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Research Article

Prevalence of MRSA nasal colonization among community members and healthcare workers at private and public hospitals in Khartoum State, Sudan.

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ABSTRACT

Background:MRSA is a growing public health threat around the world. Until now, we are still unable to control the spread of staphylococcal infection and the development of resistance.

Objectives: To determine the prevalence of MRSA nasal colonization and its antimicrobial susceptibility profile colonization among community members and healthcare workers at four private and public hospitals in Khartoum State, capital city of Sudan.

Materials and methods: A total of three hundred and seven nasal swabs were collected during March to April 2017. Isolates were identified using conventional laboratory assays. Kirby–Bauer disk diffusion method was used for antibiotic susceptibility testing and methicillin resistance was confirmed using Oxacillin disks.

Results: Of the 70 S.aureus isolated, 80% represented MRSA. The occurrence of MRSA was significantly higher among healthcare worker than community individuals [51.42% (36/70) vs. 28.57% (20/70)] (p=0.008). The overall resistance rates to commonly prescribed antibiotics in isolates were low with slightly high percentage of intermediate strains. Tetracycline was the least effective agents for hospital infections of S.aureus with sensitivity being 55%.

Conclusion: The observed pattern of susceptibility showed those medical care workers were colonized with more antibiotics-resistant S.aureus isolates than community members. Of greater significance is the finding that all community isolates were MRSA strains which indicates a growing urgency to study the colonization behavior of CA-MRSA in Sudan in order to create plausible strategies for controlling this potential pathogen.

Keywords:MRSA, S.aureus, nasal carriage, Antimicrobial susceptibility

1. Introduction

Staphylococcus aureus is a facultative anaerobic gram positive coccal bacterium.(1) It is one of the most popular human pathogen and is capable of causing wide range of infections in human.(2) Although staphylococcal disease is most often associated with skin and soft tissue infections (e.g., pimples, impetigo, boils, cellulitis, folliculitis, carbuncles, scalded skin syndrome, abscesses), S. aureus has also been implicated as a causal organism in various other diseases.(3),(4)It can causes endocarditis, osteomyelitis, conjunctivitis, pneumonia, meningitis, septicemia and toxin-mediated diseases such as toxic shock syndrome, scalded-skin syndrome, neonatal toxic shock syndrome like exanthematous disease and food poisoning.(1),(5),(6)

The most ecological niches of S.aureus strains are the anterior nares. People who are chronic carriers of S.aureus in their nose have an increased risk for subsequent infection and transmission of these bacteria. Colonization is facilitated by the anatomy of the nasal vestibule and the ability of S.aureus to resist the microbicide peptides in the mucus.(3),(6),(7), (8), (9), (10)There is a difference in nasal colonization patterns by S.aureus with unknown reasons yet. A large proportion of population (60 %) is intermittent carriers while 20 % of the population is persistent colonized with S.aureus and 20 % of population almost never carries this organism.(2),(5)Persistent carriage acts as a barrier of the acquisition of multidrug resistance strains of

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S.aureus, especially MRSA, but this barrier is reduced when carriers are treated with antibiotics.[8]So the risk factors for acquisition of MRSA include antibiotics abuse, prolonged hospitalization, intravascular instrumentation, hospitalization in an intensive care unit and the presence of other patients with MRSA colonization or infection in the hospital.[5],[8],[11]

MRSA infections are now categorized to health care-associated MRSA (HA-MRSA) and community-associated MRSA (CA-MRSA) infections. Its colonization is an important risk factor for subsequent MRSA infection. However,CA-MRSA strains and HA-MRSA strains differ in terms of epidemiology, microbiology, and clinical manifestations.[2],[9],[12] Additionally, CA-MRSA is more susceptible to antibiotics and carries staphylococcal cassette chromosome methicillin resistant locus (SCCmec) type IV or V.[12]

Infections caused by MRSA strains are increased steadily.The 2014 WHO Global Health Report on Antimicrobial Resistance reported that in all the WHO regions, MRSA prevalence of infections was above 20 % and increased the risk of morbidity and mortality,also associated with healthcare costs in terms of antibiotic therapy, isolation facilities and materials and length of hospital stay.[7],[13] In view of the rapid increase in the incidence of MRSA and the problems associated with its control and therapy, more insight into the prevalence of MRSA colonizationand its antimicrobial resistance pattern is needed to provide a useful estimate of the potential for development of S.aureus infections then improvement more effective preventive strategies.Therefore, this study was conducted to determine the prevalence of MRSA nasal colonization and its antimicrobial susceptibility profile colonization among community members and healthcare workers at four private and public hospitals in Khartoum State, capital city of Sudan.

2. MATERIALS AND METHODS

2.1. Study design and settings:

This descriptive comparative cross sectional study was carried out during the period from March to April in 2017. A total of Three hundred and fifty-seven nasal swab samples were collected randomly after obtaining verbal consent from the adult community members (n=200) and the healthcare workers, including doctors, nurses and medical technologists in the four private and public hospitals (n=157), in Khartoum State. Distribution of healthcare workers' samples from four private and public hospitals in percentage is given in Figure 1. The exclusion criteria were hospitalized community members, while the inclusion criteria was community members who were apparently healthy individuals.

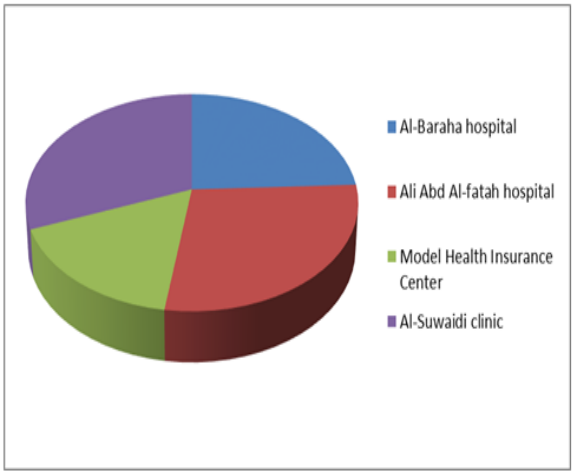

Figure 1: Percentage of hospitals' data

2.2. Sample collection:

Nasal swabs were collected from subjects by inserting sterile cotton wool swab and rotating four times for each anterior nares while applying an even pressure. Aseptic technique was followed. The swab samples were transported immediately to the Microbiology Laboratory at the Faculty of Medical Laboratory Sciences, University of Khartoum and were processed within two hours.

2.3. Microbiological study

2.3.1. Isolation and identification of S.aureus

For the isolation of S.aureus, the collected swab samples were inoculated onto Manitol salt agar plate (Oxoid, Basingstoke, England) and incubated at 37°C overnight. S. aureus colonies were selected on the basis of their colonial morphology, indirect Gram stain, catalase production, coagulase test and DNase test. [14]

2.3.2. Antimicrobial susceptibility testing

Antimicrobial susceptibility testing of S.aureus isolates was performed by the Keby-Bauer disk diffusion method using Mueller-Hinton agar plate (Difco Laboratories, Detroit, USA). Several

antibiotics used were: erythromycin (15µg), tetracycline (10 µg), oxacillin (1 µg) and Augmintin (30µg). All cultured plates were aerobically incubated at 37ºC for overnight. Isolates were considered sensitive, intermediate or resistant in compliance with Clinical and Laboratory Standards Institute (CLSI) guidelines. Zones of inhibition are measured to the nearest whole millimeter.[15]

2.3.3. Detection of methicillin resistant strains (MRSA)

A disk diffusion method with oxacillin (1 µg) was used to detect MRSA. This test was carried out along with each susceptibility testing of the isolate being performed. All the S.aureus isolates that showed Oxicillin inhibition zone diameter of (<10 mm) were considered as MRSA strains while (>13 mm) was reported as MSSA strain.[15]

3. RESULT

3.1. The prevalence of MRSA among healthcare workers and community members:

Out of the 357 nasal swab samples were examined, S.aureus was detected in 70(19.61%) of the total samples. Of these70 positive samples, 50(71.42%) isolates were collected from the healthcare workers, while 20(28.57%) isolates from the community members. A total of 56 of these 70 isolates (80%) were found to be MRSA, 36 isolates were isolated from the healthcare workers, while 20 collects from the community members.The frequency of MRSA isolates from hospital healthcare workers and community members at Khartoum State is illustrated in Table 1.

Table 1: Frequency of MRSA and MSSA isolates from hospital healthcare workers and community members at Khartoum State

Source of isolates	MRSA	MSSA	Number of isolates
Healthcare workers	36(72%)	14(28%)	50
Community members	20(100%)	0	20
Total	56(80%)	14(20%)	70

3.2. Antibiotic susceptibility test:

The results of antimicrobial susceptibility test of the S.aureus isolated from community members (n=20) and healthcare workers (n=50) are given in Tables 2-3.

Table 2: The results of antimicrobial susceptibility test of the S.aureus isolated from community members

Antibiotic	Sensitive	Intermediate	Resistance
Oxacillin(1 µg)	0	0	20(100%)
Augmentin(30µg)	15(75%)	4(20%)	1(5%)
Eryrhromycin(15µg)	18(90%)	2(10%)	0
Tetracycline(10 µg)	11(55%)	6(30%)	3(15%)

Table 3: The results of antimicrobial susceptibility test of the S.aureus isolated from healthcare workers

Antibiotic	Sensitive	Intermediate	Resistance
Oxacillin(1 µg)	14(28%)	0	36(72%)
Augmentin(30µg)	32(64%)	15(30%)	3(6%)
Eryrhromycin(15µg)	42(84%)	3(6%)	5(10%)
Tetracycline(10 µg)	37(74%)	7(14%)	6(12%)

DISCUSSION

Nasal carriage of *S.aureus* in the absence of discernible clinical disease serves as a potential source of infection for others. It has been reported as a risk factor for community acquired and nosocomial infection.[2]Until now, we are still unable to control the spread of *staphylococci* and the development of resistance. MRSA is a growing public health threat around the world. Emergence of resistance to Vancomycin is alarming.[8]In our study, the prevalence of *S.aureus* nasal colonization among healthcare workers at four private and public hospitals was 31.84% and that of healthy community individuals was 10% in Khartoum state. Similarly in one study, Alaa*et al* have found that the prevalence of *S. aureus* nasal colonization among healthcare workers at the Soba University Hospital and that of healthy community individuals were 32.7% and 6.8%, respectively.[16] Cruickshank *et al.* reported that the chances of nasal harboring *S.aureus* by hospital personnel and ward attendants were usually higher than non-hospital personnel and could be easily implicated in severe hospital infections.[17]However, worldwide studies have been reported *S.aureas* nasal colonization with different results.[9],[11],[18],[19] This may be due partly to differences in the population studied, the quality of the sampling and of the culture techniques used in these studies.[3],[8]

Methicillin, the first antibiotic of β-lactamase-resistant penicillins (Methicillin, Oxacillin, Cloxacillin, and Flucloxacillin) was introduced in 1959 and only after two years (1961) it was reported as ineffective.[1],[5]In the present study, the Oxacillin disk method was used for the detection of MRSA. According to CLSI guidelines, the mecA mediated resistance to Oxacillin can be detected by Oxacillin disk method.[5]The prevalence rate of MRSA carriage in our study was found to be 80%. In a similar study done in Sudan by Alaa*et al.* reported that the prevalence rate of MRSA carriage was 20.2%.[16] Such variation was found in different studies in Iran which is ranged from 20.48% to 90%.[12],[20] Also some of the reports show an alarmingly high incidence of MRSA infection inNebal, India and Saudi Arabia.[21],[22],[23]This alarmingly high percentage of methicillin-resistant strains along with the poorly controlled distribution and inappropriate use of antibiotics may aggravate the clinical impact of nasal *S.aureus* colonization in the coming years in Sudan.[4]

CA-MRSA is responsible for around 30 % of *S.aureus* infections in hospitals of USA.[9] Of greater significance is the finding that all community isolates were MRSA strains which indicates a growing urgency to study the colonization behavior of CA-MRSA in Sudan in order to create plausible strategies for controlling this potential pathogen.However, several studies suggest that factors associated with CA-MRSA carriage include prior antibiotic usage, contact with health care facility, poor socioeconomic conditions, and overcrowding.[9],[16] While in a hospital, colonized healthcare workers of hospital such as asymptomatic nasal and hand carriers acting as reservoirs are important sources for spreading this organism. Multiple and prolonged use of antibiotics and prolonged hospitalization are other important factors which make hospital an ideal place of transmission and perpetuation of MRSA.[9]Our result shows MRSA carriage rates were significantly higher (p=0.008) among healthcare workers than in healthy adults from the community which is in accordance with the earlier studies by Cruickshank *et al.* (1975),[17] Shakya*et al.* (2010),[1] and Alaa*et al.* (2015).[16]

Due to an increasing number of infections caused by MRSA strains, empirical therapeutic options for *Staphylococcal* infections in the hospital and community settings have become problematic.[8],[16]As shown in Tables 3-4,the overall resistance rates to commonly prescribed antibiotics in isolates were low withslightly high percentage of intermediate strains which need a higher dosage of drugs and for a longer time period.[24]The observed pattern of susceptibility showed those medical care workers were colonized with more antibiotics-resistant*S.aureus* isolates than community members which in agreement with older study done by Abdalla*et al.* in 1998.[4]In addition, our antibiotic resistances pattern among HA-MRSA almost reflects the frequency at which antibiotics prescribed over the counter in Sudan as a whole are used.[4] This mechanism of increased spreading under antibiotic pressure may have participated in the increase incidence of MRSA in hospitals worldwide.[8]Interestingly, all CA-MRSA isolates

were sensitive to Erythromycin which is in a contrary to study done by Shakya et al. (2010) which reported that HA-MRSA isolates were sensitive to Erythromycin.[1]Tetracycline was the least effective agents for hospital infections of S.aureus with sensitivity being 55%. Also Anupurba*et al.*, Kumari*et al.,* and Alaa*et al.* reported less efficacy of tetracycline against nasal*S.aureus*carriage.[11],[16], [21]Interestingly, older data has been shown that administration of tetracycline to patients colonized with a tetracycline-resistant strain of S.aureus induced the dispersal of this organism in the environment, thus contributing to further spread.[8] Also extensive studies done by Lid well *etal* reported that the staff nasal carriage of tetracycline-resistant S.aureus was significantly a greater source of staphylococci for new nasal acquisition by patients and the contribution of staff nasal organism than other antibiotic resistant-strains.[19],[25],[26]In general, African S.aureus isolates are characterized by a high proportion of resistance to penicillin, tetracycline, and co-trimoxazole, which mirrors the frequent prescription of these drugs.[27]

This study has several limitations. Firstly, we used only specimens from the anterior nares to screen for MRSA colonization despite S. aureus can be cultured from multiple sites of the skin and other parts of the body, such as axilla and pharynx. Specimens other than nasal swabs have been omitted from screening strategies due to limited time, facilities, financial support and, being easily sampled site. In addition, elegant studies have reported that the nares are the most consistent site from which this organism can be isolated.[3],[8],[9],[19]Secondly, due to the limitation of our laboratory facilities, identification of MRSA is based on the resistance to Oxicillin disc (1 μg) and not on the detection of *mecA*gene. However, a study from Shendi city in Sudan, showed absence of *mecA* gene in MRSA isolates.[28] MRSA without *mecA* gene is also being implicated in the cause of some severe infections in communities and hospitals.[9]Thirdly, information about estimation of variables related to demographics, past or current medical records such as exposure to antimicrobial agents, and lifestyle for the study groups was not available for analysis.Lastly, we selected limited antibiotic discs for susceptibility testing due to financial constraints.

CONCLUSION

Our data showed that those medical care workers were colonized with more antibiotics-resistant *S.aureus* isolates than community members.
The within hand study represents a worthy source of information for researchers about the prevalence of nasal carriage of MRSA and localantibiotic resistance pattern among healthcare workers and healthy community individuals in Khartoum state, Sudan.

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