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Frequency and Antimicrobial Pattern of *Staphylococcus aureus* Isolated From Patients with Infected Wound Attending Wad Madani Teaching Hospitals

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Abstract: Wounds are a significant cause of morbidity worldwide, studies show that for every million wound patients, at least 10.000 die from microbial infections. The skin is a vital organ that serves as a protective barrier between the human bodies on its external environment. *Staphylococcus aureus* has been known as a cause of deep-seated wound infection for close to a century, having recognized as a cause of nosocomial infection and super infection in patients receiving antimicrobial agent such as surgical cases. MRSA colonizing the anterior nares and skin of human are the major sources of surgical site infection as well as nosocomial spread. Methicillin-resistant *Staphylococcus aureus* (MRSA) are bacteria resistant to penicillin and cephalosporin classes of antibiotics and are often resistant to many other classes of antibiotics. Importance of *S. aureus* as persistent nosocomial and community acquired pathogen has become a global health concern. The aim of this study to determine the frequency and antibiotic susceptibility pattern of *S.aureus* isolated from wound infected patient attending Wad Madani Teaching Hospitals. Two hundred samples collected from patients with infected wound at the period from April to July 2016. Out of 200 samples examined, 60 (30%) isolates of *S. aureus* obtained, 116 (58%) other bacterial species and 24 (12%) samples showed no growth. The highest isolation rate of *S. aureus* 60% occurred among age group (41-60) and according to gender 32 (53.3%) *S. aureus* isolated from male and 28 (46.7%) from female. The overall profile of susceptibility pattern showed that vancomycin and linezolid were more sensitive (100%), amoxicillin and penicillin were the least sensitive (10%) and (36%) respectively. The result of the study showed that frequency of *S. aureus* isolated from infected wounds 60 (30%) and vancomycin and linezolid are more effective in the management of *S. aureus* in this locality.

Keywords: *Staphylococcus aureus*, Wad Madani Hospitals, Antimicrobial susceptibility pattern, wound infection, MRSA.

INTRODUCTION

Wounds are a significant cause of morbidity worldwide, studies show that for every million wound patients, at least, 10.000 die from microbial infections [1]. The skin is a vital organ that serves as a protective barrier between the human bodies on its external environment [2]. Breaks in the skin, such as ulcers or traumatic wounds, expose the subcutaneous tissue, providing appropriate moisture, temperature, and nutritive conditions for microbial colonization [3]. *S. aureus* has been known as a cause of deep – seated wound infection for close to a century, having recognized as a cause of nosocomial infection and super infection in patients receiving antimicrobial agent such as surgical cases [4]. MRSA colonizing the anterior nares and skin of human are the major sources of surgical site infection as well as nosocomial spread. Several trends have been identified in the epidemiology of MRSA infection increasing incidence of MRSA infection, particularly among wound infected patients [5]. Increasing proportion of nosocomial infection caused by MRSA. By the early 1990, MRSA accounted for 20-25% of *S. aureus* isolates from hospitalized patients. In1999, MRSA accounted for > 50% of *S. aureus* isolates from patients in ICU in the (national nosocomial infection "surveillance"(NNIS) system. In 2003, 59.5% of S. *aureus* isolates NNIS ICUS were MRSA [6, 7].

Methicillin-resistant *Staphylococcus aureus* (MRSA) are bacteria resistant to penicillin and cephalosporin classes of antibiotics and are often resistant to many other classes of antibiotics [8].

In MRSA, the horizontally acquired mecA gene encodes penicillin – binding protein (pBp2a), which is intrinsically insensitive to methicillin and all B-lactams that have been developed, including the

isoxazoyl penicillin (e. g Oxacillin) that superseded methicillin [9]. MRSA infections are a problem across the whole health economic, and have shown to be associated with a poorer outcome and higher mortality than similar infection caused by methicillin – sensitive strains of *S. aureus* [10, 11].

Many of these isolates are becoming multi-drug resistance and are susceptible only to glycopeptides antibiotics such as vancomycin [12].

METHODS

Samples, Setting and study period

Across-sectional, prospective study was conducted in wound infected patients attending in general surgery Hospital, orthopedics surgery Hospital, diabetic center and Maize Hospital in Wad Madani Hospitals from April to July 2016. A total of 200 wound pus swabs specimens were collected by using convenient sampling technique after getting verbal consent. Specimens were collected aseptically from all in patients and out patients during the study period attending Wad Madani Teaching Hospitals. All swab specimens were transported to medical microbiology laboratory for culture and sensitivity testing. The sample collection, culturing, staining and sensitivity testing were performed according to the standard-microbiological diagnosis for *S. aureus* [13]. The specimens were collected by the attending physician and health officer using sterile cotton swabs moistened with normal saline. The swabs socio-demographic variables (age and sex) were taken from patients record. The swab was then inoculated on blood agar and MacConkey agar plates then incubate at 37c° for 24 hours after, the plate had been left at room temperature for pigment formation, colonies were selected and checked for Gram stain, catalase test, DNase test and coagulase test then sensitivity testing was done.

Antimicrobial susceptibility test

All *S. aureus* isolates were done according to the CLSI (Clinical and Laboratory Standard Institute) by disc diffusion method [14]. From a pure culture, 3-5 selected colonies of *S. aureus* had been taken and transferred to a tube containing 5 ml sterile nutrient broth and were mixed gently. Then a homogenous suspension was formed and incubated at 37^0C until the turbidity of the suspension became adjusted to a 0.5 McFarland standard (Bacterial concentration of $1.5x10^8$ colony forming unit/ml). A sterile cotton swab was used and the excess suspension was removed by gently rotation of the swab against the surface of the tube. The swab was then used to distribute the bacteria evenly over the entire surface of the Mueller Hinton plate (pH7.2-7.4). The inoculated plates were left at room temperature to dry for 3-5 minutes and a set of 8 antibiotic discs with the following concentration were then evenly distributed on the surface of Mueller Hinton plate methicillin 5 mcg, penicillin 10 mcg, Amoxicillin 10 mcg, Amoxicillin/clavulanic acid 10 mcg, vacomycin 30 mcg, gentamicin 10 mcg, ciproflaxcin 5 mcg and Linzolid 30 mcg. The criteria used to select the antimicrobial agents to be tested were based on their availability and frequent prescription for the management of wound infections in the hospital. The plates were then incubated at 37^0C for 24 hours. Diameters of zones of inhibition round the discs were measured to nearest millimeter using to the isolates were classified as sensitive, and resistant according to standardized table supplied by the CLSI.

RESULTS

Two hundred patients with wound infection attending Wad Madani Teaching Hospitals. Out of 200 samples examined, 60 (30%) isolates of *Staphylococcus aureus* obtained, 116 (58%) other bacterial species and 24 (12%) samples showed no growth (Table-1). The highest isolation rate of *S. aureus* was 36 representing 60% occurred among age group (41-60) years, this illustrated in (Table-2).

According to gender 32 (53.3%) *S. aureus* were isolated from male and 28 (46.7%) from female, as shows in (Table-3). Looking at (Table-4) the antibiotic susceptibility test revealed that in all age groups, the isolates showed a marked sensitivity to linezolid and vacomycin.

The levels of sensitive and resistance showed that vancomycin and Linezolid were more sensitive (100%), amoxicillin and penicillin were the least sensitive (10%) and (36%), also *S.aureues* shows some levels of resistance to amoxicillin (90%), penicillin (63%) (Table-5).

Table-1: frequency of bacterial isolates

Bacterial Species	No. (%)
Staphylococcus aureus	60 (30%)
Other type of bacteria	116 (58%)
no growth	24 (12%)

Table-2: Disruption of *Staphylococcus aureus* among age groups

Age range	Number
< 20	2 (3.3%)
21 – 40	10 (16.6%)
41 – 60	36 (60%)
> 60	12 (20%)

Table-3: Distribution *Staphylococcus aureus* among gender

Gender	No. (%)
Male	32 (53.3%)
Female	28 (46.7%)
Total	60 (100%)

Table-4: Antimicrobial susceptibility pattern among age groups

Age	Isolate	Pen	Amc	Amx	Meth	Gm	Cip	Lz	Va
20		(50%)	(100%)	(50%)	(100%)	(100%)	2(100%)	2(100%)	2(100%)
1-40	0	(50%)	(50%)	(10%)	(60%)	(60%)	10(100%)	10(100%)	10(100%)
1-60	6	12(33%)	8(50%)	(5%)	2(33%)	4(66%)	30(83%)	36(100%)	36(100%)
60	2	(33%)	(50%)	(16%)	(58%)	(75%)	9(75%)	12(100%)	12(100%)

Pen: Penicillin	AMC: Amoxicillin/clavulanic acid	AMX: Amoxicillin	Meth: Methecillin
GN: Gentamicin	Cip: Ciproflaxcin	Lz: linezolid	Van: Vancomycin

Table-5: antibiotic sensitivity and resistance pattern of *Staphylococcus aureus* in Wad Mdani Teaching Hospitals

Type antibiotic (No)	Sensitive No (%)	Resistance No (%)
Penicillin 60	22 (36%)	38 (63%)
Amoxicillin/clavulanic acid 60	31 (50%)	30 (50%)
Amoxicillin 60	6 (10%)	54 (90%)
Methicillin 60	27 (45%)	33 (55%)
Gentamycin 60	43 (71)	17 (28%)
Ciproflaxcin 60	51 (85%)	9 (15%)
Linezolid 60	60 (100%)	0 (0%)
Vacomycin 60	60 (100%)	0 (0%)

DISCUSSION

This study showed the occurrence of *Staphylococcus aureus* among wound infected patients admitted to Wad Madani Teaching Hospitals. Two hundred (200) samples were analyzed; Table-1 shows the frequency of occurrence of *S. aureus* and other bacterial isolates in pure cultures.

S. aureus had the highest number of occurrence 30% (60) comparing with other types of bacteria isolated, these finding agree with previous findings investigated by Anguzu and Olila [15], the most single cause of post operative wound infection was *S. aureus* [16]. A high frequency of occurrence was recorded among the male patients (53.3%) more than female patients with (46.7%). The difference seems to be too small although statistical difference was not determined. *S. aureus* has been documented to be most liable to infected new born babies, surgical patient, old and malnourished person and patient with diabetes and other chronic disease this result find in the study of Basker *et al.*, [17].

Age group (41-60) years have the highest percentage of isolates of (60%) this result agrees with the finding of Law and Wrong 1980. The patients in age group greater than 40 years were associated with a higher surgical wound infection [18] and that was disagree with the finding of Nwnkwo and Nasiru, 2011, that observed higher frequency in age group (1-10). And older although this study concentrated on surgical wound infection [19]. Regarding the antibiotic susceptibility test revealed that in all age group the isolates showed a marked sensitivity to linezolid and vacomycin in this study was 100% which agree with the study carried out in Khartoum by Marwa *et al.*, 2016 that found vancomycin sensitivity was 100%, may be related to the low usage of this antibiotic in this study area [20]. Also *S. aureus* shows some level of resistance to amoxicycillin (90%) and penicillin (63%), this is level of resistance to penicillin was disagree observed in another study conducted by Salah *et al.*, 2012 to determined level resistance of penicillin (92%) in Khartoum state [21]. Methcillin resistance *S. aureus* was prevalence rate (60%) that disagree the prevalence rate of MRSA 74% was observed in another study conducted by Seddig , 2007 to determined prevalence of antibiotic resistance among pathogenic bacteria isolated from 3 major hospitals in Khartoum [22], resistance of antibiotic due to many factor ranging from abuse of drugs by patients and unrestricted use of broad spectrum antibiotic for prophylactic measures prior to surgery or operatively without resorting to sensitivity test. The resistance of *Staphylococcus aureus* to penicillin and other antibiotic can be due to the production of beta lactamase or possession of extra chromosomal factor, and R-factor.

CONCLUSION

The present study alarming levels of *S. aureus* cross resistance to antibiotic is essential in order to control their spread in hospital environment and transmission to the community, in addition to the important of appropriate selection of antimicrobial agents by clinicians. Intervention such as antibiotic prescribing polices and stringent infection control measure is needed to be enforced.

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