

Antibiotic Susceptibility Profile of *Staphylococcus aureus* Isolated from Clinical Samples in Nasarawa Town, Nasarawa State, Nigeria

ABSTRACT

Multidrug resistant strain of *S. aureus* is the most common cause of life-threatening hospital- and community-acquired infections. Multidrug resistant *S. aureus* infections contribute to patients' prolonged stay in the hospital, increase in total healthcare costs, morbidity, and mortality. This work was aimed at determining the occurrence and antibiotic susceptibility profile of *Staphylococcus aureus* isolated from some clinical samples (blood and urine) in General Hospital, Nasarawa, Nasarawa State, Nigeria. All the 14 samples (7 each for blood and urine) collected in this study yielded positive for *S. aureus*, which were identified by cultural appearances and confirmed using conventional biochemical tests. The antibiotic susceptibility profile of the isolates indicated that, majority of them exhibited high susceptibility to gentamycin (85.7%), ciprofloxacin (78.6%), vancomycin (71.4%), chloramphenicol (64.3%), teicoplanin (50.0%), and erythromycin (42.9%). The isolates showed high resistance to oxacillin (100%), amoxicillin (85.7%), and ceftiofloxacin (78.6%).

Key words: *Staphylococcus aureus*, clinical samples, antibiotic resistance, Nasarawa town, Nigeria.

INTRODUCTION

Staphylococcus aureus (*S. aureus*) is a Gram-positive cocci, catalase-positive, coagulase-positive, and oxidase-negative bacterium that is frequently found in the anterior nares – nose, respiratory tracts, and on the skin of healthy humans and animals (Cosgrove *et al.*, 2009). The persistence of *S. aureus* as a nosocomial and community-acquired pathogen is a cause for public health concern and studies have established *S. aureus* as an important pathogenic bacterium causing infections ranging from minor skin infections and abscesses to life-threatening diseases such as meningitis, pneumonia, endocarditis, toxic shock syndrome (TSS), and septicemia which may be rapidly fatal (Holmes *et al.*, 2005).

Antimicrobial drug resistance among bacterial pathogens is a global public health challenge. The emergence of antibiotic resistant microorganisms (e.g. *S. aureus*) is increasing extremely rapidly around the globe, creating a serious threat to the spread and treatment of infectious diseases (Oli *et al.*, 2013). *S. aureus* has the ability to acquire resistance to antimicrobial drugs through horizontal gene transfer from outside and other sources, including chromosomal mutation and antibiotic selective pressure (Nwankwo *et al.*, 2011). In view of the public health importance of *S. aureus* in human infections, this work was designed to: Isolate and identify *Staphylococcus aureus* from some clinical samples in Nasarawa town and determine the antibiotic susceptibility profile of the *Staphylococcus aureus* isolates

MATERIALS AND METHODS

The Study Area

This study was carried out in Nasarawa. Nasarawa is a town in Nasarawa State, which is located in the North-central part of Nigeria. It has an area of about 5, 704 km² with a population of 189, 835 as at the 2006 census (NBS, 2009). It is approximately 105km from Abuja, the Federal Capital Territory, 37 km from Keffi and 165 km from Lafia, the state capital. The town is located between latitude 8°21'58"N of the equator and longitude 7°5'58E of the Greenwich meridian (NBS, 2009).

Sample Collection

Fourteen (14) samples (comprising of 7 blood and urine) samples respectively, were collected from patients who had suspected *S. aureus* infections at the General Hospital, Nasarawa from August to October, 2018. The urine samples were collected into sterile, dirt-free sample bottles, while the blood samples were collected through the veins of the patients using sterile

hypothermic needles. Blood sample from each patient was emptied into separate sterile Ethylenediamine tetraacetic acid (EDTA) bottle. The bottles were labelled appropriately and transported immediately to the Microbiology Laboratory of Federal Polytechnic, Nasarawa for further processing.

Isolation and Identification of *S. aureus*

The urine samples were spun in a centrifuge at 150 rpm for 5 min after which the supernatant was discarded and the sediments were inoculated onto plates of prepared mannitol salt agar (MSA) and incubated at 37⁰C for 24 hrs. Blood samples were directly inoculated onto prepared MSA plates and incubated at 37⁰C for 24 hrs. The presumptive colonies of *S. aureus* obtained after incubation were further sub-cultured onto freshly prepared plates of mannitol salt agar (MSA) in order to obtain pure culture. These isolates were preserved for further bacterial identification. The isolates were identified as *S. aureus* on the basis of Gram staining, colony morphology on mannitol salt agar (MSA) (HiMedia®, India), beta-hemolytic patterns on blood agar enriched with 5% (v/v) sheep blood, catalase test, DNase test, and coagulase tests (Japoni *et al.*, 2004).

Antibiotic Susceptibility Test

All the *S. aureus* isolates were subjected to antibiotic sensitivity testing by standard agar disc diffusion method on Muller-Hinton agar (OXOID, England) according to the Clinical and Laboratory Standards Institute (CLSI) recommendations (CLSI, 2012). Sensitivity patterns of the isolates to: ciprofloxacin (10µg), gentamycin (10µg), clindamycin (20µg), amoxicillin (10µg), chloramphenicol (30µg), erythromycin (30µg), oxacillin (30µg), and teicoplanin (30µg) and vancomycin (30µ) (Lioflichem®, Italy), were evaluated.

After incubation, the test plates were examined for confluent growth and zone of inhibition. The diameter of each zone of inhibition was measured in millimeter (mm) using a transparent ruler on the underside of the plate. The results were interpreted using the Clinical Laboratory Standards Institute (CLSI) criteria (CLSI, 2012). Results obtained for each isolate were interpreted as: 1). Sensitive (S): if the observed zone of the inhibition diameter was equal or greater than CLSI sensitive diameter (mm); 2). Intermediate (I): if the observed zone of inhibition diameter fell within the intermediate range between the CLSI resistant and sensitive limits; 3). Resistant (R): if the observed zone of inhibition diameter was less than or equal to the CLSI resistant diameter (mm) according to the Clinical and Laboratory Standards Institute (CLSI) guideline; Performance Standards for Antimicrobial Susceptibility Testing (CLSI, 2012).

RESULTS

Purification and Identification of the *S. aureus* Isolates obtained from the Samples

Purification and identification of the fourteen (14) isolates obtained from samples showed that, all 14 (100%) samples yielded gram positive cocci in clusters by gram staining; produced yellow-coloured colonies on mannitol salt agar (MSA); produced bubbles when emulsified in drops of 3% hydrogen peroxide (H_2O_2); showed β -haemolysis on blood agar; agglutinate rabbit plasma; and gave characteristic opaque medium with clear zones around colonies on DNase agar medium (Table 1).

Table 1: Distribution of *Staphylococcus aureus* According to Sample Type

Sample Type	Number Collected	Number Positive	Prevalence (%)
Blood	7	7	100
Urine	7	7	100
Total	14	14	100

Antibiotic Susceptibility Profile of the *S. aureus* Isolates

Table 2 shows the antibiotic susceptibility profile of *Staphylococcus aureus* isolates obtained from clinical samples. The antibiotic susceptibility profile showed that, 12 (85.7%) out of the 14 isolates were susceptible to gentamycin, 11 (78.6%) were susceptible to ciprofloxacin, 10 (71.4%) were susceptible to vancomycin, 9 (64.3%) were susceptible to chloramphenicol, and 7 (50%) were susceptible to teicoplanin. The isolates exhibited high resistance to amoxicillin (85.7%) and cefoxitin (78.6%). All the 14 (100%) isolates tested showed resistance to oxacillin (Table 2).

Table 2: The Antibiotic Susceptibility Profile of *Staphylococcus aureus* Isolated from Clinical Samples in Nasarawa Town

Antibiotic	Disconc. (µg)	(n=14)		
		S	I	R
Amoxicillin	10	0(0%)	2(14.3%)	12(85.7%)
Cefoxitin	30	0(0%)	3(21.4%)	11(78.6%)
Clindamycin	2	5(35.7%)	6(42.9%)	3(21.4%)

Chloramphenicol	30	9(64.3%)	2(14.3%)	3(21.4%)
Ciprofloxacin	5	11(78.6%)	1(7.1%)	2(14.3%)
Erythromycin	30	6(42.9%)	5(35.7%)	3(21.4%)
Gentamycin	30	12(85.7%)	0(0%)	2(14.3%)
Oxacillin	30	0(0%)	0(0%)	14(100%)
Tecoplanin	30	7(50%)	4(28.6%)	3(21.4%)
Vancomycin	30	10(71.4%)	0(0%)	4(28.6%)

113 Key: S= susceptible; I= intermediate; R= resistance

114

115 **DISCUSSION**

116 From the results obtained, all the 14 (100%) clinical samples collected for this study yielded
117 positive results for *Staphylococcus aureus*. This is unlike the findings from a similar research by
118 Garba *et al.* (2017) who recorded 14.6% occurrence of *S. aureus* out of 350 clinical samples
119 collected in Zaria metropolis, Kaduna State, Nigeria. The reason for this disparity maybe
120 accounted for by the differences in the number of samples examined, sampling population and
121 demography. This study portrays *S. aureus* as being widespread in the population studied.

122 The result of the antibiotic susceptibility tests showed that, all of the 14 positive isolates obtained
123 from the samples exhibited high susceptibility to gentamicin (85.7%), ciprofloxacin (78.6%),
124 vancomycin (71.4%), and chloramphenicol (64.3%). This is in consonance with the findings of
125 Garba *et al.* (2017) in Zaria, Nigeria who reported high susceptibility of *S. aureus* isolated from
126 clinical samples to gentamycin (96.1%), ciprofloxacin (78.4%), vancomycin (76.55), and
127 chloramphenicol (82.4%). The high susceptibility of *S. aureus* reported in this study is in
128 consonance with the work of Aliyu *et al.* (2018) who reported 64.3% susceptibility of *S. aureus*
129 isolated from some hospital environments in Nasarawa State to the antibiotic. However, the

isolates were observed to have developed high resistance to oxacillin (100%), amoxicillin (85.7%), and cefoxitin (78.6%). This is in agreement with the findings of Joshua *et al.* (2015) who reported high rate of resistance of *Staphylococcus aureus* isolated from clinical samples to oxacillin (100%), cefoxitin (98%), and amoxicillin (100%). Oxacillin and amoxicillin are relatively inexpensive antibiotics which are readily available to individuals in pharmacies without prescription from authorised health personnel (Newman *et al.*, 2006), and this lends credence to the indiscriminate use of antibiotics which promotes selective pressure favouring the emergence of resistant bacteria (Levy, 2001). Not only are these resistant bacterial strains potential causes of recurrent infections but they are also reservoirs of resistance genes that could be transferred to other pathogens. For these reasons, the antibiotic susceptibility trends seen in the *S. aureus* isolates may also occur in other bacterial pathogens.

The presence of multiple drug resistant strains of *S. aureus* among the isolates may be attributed to antibiotic misuse arising from self-medication in suspected bacterial infections (Newman *et al.*, 2006). Self-medication prevents early reporting of patients to hospitals at the onset of disease symptoms, except where complications have occurred. Also, some other factors such as unnecessary prescriptions and substandard antibiotics could lead to the emergence of antibiotic resistance among organisms (Newman *et al.*, 2006).

Conclusion

The results obtained in this study portray *S. aureus* as being widespread in the studied population. It is apparent in this work that, isolates of *S. aureus* were resistant to most commonly prescribed antibiotics except for ciprofloxacin, gentamycin, ciprofloxacin, vancomycin, and

chloramphenicol to which the isolates were observed to be highly susceptible. High levels of antimicrobial resistance to oxacillin, amoxicillin, and cefoxitin were observed. Thus, this finding suggests the need for antibiotic stewardship and sensitivity test on pathogens before administration of antibiotics. This measure can help lower the burden of antimicrobial resistance and solve a public health problem or reduce it drastically.

References

- Aliyu, Y., Jibril, U.Y., Jibrin, S.M. and Salawu, E.M. (2018). Occurrence and antibiotic resistant phenotypes of *Staphylococcus aureus* isolated from some hospital environments in Nasarawa Town, Nasarawa State, Nigeria. *European Journal of Pharmaceutical and Medical Research*, 5(11):20-26.
- Clinical and Laboratory Standards Institute (CLSI) (2012). Performance Standards for Antimicrobial Susceptibility Testing. Eighteenth Information Supplement, 28(1):34-52.
- Cosgrove, S.E., Qi, Y., Kaye, K.S., Harbath, S., Karchmer, A.W. and Carmell, Y. (2005). "The impact of methicillin resistance in *Staphylococcus aureus* bacterium in patients' outcome: mortality, length of stay, and hospital charges". *Infection, Control, and Hospital Epidemiology*, 26(2):166-174.
- Garba, S., Josiah, A.O. and Busayo, O.O. (2017). Antibiotic susceptibility profile of *Staphylococcus aureus* from clinical isolates in Zaria metropolis, Kaduna. *Nigerian Journal of Pharmaceutical and Medical Research*, 2(2):116-120.
- Holmes, A., Ganner, M., McGuanes, M., Pitt, T.L. and Cookson, B.D. (2005). *Staphylococcus aureus* isolates carrying Panton-Valentine Leukocidin genes in England and Wales: frequency, characterization, and association with clinical disease. *Journal of Clinical Microbiology*, 43:2384-2390.
- Japoni, A., Alborzi, A., Rasouli, M. and Pourabbas, B. (2004). Modified DNA extraction for rapid PCR detection of methicillin resistant staphylococci". *Iran Biomed J*, 8(3):161-165.
- Joshua, B., Owolabi, R. and Olorioke, C. (2015). Prevalence and antimicrobial susceptibility profile of methicillin resistant *Staphylococcus aureus* and coagulase-negative

181 staphylococci isolated from apparently healthy university students in Ota, Nigeria.
182 *Journal of Natural Sciences Research*, 5(24).

183 Levy, S. (2001). Antibiotic resistance: Consequences of inaction. *Clinical Infectious Diseases*,
184 33(3):S124-S129.

185 National Bureau of Statistics (2009). Federal republic of Nigeria, 2006 population census.
186 <http://nigeriastatgov.ng/connection/pop2006.pdf>.

187 Newman, M.J., Frimpong, E., Asamoah-Adu, A., Sampane-Donkor, E. (2006). Resistance to
188 antimicrobial drugs in Ghana. *The Ghanaian – Dutch collaboration for Health Research*
189 *and Development*, pp. 1-6.

190 Nwankwo, E.O. and Nasiru, M.S. (2011). Antibiotic sensitivity patterns of *Staphylococcus*
191 *aureus* from clinical isolates in a tertiary health institution in Kano, North-Western
192 Nigeria. *Pan African Med. J.*, 8:4.

193 Oli, A.N., Nweke, J.N., Ugw, M.C., Anagu, L.O., Oli, A.H. and Esimone, C.O. (2013).
194 Knowledge and use of disinfection policy in some government hospitals in South-East,
195 Nigeria. *British Journal of Medicine and Medical Research*, 3(4): 1097 - 1108 (3): 183 -
196 87.