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Prevalence and Antibiogram of Penicillinase-Producing Staphylococcus aureus among Nigerian Students

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Abstract: This study was carried out to determine the presence of penicillinase producing strains of *Staphylococcus aureus* in the nose and ear of apparently healthy students of Bells University of Technology, Ota. 160 nasal and ear swab samples were randomly collected and screened for the presence of penicillinase producing strains of *S. aureus* using standard microbiological procedures. Of the 160 samples collected, 62 (38.8%) yielded growth of *S. aureus*. Male students harboured majority of the bacteria (26.9%) while 11.9% of *S. aureus* were found among samples collected from female students. 28 (17.5%) of the sixty-two *S. aureus* isolates were penicillinase positive, while 34 (21.3%) were non-penicillinase producing strains. Antibiotic sensitivity test of species of *S. aureus* showed highest sensitivity to gentamicin and chloramphenicol and moderate sensitivity to augmentin, amoxicillin and tetracycline, but resistant to cotrimoxazole, cloxacillin and erythromycin. The threatening prevalence of penicillinase positive strains of *S. aureus* among students of Bells University of Technology underscores the strong need for establishing efficient and effective antibiotic policy in the school community to reduce the level of drug abuse.

Key words: Nasal · Ear · Non-Penicillinase · Antibiotic · Harboured

INTRODUCTION

Staphylococcus aureus has long been recognised as a major pathogen causing a wide range of conditions from mild skin infections to severe bacteraemia, which may lead to further complications such as endocarditis, metastatic infections and septicaemia [1]. About 20-60% of humans carry relatively large populations of this organism in their noses; many of these people are also skin carriers [2].

Humans are a natural reservoir for *S. aureus* and asymptomatic colonization is far more common than infection. Colonization of the nasopharynx, perineum, or skin, particularly if the cutaneous barrier has been disrupted or damaged, may occur shortly after birth and may re-occur anytime thereafter [3].

Carriage of *S. aureus* in the nose appears to play a key role in the epidemiology and pathogenesis of infection [4]. *S. aureus* nasal carriage has been extensively studied in patients and healthy individuals [4, 5]. There appear to be three main classes of carriers: the persistent carriers, the persistent non-carriers and the intermittent carriers [5]. The persistent carriers are those

who keep a particular type of *S. aureus* for a long time, intermittent carriers sporadically harbour *S. aureus* occasionally, while the non-carriers are those who do not harbour staphylococcal diseases for a certain period [6, 7].

Methicillin resistance *Staphylococcus aureus* (MRSA) infections in the absence of identified risk factors have been reported infrequently [8, 9]. Since methicillin-resistant *S. aureus* (MRSA) was first reported, it has become endemic in hospitals and communities around the world [10]. The recent emergence of a highly virulent community-associated MRSA (CA-MRSA) and vancomycin-resistant, intermediate-resistant, or hetero-resistant *S. aureus* further heightens public health concerns [11-14]. Prevention of *S. aureus* infection and reduction of the spread of virulent and resistant strains are therefore of great importance.

In the early 1950s, penicillinase-producing strains were universally present in hospital while communityassociated isolates of *S. aureus* were considered to be largely penicillin susceptible. However, over the past few years, community-associated *S. aureus* infections are not only resistant to penicillin but to all other β -lactam

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antibiotics [15]. More so, it is known that epidemic strains of *S. aureus* are commonly resistant to many antimicrobial drugs thereby making the choice of appropriate therapy difficult [15]. The selection of an antibiotic panel for susceptibility testing is based on the commonly observed susceptibility patterns and is revised periodically [16].

This study was carried out to determine the prevalence of penicillinase-producing and antimicrobial susceptibility pattern of community associated *S. aureus* strains isolated from nasal and ear samples amongst apparently healthy students of Bells University of Technology (Bellstech), Ota, Nigeria.

MATERIALS AND METHODS

Study Population: In total, 80 volunteer apparently healthy undergraduate students made up of 40 males and 40 females were randomly sampled in the Bells University of Technology, Ota, Nigeria. These students reported that, they had not taken antibiotic or nasal spray for at least three weeks before the collection of the samples.

Collection of Samples: Nasal and ear samples were randomly collected from 80 apparently healthy students using sterile swab stick moistened with physiological saline. A total of 160 samples were collected (40 nasal and 40 ear samples from male students and 40 nasal and 40 ear samples from females).

Isolation and Identification of Bacteria: The collected nasal and ear swab samples were inoculated aseptically into nutrient broth and incubated at 37°C for 18-24 hours. It was then sub cultured onto mannitol salt agar plates and incubated at 37°C for 18-24 hours. The morphology and cultural characteristics of the *S. aureus* strains were studied. Biochemical tests were carried out based on the conventional microbiological methods according to Cowan and Steel [17].

Beta-Lactamase (Penicillinase) Production Test: Modified iodometry method of Odugbemi *et al.* [18] was used. Cut strips of starch papers were soaked for 10 minutes in a solution of benzyl penicillin or penicillin G (100 IU/ml) and then spread smoothly in a Petri-dish. Each strip was used to test 2 isolates. Test organisms were transferred to surface of the paper using fine wire loop (2mm diameter) at about 2 cm apart. The plates were incubated at 37°C for 30 minutes after which the incubated paper was flooded with $\frac{1}{2}$ diluted Lugol's iodine and drained off immediately. Penicillinase production was manifested by clearing around the organisms on starch paper, whereas the non penicillinase producer remained blue black.

Antimicrobial Sensitivity Test: The disc diffusion method of CLSI [19] was used. Turbidity of the inoculums of the *Staphylococcus aureus* strains was compared with 0.5 McFarland standards and each of the samples was then swabbed onto the surface of sterile nutrient agar plates. The antimicrobial discs which consisted of gentamicin (10µg), cloxacillin (5µg), augmentin (30µg) amoxycillin (25µg), erythromycin (5µg), tetracycline (10µg), cotrimoxazole (25µg) and chloramphenicol (30µg) were placed on the inoculated surface. After 30 minutes, the plates were inverted and incubated for 18-24 hours at 37°C. Then the zone of inhibition was measured in (mm) and the interpretation chart was used to determine the sensitivity patterns of the *S. aureus* strains.

Analysis of Data: Analysis of variance using student's t-test was used to determine the prevalence of penicillinase-producing *S. aureus* among volunteer male and female students.

RESULTS

The frequencies of *S. aureus* isolates were equal in both nose and ear samples (representing 50% in each of the samples). Sixty two (38.8%) isolates yielded *S. aureus*: 43 (26.9%) from the male students and 19 (11.9%) from the female students, while 37 (23.1%) and 61 (38.1) of other *Staphylococcus* species occurred in male and female respectively. The occurrence of penicillinase positive *S. aureus* strains was 17.5% of the total population, while penicillinase negative *S. aureus* strains was 21.3% of the total population and the remaining 61.3% were other species of *Staphylococcus*. Of the 62 isolates that yielded *S. aureus*, the frequencies of penicillinase positive *S. aureus* in male and female students were 21 (33.9%) and 7 (11.3%) respectively.

Antibiotic sensitivity test of *Staphylococcus aureus* isolates revealed that over 90% of the strains were sensitive to gentamicin indicating that gentamicin was the most effective antibiotics tested against these isolates, while less than 36% of the *S. aureus* isolates were sensitive to cotrimoxazole indicating that cotrimoxazole was the most resisted antibiotic tested (Figure 1). Figure 2 depicts the antibiotic sensitivity pattern of penicillinase producing *S. aureus* strains.



Fig. 1: Antibiotic Sensitivity Pattern of *Staphylococcus aureus* Strains.

AUG = Augmentin, AMX= Amoxycillin, ERY= Erythromycin, TET= Tetracycline CXC= Cloxacillin, GEN= Gentamicin, COT= Cotrimoxazole, CHL= Chloramphenicol



Fig. 2: Antibiotic Sensitivity Pattern of Penicillinase Positive *Staphylococcus aureus* Strains. AUG = Augmentin, AMX= Amoxycillin, ERY= Erythromycin, TET= Tetracycline CXC= Cloxacillin, GEN= Gentamicin, COT= Cotrimoxazole, CHL= Chloramphenicol

DISCUSSION

From the results obtained, it was found that the carrier rate of *Staphylococcus aureus* in the study community is in agreement with reports by some earlier authors, who indicated that approximately 10-75% of a healthy population harbour *Staphylococcus* while a carrier rate as high as 40-70% has been estimated for pathogenic *Staphylococcus* particularly among hospital personnel [20].

In this study, it was found that 38.8% of the studied population harbour S. aureus. The reported carrier rate of coagulase positive organisms varies pathogenic considerably from location to location and from one community to another [21]. The differences within community may depend on the level of hygiene and general sanitation as well as the knowledge of the epidemiology of infectious organism involved. The carrier rate was higher in the male than the female which could probably result from hygienic level of the gender involved. The males are more involved in activities which bring them together and this facilitates spread either by direct or indirect contact with each other. In addition, they sweat a lot from their engagement in sporting activities which enhances Staphylococcus aureus proliferation because it has an affinity for sodium chloride and metabolizes speedily in the presence of sodium chloride as described by Cheesbrough [22].

The occurrence of *S. aureus* was the same in both nasal and ear cavity of carriers; which was reported to be directly related to contamination of the environment. It can be observed that *S. aureus* in the air, on objects and surface areas increases when carriers speak, sneeze or cough. It is not unusual to regard *S. aureus* as a normal flora in some parts of the body such as the upper respiratory tract and the skin [4, 23, 24].

The penicillinase producing ability of *Staphylococcus aureus* isolates from apparently healthy students of Bells University of Technology in this study was 17.5%. The possession of the enzyme is of great clinical significance in the treatment of staphylococcal infection with beta-lactam drugs. Penicillinases are capable of inactivating penicillin G, penicillin V, ampicillin and some other penicillins [23].

It can be concluded that penicillinase production among *S. aureus* isolates obtained from healthy individuals in this study is of serious therapeutic implication as these resistance strains gradually replace the virulent or less virulent strains which cannot synthesise penicillinase (beta-lactamase). Besides this, students who are carriers can even spread these resistance strains among themselves by day to day direct and indirect contact.

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