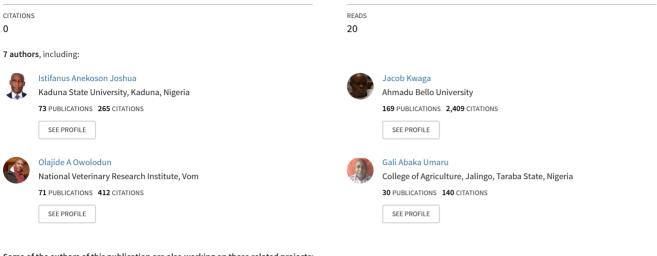
See discussions, stats, and author profiles for this publication at: https://www.researchgate.net/publication/365904655

Phenotypic Characterisation of Staphylococcus aureus Isolated from Patients in Healthcare Institutions in Zaria Metropolis, Kaduna State, Nigeria

Article in West African journal of medicine · November 2022



Some of the authors of this publication are also working on these related projects:

Project

Lassa virus and Hepatitis E virus View project

STAPHYLOCOCCUS AUREUS SEQUENCE TYPE (ST) 1 ISOLATED FROM SUB CLINICAL MASTITIS IN SETTLED FULANI HERDS IN KADUNA STATE, NIGERIA View project

VOLUME 39, NUMBER 11 November 2022 ISSN 0189 - 160X



WEST AFRICAN JOURNAL OF MEDICINE

ORIGINALITY AND EXCELLENCE IN MEDICINE AND SURGERY



OFFICIAL PUBLICATION OF THE WEST AFRICAN COLLEGE OF PHYSICIANS *AND* WEST AFRICAN COLLEGE OF SURGEONS







TABLE OF CONTENTS

GENERAL INFORMATION INFORMATION FOR AUTHORS EDITORIAL NOTES – The Stroke Epidemic and Associated Co-morbidities by Prof. Gregory E. Erhabor	1C 1F
ORIGINAL ARTICLES	1111
 10-Year Risk of Developing Type 2 Diabetes Mellitus – A Survey of Rural Communities in Southern Nigeria	1113
Challenges of Case Management of COVID-19 in University of Uyo Teaching Hospital: A One-Year Experience I. P. Oloyede, A. Onukak, O. O. Motilewa, A. Ekuma, S. Udoette, C. Eyo, E. K. Abudu, V. A. Umoh, E. Bassey, E. Peters	1119
Fungal Nail Infections amongst Cassava Farmers and Processors in Southwest Nigeria O. O. Ayanlowo, R. O. Oladele	1127
Immunohistochemical Study and Clinicopathologic Correlation of Cox-2 and Her-2 Expression in Colorectal Carcinoma: A 5-Year Retrospective Study L. A. Odukoya, K. B. Badmos, G. Khramtsova, L. A. Adebayo, O. I. Olopade, F. B. Abdulkareem	1134
 L. A. Odukoya, K. B. Badmos, G. Khramisova, L. A. Adebayo, O. I. Olopade, F. B. Abdulkareen The Impact of Co-Morbidities on the Pattern of Blood Pressure Control in Elderly Hypertensives in Nigeria	. 1141
Phenotypic Characterisation of <i>Staphylococcus aureus</i> Isolated from Patients in Healthcare Institutions in Zaria Metropolis, Kaduna State, Nigeria I. A. Joshua, F. J. Giwa, J. K. P. Kwaga, J. Kabir, O. A. Owolodun, G. A. Umaru, A. G. Habib	1148
The Relationship between Adolescents' Family Background, Perceived Self-Concept and Health Seeking Behaviour in an Urban City of South-Western Nigeria	1156
Awareness and Adherence to COVID-19 Preventive Measures among Oral Health Care Workers in Nigeria L. L. Enone, A. Oyapero, J. O. Makanjuola, R. O. Ojikutu	1165
Short Term Visual and Refractive Outcome following Surgical Intervention for Posterior Capsule Opacification (PCO) in Children in a Tertiary Eye Hospital Q. I. Sazzad, M. Hossain, H. Alimi, M. Khatun, M. R. Chowdhury, S. Toufique, S. M. Naznin	1174
Preferences, Utilization and Factors affecting Use of Contraceptives among Women attending Primary Health Care Facilities in Delta State, Southern Nigeria D. T. Obong, N. S. Awunor, P. G. Oyibo	1180
Prevalence of Hyponatremia in Acute Stroke Patients in a Federal Teaching Hospital, Abakaliki, Nigeria C. O. Eze, O. F. Afolabi, A. U. Kalu	1188
An Evaluation of Renal Care received by Human Immunodeficiency Virus (HIV) Patients admitted in a Tertiary Hospital in Sierra Leone	1193
Assessment of <i>MTR</i> Rs1805087 SNP as Possible Modifier of Sickle Cell Disease Severity in a Nigerian Population V. O. Osunkalu, A. A. Ogbenna, N. O. Davies, F. O. Olowoselu, O. E. Aiyelokun, O. J. Akinsola, I. A. Taiwo	1198
Quest to Improve Management of Prostate Cancer in West Africa: Development of a Clinical Audit Tool S. O. Osaghae	1205
CLINICAL PERSPECTIVE Roll Back Stroke: The Way Forward for Physicians and Patients Y. Ogun, A. Morawo	1209
INDEX TO VOLUME 39, NO. 11, 2022 Author Index	1215
Subject Index	
E-PUBLISHED Identification of the new progress on Pyrazole Derivatives Molecules as Antimicrobial and Antifungal Agents F. E. Bennani, L. Doudach, Y. El rhayam, K. Karrouchi, Y. Cherrah, A. Tarib,M. Ansar, M. E. A. Faouzi	1217





ORIGINAL ARTICLE

Phenotypic Characterisation of *Staphylococcus aureus* Isolated from Patients in Healthcare Institutions in Zaria Metropolis, Kaduna State, Nigeria

Caractérisation Phénotypique des Staphylococcus aureus Isolés Des Patients dans les Établissements de Santé de la Métropole de Zaria, État de Kaduna, Nigeria

^{1*}I. A. Joshua, ²F. J. Giwa, ³J. K. P. Kwaga, ³J. Kabir, ⁴O. A. Owolodun, ⁵G. A. Umaru, ⁶A. G. Habib

ABSTRACT

BACKGROUND: *Staphylococcus aureus* is a cosmopolitan and pathogenic microorganism associated with various diseases spectra and antimicrobial resistance of public health importance. Aim: This study determined the phenotypic characteristics of *S. aureus* isolated from patients in healthcare institutions in Zaria metropolis.

STUDY DESIGN: A cross-sectional hospital-based study was carried out in 5 healthcare institutions. Four hundred and twenty clinical samples were collected and analyzed.

RESULTS: Majority of the patients (54.3%) were within the age range 21–40 years and mean age of 26.04 ± 12 years. Approximately, 70% of the respondents had history of antibiotic use prior to consultation in the hospitals and were self-prescribed, and 91.2% were outpatients. The most commonly abused antibiotics were ampicillin-cloxacillin (19.5%) and cotrimoxazole (10.0%), and the mean duration of their use was 3.5 ± 1.3 days. The detection rate for *S. aureus* was 10% and 5.2% for MRSA. The *S. aureus* isolates showed the highest frequency of resistance against ampicillin 42 (100%), followed by penicillin G 39 (92.9%) and least was to gentamicin 5 (11.9%). The frequency of resistance for the MRSA were ampicillin 22 (100%), penicillin G 21(95.5%) and least was to gentamicin 2 (9.1%). The minimum inhibitory concentrations of oxacillin were greater than 128 µg/ml.

CONCLUSION: The detection rate of *S. aureus* and MRSA strains are of great public health concern which requires continuous health education on rational use of antibiotics among others. **WAJM 2022; 39(11): 1148–1155.**

Keywords: S. aureus, MRSA, Phenotype, Antimicrobial resistance, Hospital patients, Nigeria.

RÉSUMÉ

CONTEXTE: Staphylococcus aureus est un micro-organisme cosmopolite et pathogène associé à divers spectres de maladies et à une résistance aux antimicrobiens d'importance pour la santé publique. Objectif : Cette étude a permis de déterminer les caractéristiques phénotypiques de S. aureus isolé chez des patients dans des établissements de santé de la métropole de Zaria.

PLAN DE L'ÉTUDE: Une étude transversale en milieu hospitalier a été menée dans 5 établissements de santé. Quatre cent vingt échantillons cliniques ont été recueillis et analysés.

RÉSULTATS : La majorité des patients (54,3 %) étaient âgés de 21 à 40 ans et l'âge moyen était de $26,04 \pm 12$ ans. Environ 70 % des répondants avaient des antécédents d'utilisation d'antibiotiques avant la consultation dans les hôpitaux et étaient auto-prescrits, et 91,2 % étaient des patients externes. Les antibiotiques les plus fréquemment utilisés étaient l'ampicillinecloxacilline (19,5 %) et le cotrimoxazole (10,0 %), et la durée moyenne de leur utilisation était de $3,5 \pm 1,3$ jours. Le taux de détection de S. aureus était de 10 % et de 5,2 % pour le SARM. Les isolats de S. aureus ont montré la plus grande fréquence de résistance à l'ampicilline 42 (100%), suivie de la pénicilline G 39 (92,9%) et la plus faible à la gentamicine 5 (11,9%). La fréquence de résistance pour le SARM était de 22 (100%) pour l'ampicilline, 21 (95,5%) pour la pénicilline G et 2 (9,1%) pour la gentamicine. Les concentrations minimales inhibitrices de l'oxacilline étaient supérieures à 128 μ g/ml.

CONCLUSION: Le taux de détection des souches de S. aureus et de SARM est un grand problème de santé publique qui nécessite une éducation sanitaire continue sur l'utilisation rationnelle des antibiotiques entre autres. **WAJM 2022; 39(11): 1148–1155.**

Mots clés: *S. aureus*, MRSA, phénotype, résistance aux antimicrobiens, patients d'hôpitaux, Nigeria.

¹Department of Community Medicine, Faculty of Clinical Sciences, College of Medicine, Kaduna State University, Kaduna, Nigeria. ²Department of Medical Microbiology, Faculty of Basic and Clinical Sciences, College of Health Sciences, Ahmadu Bello University, Zaria, Kaduna State, Nigeria. ³Department of Veterinary Public Health and Preventive Medicine, Ahmadu Bello University, Zaria, Nigeria. ⁴Biotechnology Division, National Veterinary Research Institute, Vom, 930001, Plateau State, Nigeria. ⁵Department of Animal Health, College of Agriculture, PMB 1025, Jalingo, Taraba State, Nigeria. ⁶Infectious and Tropical Diseases Unit, Department of Medicine, Bayero University, Kano, Nigeria.

^{*}*Correspondence:* Dr. Istifanus Anekoson Joshua, Department of Community Medicine, Faculty of Clinical Sciences, College of Medicine, Kaduna State University, Kaduna, Nigeria. E-mail: dristifanus@yahoo.com Phone No: +234 8037039752.

Staphylococcus aureus Isolated from Patients in Healthcare Institutions

INTRODUCTION

Staphylococcus aureus (S. aureus) is the most pathogenic species of Staphylococcus, causing various disease spectra.¹⁻³ It is able to elaborate numerous virulence factors, which include capsule, penicillin binding protein A, mec A, Panton-Valentine leukocidin, fem B, catalase, coagulase, hae-molysins, enterotoxins, exfoliatins, among others.⁴ S. aureus develops resistance to commonly used antimicrobials.⁵ Methicillin-resistant S. aureus (MRSA), which is usually multidrug-resistant, has been a serious challenge for clinicians for more than half a century.⁶ MRSA can spread from human to human, from animal to human and from human to animal with public health implications.7-9 Most MRSA infections are expensive to treat and eradicate; leading to poor clinical outcomes.5,10 Antibiotic abuse/ misuse is still a problem in Nigeria.^{11,12}

There is paucity of information on the occurrence, distribution and characteristics of the organism in Zaria. The few studies that have been reported are of limited scope. Due to the changing epidemiology of MRSA, accurate information on the scope and burden of MRSA in Zaria is needed to set priorities for effective infection prevention and control.

The aim of this study was to determine the phenotypic characteristics of *S. aureus* isolated from patients in healthcare institutions in Zaria metropolis, Nigeria.

METHODS

Study Area and Healthcare Institutions

Zaria is a heterogeneous city with a population of about 1,490,000 and located in the north western part of Nigeria.¹³ It is positioned between latitude 110⁰ 3'N and 704⁰ 2'E. The climate is tropical continental in nature.¹⁴

The five selected healthcare facilities for the study were Ahmadu University Teaching Hospital (ABUTH), Zaria; Ahmadu Bello University (ABU) Medical Centre, Samaru; Saint Luke's Hospital (SLH), Wusasa; Major Ibrahim Bello Abubakar Hospital (MIBA), Sabon Gari and Gambo Sawaba Hospital (GSH), Kofar Gayan, Zaria City. They were purposively selected to reflect all the areas in the metropolis; and they are hospitals in which culture and sensitivity testing are being conducted. All the hospitals provide secondary health services, except ABUTH, Zaria which provides tertiary health services.

Sources of Clinical Samples

The calculated minimum sample size of four hundred and twenty participants was based on the set p value of 0.5¹⁵ since there was no previously documented value of p among the general population in the study area during the design of the study.

After the administration of the questionnaire by the researchers, clinical samples were collected from both inpatients and out-patients from the five hospitals. Only one sample was collected per patient, and they included wound swabs, ear swabs, eye swabs, sputum, seminal fluid, urine, pus, and blood. The period for the sample collection, processing, and phenotypic characterisation was between November, 2013 and December, 2016.

Isolation and Identification of *Staphylococcus aureus*

The isolates detected from the 420 clinical samples were screened using selective media, namely mannitol salt agar (MSA) and Baird Parker agar supplemented with egg yolk and potassium tellurite (BPA). The isolates were inoculated unto MSA (Oxoid, UK) plates and BPA plates (Oxoid, UK) and incubated at 37°C for 24 to 48 hrs. Then the plates examined for growth of colonies with characteristic golden colour for those inoculated unto MSA, and dark gray to black, tiny, shiny, convex with clear halos surrounding colonies for those inoculated unto BPA.

Sixty four of the 86 isolates were identified to be *S. aureus* after further testing- gram staining, catalase, coagulase (slide and tube), DNase and fermentation of glucose, xylose, maltose and mannitol and further testing with Microbact 12S identification system (Oxoid Ltd., Basingstoke, UK). Further tests carried out were PBP2a latex agglutination test, oxacillin resistance screening agar base (ORSAB), and minimum inhibitory concentration (MIC) for oxacillin using E-test (Biomérieux, France). Antimicrobial susceptibility testing was carried out by the modified disk diffusion method according to Clinical and Laboratory Standard Institute (CLSI) guidelines.¹⁶

Microbact 12S Identification System

The 64 isolates were subjected to further identification using MicrobactTM Staphylococcal 12S identification system test to confirm Staphylococcus species. The test was conducted according to the Manufacturer's instructions (Oxoid Ltd., Basingstoke, UK). The interpretation of the results was aided by colour charts provided in the test kit. Results were entered on the report form and each block of three reactions converted into a numeric value. The three numbers were then added together to obtain five digits of the MicrobactTM code which was entered into the computer aided software for identification.

Minimum Inhibitory Concentrations of Oxacillin

The minimum inhibitory concentrations (MIC) of oxacillin were determined using the Epsilometer strip (E strip) (Oxoid Ltd., Basingstoke, UK). This was carried out as described by the manufacturer and interpreted using the CLSI guidelines¹⁵ as follows: A 0.5 McFarland standard bacterial suspension was made from an overnight culture and spread on Mueller-Hinton agar using sterile swabs. Antibiotic E-test strips were thereafter placed on each plate with a sterile forceps. The plates were incubated for 24 hrs at 37°C. After the period of incubation, an eclipse corresponds to the antimicrobial concentration no longer inhibitory to the growth of the organism. The corresponding concentrations of antibiotic at the point of intersection on the strip were read as the MIC in μ g/ml. The MIC values were recorded and results interpreted as susceptible (S), intermediate (I) and resistant (R) according to CLSI criteria.

Guidelines of the CLSI¹⁶ was used to interpret the growth inhibition zones and classification of isolates as susceptible, intermediate and resistant strains. *S. aureus* ATCC 25923 and 43300 were used for quality control for *S. aureus* and MRSA strains respectively.

Detection of Penicillin Binding Protein (PBP2a) by Latex Agglutination Test

This is a rapid test latex agglutination assay for detecting PBP2a in Staphylococci as an aid in identifying MRSA and methicillin-resistant coagulase-negative Staphylococci. The test was conducted according to the manufacturer's instructions (Oxoid Ltd., Basingstoke, UK) as described below:

PBP2a Extraction Procedure: Four drops of the extraction reagent 1 was added to the micro centrifuge tube. Five micro litre (5ul) of the suspension was picked using 5ul loop and suspended in the micro centrifuge tube, vortexed and observed for clumps. The tube was placed into boiling water and heated for 3 minutes. The micro centrifuge was removed and allowed to cool at room temperature. One drop of the extraction reagent 2 was added to the tube and mixed thoroughly. The tube was then centrifuged at 3000 rpm at 15cm rotation radius and the supernatant separated.

Latex Agglutination Procedure: One circle of the test card was labeled for each suspension for testing with test latex and another for testing with control latex. The latex reagent was mixed thoroughly by inversion several times, and one drop of latex test and control test was added to each of the labeled circles. Five (5) micro litre of the supernatant was placed on the test circle and control circle, and then mixed thoroughly with a mixing stick. The card was then rocked for 3 minutes and agglutination was observed under lighting conditions. The results for the tests and control reactions were then recorded.

Interpretation of Results: No agglutination in either latex reagent within 3 minutes- PBP2a negative (MSSA); Agglutination seen with the control/latex within 3 minutes- Positive (MRSA).

Oxacillin Resistance Screening

All confirmed coagulase positive S. aureus were screened for oxacillin resistance by growing on Oxacillin Resistance Screening Agar Base (ORSAB) (Oxoid Ltd, Basingstoke, United Kingdom). The isolates were grown in broth enrichment comprising of Mueller-Hinton broth supplemented with 6.5% NaCl, prepared according to the manufacturer's instruction.

All confirmed coagulase-positive *S. aureus* were screened for methicillin/ oxacillin resistance by growing on Oxacillin Resistance Screening Agar Base (ORSAB) (Oxoid, Basingstoke, United Kingdom).

The isolates were grown in broth enrichment comprising of Mueller-Hinton broth supplemented with 6.5% NaCl, prepared according to the manufacturer's instructions. All inoculated plates were then incubated at 37° C for 24 hr. Then the medium was prepared according to the manufacturer's instructions, and all plates were inoculated with the samples. The plates were then incubated at 37° C for 24 hr. Growth of colonies that showed blue coloration was considered MRSA, while those with no growth or colonies with colour other than blue were considered as negative.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing of S. aureus isolates was done by the modified Kirby-Bauer disk diffusion method¹⁷ according to CLSI guidelines. The antimicrobials used and their concentrations are ampicillin (AMP10µg), amoxicillin-clavulanate (AMC30µg), cefoxitin (FOX30µg), chloramphenicol (C30µg), ciprofloxacin (CIP5µg), erythromycin (E5µg), gentamicin (CN10µg), oxacillin (OX5µg), penicillin G (P10IU), cotrimoxazole (SXT30µg) and tetracycline (TE30µg) (Abtek Biologicals Ltd, Liverpool, U.K). All antimicrobial susceptibility testing were performed by preparing a 0.5 McFarland standard suspension of each isolate and with the aid of a sterile swab stick, the inoculum was spread evenly onto prepared Mueller-Hinton agar plate. With the aid of a disk dispenser, 8 antibiotics each were dispensed onto each Mueller-Hinton agar plate. The plates were incubated at 37°C for 24hrs. After incubation, zones of inhibition around each antibiotic disc were measured to the nearest millilitre and recorded as susceptible (S), intermediate (I) and resistant (R) based on the CLSI guideline.¹⁶ *S. aureus* ATCC 25923 was used as control in each test run.

Multiple Antibiotic Resistance Index

Multiple antibiotic resistance (MAR) index was calculated as a/b, where 'a' represents the number of antibiotics to which the isolates were resistant and 'b' represents the total number of antibiotics to which the isolate was exposed.¹⁸

Storage of S. aureus Isolates

All isolates positive for coagulase, catalase, gram stain, sugar fermentation, DNase was stored on nutrient agar after preparation. The nutrient agar medium was prepared according to the manufacturer's instruction. The *S. aureus* isolates were also stored at -20° C in 15 % glycerol in BHI broth, in addition to those kept at 4°C on nutrient agar slants for further studies.

Data Management and Analysis

This was carried out using SPSS version 21 and Chi-square test used to determine the relationship between categorical variables. P < 0.05 was considered statistically significant.

Ethical Clearance

Ethical clearance was sought and obtained from Health Research Ethics Committee of Ahmadu Bello University Teaching Hospital (ABUTH) Zaria (ABUTH/HREC/A22/2012). Informed consent was obtained from each patient after thoroughly explaining the aim of the study. In the case of children, assent was obtained from the parent or guardian. All information collected was treated as highly confidential.

RESULTS

All the 420 interviewer-administered semi-structured questionnaires were analyzed giving a response rate of 100 %. Of the 420 clinical samples collected; 86 were *Staphylococcus* species (64 *S. aureus* and 22 CoNS).

The age of the respondents ranged from 1-60 years, however, more than half of the respondents (54.3%) were within 21-40 years, mean was 26.04 ± 12 years.

More than half (58.6 %) were females, about 2/3 (36.0 %) had secondary education and almost 70 % gave history of use of antibiotics before seeking medical care at the hospitals, 91.2 % were outpatients and only 6 % had history of surgery (Table 1). The mean duration for in-patient admission (N=37) were: 4.2 ± 2.4 days with range of 2–14 days.

The most commonly used antibiotic by the patients was ampicillin-cloxacillin (27.9%). About 13% of the respondents

 Table 1: Socio-demographic Characteristics of the Patients (N=420)

Variable	Frequency	Percent (%)	
Age (in years)			
1–10	56	13.3	
11–20	85	20.2	
21–30	175	41.7	
31-40	53	12.6	
41–50	28	6.7	
51-60	23	5.5	
Sex			
Male	174	41.4	
Female	246	58.6	
Religion			
Christianity	178	42.4	
Islam	242	57.6	
Marital Status			
Child	74	17.6	
Divorced	1	0.2	
Single	86	20.5	
Married	245	58.3	
Widow	6	1.4	
Widower	8	1.9	
Educational Status			
Child	69	16.4	
No formal education	78	18.6	
Primary	83	19.8	
Secondary	151	36.0	
Tertiary	39	9.3	
Occupation			
Applicant	10	2.4	
Civil servant	57	13.6	
Student	68	16.2	
Child	72	17.1	
Business man	12	2.9	
Business woman	1	0.2	
Trader	32	7.6	
Farmer	30	7.1	
Artisan	3	0.7	
Housewife	135	32.1	
History of use of Antibiotics			
Yes	293	69.8	
No	127	30.2	
Type of Patient			
Out-patient	383	91.2	
In-patient	37	8.8	
History of Surgery			
Yes	25	6.0	
No	395	94.0	

did not know the names of the antibiotics they took (Table 2). Of the 293 patients (69.8 %) who used antibiotics, the mean duration of use was 3.5 ± 1.2 days, with antibiotic use range of 1-7 days and mode of 3 days. The commonest surgery the patients underwent was appendectomy (28 %) (Figure 1). The overall detection rate for S. aureus was 10.0 % and 5.2 % for MRSA (Table 3). The highest frequency of resistance by the S. aureus clinical isolates was to ampicillin (100%), and least to gentamicin (11.9 %) (Figure 2). MRSA clinical isolates displayed the highest rate of resistance to ampicillin (100%), and least to gentamicin (9.1 %) (Figure 3). Penicillin binding protein 2a were detected in 15 out of the 22 MRSA isolates (68.2 %) (Table 4). All the 22 MRSA clinical isolates (100%) were resistant to oxacillin. The multiple antibiotic resistance indices ranged from 0.2 to 0.9 for the S. aureus, 9.5 % of the isolates had MAR index of 0.2, 0.3 - 0.9 was 90.5 %.; 0.6 MAR index had the highest percentage of 28.5 %, followed by 0.7 with 19.5 % and 0.4 with 16.6% (Table 5).

DISCUSSION

Majority of the respondents were within the age bracket of 21–40 years, 58.6% females, 58.3% married and 36% had secondary education. This means that all ages groups of both sexes were infected by the organism. Even the highly educated were not spared from infections caused by *S. aureus*.

More than two-third of the respondents gave history of antibiotics usage prior to seeking medical attention in the hospitals. The variables are risk factors for S. aureus infection and development of antibiotic resistance. Studies by researchers reported widespread use of self-prescribed antibiotics in Nigeria.11,12 This implies that wide spread use of unprescribed antibiotics has been a public health problem in the country for a very long time, and it is still a challenge today. Inexpensive antibiotics in developing countries (Nigeria inclusive) are widely available without prescription from authorized health institutions, doctors, and pharmacies. Unauthorized patent medicine vendors and other institutions

Table 2: Antibiotics used by the Patients before Consultations (N=293)

Antibiotic	Frequency	Percentage (%)	
Name unknown	38	12.9	
Amoxicillin	16	5.5	
Ampicillin	27	9.2	
Ampicillin-cloxacillin	82	29.9	
Amoxicillin-clavunate	7	2.3	
Ciprofloxacin	12	4.1	
Chloramphenicol	4	1.4	
Cefuroxime	2	0.7	
Erythromycin	5	1.7	
Ceftriaxone	3	1.0	
Tetracycline	32	10.9	
Gentamicin	1	0.3	
Cotrimoxazole	42	14.3	
Spectinomycin	1	0.3	
Metronidazole and ampicillin-cloxacillin	8	2.7	
Gentamicin and ampicillin-cloxacillin	1	0.3	
Augmentin and gentamicin	1	0.3	
Penicillin injection	6	2.0	
Doxycycline	5	1.7	

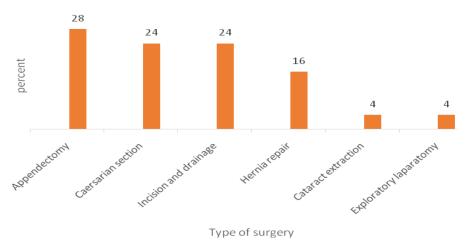


Fig. 1: Types of Surgeries the Patients had (N=25)

 Table 3: Detection Rate of S. aureus and Methicillin Resistant S. aureus (MRSA)

 from Clinical Samples

Hospital	No. of Samples Examined	No. (%) Positive for <i>S. aureus</i>	No. (%) Positive for MRSA
ABUTH	140	20(14.3)	9(2.1)
Sickbay	70	7(10.0)	4(0.9)
MIBA	70	7(10.0)	5(1.2)
St Luke's Hospital	70	5(7.1)	2(0.5)
Gambo Sawaba Hospital	70	3 (4.3)	2(0.5)
Total	420	42 (10.0)	22 (5.2)
Test Statistics			
χ^2		17.668	8.066
df		4	4
P value		=0.819	=0.427

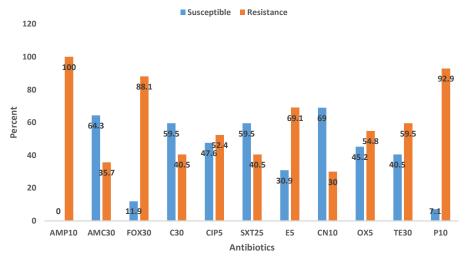
also contributes to the irrational use of antibiotics.¹⁹ The public health consequence of this is the development of drug resistance by the microorganisms. In Nigeria, 53% of respondents in a survey in Benin took incomplete regimen of antibiotics, a significant proportion of which were self-prescribed for unspecified ailment.²⁰

In this study, ampicillin-cloxacillin and cotrimoxazole were the most commonly used antibiotics by the patients. This could be as a result of the availability and low cost of these drugs, a finding which is similar to studies that reported cotrimoxazole (40%) and penicillin (11.8 %) to be commonly abused in Gambia and Gabon.^{21,22} Nigeria shares similar demographics with these two countries which could account for the findings. Appendectomy was the most common surgery reported by the patients. This may be because majority of the patients being within the range of 20-40 yrs. However, appendectomy and other surgeries if not carried out under aseptic conditions could result in wound infection and subsequent colonization by MRSA.

The mean duration of antibiotic use is very important. The mean of 3.5 ± 1.2 days means majority of the respondents took the drugs for a very short time which is a risk factor for the development of drug resistance.^{11,12}

A study in Kano reported *S. aureus* detection rate of $28.5 \%^{23}$ which is higher than the result of this study (10 %), probably because 38 (62.0 %) of the isolates in the Kano study were from inpatients and 15 (38 %) from out-patients²³ whereas in this study only 8.8 % of the respondents were inpatients. Similar studies from Jos and Ilorin reported higher rates of 43% and 34.7% respectively.^{24,25}

The global problem of antimicrobial resistance is particularly pressing in developing countries; where infectious disease burden is high and cost constraints prevent the wide spread application of newer and more expensive agents.²⁶ The isolated *S. aureus* showed different degrees of susceptibility to the 14 commonly used antibiotics with penicillin having resistance ranging between 92.9 % and 100 %. Gentamicin



AMP10: Ampicillin 10µg, AMC30-Amoxicillin-clavulanate 30µg, FOX30-Cefoxitin 30µg, C30-Chloramphenicol 30µg, CIP5-Ciprofloxacin 5µg, SXT25-cotrimoxazole 25µg, E5-Erythromycin 5µg, CN10-Gentamycin 10µg, OX5-Oxacillin 5µg, TE30-Tetracycline 30µg, P10-Penicillin G 10IU



Table 4: Detection Rate of pbp2a among the MRSA Isolates in the 5 Hospitals (n=22)

Hospital	No Pbp2a Positive (%)	No pb2a Negative (%)	Total
ABUTH	7(77.8)	2 (22.2)	9
Gambo Sawaba Hospital	2(100)	0(0)	2
Major Ibrahim Bello Abubakar Hospital	4(80)	1(20)	5
Medical Centre ABU	1(25)	3(75)	4
St Luke's Hospital	1(50)	1(50)	2
Total	15 (68.2)	7 (31.8)	22 (100)
Statistics $\chi^2 = 7.876$	df=4	p=0.446	

120

The relationship between antibiotic use and microbial resistance is complex. Over-use of antibiotics in humans and animals for treatment leads to release of antibiotics and antibiotic-resistant strains into the environment.²⁸ A study showed an annual antibiotics production of 210 million kilograms in China and 46.1 % were used in livestock industries,²⁹ which is of great public health significance nationally and internationally.

The multiple antimicrobial resistance (MAR) indices of S. aureus ranged between 0.2 and 0.9; with majority (28.5%) being 0.6. This agrees with the report of a study in 2016.30 A study stated that MAR index value of greater than 0.2 indicates existence of isolates from high risk contaminated sources with frequent use of several antibiotics, while values <0.2 shows bacteria from sources with less antimicrobial usage.³¹ This will lead to the release of antimicrobials and antimicrobial resistant bacterial strains into the environment28 which will increase morbidity, mortality and cost of treatment of infections caused by the isolates.³² The findings here imply that the burden of antimicrobial resistance in the study population is high and this may directly or indirectly increase resistance of S. aureus and other bacteria in the study area and population. The nasal cavity is the natural ecological niche of Staphylococci, and the nasal carriage of

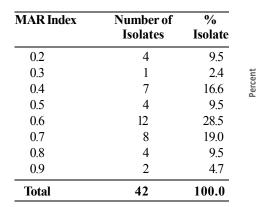
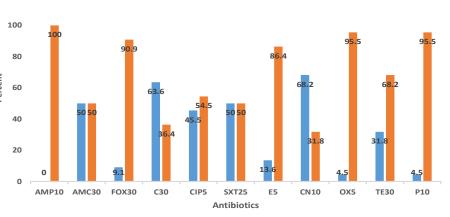


Table 5: Multiple Antibiotic Resistance(MAR) Index



Resistance

Susceptible

had the least resistance exhibited by the isolates. Penicillin is among the most commonly abused antibiotics in Nigeria. This agrees with findings of other studies in Nigeria.^{12, 27}

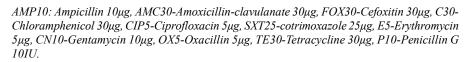


Fig. 3: Antibiotic Susceptibility Profile of MRSA from Clinical Samples (N=22)

West African Journal of Medicine Vol. 39, No. 11, November, 2022

multidrug-resistant *S. aureus* is thought to be a major risk factor of staphylococcal transmission among humans.^{33,34}

The detection rate of 5.2 % MRSA in this study is low compared to 12.5 % in a study carried out in ABUTH, Zaria.35 The high prevalence of MRSA in the teaching hospital could be as a result of the fact that a significant number of the study populations were in-patients and the hospital is a major referral centre in the region. However, there was no statistically significant difference between the detection rates of S. aureus (p=0.819) and MRSA (p=0.427) from the clinical samples. Fifty percent of the MRSA isolates were resistant to amoxicillin-clavunate which agrees with the findings of the study in Kano (Nigeria),²³ but at variance with the figure of 96 % reported in West Bengal (India).10 Similarly, the resistance of the MRSA isolates to penicillin and ampicillin (95.5 % and 100 % respectively) agrees with the findings by other researchers.^{12,36} Gentamicin showed the least resistance by the organism probably because the preparation is in injectable form, so it is not easily abused. Misuse and overuse of drugs, together with lack of control measures in Nigeria, might be responsible for the significant MRSA rates.37

Infections due to MRSA have been associated with a variety of clinical manifestations, including mild skin infection, pneumonia, and sepsis. Furthermore, there have been reports of increasing mortality associated with MRSA bacteraemia and ventilatorassociated MRSA pneumonia. Generally, the main prognostic factor in patients with MRSA infections is inadequate antibiotic therapy.³⁸ MRSA is usually transmitted by direct skin to skin contact with colonized or infected individuals and occasionally via fomites. Five Cs have been implicated in MRSA outbreaks namely: contact, lack of cleanliness, compromised skin integrity, contaminated object, and crowded living conditions.^{39,40} Other risk factors for MRSA include Acquired Immune Deficiency Syndrome (AIDS), diabetes, frequent hospital admission, renal replacement therapy (dialysis) and advancing age.39

The limitations of the study included the industrial strike action embarked upon by the hospital staff during the period of data collection affected the total number of the inpatients. Secondly, there was nonavailability of some antimicrobial disks such as imipenem, linezolid and teicoplanin during the research, so they were not included in the antimicrobial susceptibility testing.

CONCLUSION

Majority of the persons infected with *S. aureus* were within the productive age group; with positive history of antibiotic misuse and abuse. The detection rate for *S. aureus and* MRSA were 10.0% and 5.2% respectively. Both the *S. aureus* and MRSA strains showed various degrees of resistance to ampicillin, penicillin, cefoxitin and oxacillin, among others. There should be education on antimicrobial stewardship, periodic clinical and laboratory auditing in hospitals and the concept of one health should be encouraged by the various professional bodies.

Conflict of Interest

No conflict of interest by the authors.

REFERENCES

- 1. Mathanraj S, Sujatha S, Sivasangeetha K, Parija S.C. Screening for methicillinresistant *Staphylococcus aureus* carriers among patients and health care workers of a tertiary care hospital in South India. *Indian Journal of Medical Microbiology* 2009; **27:** 62-64.
- 2. Lulitanond L, Teroyo I, Shanshuang L, Xiao H, Xiao XM, Chulapan E, *et al.* ST9 MRSA strains carrying a variant of type IX SCC*mec* identified in the Tai community. *Infectious Diseases* 2013; **13**: 214–217.
- Bazzi AM, Rabaan AA, Mahmoud F, Al-Tawiq, J. Prevalence of Panton Valentine Leukocidin-Positive methicillin susceptible S. aureus infections in Saudi Arabian Hospital. Journal of Infection and Public Health 2015; doi.org/10.1016/j.jiph.2015.01.010.
- Lozano C, Mari A, Aspiroz C, Gomez-Sanz E, Ceballos S, Furto-no B, *et al.* Nasal carriage of coagulase positive staphylococci in patients of a primary health care centre: genetic lineages

resistance and virulence genes. Enfermedades Infecciosasy Microbiologia Clinica. 2015; **33:** 391–396.

- Harkins CP, Pichon B, Doumith M, Parkhill JH, Alexander T, Herminia D, *et al.* Methicillin-resistant *Staphylococcus aureus* emerged long before the introduction of methicillin into clinical practice. *Genome Biology.* 2017; 18: 130: 1–11.
- Fall C, Abdoulaye S, Vincent F, Moustapha N, Mbacke S, Federic L, *et al.* Epidemiology *S. aureus* in pigs and farmers in the largest farm in Dakar, Senegal. *Foodborne pathogens and Diseases.* 2012; **9:** 962–965.
- Lu PL, Pen CF, Chang YH, Ma L, Siu LK. Risk factors and molecular analysis of community methicillin-resistant *S. aureus* carriage. *Journal of Clinical Microbiology*. 2005; 43: 132–139.
- 8. Huber H, Kuller S, Giezendenner N, Stephen R, Zweifel C. Prevalence and characteristics of methicillin-resistant *S. aureus* in humans in contact with farm animals, livestock and in foods of animal origin. *Switzerland and Eurosurveillance*. 2009; **15:** 1–4.
- Kwoji ID, Tambuwal FM, Abubakar MB, Yakubu Y, Musa JA, Jauro S, et al. Antibiotic sensitivity patterns of methicillin-resistant Staphylococcus aureus isolated from chickens in poultry farms in Sokoto, Nigeria. Advances in Animal and Veterinary Sciences. 2018; 6: 8–11.
- Karmakar A, Dua P, Ghosh C. Biochemical and molecular analysis of S, aureus clinical isolates from hospitalised patients. Canadian Journal of Infectious Diseases and Medical Microbiology. 2016; 26: 1–7.
- Odugbemi T. The use and abuse of antibiotics. Nigeria Medical Practitioner. 1980; 1: 4–8.
- Shittu AO, Okon K, Adesida S, Oyedara O, Witte W, Strommenger B, *et al.* Antibiotic resistance and molecular epidemiology of *S. aureus* in Nigeria. *BMC Microbiology*. 2011; 11: 1–8.
- National Population Commission (NPC) 2006. Population Census of the Federal Republic of Nigeria. Abuja, Nigeria: National Population Commission; 2007.
- 14. Mortimore, M.J. Zaria and its region. Annals of the Association of American Geographers 1970; 60: 73–80.
- 15. Ibrahim T. Research Methodology and Dissertation writing for Health and Allied Health Professionals, 2000. Cress Global Link Limited Publisher, Abuja, Nigeria.

I. A. Joshua and Associates

- CLSI- Clinical and Laboratory Standard Institute (2011). Performance standards for antimicrobial susceptibility testing, 220th Informational supplement, Available from:clsi.org/source/orders/ free/m100-s20.pdf (Accessed 10 January, 2018).
- Bauer WA, Kirby WMM, Sherris JC, Truck M. Antibiotic susceptibility by standardized single disk method. *American Journal of Clinical Pathology*. 1996; 45: 493–496.
- Apun K, Chong YL, Abdullahi MT, Micky V. Antimicrobial susceptibilities of *Escherichia coli* isolates from food animals and wildlife animals in Sarawak, East Malaysia. *Asian Journal of Animal* and Veterinary Advances 2008; 3: 409–416.
- Sharma BC, Rai B. Incidence of multidrug resistance in Escherichia coli Strains isolated from three lakes of tourist attraction (Mirik lake, Jorepokhari lake and Nakhapani lake) of Darjeeling hills, India. Indian Journal of Fundamental Applied Life Sciences. 2012; 2: 108–114.
- Yah SC, Yusuf OE, Eghafona NO. Pattern of antibiotic usage by adult population in the city of Benin, Nigeria. *Scientific Research Essay* 2008; 3: 081– 085.
- 21. Risk R, Naismith H, Burnett A. Moore SE, Cham M, Unger S, *et al.* Rational prescribing in paediatrics in a resourcelimited setting. *Archives of Diseases in Childhood.* 2013; **98:** 503–509.
- Alabi AS, Frielinghaus L, Kaba H, Koster K, Huson MAM, Kahl BC, *et al.* Retrospective analysis of antimicrobial resistance and bacterial spectrum of infection in Gabon, Central Africa. *BMC Infectious Diseases.* 2013; 13: 455.
- 23. Nwankwo EO, Nasiru MS. Antibiotic sensitivity pattern of *Staphylococcus aureus* from clinical isolates in a tertiary health institution in Kano, North western Nigeria. *Pan Africa Medical Journal.* 2011; **8:** 1–12.

- 24. Ikeh EI. Methicillin resistant Staphylococcus aureus at Jos University Teaching Hospital (JUTH). African Journal of Clinical Experimental Microbiology. 2003; **4:** 52–55.
- 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 African Journal of Medicine. 2005; 24: 100– 106.
- Osundiya OO, Oladele RO, Oduyabo OO. Multiple antibiotic resistance index of *Pseudomonas* and *Klebsella* spp in Lagos University Teaching Hospital. *African Journal of Clinical and Experimental Microbiology*. 2013; 14: 164–168.
- 27. Momoh AH. Nasal carriage and molecular characterisation of *S. aureus* in pigs and occupationally exposed humans in Jos Metropolis, Plateau State, Nigeria. PhD dissertation submitted to School of Postgraduate Studies, Ahmadu Bello University, Zaria, Nigeria, 2015.
- Ghafur AK. An obituary on the death of antibiotics. *Journal of Association of Physicians*. 2010; 1: 58: 143–144.
- Hvistendahl M. Public health. China takes aim at rampant antibiotic resistance. *Science*. 2012; **336**: 795–797.
- Adenaike O, Olonitola OS, Ameh JB, Whong CMZ. Multi-drug resistance and multiple antibiotic resistance index of *Escherichia coli* strains isolated from retailed smoked fish. *Journal of Natural Sciences Research*. 2016; 6: 7–10.
- Thenmozhi S, Rajeswari P, Suresh Kumar T, Saipriyanga V, Kalpana M (2014) Multi-drug resistant patterns of biofilm forming aeromonas hydrophila from urine samples. *Int. J. Pharm Sci Res.* 2014; 5: 2908–2918.
- 32. Kleven RN, Edwards JR, Tenover FC, McDonald LC, Huran T, Gaynes R. Changes in the epidemiology of methicillin resistant *Staphylococcus aureus* in intensive care unit in US

hospitals, 1992–2003. Clinical Infectious Diseases. 2007; **42:** 389–391.

- 33. Abulreesh HH, Organji SR. The prevalence of multidrug resistant staphylococci in food and the environment of Makkah, Saudi Arabia. *Research Journal of Microbiology*. 2011; 6: 510–523.
- Bettin A, Causil C, Reyes N. Molecular identification and antimicrobial susceptibility of *Staphylococcus aureus* nasal isolates from medical students in Cartagena, Colombia. *Brazil Journal of Infectious Diseases*. 2012: 329–334.
- 35. Abdulaziz MM, Olayinka A. Nasal carriage of MRSA among health workers in high-risk units in a tertiary hospital in north western Nigeria. Poster presentation at the 17th International congress on infectious diseases. *International Journal of Infectious Diseases* 2016; 1: 477–479.
- 36. Falagas ME, Karageorgopoulos DE, Leptidis J, Korbila IP. MRSA in Africa: filling the global map of antimicrobial resistance. *PLoS One.* 2013; **8**: e68024.
- Kesah C, Ben Redjeb S, Odugbemi TO, Boye CSB, Dosso M, Ndinya Achola JO, et al. Prevalence of methicillinresistant Staphylococcus aureus in eight African hospitals and Malta. Clinical Microbiology and Infection 2003; 9: 153–156.
- Ippolito G, Leone S, Lauria FN, Nicastri E, Wenzel RP. Methicillin-resistant Staphylococcus aureus: the superbug. International Journal of Infectious Diseases. 2010; 1454: S7–S11.
- DeLeo FR, Otto M, Kreishwirth BN, Chambers HF. Community-associated methicillin resistant *S. aureus. Lancet.* 2010; **375:** 1557–1568.
- Goyal A, Diwakar MK, Bhooshan SS., Goyal SS, Agrawal A. Prevalence and antimicrobial susceptibility pattern of Methicillin-resistant *Staphylococcus aureus* (MRSA) isolates at a tertiary care hospital in Agra, North India – A systemic annual review. *Journal of Dental and Medical Sciences*. 2013; 11: 80–84.