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PREVALENCE OF METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS IN WOUND INFECTIONS OF PATIENTS ATTENDING SELECTED HOSPITALS IN ESAN CENTRAL, EDO STATE. NIGERIA

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ABSTRACT

This study investigates the prevalence of Methicillin Resistant *Staphylococcus aureus* (MRSA) infection associated with wound patients from selected hospitals in Esan-Central, Edo State. A total of 152 wound swabs specimens were collected and analyzed. The study population comprised 88 males and 64 females. Bacterial isolates were identified using Gram staining and biochemical tests, including catalase and coagulase; antibiotic susceptibility tests was done using Kirby Bauer disc diffusion method. 38 (25%) *Staphylococcus aureus* isolates were identified, out of which 31 (81.58%) were methicillin resistant and 7 (18.42%) were methicillin sensitive. In this study, the overall prevalence of MRSA in wound infection was 20.39%. The prevalence of MRSA in wound infection varied insignificantly ($p>0.05$) with age, with the highest respondents of ≥ 70 years old 1 (100%), followed by patients within the age range of 40-49years old 7 (25.0%), while those within age groups 30-39, ≤19, 60-69, 20-29 and 50-59 old had prevalence 9 (22.50%), 4 (22.22%), 2 (20.0%), 6 (15.38%), 2 (12.50) respectively MRSA infection amongst male was higher 21 (23.86%) than females 10 (15.63%) although not statistically significant ($p>0.05$). There was also no significant variation of MRSA infection based on occupation, although farmers had the highest prevalence, 9(32.14%) followed by teachers 6(28.57%). The results suggest the importance and role of diagnostic clinical microbiology in determining appropriate antimicrobial therapy thus limiting the emergence and cross transmission of antimicrobial resistant bacteria and in turn decreased morbidity and mortality.

KEYWORDS: Methicillin, Resistant, Staphylococcus, Wound, Infection, Nigeria.

INTRODUCTION

Wound infection is described as the deposition and multiplication of bacteria on a wound site causing an associated host reaction. Wound infection is the result of successful invasion and proliferation by one or more species of microorganism, to a level that involves a local and / or systemic response in the host resulting in the formation of pus.[1] Burn wounds, surgical sites, bite wounds, acute soft tissue infections, diabetic foot ulcers; leg and pressure ulcer infections are all examples of wound infections.[2,3] Wound infections are one of the most common hospital acquired infections and are an important cause of morbidity and account for 70-80% mortality.[4,5] Bacterial wound infections are economically important because they can slow down the healing process, lead to wound breakdown, prolonged hospital stay and increase in the cost of treatment.[6] Most wound infections are caused by bacterial colonization, initiating either from the normal flora on the skin, or bacteria from other parts of the body or the environment.[7]

Staphylococcus aureus is a gram positive and commensal bacterium that colonizes 30% of healthy individuals from different body parts; it is also a member of the normal flora of the skin.[8] *Staphylococcus aureus* is a common hospital and community acquired pathogen known to be frequently associated with wound infections.[9] Methicillin-resistant *Staphylococcus aureus* (MRSA) are isolates of *Staphylococcus aureus* which have acquired genes encoding antibiotic resistance to all penicillins including methicillin. This resistance is mediated by an altered penicillin binding protein (PBP2a) which is encoded by the MecA gene.[10] Numerous studies have shown that *Staphylococcus aureus* is among the most frequently encountered bacteria in Microbiology Laboratories in Nigeria.[11,12] Studies by[13] reported that S. aureus, P. aeruginosa and Proteus spp are the most common bacteria found in wound infections in Nigeria.

Wound infection continue to be a source of concern in clinical practice as they cause delayed or poor wound healing, leading to prolonged hospital stay and increased cost of hospitalization. Bacterial resistance to medications has made controlling wound infections more difficult, particularly in infections caused by MRSA and polymicrobial flora.[14] The condition is serious in developing countries, like Nigeria owing to lack of antibiotic stewardship amongst other factors. This study aims at the identification of isolates of *Staphylococcus aureus*, obtaining the antibiotic susceptibility pattern of MRSA isolates and determining the prevalence of MRSA strains from the wound samples in the study population.

MATERIALS AND METHODS

Study Area

This study was carried out in Esan Central and Esan West, Edo state. Esan Central is a Local Government Area of Edo Stat, Nigeria. The administrative headquarters is located in the town of Irrua and it has an area of 253km² and a population density of 545.1/km². Esan West Local Government Area has its headquarters in the town of Ekpoma and it lies between the latitude 60 43' and 60 45' North of the equator and longitudes 60 6' and 60 8' East of the Greenwich Meridian. It has an area of 502km and a population density of 333.3/km. it has an estimated population of over 190,000 people which consists of an adult male population of over 60,000 and adult female population of over 60,000.[15]

Edo state is a Nigerian state in the South-South geopolitical zone of the country. Edo state is the 22nd largest state by land mass, with a land area of 19,559km². Edo state borders Kogi state to the northeast, Anambra state to the east, Delta state to the southeast and south-south and Ondo State to the west.

Ethical Consideration

Ethical approval for the collection of wound swabs from patients was taken from the Health Research Ethics Committee Irrua Specialist Teaching Hospitals (ISTH). The ethical principle of scientific research as well as the related national laws and regulations was strictly adhered to, privacy and confidentiality was ensured.

Inclusion Criteria

The included group in this study are all patients with wound of various causes, from different hospitals.

Exclusion Criteria

The included group in this study are all patients without any form of wound.

Study Population

The target population for the study include in-patients and out-patients attending four selected hospitals in Edo state namely; Irrua Specialist Teaching Hospital (ISTH), Eromonsele Hospital, General Hospital Ekpoma and Ofure Medical Center.

Sample Collection and Transportation

The study sampled one hundred and fifty-two (152) wound cases collected by nurses randomly using commercially available sterile cotton swabs. The samples were preserved aseptically in sterile normal saline and then it was transported to the microbiology laboratory.

Isolation of Organism

The preserved specimens were placed on MacConkey agar, blood agar and Mannitol Salt Agar (MSA) plates using the streak plate method and incubated aerobically at 37˚C for 24-48 hours.

Characterization and Identification of Staphylococcal Isolates

This was done to identify the presumptive Staphylococcus isolates obtained from the different wound specimens analyzed. Characterization was done phenotypically on the basis of their colonial morphology,

microscopy (Gram stain) and biochemical attributes observed after performing catalase test and coagulase test for each of the isolates.

Antimicrobial Susceptibility Testing

The antimicrobial susceptibility testing of all identified isolates was done according to the criteria of the clinical and Laboratory Standards Institute method[16] using the Kirby Bauer disc diffusion method. Diameters of the zone of inhibition around the discs was measured using a transparent ruler, and the isolates was classified as sensitive, intermediate and resistant according to the standardized table supplied by.[16]

Interpretation of Zone Sizes

Using the Interpretative Chart, the zones sizes of each antibiotic were interpreted and the isolate reported as 'Resistant', 'Intermediate' or 'Susceptible'.[16]

Resistant

A pathogen reported as 'resistant' implies that the infection it has caused will not respond to treatment with the drug to which it is resistant irrespective of dose or site of infection. All strains showing a heaped-up zone edge, regardless of the size of the inhibition zone, was reported as '*Resistant*'.[16]

Intermediate

A pathogen reported as intermediately susceptible suggests that the infection it has caused is likely to respond to treatment when the drug is used in larger doses than normal or when the drug is concentrated at the site of infection.[16]

Susceptible

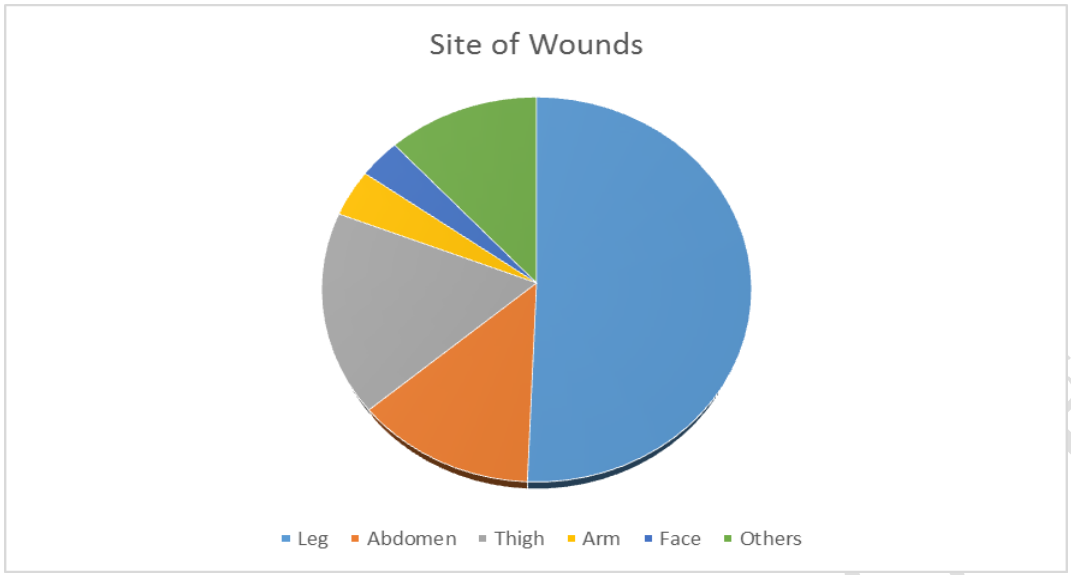
A pathogen reported as susceptible suggests that the infection it has caused is likely to respond to treatment when the drug to which it is susceptible is used in normal recommended doses and administered by an appropriate route.[16]

RESULTS

Table 1 and Figure 1 & 2 shows the socio-demographic data (age, gender, site of wound, causes of wound and occupation) of the study population. A total of 152 patients were examined for wound infection. Most of the respondents were within 30-39 years (26.32%) of age, followed by 20-29 years (25.66%), 40-49 (18.42%), 0-19 years (11.84%), 50-59 years (10.53%), 60-69 years (6.58%) and ≥70 years (0.66%). Based on gender most of the respondent were males (57.89%), while the others were females (42.11%). The most common site of wound on respondents was leg (50.66%), followed by thigh (17.11%), Abdomen (13.16%), others (11.84%), Arm (3.95%), and Face (3.29%). Most of the respondent were Civil servants (26.97%), followed by Students (19.74%) then Farmers (18.42%), Teachers (13.82%) and others (21.05%). Most of the respondents has cause of wound to be from accident (63.82%) followed by surgery (22.37%), burn (7.24%), cut (1.32%), and others (6.59%).

Table 1: Socio-Demographic Data of The Population (N=152).

		Frequency	Percentage
Age (years)	≤19	18	11.84
	20-29	39	25.66
	30-39	40	26.32
	40-49	28	18.42
	50-59	16	10.53
	60-69	10	6.58
	≥70	1	0.66
	TOTAL = 152		
Gender	Male	88	57.89
	Female	64	42.11
	TOTAL = 152		
Occupation	Civil Servant	41	26.97
	Farmer	28	18.42
	Teachers	21	13.82
	Student	30	19.74
	Others	32	21.05
	TOTAL = 152		



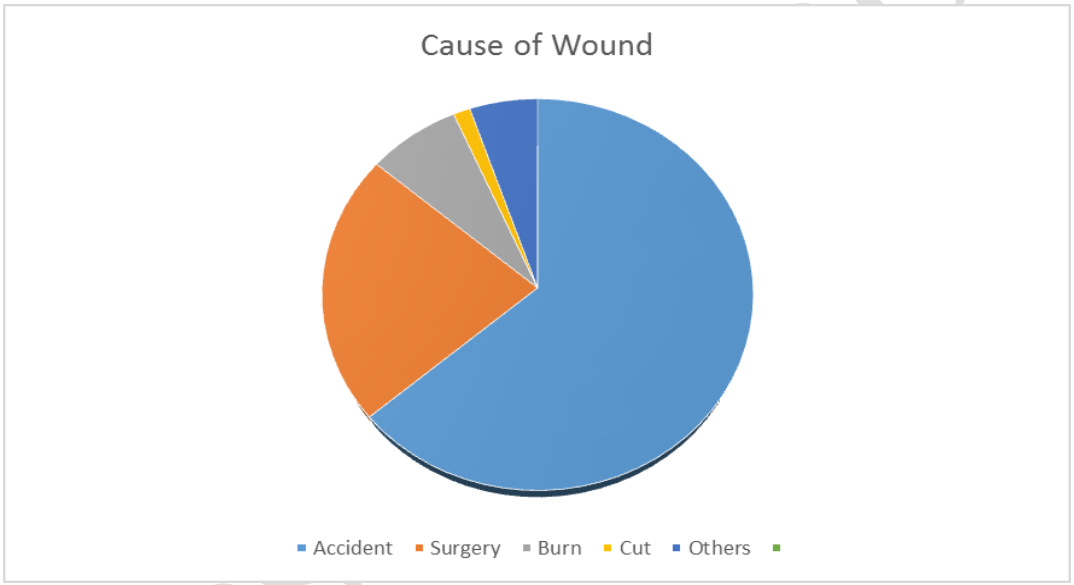


Table 2: Distribution of *S Aureus* and Mrsa Among Patients With Wound Infection In Relationship With The Selected Hospitals.

HOSPITALS	NUMBER EXAMINED	NO. OF *S. AUREUS*	NO OF MRSA	NO OF MSSA
ISTH	100	28 (28.0%)	24 (24%)	4 (4%)
GENERAL HOSPITAL	32	7 (21.88%)	6 (18.75%)	1 (3.13%)
OTHERS	20	3 (15%)	1 (5%)	2 (10%)
TOTAL (%)	152 (100%)	38 (25%)	31 (20.39%)	7 (4.61%)

Table 3: Distribution of Mrsa In Wound Infection Based on Age Range.

AGE (YEARS)	NUMBER EXAMINED	NO. OF *S AUREUS* ISOLATED (%)	NO. OF MRSA (%)	NO. OF MSSA (%)
≤19	18	7 (38.9)	4 (22.22)	3 (16.67)
20-29	39	8 (20.51)	6 (15.38)	2 (5.13)
30-39	40	10 (25.0)	9 (22.50)	1 (2.50)
40-49	28	8 (28.57)	7 (25.0)	1 (3.57)
50-59	16	2 (12.50)	2 (12.50)	0
60-69	10	2 (20.0)	2 (20.0)	0
≥70	1	1 (100)	1 (100)	0
TOTAL	**152**	**38 (25)**	**31 (20.39)**	**7 (14.60)**

Table 4: Distribution of Mrsa in Wounds Infection Based on Gender.

GENDER	NUMBER EXAMINED	NO. OF *S AUREUS* ISOLATED (%)	NO. OF MRSA (%)	NO. OF MSSA (%)
MALE	88	26 (29.55)	21 (23.86)	5 (5.68)
FEMALE	64	12 (18.75)	10 (15.63)	2 (3.13)
TOTAL	**152**	**38 (25)**	**31 (20.39)**	**7 (14.60)**

Table 5: Distribution of Mrsa in Wound Infection from Different Sites of Wound.

SITE OF WOUNDS	NUMBER EXAMINED	NO. OF *S AUREUS* ISOLATED (%)	NO. OF MRSA (%)	NO. OF MSSA (%)
LEG	77	24 (31.17)	19 (24.68)	5 (6.85)
ABDOMEN	20	7 (35.0)	6 (30.0)	1 (5.0)
THIGH	26	2 (7.69)	2 (7.69)	0
OTHERS	18	5 (27.78)	4 (22.22)	1 (5.56)
TOTAL	**152**	**38 (25)**	**31 (20.39)**	**7 (14.60)**

Table 6: Distribution of Mrsa in Wound Infection Based on Cause of Wound.

CAUSES OF WOUND	NUMBER EXAMINED	NO. OF *S AUREUS* ISOLATED (%)	NO. OF MRSA (%)	NO. OF MSSA (%)
ACCIDENT	97	24 (24.74)	19 (19.59)	5 (5.15)
SURGERY	34	9 (26.47)	8 (23.53)	1 (2.94)
BURN	11	1 (9.09)	1 (9.09)	0
OTHERS	10	4 (40.0)	3 (30.0)	1 (10.0)
TOTAL	**152**	**38 (25)**	**31 (20.39)**	**7 (14.60)**

Table 7: Distribution of Mrsa in Wound Infection Based on Occupation.

CAUSES OF WOUND	NUMBER EXAMINED	NO. OF *S AUREUS* ISOLATED (%)	NO. OF MRSA (%)	NO. OF MSSA (%)
CIVIL SERVANTS	41	14 (11.66)	10 (24.39)	4 (9.76)
FARMERS	28	11 (39.27)	9 (32.14)	2 (7.14)
TEACHERS	21	6 (28.57)	6 (28.57)	0
STUDENTS	30	3 (10.0)	2 (6.67)	1 (3.33)
OTHERS	32	4 (12.50)	4 (12.5)	0
TOTAL	**152**	**38 (25)**	**31 (20.39)**	**7 (14.60)**

Table 8: Antibiotic Resistance Pattern of Mrsa from Wound Infection Resistance Pattern (%).

ANTIBIOTICS	*S. AUREUS* (N=38)	MRSA (N=31)	MSSA (N=7)
CEFOTAXIME	35 (92.1%)	29 (93.5%)	6 (85.7%)
OFLOXACIN	17 (44.7%)	16 (51.6%)	1 (14.2%)
AZITHROMYCIN	23 (60.5%)	21 (67.7%)	2 (28.6%)
GENTAMYCIN	25 (65.8%)	23 (74.2%)	2 (28.6%)
CEFOXITIN	31 (81.6%)	31 (100%)	0
ERYTHROMYCIN	28 (73.7%)	25 (80.6%)	3 (42.8%)
CEFUROXIME	30 (78.9%)	27 (87.1%)	3 (42.8%)
AVERAGE	**27 (71.16%)**	**24.6 (79.26%)**	**2.4 (34.76%)**

DISCUSSION

This study identified isolates of *Staphylococcus aureus*, obtained the antibiotic susceptibility pattern of MRSA isolates and also determined the prevalence of MRSA strains from various wound samples collected from patients in Esan central, Edo State. Wound infections are costly complications that increase morbidity and mortality in hospitalized patients.[17] reported that S. *aureus*, *P. aeruginosa* and *Proteus spp.* are the most common bacteria implicated in wound infections in Nigeria. MRSA is spread throughout many hospitals and health care institutions and it is the most commonly isolated antimicrobial resistant pathogen condition.[18]

The overall prevalence of MRSA in wound infection from this study was 20.39%, from the 38 Staphylococcus aureus isolates obtained, the prevalence of MRSA was 81.59%.[19] had reported an MRSA prevalence of 43.5% in Jos, Nigeria, of which 81% was from in-patients.[20] reported 28.6% prevalence in Kano, Nigeria with 62% from in-patients.[21] had earlier reported a prevalence of 34.7% in Ilorin with 70.6% from in-patients These findings indicates an increase in the trend of MRSA prevalence in Nigeria. Unhygienic conditions and non-adherence to or lack of relevant antibiotic policy have been suggested as possible reasons for these high carriage rates.[22] This study Staphylococcus aureus

shows that microbial wound infection has no significant variation with age, although the prevalence was higher in males (23.86%), compared to the females (15.63%). However, other studies reported gender variation and concluded that gender and age does not contribute to the pattern of microbial wound infection.[23,24] [25] also reported that age have no bearing on wound infections with persons in their fouth to fifth decades of life being more prone to microbial wound infection. This study also shows that microbial wound infection has no significant variation with age.

The prevalence of MRSA observed in this study and those of the in-patients in other studies indicate that MRSA continues to be a menace in Nigerian hospitals and that the spread is nocosomial. This may not be unconnected with the hospital environment, for example, arrangement of patients in rooms and wards facilitates the transfer of these organisms among in-patients and even health care providers. Unhygienic conditions and non-adherence to or lack of a relevant antibiotic policy have been suggested as possible reasons for these high carriage rates.[22] Antibiotic susceptibility of isolates to commonly used antibiotics was very low; Ofloxacin and Azithromycin were the most potent antimicrobial agents observed in our study. The multiple resistance of *Staphylococcus aureus* isolates to commonly used antibiotics in the locality of this study calls for immediate action on the controlled use of antimicrobials in hospitals and the need to monitor resistance. Good antimicrobial use is necessary for effective wound management. The susceptibility rate of bacterial isolates observed in this study agrees with the reports of.[23,24,25,26] [23] also reported that high level of antibiotic abuse in Nigeria arise from self-medication which is associated with inadequate dosage and failure to comply with treatment regimen. These antibiotics are sold over the counter with or without prescription.

CONCLUSION

Staphylococcus aureus has become an important hospital and community acquired pathogen. This problem is further compounded by development of methicillin resistance. Resistance to this antibiotic implies resistance to all β-lactam antibiotics, the most important group of antibiotics to treat staphylococcal infection. Hospital infection by MRSA strains has not only caused therapeutic problems, but also put tremendous pressure on resources controlling their spread. Therefore, it is important that diagnostic microbiology laboratories identify the organism accurately, which helps in determining the appropriate antimicrobial therapy. Consequent benefits will be shorter hospital stay, lower hospital cost (by preventing unnecessary use of Vancomycin-glycopeptides and isolation precautions), limiting the emergence and cross transmission of antimicrobial resistant bacteria and in turn decreased morbidity and mortality.

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