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RESEARCH PAPER

THE PREVALENCE AND RESISTIVITY PATTERN OF *STAPHYLOCOCCUS AUREUS* ISOLATES FROM APPARENTLY HEALTHY UNIVERSITY STUDENTS IN EKPOMA, EDO, NIGERIA

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ABSTRACT

Based on reported cases of increased multi-antibiotic resistance of *Staphylococcus aureus*, this study investigates the prevalence and resistivity pattern of *Staphylococcus aureus* isolates from ear and nasal swabs of apparently healthy students. A total of 100 samples comprising 50 nasal and 50 ear swabs, were collected randomly from students of Ambrose Alli University, Ekpoma, Edo State, Nigeria. The samples were inoculated on Mannitol salt agar and then incubated for a period of 18-24 hours following standard laboratory procedures. The results showed that the isolates obtained from cultures of nose and ear swabs included *Staphylococcus aureus* (66.70%; 44.40%), *Staphylococcus epidermidis* (55.6%; 33.3%), and *Staphylococcus saprophyticus* (60%; 40%) respectively. However, the level of resistivity to Methicillin by *Staphylococcus aureus* amongst the sampled population was observed to be 44.4% for ear swabs and 66.7% for nose swabs. Interestingly, microbial resistance was higher for Ampicillin than Methicillin, while Tetracycline, among other antibiotics, was the most effective to both ear and nose isolates. Thus, the treatment for *Staphylococcus aureus* with Methicillin and other related antibiotics should be limited or controlled by susceptibility test results. It is also recommended that complete doses of appropriate antibiotics should always be taken to avoid the emergence of resistant strains.

Keywords: Methicillin, Staphylococcus aureus, Antibiotics, Antibiotic resistance,

INTRODUCTION

Staphylococcus aureus -the most important pathogenic organisms in the genus *Staphylococcus*, is a Gram-positive, catalase positive, coagulase positive, non-motile coccus bacterium that causes a variety of human infections in all age groups (Boyce, 1981). It has overcome most of the therapeutic agents that have been developed in recent years and hence, antimicrobial chemotherapy for this species has always been empirical (Jun *et al*., 2004). Its mechanism of resistance to betam lactam and the fluoroquinolones has been documented (Kloos, 1998).

It has also been reported that *S. aureus* strains have a wide variety of multi-drug resistant genes on plasmids, which can be exchanged and spread among different species of Staphylococci and can be transferred to new bacterial hosts (Neihart et al., 1988). This phenomenon is more worrisome in developing countries such as Nigeria, where antimicrobial drugs are readily available to consumers across the counter with or without prescription from a medical practitioner (Nnochiri, 1973; Adekeye, 1979). The most notable example is the emergence of Mehticillin resistant *Staphylococcus aureus* (MRSA), which was reported just one year after the launch of Methicillin (Qureshi *et al*., 2004).

MRSA is a major nosocomial pathogen causing significant morbidity and mortality (Sachdev *et al*., 2003). This pathogen causes various types of diseases and infections ranging from minor skin to soft tissues infections including immune-compromised patients due to its ability to survive in different growth conditions (Lowy, 1998; Lowy, 2003). It has also been reported that the predominant mode of patient-to-patient transmission in hospitals are infected or colonized patients and transient hand carriage on the hands of health care workers (McDonald, 1997). On the other hand, prolonged hospital stay, indiscriminate use of antibiotics, lack of awareness, and receipt of antibiotics before coming to the hospital have also been adjudged as possible predisposing factors of MRSA emergence (Anupurba *et al*., 2003). In addition, with increasing migration throughout the world, transmission of multi-resistant super bugs from one country to another has became a possibility (Ang *et al*., 2004)

Despite the global incidence of MRSA coupled with the increasing multi-antibiotic resistance, it is worrisome to note that the spread of *Staphylococcus aureus* in developing countries is still been underestimated. This study o therefore, is designed to establish the prevalence and resistivity pattern of *staphylococcus aureus* isolates from ear and nasal swabs of apparently healthy students of Ambrose Alli University, Ekpoma, Nigeria.

MATERIALS AND METHODS

Study area and population: This study was carried out in Ekpoma, located in Esan West Local Government Area of Edo State, Nigeria.

Subjects: A total of 100 apparently healthy students of Ambrose Alli University students (50 males; 50 females) served as the study population.

Study Duration: This study was carried out within a period of two months.

Sample collection: The materials used are swab sticks, slides, petri dishes, wire loops, hydrogen peroxide, normal saline, human plasma, crystal violet, lugol's iodine, acetone, neutral red, Bunsen burner, sensitivity discs (Methicillin, Tetracycline, Ciprofloxacin and Ampicillin). Nasal and ear swabs were collected from 50 males (25 ear and 25 nasal swabs) and 50 females (25 ear and 25 nasal swabs) and examined. This was done by inserting a sterile swab stick each inside each nostrils and ear of each subject. After this, the swab sticks were taken to the laboratory for culturing in Nutrient agar and then Manitol salt agar for specificity.

Sample Analysis: Isolation and characterization of bacteria: The swab specimens were inoculated on nutrient agar and subsequently sub-cultured in mannitol salt agar (Difco) as to obtain discrete colonies. The plates were incubated at 37^{o}C for 24 hours under aerobic conditions. After 24 hours of incubation, the culture plates were examined recording the appearance, size, colour, and morphology of the colonies. The bacterial isolates were identified using standard bacteriogical procedures, including Gram stain, microscopic examination and biochemical tests as described by Cheesbrough (2004). Isolates that were gram-positive cocci, catalase positive and coagulated human plasma were considered *Staphylococcus. aureus* in this study.

Antibiotics sensitivity test: Commercially prepared antibiotics discs such as methicillin, tetracycline, ciprofloxacin and ampicillin were used to test the susceptibility of *staphylococcus* isolates obtained. The test isolate was streaked aseptically using a sterile wire loop and spread on a nutrient agar plate uniformly. The antibiotic discs were aseptically placed on the plate; this was incubated at 37^{o}C for 24 hours and examined for zones of inhibition around each antibiotic. The zones of inhibition were measured in millimetres and recorded. Antibiotic zone with less than 10mm in diameter were recorded as being resistant (R) by the organism while those with diameter of 10mm and above were recorded as sensitive (S).

Data analysis: Chi- square statistic was used to determine the differences in the level of resistance by *staphylococcus aureus* to methicillin.

RESULTS

The result showed that of the 50 nasal swabs examined, 26 samples yielded positive for *staphylococcus* species and also, of 50 samples of ear swabs examined, 13 yielded positive for *staphylococcus* species as seen in table 1.

The number of *staphylococcus aureus* isolated from nose was 26 (52%) and from ear was 13 (26%) - table 1. Among the *staphylococcus* species isolated, the prevalence of *Staphylococcus aureus* was higher.

Exposure of *Staphylococcus aureus* to standard concentrations of different antibiotics by agar diffusion method on nutrient agar plates, showed that 6(46.2%) from the ear swabs and 10 (38.5%) from nasal swabs were found resistant to Methicillin (see table 2).

Table 1: Isolates obtained from cultures of ear and nose swabs of Ambrose Alli University students.

Isolates	**Ear**	**Nose**
Staphylococcus aureus	13(44.4%)	26(66.7%)
Staphylococcus epidermidis	4(33.3%)	5(55.6%)
Staphylococcus saprophyticus	2(40%)	3(60%)
Total	**19(35.8%)**	**34(64.2%)**

Table 2: Antibiotic resistance pattern of *Staphylococcus aureus* to methicillin

Samples	Number of *staphylococcus aureus* isolates	Met (%)	Tet (%)	Pen (%)	Ap (%)	Cip (%)
Nose	**26**	**10(38.5%)**	**3(11.5%)**	**1(7.7%)**	**12(92.3%)**	**1(7.7%)**
Ear	**13**	**6(46.2%)**	**1(7.7%)**	**9(34.6%)**	**25(96.2%)**	**4(15.4%)**
Total	**39**	**16(41.0%)**	**3(7.7%)**	**10(25.6%)**	**37(94.9%)**	**5(12.8%)**
X^2		**12.154**	**51.231**			
P.value		**<0.05**	**<0.05**			

Key: Met = Methicillin; Tet =Tetracycline; Pen=Penicillin; Ap=Ampicillin; Cip=Ciprofloxacin

DISCUSSION

Methicillin-resistant *Staphylococcus aureus* (MRSA) has been proven to be one of the most worldwide spread nosocomial pathogen of the 20th century (Nimmo *et al.*, 2000). The observed prevalence of *Staphylococcus* in this study, as compared to *Staphylococcus epidermidis* and *Staphylococcus saprophyticus*, is in line with the findings from a study by Doig, (1981). It is also in agreement with the reports by Nester *et al*. (2001) that 20 % of healthy adults have continually positive nasal cultures for a year or more, while over 60 % will be colonized at some time during a given year. Also, the colonization rate may range from 10 % or more than 40 % in a normal adult population (Kloos, 1998).

Furthermore, the percentage resistant to methicillin as observed in this study, is of great concern that has been widely reported worldwide (Fridkin, 2001; Hiramatsu *et al.*, 1997), and in Nigeria communities (Ikeh, 2003; Onanuga *et al.*, 2005; Olayinka *et al*., 2005). Judging by the number of *staphylococcus aureus* isolated from nose and ear, one can conclude that the nasal carrier rate in this study was higher than what earlier workers reported in normal populations (Osuide *et al*., 1996). This may be attributed to the function of the nasal cavity as the route for air passage, making it more prone to dust carrying *Staphylococcus aureus*. Additionally, some individuals touch their nose more frequently, thereby transferring bacteria from their hands to nostril. On the other hand, it can be said that fewer *Staphylococcus aureus* colonized the ear probably because individuals also touch their ear less often than their nose (Chigbu and Ezeronye, 2003).

In addition, the observation that resistance to Ampicillin was greater, supports the findings by Gross- Schulman *et al.*, 1998) that MRSA strains are equally resistant to all beta-lactam antibiotics. The high level of resistance observed for Ampicillin is also in agreement with the reports by Onanuga *et al.*, (2005) who observed a resistivity of 91.7 % and 100 % to Ampicillin respectively in their two different studies. This can be attributed to antibiotic abuse in the developing countries such as Nigeria, where strict regulatory policies are not adequately implemented and as such, have rendered commonly used antibiotics completely ineffective in the treatment of *Staphylococcus aureus* infections (Odugbemi, 1981).

Although resistance to ciprofloxacin has been reported to be high (Qureshi et al., 2004), the observation on the resistant pattern of *Staphylococcus aureus* to ciprofloxacin agrees with the findings from a study by Chigbu and

Ezeronye (2003) in which Cloxacillin, ciprofloxacin, and lincomycin, recorded high sensitivity. This may be because they are relatively new. Similarly, the observation that there was a low resistance of *Staphylococcus aureus* to penicillin, contradicts the findings from a study by Rajaduraipandi et al. (2006) whereby all the carrier strains were resistant to penicillin and most of them were resistant to ampicillin.

In conclusion, based on the fact that the resistance or sensitivity of MRSA towards commonly used antibiotics is recognized to be diverse from region to region (Rajaduraipandi et al 2006), our findings suggest therefore, that there is a high prevalence of multi-drug resistance MRSA among apparently healthy students without any healthcare risk factor, and that there is the need for continuous surveillance of antibiotic sensitivity pattern of *Staphylococcus aureus* with a view to achieving effective therapy.

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AUTHOR(S) CONTRIBUTION

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