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Phenotypic Detection of Methicillin and Inducible Clindamycin Resistant *Staphylococcus Aureus*: An Effort to Combat Antimicrobial Resistance and Improve Patient Safety in Gombe, Nigeria

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Abstract:

Introduction: Antimicrobial resistance (AMR) is a major threat to patient safety. Methicillin and inducible clindamycin resistant *Staphylococcus aureus* are important multidrug resistant organisms (MDROs). Timely reporting of MDROs is necessary for rational antibiotic prescription and in combating AMR. We present the prevalence and distribution of Methicillin and inducible clindamycin resistant (iCR) isolates of *Staphylococcus aureus* from Gombe Nigeria.

Materials and Methods: This descriptive cross-sectional study included 260 isolates of *Staphylococcus aureus* from clinical specimens in Federal Teaching Hospital Gombe (FTHG). Isolates identification was done using conventional biochemical methods. Methicillin resistance was detected by cefoxitin disc diffusion method while iCR isolates by erythromycin and clindamycin disc approximation test (D-test). Data analysis was done using SPSS version 23.0.

Results: Methicillin resistant *Staphylococcus aureus* (MRSA) was detected in 178 (68.5%) isolates while 214 (82.3%) were iCR (D-test positive). Among the MRSA, 87.1% were also iCR while 72.4% of the iCR isolates were MRSA. There was significant association between MRSA and iCR ($p = 0.03$), MRSA and clindamycin resistance ($p = 0.05$) and MRSA and erythromycin resistance ($p < 0.01$).

Conclusion: Prevalence of MDR *Staphylococcus aureus* is high in Gombe Nigeria. Antimicrobial stewardship programme (ASP) and good Infection Prevention and Control (IPC) are necessary in combating AMR and improving patient safety.

Keywords: Methicillin Resistant *Staphylococcus aureus* (MRSA), Inducible Clindamycin Resistant *Staphylococcus aureus* (iCRSA), Antimicrobial Resistance (AMR), Patient Safety, Nigeria

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Introduction

The emergence of antimicrobial resistance (AMR) posed one of the greatest challenges to patient safety and modern medicine (1). Mortalities due to multidrug resistant pathogens have continued all over the globe with worsening indices in developing countries where regulations guiding the use and sale of antimicrobials are suboptimal (2). The economic impact of AMR has continued to exert negative effects on both countries and families due to increased cost of care (2). *Staphylococcus aureus* has remained one of the most common and clinically relevant bacterial pathogens responsible for both local and systemic infections in both community and healthcare settings (3). There is a growing prevalence of multidrug resistant *Staphylococcus aureus* such as iCR and MRSA across the globe leading to poor patient outcome with more difficult and expensive treatment options (3). Multidrug resistant (MDR) *Staphylococcus aureus* has been ranked by the World Health Organisation (WHO) as one of the high priority pathogens that urgently require newer drugs for treatment (4). The presence of MRSA and co-resistance with iCR pathogens is associated with worsening of clinical presentation and increased frequency of treatment failure (5).

Diagnostic stewardship relies on good relationship between clinical laboratories and other clinicians/clinics for accurate diagnosis that will guide in providing better patient care for the best outcome and patient safety (6). This has become more critical in infectious diseases in view of the growing global threat of AMR which remains a major challenge to IPC and patient safety. (6) Phenotypic detection of both MRSA and inducible clindamycin resistance could be easily and realistically carried in most medical microbiology laboratories in low resource setting like Nigeria at a minimal or no extra cost (7, 8). Most of the studies from Nigeria and other developing countries utilized phenotypic detection methods in identifying the MDR pathogens (5, 9–11).

Although there were few reports on MDR *Staphylococcus aureus* from North Eastern Nigeria, virtually all the studies were carried about a decade ago and only on MRSA (12,13). Many other studies some of which also reported findings on inducible clindamycin resistance were from other parts of Nigeria (11). This study reported the prevalence and relationship of MRSA and inducible clindamycin resistance among clinical isolates of *Staphylococcus aureus* in Gombe Nigeria.

Methods

A descriptive cross-sectional study was conducted using a total of 260 non-repeat isolates of *Staphylococcus aureus* from clinical specimens (urine, swabs, blood, aspirates and seminal fluids) received in the department of medical microbiology and immunology, FTHG between May and October 2020. Identification of *Staphylococcus aureus* was done manually using biochemical methods (Gram staining, catalase and coagulase tests) and methicillin resistance was detected by cefoxitin disc diffusion method and inducible clindamycin resistance by erythromycin and clindamycin disc approximation test (D-test). Isolates with cefoxitin zone of inhibition ≥ 22 mm was regarded as methicillin susceptible while those with ≤ 21 mm was interpreted as methicillin resistant (14). Inducible clindamycin resistant phenotypes were identified based on resistance to erythromycin and having a clindamycin zone ≥ 21 mm with a D-shaped zone (14). Testing and interpretations were all based on Clinical and Laboratory Standards Institute (CLSI) performance standards for antimicrobial susceptibility testing (14). *Staphylococcus aureus* strains ATCC 25923 (MSSA), ATCC 43300 (MRSA) and ATCC 25923 (D-test positive) were used for quality control based on CLSI guidelines (14).

Ethical clearance for the study was obtained from the research and ethics committee (REC) of FTHG.

Data analysis was done using statistical package for social sciences (SPSS) version 23.0 software. Chi-square test was used in analyzing categorical variables ($P < 0.05$ was considered statistically significant).

Results

Of the 260 isolates, majority were from females (59.2%) and more than two third (79.2%) came from out-patients. More than half of the isolates were recovered from patients between the ages of 21 and 40 years (figure 1). Distribution of different resistant isolates based on type of specimen, gender of patients and origin of the isolates (in-patients and out-patients) were as highlighted in table 1, figure 2 and figure 3. Prevalence of MRSA was 68.5% while 82.3% were iCR. Among the MRSA, 87.1% were also iCR while 72.4% of iCR isolates were MRSA. Individual resistance to erythromycin and clindamycin was found among 86.5% and 85.8% of the isolates respectively. As shown in table 2, there was significant association between MRSA and iCR isolates

($p = 0.003$), MRSA and individual clindamycin resistance ($p = 0.005$) and MRSA and individual erythromycin resistance ($p < 0.001$). There was no significant association between MRSA and origin of isolates (in-patients and out-patients) and gender of patients.

Figure 1: Age distribution of patients from whom the isolates were recovered.

Figure 2: Pattern of resistance among isolates from in-patients and out-patients.

Figure 3: Pattern of resistance among isolates from males and females.

Table 1: Distribution of different resistant isolates based on type of specimen

Type of specimen	Number of isolates (%)	MRSA	iCR	Erythromycin resistance	Clindamycin resistance
Urine	157 (60.4)	108 (68.8)	134 (85.4)	140 (89.2)	139 (88.5)
Swabs	47 (18.1)	30 (63.8)	34 (72.3)	35 (74.4)	37 (78.7)
Seminal fluid	22 (8.5)	17 (73.3)	20 (90.9)	22 (100)	20 (90.9)
Aspirates	21 (8.0)	12 (57.1)	15 (71.4)	16 (76.1)	16 (76.1)
Blood	13 (5.0)	11 (84.6)	11 (84.6)	12 (92.3)	11 (84.6)
Total	260 (100)	178 (68.5)	214 (82.3)	225 (86.5)	223 (85.8)

Table 2: Association between MRSA, other resistant patterns (iCR, erythromycin resistance clindamycin resistance), of patients and origin of isolates (in-patients and out-patients)

	MRSA (N; %)	MSSA (N; %)	Total (N; %)	X	P value
iCR (D-test positive)	155 (72.4)	59 (27.6)	214 (100)	8.822	0.003
No iCR (D-test negative)	23 (50.0)	23 (50.0)	46 (100)		
Total	178 (68.5)	82 (31.5)	260 (100)		
Erythromycin resistant	165 (73.3)	60 (26.7)	225 (100)	18.373	<0.001
Erythromycin sensitive	13 (37.1)	22 (62.9)	35 (100)		
Total	178 (68.5)	82 (31.5)	260 (100)		
Clindamycin resistant	160 (71.7)	63 (28.3)	223 (100)	7.843	0.005
Clindamycin sensitive	18 (48.6)	19 (51.4)	37 (100)		
Total	178 (68.5)	82 (31.5)	260 (100)		
Gender					
Males	75 (70.8)	31 (29.2)	106 (100)	0.436	0.509
Females	103 (57.9)	51 (33.1)	154 (100)		
Total	178 (68.5)	82 (31.5)	260 (100)		
Source of isolates					
In-patients	37 (68.5)	17 (31.5)	54 (100)	0.000	0.992
Out-patients	141 (68.4)	65 (31.6)	206 (100)		
Total	178 (68.5)	82 (31.5)	260 (100)		

Discussion

This study identified high rates of both MRSA and inducible clindamycin resistance among isolates from clinical specimens in FTHG, Nigeria. The prevalence of MRSA in this study (68.5%) is higher than earlier studies of 12.5% and 8.0% from Gombe/North-eastern Nigeria (12, 13). This could be attributable to variation in time/methodology and changes in prescribing habits of clinicians and pattern of resistance among the microorganisms as reported in other studies across the globe (15, 16). However, differences (both higher and lower prevalence) with other Nigerian (9, 11) and foreign (5, 10, 17) studies have been observed in both MRSA and inducible clindamycin resistance. These disparities are mainly due to variation in site and time of the studies while few are due to differences in the detection method. Most of the studies used phenotypic detection methods and also emphasized the need for routine reporting of these common MDROs

to guide in rational use of antimicrobials and for good antimicrobial stewardship that will ensure patient safety (18, 19). Association of both MRSA and inducible clindamycin resistance with individual resistance to erythromycin and clindamycin as observed in this study is similarly reported by several other studies in Nigeria and other countries (5, 9–11, 17, 20). This further affirms the need for routine testing and reporting of common MDROs to appropriately guide prescribers in both targeted and empirical anti biotherapy, and to combat antimicrobial resistance for improved healthcare quality and patient safety.

The pattern of distribution of isolates among different specimens which revealed urine being the most frequent followed by swabs before seminal fluid, aspirates, and blood is a reflection of the frequency/predominance of these specimens in the laboratory requests. Similarly, several other studies have noted differences in pattern of distribution of isolates among different specimens

but mostly without statistical significance and mainly due to disparities in location, patient distribution, type of study and practices among requesting physicians based on training and guidelines (5, 10, 11, 20). Variations in gender and age distribution did not also reveal any statistical significance probably due to similarities in habits/exposure with prevalence of the isolate among the different groups.

Although there is no significant difference in pattern of resistance between isolates from in and out-patients, most were from out-patients. This is probably because most microbiological investigations are requested at the points of hospital entry and in addition to poor utilization of clinical microbiology laboratory with over-reliance on empirical anti biotherapy among Nigerian prescribers (21, 22). This also points to the danger of having MDROs circulating within the communities mainly due to poor regulation in the country, absence/poor adherence to antibiotic guidelines among HCWs and misuse/abuse of antibiotics among especially general public in Nigeria (22, 23).

Phenotypic detection was used in this study largely because it is relatively cheap and easy to conduct on routine basis even in low resource settings like ours (7, 8). This can be timely advanced to requesting clinicians and other prescribers for better antimicrobial stewardship and improvement in patient safety. However genotypic detection has the additional advantage of identifying the resistant genes in the pathogens and has been reported by some studies mainly for research purposes in different parts of the world (24–26).

Conclusion

The prevalence of MDR *Staphylococcus aureus* (MRSA and iCR) is high in Gombe Nigeria. Significant association between MRSA and iCR and individual erythromycin/clindamycin resistance was observed. Routine and timely reporting of findings to prescribers is necessary for rational anti biotherapy to combat AMR and improve IPC/patient safety. National and local guidelines/policies are important and should target both hospital and community-based utilization of antimicrobials with the concept of “one health” for better patient safety. Development of national/local guidelines, their wide dissemination is important and should be emphasized.

Conflict of Interest

The authors declare no conflict of interest.

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