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Susceptibility Patterns of Staphylococcus Aureus Isolates from Clinical Specimens to the Penicillin's

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Abstract: ***Staphylococcus aureus*** **is a major pathogen causing a variety of infections ranging from mild skin infections to life threatening systemic illness. Antibiotic resistance by Methicillin Resistant Staphylococcus Aureus (MRSA) has grown to be common, and resistant to almost all antibiotics. The objective of the study was to see the antimicrobial susceptibility pattern of *S. aureus* isolates from different clinical samples against various brands of penicillins in Ebonyi State Teaching Hospitals of Abakaliki, Ebonyi State, Nigeria. A cross-sectional study was conducted from September to October 2008 among patients attending Ebonyi State Teaching Hospital, Abakaliki, Nigeria. A total of 200 *Staphylococcus aureus* isolates were obtained from patients with infections. They were cultured on mannitol salt agar media, a selective medium for *S. aureus*. Isolated strains of *S. aureus* were tested for antibiotic susceptibility patterns using standard disc diffusion technique and interpretation was done based on Clinical and Laboratory Standard Institute criteria.The resistance patterns of this isolates were ranked in decreasing order: penicillin V (81%), Ampicillin (63%), Amoxycillin (56%), Ampiclox (30%), Flucloxacillin (20%) and Cloxacillin (18%). It was observed that Cloxacillin, Flucloxacillin and Ampiclox showed better sensitivity patterns, the incorporation of beta lactam inhibitors like clavulanic acid present in these antibiotics have made them more effective.The rate of antimicrobial resistance is to conventional antibiotics is very high, continuous surveillance on antimicrobial susceptibility testing of *S. aureus* is essential for the detection of emerged resistant strains of *S.aureus* especially MRSA.**

Keywords: ***S. aureus*****, MRSA, Resistance, Penicillins.**

1. INTRODUCTION

Staphylococcus aureus is one of the most common pathogens causing a variety of infections ranging from mild skin infections to life threatening systemic illness such as pneumonia, endocarditis, septic arthritis, subcutaneous or visceral abscesses [1]. It is also responsible for toxicoses, including food poisoning and toxic shock syndrome [2]. After the development of antibiotics, a general belief arose that the problem of bacterial infections would be solved. Before the introduction of penicillin in the late 1940s, Staphylococcal septicaemia was associated with an extremely high mortality rate. Penicillin dramatically improved the prognosis of this infection [3]. However, penicillin resistant strains were discovered shortly and penicillin became ineffective both in the hospital and community settings [4, 5]. The development of beta-lactamase resistant penicillins such as methicillin and oxacillin in the early 1960s once again revolutionized the treatment of Staphylococcal infections. Pathogens evolved sophisticated mechanisms of drug resistance by producing an enzyme called beta lactamase that destroys antimicrobial agents. As a result of these, antimicrobial resistance emerged as

one of the most serious health threats causing significant number of deaths worldwide every year, This has been pointed out in reports of the European Medicines Agency and by the U.S. Centre for Disease Control and Prevention (CDC) in the United States [6] and this have prompted widespread efforts to develop new antibacterial with the addition of inhibitor to combat the effect of betalactamase. The prevalence of *Staphylococcus aureus* resistant to conventional antibiotics has been increasing at high levels in some hospitals [7, 8] and it is still a challenge for clinicians till present day. Also, the wide spectrum of antibiotics in the hospital environment has led to the development of increased resistance to these antimicrobial agents. Globally, *S. aureus* is one of the principal microorganisms involved in nosocomial infections, and strains resistant to methicillin (MRSA) represent 15-45% of all *S. aureus* isolated [8, 9]. Several trends have been identified in the epidemiology of MRSA infections: increasing incidence of MRSA infections, particularly among surgical patients and also increasing proportion of nosocomial infections caused by MRSA. Infections caused by these bacteria are treated mainly with vancomycin. The objectives of the study were to see the antimicrobial susceptibility pattern of *S. aureus* isolates from different clinical samples against various brands of Penicillins in Ebonyi State teaching hospitals of Abakaliki, Ebonyi State, Nigeria.

2. MATERIALS AND METHODS

Sampling and location:

This study was executed at the Department of Medical Microbiology and Parasitology in Ebonyi State University Teaching Hospital from September to October 2008. A total of 200 clinical samples originating from patients in the hospital confirmed to be *Staphylococcus aureus* were used for this study. Informed consent was obtained from all patients that participated in this study. The clinical samples consisted of urine, pus, eye swab, wound swab, ear swab, high vagina swab, endocervical swab and semen.

Culture and Identification:

All clinical samples were inoculated directly on mannitol salt agar (Oxoid) and sub cultured to blood agar (Oxoid) and MacConkey agar (Oxoid). The plates were then incubated at 37 °C for 20 hours. All positive cultures were identified by their characteristic appearance on media, gram staining reactions and the pattern of biochemical tests. *S. aureus* was identified based on characteristic yellow colonies on mannitol salt agar; β hemolytic colonies with yellowish pigment on blood agar; gram positive cocci singly in pair, in short chain or clusters, catalase positive and coagulase production and mannitol fermentation.

Antimicrobial susceptibility testing:

The *Staphylococcus aureus* isolates were subjected to susceptibility testing by disc diffusion technique described by Kirby-Bauer according to the Clinical Laboratory Standards International (CLSI) guidelines with quality controls (*Staphylococcus aureus* ATCC 29213) [10]. The antimicrobial tested against isolates included ampicillin (10µg), amoxicillin (10µg), cloxacillin (30µg), flucloxacillin (10µg), penicillin V (10 units/10 IU) and Ampiclox (10µg). Antimicrobial sensitivity patterns were determined on the basis of the size of the zone of inhibition and were interpreted at sensitive, intermediate or resistant.

3. RESULTS

A total of 200 clinical samples originating from patients in the hospital confirmed to be *Staphylococcus aureus* were examined. The isolates were isolated from urine (n=10), pus (n=20), eye swab (n=10), wound swab (n=60), ear swab (n=20), high vagina swab (n=20), endocervical swab (n=56) and semen (n=14). Table 1 showed the sensitivity patterns of the isolates to the antibiotics *S. aureus* showed the highest resistance of 81% to Penicillin V, 16% to be intermediate and 3% to be sensitive. It was observed that 63% of the isolate were resistant to Ampicillin, 23% intermediate, and 14% to be sensitive. Amoxycillin showed a resistance of 56%, 24% intermediate and 20% to be sensitive. Ampiclox showed a resistance of 30%, 21% to be intermediate and 49% were sensitive. Flucloxacillin showed a high sensitivity rate of 58%, intermediate 22% and resistance of 20%. Cloxacillin had the highest sensitivity rate of 60 %, intermediate 22% and resistance of 18%. (Figure 1)

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Table 1: Sensitivity patterns of the isolates to the antibiotics tested. The results were interpreted as either resistant, intermediate or sensitive according to the recommendations of the CLSI (CLSI, 2007)

	Ampicillin (30µg)			Amoxycillin (30µg)			Ampiclox (30µg)			Flucloxacillin (30µg)			Cloxacillin (30µg)			PenicillinV (10µg)		
	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S
ECS	34	12	10	26	16	14	12	4	40	8	8	40	6	10	40	42	10	4
WS	40	10	10	44	16	6	20	10	30	6	12	42	10	2	48	50	8	2
SEMEN	10	0	4	6	2	6	6	2	6	2	4	6	0	8	6	6	8	0
HVS	12	8	0	10	4	6	4	6	6	4	6	10	4	10	6	0	10	0
EAR SWAB	4	4	2	4	4	2	2	6	2	2	4	4	2	4	4	10	0	0
EYE SWAB	4	4	2	4	4	2	2	4	4	4	2	4	4	2	4	5	2	0
PUS	16	4	0	14	4	2	12	4	4	12	4	4	10	4	6	18	2	0
URINE	6	4	0	4	2	2	2	4	4	2	4	4	0	4	6	8	2	0
TOTAL	126	46	28	112	48	40	60	42	98	40	44	116	36	44	120	162	32	6

ECS= Endocervical swab, WS= Wound swab, HVS= High vagina swab, R= Resistant, I= Intermediate and S= Sensitive

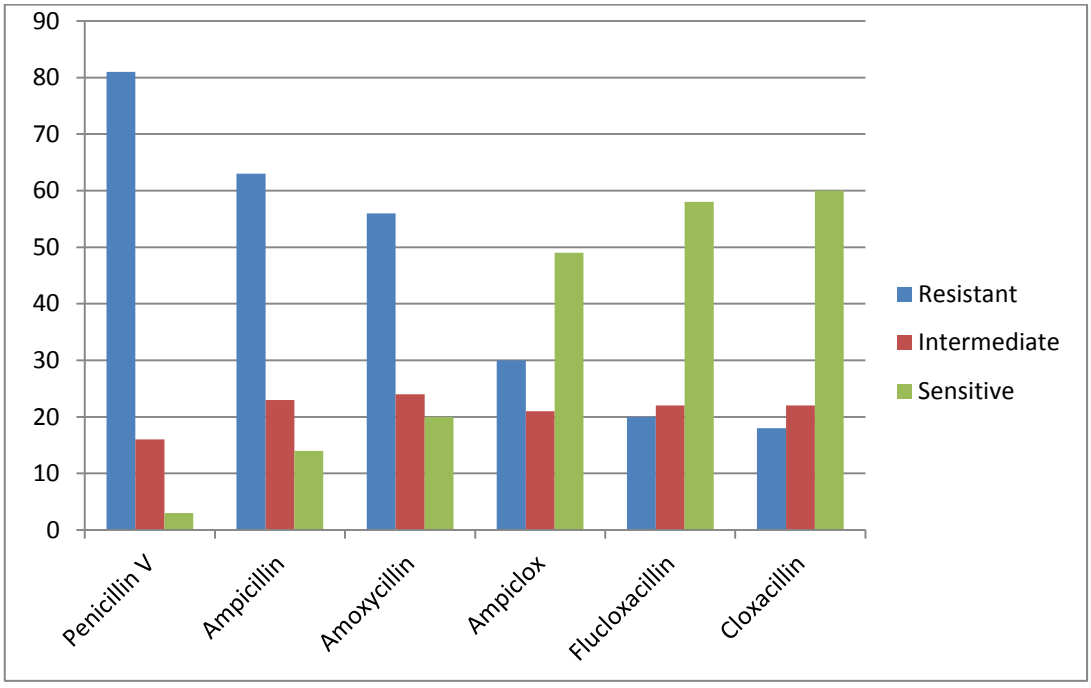

Figure 1: A bar chart showing the susceptibility patterns of isolates to the Penicillins

4. DISCUSSION

S. aureus has long been recognized as an important pathogen in human disease and is the most common cause of nosocomial infections [11]. The treatments of infections caused by *S. aureus* have always been limited nowadays due to the alarming rate of resistance to the conventional antibiotics we have currently. These organisms are capable of producing betalactamase, an enzyme that destroys the active part of pencillins and cephalosporins which act on the cell wall of the bacteria by inhibiting the production of PBPa. Inhibitors were incorporated to new generations' penicillins such as clavulanic acid but after some time bacteria were able to circumvent the action of these inhibitors and they were named as extended betalactamase. Greater than 50% resistance was observed to ampicillin, amoxicillin, penicillin V among *S. aureus* isolates (see Figure 1). The high resistance to penicillin and the total susceptibility to vancomycin are commonly noted for *S. aureus* isolated at different hospitals worldwide [12, 13, 14, 15,16]. Hoerlle and Brandelli [13] reported that *S. aureus* isolates presented a resistance rate to ampicillin and penicillin of 88%, against 91% in 2003 and

100% in 2004. These facts have already been observed in other hospitals [9, 12]. Many factors may have contributed to such level of resistance, including misuse of antibiotics by health professionals, unskilled practitioners and lay persons. In Third world countries like Nigeria it is a common practice that antibiotics can be purchased without prescription, which leads to misuse of antibiotics by the public, thus contributing to the emergence and spread of antimicrobial resistance. Other causal factors could be poor hospital hygienic conditions, accounting for the spread of resistant bacteria and inadequate surveillance, i.e. lack of information from routine antimicrobial susceptibility testing of bacterial isolates and surveillance testing of bacterial isolates and surveillance of antibiotic resistance, all of which are crucial for good clinical practice and for rational policies against antibiotic resistance [17]. It was observed that Cloxacillin, Flucloxacillin and Ampiclox showed better sensitivity patterns, reason could have been the incorporation of beta lactam inhibitors like clavulanic acid present in these antibiotics

5. CONCLUSION

The rate of antimicrobial resistance is to conventional antibiotics is very high, high level of resistance was observed to Penicillin V, Ampicillin and Amoxycillin than Ampiclox, Flucloxacillin and Cloxacillin. Continuous surveillance on antimicrobial susceptibility testing of *S. aureus* is essential for the detection of emerged resistant strains of *S.aureus* to Methicillin (MRSA) and the development of appropriate therapeutic strategies for empirical treatment and prophylaxis and in cases when sensitivity are done, the narrowest spectrum with highest efficacy should be administered. Information from these surveys will be a valuable for programs and policy decisions to forestall the emergence and spread of antimicrobial resistance of *S.aureus* in Abakaliki and in Nigeria as a whole.

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