Female	37	37.0		
Total	**100**	**100.0**		

Table 2. Distribution of *Staphylococcus aureus* according to age group

Age group (years)	Frequency	Percentage (%)	X^2	P-value
1-10	28	28	81.317	0.000
11-20	37	37		
21-30	10	10		
31-40	16	16		
41-50	9	9		
Total	**100**	**100**		

Table 3. Distribution of *Staphylococcus aureus* according to source of isolates

Type of specimen	No. tested	percentage %
Nasal	9	9.0
Urine	32	32.0
Wound	23	23.0
Pus	9	9.0
HVS	6	6.0
Semen	9	9.0
Ear	12	12.0
Total	**100**	**100.0**

Table 4. Antibiotic susceptibility pattern of *Staphylococcus aureus* isolates

Antibiotic	Sensitive (%)	Resistant (%)
Clindamycin	40	60
Quinupristin/Dalfopristin	46	54
Cefoxitin	34	66
Tetracycline	58	42
Sulphamethoxazole/Trimethoprim	58	42
Erythromycin	57	43
Ciprofloxacil	64	36
Gentamicin	71	29

Table 5. Antibiotic susceptibility profile of methicillin resistant *Staphylococcus aureus* (MRSA)

Antibiotic	Sensitive (%)	Resistant (%)
Cefoxitin	0.0 (0.0)	66 (100.0)
Clindamycin	22(33.3)	44 (66.7)
Quinupristin/Dalfopristin	28 (42.4)	38 (57.6)
Erythromycin	39 (59.1)	27 (40.9)
Tetracycline	34 (51.5)	32 (48.5)
Sulphamethoxazole/Trimethoprim	38 (57.6)	28 (42.4)
Ciprofloxacin	36 (54.5)	30 (45.5)
Gentamicin	39 (59.1)	27 (40.9)

found to be low (43.0%) in studies conducted in Jos Nigeria [17] compared to the present study. This may be associated to the ever increasing prevalence of MRSA; in Nigeria prevalence of MRSA ranging between 37.4% and 72.1% has been reported [18,19].

4. CONCLUSION

In this study, greater numbers of *Staphylococcus aureus* isolates were obtained from male subjects than female subjects and the highest frequency of *Staphylococcus aureus* isolates was observed in the age group 11-20 years. A relatively high prevalence of MRSA of 66% was recorded. The study has shown that *S. aureus* isolates were moderately sensitive to the antibiotics tested and that Gentamicin and Ciprofloxacin were the most active antibiotics against *Staphylococcus aureus*.

ETHICAL APPROVAL

Ethical approval to conduct this study was obtained from the ethics and Research committee of Specialist Hospital, Sokoto in accordance with the clinical and university standard.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Yassin NA, Mohammed HH, Ahmad AM. Antibiograming profiles of *Staphylococcus aureus* isolated from various clinical specimens in Duhok City Iraq. Adv Trop Med Public Health Int. 2013;3(1):25-31.
2. Jombo GT, Akpan S, Epoke J, Akaa PD, Eyong KI. Methicillin resistant *Staphylococcus aureus* and their antibiotic sensitivity pattern in Kano, Nigeria. J Med Sci. 2010;1(3):043-046.
3. Onanuga A, Awhowho GO. Antimicrobial resistance of *Staphylococcus aureus* strains from patients with urinary tract infections in Yenagoa, Nigeria. J Pharm Bioallied Sci. 2012;4(3):226-30.
4. Speziale P, Pietrocola G, Provenzano M, Visai L. Structural and functional role of *Staphylococcus aureus* surface components recognising adhesives matrix molecule of the host. J Nat Med Assoc. 2009;4(2):1337-352.
5. Karchmer AW. From theory to practice: Resistance in *Staphylococcus aureus* and new treatments. Clin Microbiol Infect. 2006;12(8):15-21.
6. Mincey BA, Parkulo MA. Antibiotic prescribing practices in a teaching clinic: Comparison of resident and staff physicians. South Med J. 2001;94(4):365-69.
7. Todar K. Bacterial resistance to antibiotics. Textbook of bacteriology. 4th Ed. Edinburg: Churchill Livingstone; 2011.
8. Aboderin OA, Abdu A, Odetoyin BW, Lamikanra AS. Antimicrobial resistance in *Escherichia coli* strains from urinary tract infections. J Nat Med Assoc. 2009;101(12): 1268-273.
9. Chessbrough M. District laboratory practice in tropical countries. 2nd Ed. Cambridge: University Press. 2009;62-70:105-114.
10. Clinical and Laboratory Standard Institute. Performance standards for antimicrobial disc susceptibility tests. 11th Ed. CLSI Document M02-A11. 2012;323-337.
11. Kumurya AS, Ado ZG. Detection of clindamycin resistance among methicillin resistant *Staphylococcus aureus* isolates in Kano, Nigerian. Access J Microbiol. 2017;1(2):34-40.
12. Nwankwo EOK, Abdulhadi S, Magaji A, Ihesiulor G. Methicillin resistant *Staphylococcus aureus* and their antibiotic sensitivity pattern in Kano, Nigeria. Afr J Clin Exp Microbiol. 2010;11(1):129-36.
13. Onanuga A. Prevalence and susceptibility pattern of methicillin- resistant *Staphylococcus aureus* isolates among healthy women in Zaria, Nigeria. Afr J Biotechnol. 2005;4:1321-324.
14. Odugbemi T. The use and abuse of antibiotics. Nig Med Pract. 1998;1(1):4-8.
15. Onanuga A, Temedie TC. Nasal carriage of multi-drug resistant *Staphylococcus aureus* in healthy inhabitants of Amassoma in Niger delta region of Nigeria. Afr Health Sci. 2011;11(2):176-81.
16. Kumurya AS, Ado ZG. Detection of clindamycin resistance among methicillin resistant *Staphylococcus aureus* isolates in Kano, Nigerian. Access J Microbiol. 2015;1(2):34-40.
17. Ikeh EI. Methicillin resistant *Staphylococcus aureus* at Jos University Teaching Hospital (JUTH). Afr J Clin Exp Microbiol. 2003;4(1):52-55.
18. Taiwo SS, Bamidele M, Omonigbehin EA, Akinsinde KA, Smith SI, Onile BA, et al. Molecular epidemiology of methicillin resistant *Staphylococcus aureus* in Ilorin, Nigeria. West Afr J Med. 2005;24(2):100-6.
19. Onanuga A, Oyi AR, Olayinka BO, Onaolapo JA. Prevalence of community-associated multi-resistant *Staphylococcus aureus* among healthy women in Abuja, Nigeria. Afr J Biotech. 2005;4(9):942-45.

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