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Prevalence and Antibiogram of Staphylococcus Aureus Isolated from Clinical Samples in Sokoto Metropolis

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Abstract:
The importance of Staphylococcus aureus as a persistent nosocomial and community acquired pathogen has become a global health concern. The aim of this research is to isolate Staphylococcus aureus from samples sent for microbiological test in major government hospitals in Sokoto. A total of 410 clinical samples were screened for the presence of S. aureus using standard microbiological techniques. Antibiotic sensitivity profiling was carried out by disc diffusion. Of the 410 samples, 93 (22.7%) yielded growth of S. aureus, the highest sensitivity was observed on nitrofurantoin (96.7%), all the isolates were resistant to ceftazidime, cloxacillin and augment in, 36.6% were resistant to cefoxitin while only 1.1% was resistant to vancomycin. Vancomycin could still be a drug of choice in the treatment of MRSA infections. There is the need for antibiotic surveillance in the study area to prevent the spread of resistance among bacteria.

Keywords: *Prevalence, staphylococcus aureus, antibiotics, sokoto*

1. Introduction

Staphylococcus aureus is a Gram-positive bacterium that belongs to the family Staphylococaceae and is often found as a commensal on the skin, skin glands and mucous membranes particularly in the nose of healthy individuals (Plata et al.,2009). It is a versatile human pathogen causing infections ranging from relatively mild skin and soft tissue infections to life threatening sepsis, pneumonia, osteomyelitis, endocarditis as well as toxin mediated syndromes such as toxic shock syndrome and food poisoning (Shittu et al.,2011).

It is one of the important pathogens in many countries causing infection in hospitals and the community. It causes a variety of diseases, ranging in severity from boils and furuncles to more serious diseases such as septicaemia, pneumonia and endocarditis (Lowy, 1998). Virulent strains of these bacteria are responsible for the majority of Hospital Acquired Infections (HAIs) and can cause severe disease that can be fatal (Deleo and Otto, 2009).

More than 90% of Staphylococcus strains are resistant to penicillin (Chambers, 2001), followed by increasing resistance to methicillin, aminoglycosides, macrolides and lincosamide (Levin et al.,2005; Nalwoga et al., 2016; Schmitz et al.,2000). Staphylococci have a record of developing resistance quickly and successfully to antibiotics. This defensive response is a consequence of the acquisition and transfer of antibiotic resistance plasmids and the possession of intrinsic resistance mechanisms (Kloos, 1998).

2. Materials and Methods

A total of 410 samples comprising of Blood cultures, wound swabs, ear swabs and body fluids collected from the laboratories of Usmanu Danfodiyo University teaching hospital, Specialist hospital and Maryam Abacha women and children hospital all in Sokoto were inoculated onto Mannitol salt agar (MSA). S. aureus colonies on Mannitol salt agar were surrounded by a yellow halo, due to mannitol fermentation. Colonies range in size from medium to large (0.5-1.5 µm), smooth, entire edges, slightly raised elevation and opaque.

Gram stain, Catalase, Coagulase (slide and tube) and DNAse activity were carried out on the young colonies to identify S. aureus. Antibiotic susceptibility testing was then carried out by first standardizing the innoculum to 0.5 McFarland standard as described by CLSI,2012. Already dried Mueller-Hinton agar (MHA) plates were then inoculated by streaking the swab over the entire surface of the agar and excess surface moisture allowed to evaporate before the antibiotic-impregnated disks (Oxoid, England) comprising Rifampin(5µg), Chloramphenicol(30µg), Tetracycline(30µg), Nitrofurantoin(200µg), Erythromycin(15µg), Ofloxacin(5µg), Augmentin(30µg), Gentamicin(10µg), Cloxacillin(5µg), Cefuroxime(30µg), Ceftriaxone(30µg), Clindamycin(2µg), Quinupristin/Dalfostrin(15µg) Ceftazidime(30µg) Cefoxitin(30µg) and Vancomycin(30µg) were applied and the plates incubated at 37 °C. After the prescribed period of incubation, the zone of inhibition was measured using a metre rule (against transmitted light) and the results interpreted using the CLSI 2014 guideline.

3. Result

Table 1 shows the prevalence of Staphylococcus aureus in the three centers and the number of samples collected in each center. The highest prevalence of Staphylococcus aureus was seen in Specialist hospital (30.1%) and lowest In Maryam Abacha women and children hospital with (12%). Table 2 shows the distribution of S. aureus by Gender and Age in the three Study Centers. Table 3 shows the distribution of the resistant isolates in the different types of the samples collected with 100% prevalence seen in Endo Cervical swab, Semen, Blood, Peritoneal fluid, Catheter tip and Burn. This is followed by High Vaginal swab with 75% and the least prevalence seen in Sputum with 12.5%. Table 4 shows the susceptibility pattern of the Staphylococcus aureus isolates to different antibiotics. The highest sensitivity was seen on Nitrofurantoin, in which 90 (96.7%) out of the 93 isolates were sensitive with an inhibition zone of ≥17mm followed by Quinupristin and Dalfostrin with 89 (95.7%) with an inhibition zone of ≥19mm and then Chloramphenicol with 80 (86%) with an inhibition zone of ≥18, while all the isolates were resistant to Ceftazidime, Cloxacillin and Augmentine. Table 5 depicts the susceptibility pattern of the isolates to Cefoxitin and Vancomycin. For the Cefoxitin, 34(36.6%) of the isolates were resistant with a zone of inhibition ≤21mm while 20(21.5%) with a zone of inhibition 15 and 16mm of the isolates showed Vancomycin intermediate sensitivity (VISA) and only 1(1.1%) isolate with a zone of inhibition 14mm showed resistance to Vancomycin.

4. Discussion

The importance of Staphylococcus aureus as a persistent nosocomial and community acquired pathogen has become a global health concern. In this study, out of a total of 410 non-repetitive clinical specimens collected from patients attending three hospitals in Sokoto metropolis, a total of 93 were positive for S. aureus, giving a prevalence of 22.7%. This agrees with findings of Obiazi et al., (2007), who obtained a prevalence of 20.8% in Irrua Nigeria. However, these reports are comparatively lower than what was reported by Charles et al., (2015), Nwoire et al., (2013) and Chibuike et al., (2013) where a prevalence of 84% in Anyingba, 60.4% in Ebonyi and 82.1% in Abia were respectively documented. This pattern of prevalence shows that S. aureus infection is probably influenced by the geographical location of the patients.

The antibiogram pattern of isolates showed a 100% resistance to ceftazidime, cloxacillin and augmentine, while the most potent of the antibiotics tested were nitrofurantoin, quinopristin/dalfostrin and chlorampenicol with 96.7%, 95.7% and 86% respectively. However, Charles et al., (2015) in Anyingba reported 54% resistance to erythromycin and augmentine, and sensitivity to gentamicin, ofloxacin and ciprofloxacin as 100%, 81.8% and 72.7% respectively. The pattern of resistance shown by S. aureus to many groups of antimicrobial agents in this research represents a serious concern in therapeutic option available to the clinician in managing such infections and further confirms various literatures that S. aureus is a multidrug resistant bacterium.

Methicillin resistance- the marker of multidrug resistance showed a prevalence of 36.6% among the S. aureus isolates and is similar to findings of Adetayo et al., (2014) and Angela et al., (2015), who reported a prevalence of 30.4% in Ibadan and 31.4% in Brazil respectively. The rate 36.6% from this study is however lower than what was reported by Onemu and Ophori (2013) in Benin City with 79%, indicating that MRSA is ever increasing. It is clear that MRSA has become a global nosocomial pathogen with attendant therapeutic problems and warrant urgent infection awareness, considering the common practice of unregulated sale of antimicrobial agents and movement of people which may result in rapid dissemination. In this study, sensitivity to vancomycin was established with 77.4% of the isolates being Vancomycin sensitive, 21.5% Vancomycin intermediate resistant and only 1 of the samples was Vancomycin resistant.

Other researches, Kumurya and yahaya (2016) reported 14.6% VRSA in Kano and Moses et al., (2013) reported 5.4% VRSA in Abakaliki all in Nigeria. Similar researches conducted in Brazil by Angela et al., (2015) reported 2.8% VRSA and Chakraborty et al., (2011) reported 26.6% VRSA in India all contradicts the findings of this work. The present study detected 77.4% VSSA and 21.5% VISA and is supported by findings of Chakraborty et al., (2011) in Pakistan, who reported 62% VSSA; 38% VISA and Venababu et al., (2011) who also reported 93.57% VSSA; 4.47% VISA. The implication of VISA seen is the

variable susceptibility patterns for the strains, and there are no uniform recommendations for treatment of infections due to this organism (CDC, 2013). The high sensitivity of vancomycin seen in this study may be because it remains as the only active antibiotic against all strains of S. aureus and MRSA in particular and therefore the most reliable therapeutic agent as demonstrated by literature.

5. Conclusion

This study highlights the need for antibiotic susceptibility testing with a view to selecting appropriate antibiotic therapy so as to prevent treatment failures as all the isolates were resistant to one or more antibiotics with nitrofurantoin being the most potent in terms of activity. This study concludes that vancomycin still remains a potent drug of choice for the treatment of MRSA infections.

Collection Centers	Number of Samples Examined from Different Centers (%)		
	Samples Examined	Sample Positive	Sample Negative
Usmanu Danfodiyo University teaching hospital	187	35 (18.70)	152 (81.30)
Specialist hospital Sokoto	173	52 (30.10)	121 (69.90)
Maryam Abacha Women and children hospital	50	6 (12.00)	44 (88.00)
Total	410	93 (22.70)	317 (77.30)

Table 1: Prevalence of Staphylococcus Aureus in the Study Centers

Gender	Number of Samples Examined (%)			
	Samples Examined	No. Positive	No. Negative	P Value
Males	58	21 (36.2)	37 (63.8)	0.928
Females	35	13 (37.1)	22 (62.9)	
Total	93	34 (36.6)	59 (63.4)	

Table 2: Distribution of S. Aureus by Gender and Age in the Three Study Centers
$X^2 = 0.008$

Sample Type	Number of Isolates Examined for Methicillin Resistance (%)			
	Isolates Examined	MRSA	MSSA	P Value
Wound	25	12 (48.0)	13 (52.0)	0.008
Sputum	16	2 (12.5)	14 (87.5)	
High Vaginal Swab	2	1 (50.0)	1 (50.0)	
Endocervical Swab	1	1 (100.0)	0 (0.0)	
Ear Swab	14	2 (14.3)	12 (85.7)	
Urine	26	9 (34.6)	17 (65.4)	
Semen	1	1 (100.0)	0 (0.0)	
Blood	1	1 (100.0)	0 (0.0)	
Peritoneal Fluid	1	1 (100.0)	0 (0.0)	
Pus	4	3 (75.0)	1 (25.0)	
Catheter Tip	1	1 (100.0)	0 (0.0)	
Burn	1	0 (0.0)	1 (100.0)	
Total	93	34 (36.6)	59 (63.4)	

Table 3: Distribution of MRSA by Sample Type in the Study Centers

Antibiotic	Content (µg)	Antibiogram of the Isolates (Mean Zone of Inhibition), N = 93		
		Sensitive	Intermediate	Resistant
Rifampin	5	79(29.00)	7(18.00)	7(9.70)
Chloramphenicol	30	80(25.30)	2(15.00)	11(7.10)
Tetracycline	30	28(23.80)	0(0.00)	65(7.60)
Nitrofurantoin	200	90(23.50)	2(15.50)	1(13.00)
Erythromycin	15	4(24.00)	47(22.66)	42(5.20)
Ofloxacin	5	59(23.30)	3(14.00)	31(6.20)
Augmentine	30	0(0.00)	0(0.00)	93(6.10)
Gentamicin	10	61(20.35)	2(13.00)	30(6.46)
Cloxacillin	5	0(0.00)	0(0.00)	93(6.10)
Cefuroxime	30	7(21.60)	25(16.00)	61(6.26)

Antibiotic	Content (µg)	Antibiogram of the Isolates (Mean Zone of Inhibition), N = 93		
		Sensitive	Intermediate	Resistant
Ceftriaxone	30	30(23.25)	23(18.20)	40(6.69)
Clindamycin	2	72(24.37)	4(14.57)	17(6.66)
Quinupristin-Dalfostrin	15	89(24.10)	4(16.50)	0(0.00)
Ceftazidime	30	0(0.00)	0(0.00)	93(6.10)

Table 4: Susceptibility Pattern of the Staphylococcus Aureus Isolates to Different Antibiotics

Antibiotic	Content	Antibiogram of the Isolates (Mean Zone of Inhibition), N = 93		
		Sensitive	Intermediate	Resistant
Cefoxitin	30µg	59 (25.33)	0 (0.00)	34 (9.80)
Vancomycin	30µg	72 (18.30)	20 (15.75)	1 (14.00)

Table 5: Susceptibility Pattern of the Staphylococcus Aureus Isolates to Cefoxitin and Vancomycin

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