African Journal of Microbiology Research Vol. 5(6) pp. 671-674, 18 March, 2011
Available online http://www.academicjournals.org/ajmr
DOI: 10.5897/AJMR10.644
ISSN 1996-0808 ©2011 Academic Journals

Full Length Research Paper

Antibiotic drug resistance of hospital acquired *Staphylococcus aureus* in Andra Pradesh: A monitoring study

Rabbani Syed[1,2]*, Giri Prasad[1], Farha Deeba[1,2], Rani D.[1] , Kaiser Jamil[1,2] and Ali A. Alshatwi [3]

[1]Department of Genetics, Bhagawan Manahir Hospital and Research Centre, Hyderabad, India.
[2]Central Research Institute for Unani Medicine, Hyderabad, India.
[3]Molecular Cancer Biology Research Lab(MCBRL) Dept. of Food Science and Nutrition. College of Food and Agriculture Sciences King Saud University.

Accepted 1 March, 2011

Nosocomial infections are one of the occupational biohazards that affect the health of individuals with or without predisposing factors. *Staphylococcus aureus* is associated with significantly higher mortality and is associated with community-acquired serious nosocomial infections because strains generally show multiple drug resistance, which limits treatment possibilities. A total of 1800 patients in the state of Andhra Pradesh were screened for the presence of *Staphylococcus* species and were tested for antibiotic resistance. The results indicated that among ten antibiotics used in the present study, Amikacin and Azithromycin should be the drug of choice to treat *S. aureus* infection. It was observed that the resistance of most of the antibiotics tested showed increased resistance with increasing age. These results suggest that clinicians should consider age as an important factor while prescribing these antibiotics.

Key words: Nosocomial, antibiotic resistance, *Staphylococcus aureus.*

INTRODUCTION

Nosocomial infections are one of the occupational biohazards that affect the health of individuals with or without predisposing factors. Sanitizing surfaces is often an overlooked critical component of breaking the cycle of infection in health care environments.

Nosocomial infections are the infections acquired during hospital stay. These infections are found in 5 to 15% (two million cases are estimated annually) of hospitalized patients and can lead to complication in 25 to 33% of those admitted in ICU (Parvez, 2011).

Nosocomial infections, as they are referred to, are not uncommon even in most advanced countries. In fact, the center for disease control (CDC) estimates that 4.5% of all hospital admissions in the U.S. are due to infections acquired from hospital stays.

Studies conducted in hospitals in Delhi and Mumbai report figures as high as 30% (The Hindu, 2007). Reports on antibiotic resistance have grown to be increasingly common and pathogens that are resistant to almost all antibiotics have been found. It has become painfully obvious that antibiotic resistance is reaching a crisis stage and some clinicians have even forecasted a return of the devastating diseases of the pre-antibiotic era. Resistant strains of *Staphylococcus aureus* are devastating human pathogens, sometimes referred to as "modern Ghengis Khan". Resistance to methicillin is often accompanied by resistance to many other antibiotics (Locksely et al., 1982). According to the data published by the US Centers for Disease Control and Prevention (CDC) as part of their national nosocomial infection surveillance (NNIS) System, well over half of all intensive care unit isolates from documented infections are caused by methicillin-resistant *S. aureus* (MRSA) (NINS, 2003). The level of antibiotic resistance in a given community or hospital can be predicted by these important measures, such as: the proportion of resistant organisms introduced from outside the population, the extensive use of anti microbial agents and the proportion that spreads from

*Corresponding author. E-mail: rabbanisyd@gmail.com.

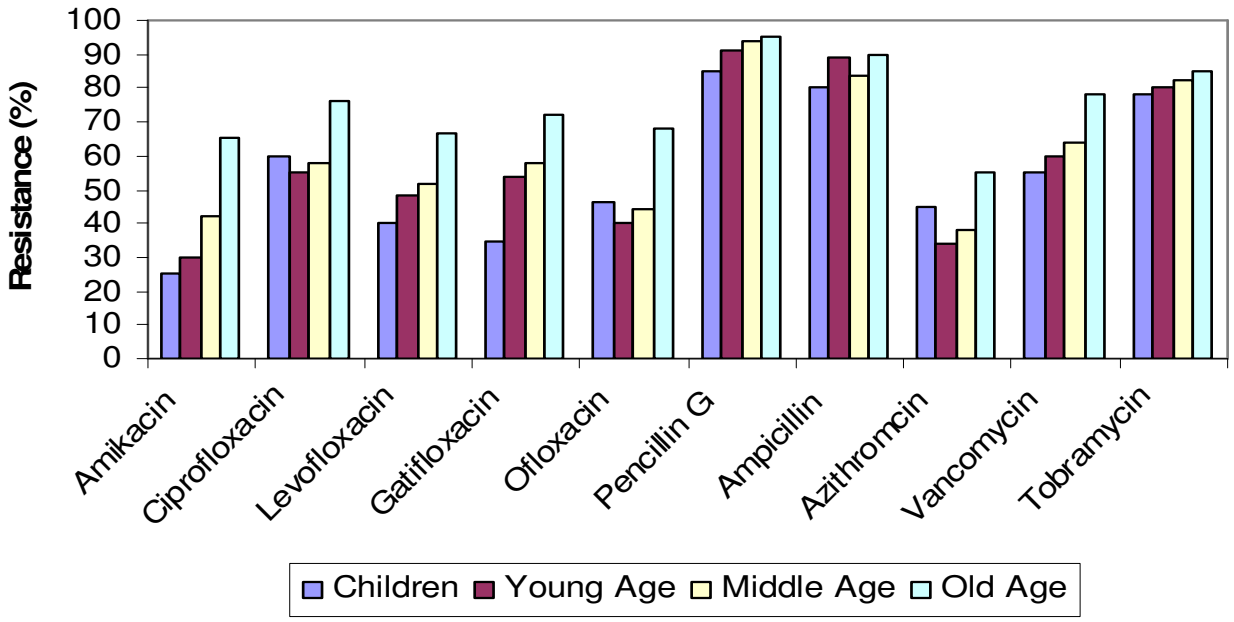

Figure 1. Details of results for antibiotic resistance pattern among different age groups in the study population.

one person to another (Wenzel and Edmond, 2000; Edmond, 2000). The type, as well as severity, of *S. aureus* infections and its response to antibiotic treatment are dictated by the specific suite of virulence and antibiotic resistance associated genes carried by the strain of the *S. aureus* causing the infection (Peacock et al., 2002).

Hence, there is need to screen individuals exposed to such infection. In the present study, we assessed the multiple drug resistance profile of *S. aureus* isolated from the pus sample of hospitalized patients in Andhra Pradesh.

METHODOLOGY

Collection of samples

A total of 1800 patients' pus samples, collected from orthopedics and burns units of general and private hospitals in Kavali and Nellore districts of Andhra Pradesh, were studied over a period of two years from January 2007 to January 2009. Informed consent was obtained from all patients as required by the ethics committee used in this study. Pus samples were screened for *S. aureus* after obtaining the informed consent and approval by the institute's ethics committee. Clinical details, including duration of hospital stay, diagnosis, antibiotic intake and presence of other medical illnesses, were recorded for the inpatients. Using pre-moistened sterile cotton swabs, specimens were collected from the anterior nares, palms and web spaces of patients. The samples were divided into four age groups: children (0 to 14), young age (15 to 35), middle age (35 to 50) and old age (>50), while the swab samples collected were screened for *S. aureus*, using the conventional microbiological methods.

Media and culture conditions

The nutrient agar media was used as the general media, while the blood and MacConkeys (Merck) agar were used for isolation and identification of bacterial cultures. The incubations were done at 37°C for 18 h throughout the investigation.

Screening and identification of *S. aureus*

Primary screening

Primary screening of *S. aureus* was conducted by observing the following characters:

A. Based on colony morphology: Circular, golden yellow and white pigmented colonies were selected on a blood agar media, whereas on the nutrient agar plate, yellowish white colour colonies were selected.

B. Based on haemolytic pattern: The ß-haemolytic bacteria were identified on a blood agar and were isolated.

The primarily screened strains were subjected to secondary screening. The isolated bacteria were subjected to gram staining as per the modification made by Hucker. To confirm the strains of *S. aureus*, biochemical tests, like catalase and coagulase tests, were conducted.

Antibiotic susceptible test

The antibiotic susceptibility pattern of all isolated *S. aureus* was tested in 10 antibiotics (Figure 1), determined by the modified Kirby-Bauer disk diffusion technique. The standardized 10^8 cell/cfu over night culture of each isolate was used to flood the surface of Muller Hinton agar plate and the excess of it was drained off and dried, while the Petri dish lid was in place. The standard antibiotic discs

were then aseptically placed at reasonable equidistance on the inoculated Muller Hinton plates and were allowed to stand for 1 h. The plates (prepared in duplicate for each isolate) were then incubated at 37°C for 18 h (Ehinmidu, 2003). The diameter of the zone of inhibition produced by each antibiotic disc was measured and recorded, and the isolates were classified as "resistant" or "sensitive", based on the standard interpretative chart updated according to the current NCCLS standard (Cheesbrough, 2002) and Fluka zone interpretative chart in accordance with WHO requirements.

RESULTS

By the analysis of conventional microbiological methods, the isolated strains were confirmed by the presence of round, violet colored cells arranged in clusters as gram positive cocci. The isolated strain was found to be catalase and coagulase positive.

Based on the aforementioned results, the isolated stains were confirmed as *S. aureus*. The drug resistance patterns of *S. aureus* isolates from clinical samples were found to be highly variable. Almost all the strains were resistant to Penicillin-G, Ampicillins and Tobramycin. However, all strains recorded sensitivity to Amikacin and Azithromycin, followed by Ofloxacin, Levofloxacin and Gatifloxacin.

S. aureus isolates were assessed in the children's group. The results of the antibiotic susceptibility in children (Age 0 to 14) showed that *S. aureus* isolates were highly resistant to Penicillin-G (85%), Ampicillin (80%), Tobramycin (78%) and Ciprofloxacin (60%), but showed or presented medium resistance to Vancomycin (55%), Ofloxacin (46%), Azithromycin (45%) and Levofloxacin (40%). Moreover, the least resistance was recorded with Amikacin (25%) (Figure 1). The trend is as follows:

Penicillin (85%) > Ampicillin (80%) >Tobramycin (78%) > Ciprofloxacin (60%) > Vancomycin (55%) > Ofloxacin (46%) > Azithromycin (45%) > Levofloxacin (40%) > Amikacin (25%).

In the case of the young age group (15 to 35) (Figure 1), *S. aureus* isolates showed the resistance trend as follows:

Penicillin (91%) > Ampicillin (89%) > Tobramycin (80%) > Vancomycin (60%) > Ciprofloxacin (55%) > Gatifloxacin (54%) > Levofloxacin (48%) > Ofloxacin (40%) > Azithromycin (34%) > Amikacin (30%).

The resistance pattern of *S. aureus* isolates for the middle age group (35 to 50) reveals the least resistance antibiotic as Amikacin (42%) and the highest resistance antibiotic as Penicillin G (94%). In this age group, Azithromycin (38%) actively inhibited *S. aureus* growth as well.

DISCUSSION

The widespread use of antibiotics has been responsible for the development of numerous problems, including the emergence of multi drug resistance bacteria, increased number of nosocomial (hospital) and community acquired infections and increased health care costs (Snyder et al., 2000). Rising to the challenge posed by hospital acquired infections, which are emerging as a global health concern over 1.4 million people worldwide are suffering from hospital acquired infections (HAIs) or nosocomial infections. In this study, in almost all age groups, *S. aureus* isolates were resistant to Penicillin-G. The indiscriminate use of antibiotics may be a cause for this multidrug resistance. Among the ten drugs used in the present study, Amikacin is the best choice for the treatment of *S. aureus* infection in all age groups. *S. aureus* is capable of causing a variety of human infections, including fatal invasive and toxic conditions and also possesses a differential ability to spread and cause hospital associated outbreaks of infections (Baird, 1996; Aucken, 2002).

Reports from the International Infection Control Consortium (INICC) surveillance study show that nosocomial infection is markedly higher in the ICUs of the INICC hospitals (Rosenthal et al., 2010). The emergence of methicillin resistant *S. aureus* (MRSA) strains, with multidrug resistance, has posed a challenge in the treatment of infection (Van Belkum and Vertburgh, 2001). In India, nosocomial infection rate is over 25% and is responsible for more mortality than any other form of accidental death. The prospective observational study describes that isolates of *Acinetobacter*, *Pseudomonas*, *Klebsiella* and *Escherichia coli* are resistant to the third generation cephalosporins, and it also states that the increased duration of the time spent in intensive care units and days of intervention are associated with the incident (Shabina et al., 2008). *S. aureus* is a pathogen of greater concern because of its virulence (Chambers, 2005), its ability to cause a diverse array of life threatening infections, and its capacity to adapt to different environment conditions (Lowy, 1998, 2003). In this study, the highest level of resistance is observed in Penicillin-G and Ampicillin in all age groups, which is in agreement with the reports given by Umolu et al. (2002) and Ehinmidu (2003).

So, in the present investigation, the variation that occurs in the antibiotic sensitivity pattern of *S. aureus* confirms the emergence of antibiotic resistance. The resistance in bacterial pathogens to antibiotics increases the chance of severe infections in human beings. However, the data indicate that among ten antibiotics used in the present study, Amikacin and Azithromycin should be the drug of choice to treat *S. aureus* infection. For proper treatment, the physician should perform the antibiotic sensitivity test before antibiotic treatment. It is also observed that resistance of most of the antibiotics

tested shows increased resistance with increasing age. These results suggest that clinicians should consider age as an important factor while prescribing antibiotics.

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