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Prevalence and Resistance pattern of *Staphylococcus aureus* isolated from a Tertiary Care Centre in Pudhucherry

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Abstract

Objectives: To study the prevalence and resistance pattern of *Staphylococcus aureus* isolated from abscesses from a tertiary care centre.

Materials and Method: A retrospective study was done at Department of Microbiology, Aarupadai Veedu Medical College and Hospital on 465 isolates from pus samples of which 218 isolates were identified as *Staphylococcus aureus* upon standard techniques. The samples were obtained from patients with abscess from various sites like foot, legs, hands, surgical sites, skin ulcers, injection wounds of both inpatients and outpatients of all age groups and both gender.

Results: Overall rate of culture positive abscesses were 35.6% (n=465/1306). *Staphylococcus aureus* is the predominant pathogen isolated contributing to 46.88% (n=218/465) isolates followed by *Pseudomonas aeruginosa* 15.4% (n=72/465), *Escherichia coli* 11.1% (n=52/465), *Klebsiella spp* 6.5% (n=30/465), *Proteus spp* 5.3% (n=25/465), *Enterococcus spp* 4.3% (n=20/465), *Streptococcus spp*2.7% (n=13/465). The highest resistance were observed for Penicillin 98% (n=214/218), followed by Methicillin 79.3% (n=173/218), Clindamycin 73.6% (n=160/218), Streptomycin 72.6% (n=158/218), Amoxyclav 65% (n=142/218), Chloramphenicol 41.8% (n=91/218). Majority of *Staphylococcus aureus* isolates were sensitive to Vancomycin 97% (n=211/218), Gentamicin 95% (n=207/218), Ciprofloxacin 85% (n=185/218), Erythromycin 78.1% (n=170/218), Oxacilin 52% (n=113/218), Ampicilin 61.3% (n=134/218).

Conclusion: *Staphylococcus aureus* is the predominant pathogen isolated in our study. Higher rate of isolation were seen among skin ulcers as they are more exposed to environment. Following them were from diabetes patients, since *Staphylococcus aureus* play vital role in causing varied ulcers because of their lower immunity status. Resistance rates were high for Penicillin, Methicillin, Amoxyclav, Clindamycin and Chloramphenicol because of the abundant use in routine practice. Judicial use of antibiotics, better hygiene and good clinical practices should be followed to prevent the severity of these infections.

Keywords *Staphylococcus aureus*, Prevalence, Antibiotic Resistance

Introduction

Staphylococcus aureus bacteria is a widespread commensal bacteria accounting for 30% of population.(Alsterholm et al.,2017) People with diabetes, cancer, tissue necrotizing pneumonia, sepsis, eczema, vascular diseases and lung diseases are at higher risk of infection by *Staphylococcus aureus*.

Staphylococcus aureus also becomes resistant to many antibiotics. Most common were the Methicillin-Resistant *Staphylococcus aureus* (MRSA), Vancomycin-Intermediate *Staphylococcus aureus* (VISA) and Vancomycin-Resistant *Staphylococcus aureus* (VRSA). Worldwide, these three resistant types are the major cause of community and hospital acquired infections. Community- associated MRSA has primarily been described as a cause of skin and soft tissue infections, but it has also been associated with sepsis and tissue necrotizing pneumonia. (Bauer et al., 1996; Centre for disease control and prevention, 2001; Chai Lee et al.,2017)

Staphylococcus are the primary cause of skin and soft tissue infections such as abscesses, furuncles and cellulitis. The infection rate is much higher in hospitalized patients (60-70%) than among community people (30-50%).(Diekema et al.,2001) So, the treatment becomes much more challenging since many of the *Staphylococcal* species are multidrug resistant (MDR).

There is a extensive increase of multidrug-resistant healthcare-associated MRSA cases has reduced the number of available therapeutic options for these infections. Potential antibiotics include daptomycin, linezolid, quinupristin/dalfopristin, trimethoprim/sulfamethoxazole and vancomycin. Glycopeptides, in particular vancomycin, are the treatment of choice for serious infections. (Dufour et al.,2001)

In current scenario, though combination of antibiotics for the treatment options exists, resolve to some extent, but resistance against modern chemotherapies pose a challenge to the medical practices. Second-line drugs like glycopeptides are the current choice of drugs for the treatment for VRSA infections. However, they are more expensive and causes severe side effects, hence monitoring during treatment is advisable, which increases the cost even further.(Kluytmans and Belkum, 1997)

Approximately, there are 50% to 60% of individuals who are intermittently or permanently colonized with *Staphylococcus aureus* and hence the

potential for infections is relatively high. This study is to determine the prevalence and resistance rates of *Staphylococcus aureus* obtained from abscess from various sites and also to provide a clear outline about the prevention of infection, efficacy of treatment and hygiene which is the need of the hour.

Materials and Method

A retrospective study were done on 465 isolates obtained from patients with abscess from various sites such as legs, toes, abdomen, foot ulcers, hands, skin, breast, gluteum, clavicle, etc, during a period of one year from January 2016 to December 2016, from a total of 1306 specimens received at Department of Microbiology, Aarupadai Veedu Medical College and Hospital were included in this study.

The samples were collected from both inpatients and outpatients of all age groups and both gender. The study protocol was approved by the Institutional Ethics Committee (IEC) after which the study was initiated. Verbal informed consent was obtained from the patients before including into the study.

Pus was collected from the deep site of the lesion after superficial cleaning of that particular site either using syringe where ever possible or 2 sterile swabs when the pus was minimum. The pus specimens were transported immediately to the laboratory.

Investigations: The specimens were processed as per standard procedure. Gram's stain were performed for all the specimens. They were then inoculated into the Blood Agar and MacConkey agar and incubated at 37º C for 24- 48 hrs. The isolated organism were further identified by standard techniques.

Antimicrobial susceptibility testing for *Staphylococcus aureus*: Antimicrobial susceptibility testing was performed on Mueller-Hinton agar as per CLSI guidelines. The susceptibility of isolates to various antibiotics was done by Kirby-Bauer disc diffusion method using commercial disks.(Kumari et al.,2016) For antibiotic susceptibility technique, Mueller-Hinton agar incorporated with 4.0% Nacl and antibiotic discs were used and incubated at 37ºC for 24-48 hours. The following antibiotics were tested: Amoxyclav (5µg), Ampicillin (5µg), Chloramphenicol (30µg), Clindamycin (2µg), Erythromycin (10µg), Gentamicin (10µg), Oxacillin (5µg), Penicillin G (10µg), Streptomycin (10µg), Vancomycin (30µg), Methicillin (5µg), Ciprofloxacin (5µg).

Results

Overall rate of culture positive abscesses were 35.6% (n=465/1306). *Staphylococcus* is the predominant pathogen isolated contributing to 46.88% (n=218/465) isolates followed by *Pseudomonas aeruginosa* 15.4% (n=72/465), *Escherichia coli* 11.1% (n=52/465), *Klebsiella spp* 6.5% (n=30/465), Proteus spp 5.3% (n=25/465), Enterococcus spp 4.3% (n=20/465), *Streptococcus spp* 2.7% (n=13/465). This is illustrated in Fig. 1.

The rate of infection is greater in abscesses from skin ulcer followed by diabetic foot ulcers, injection abscesses, surgical infection, wound infection. A total of 218 isolates were *Staphylococcus aureus* isolates out of 465 samples. These 218 isolates were subjected to antibiotic susceptibility test against 12 antimicrobial drugs.

Majority of *Staphylococcus aureus* isolates were sensitive to Vancomycin 97% (n=211/218), Gentamicin 95% (n=207/218), Ciprofloxacin 85% (n=185/218), Erythromycin 78.1% (n=170/218), Oxacilin 52% (n=113/218), Ampicilin 61.3% (n=134/218).

The highest resistance were observed for Penicillin 98% (n=214/218), followed by Methicillin 79.3% (n=173/218), Clindamycin 73.6% (n=160/218), Streptomycin 72.6% (n=158/218), Amoxyclav 65% (n=142/218), Chloramphenicol 41.8% (n=91/218). The results are represented as antibiogram of *Staphylococcus aureus* to different antibiotics in Fig. 1.

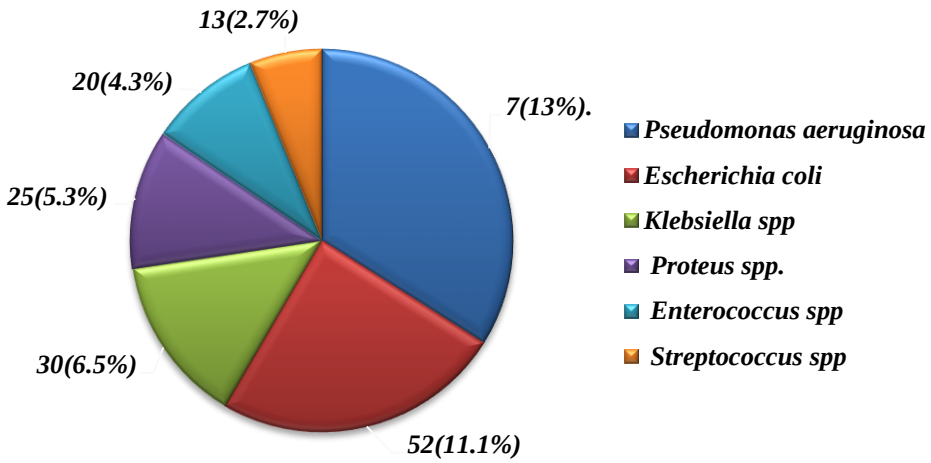

Fig. 1: Positive Culture Isolates from Abscesses

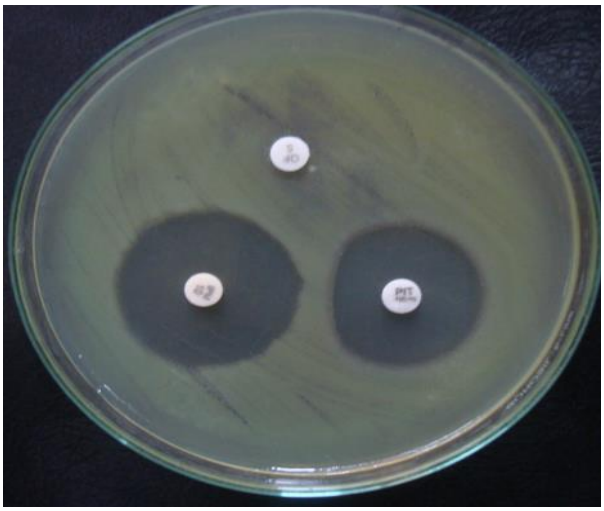

Fig. 2: Antibiotic sensitivity result of a *Staphylococcus aureus* isolate grown in Muller Hinton Agar

Discussion

Among 1306 samples collected from different abscesses for culture and sensitivity, 1104 showed aerobic growth whereas 202 did not show any growth. This may be suspected as anerobic bacteria or slow growing bacteria or due to immune system involvement.(Kumari et al.,2016; Liu et al.,2011; Loomba et al.,2010; Mehta et al.,1998; Mendem et al.,2016). This is similar to a study carried out by LF McCaig and Lc Mc Donald.

This study is done by collecting pus from various sites of abscesses. Abscess is a collection or pocket of infection which is formed at the site of injury. It is usually filled with pus. The area surrounding the abscess is usually swollen, red, painful, and the skin surrounding the abscess will be warmth to touch. Most skin infections resolve without treatment, however, some infections require incision and drainage or some antibiotic treatment to resolve the infection. (Mrsa,2003; Naimi et al.,2003; Said-Salim et al.,2003) Skin infections when untreated may develop into a more serious life threatening infections such as bone infections or blood infections. There is a possibility for longer lasting severe infections with resistant *Staphylococcus aureus* if the initial antibiotic prescribed is not capable of killing the bacteria. (Naimi et al.,2003; Said-salim et al.,2003)

Higher rate of isolation were seen among skin ulcers as they are more exposed to environment. Following them were from diabetes patients, since *Staphylococcus aureus* plays a vital role in causing varied ulcers because of their lower immunity status. Resistance rates were high for Penicillin, Methicillin, Amoxyclav, Clindamycin and Chloramphenicol because of the abundant use in routine practice.

The infection rates were higher among males (65%) than among females (35%). This is similar to a study carried out by Tebelay Dilnessa and Adane Bitew. The infection rates varied according to race, sex and age. These variations are due to low socio-economic status, failure to finish full course of antibiotics, improper hygiene, over use of antibiotics without prescription. Also indiscriminate use of antibiotics in high age groups (>30 years) is responsible for slow development of drug resistant *Staphylococcus aureus*. Also significant variation in infection rates is more in deep seated lesions compared to superficially infected wounds that correlated with the other studies.(Said-salim et al.,2003; Saiman et al., 2003; Shakya et al., 2010; Shrestha et al.,2009)

Also the infection rates were high from August to December from skin and soft tissue infected patients which is similar to findings of study in other places too. (Stacey et al., 2010; Udaya et al.,1997) In these months *Staphylococcus aureus* colonize more rapidly due to favourable conditions and also due to endogenous autoinfection. A study carried out by Mehta AP, Seth K in a tertiary care centre also suggests this. So, the patients as well as health care professionals should be advised to maintain personal hygiene by washing hands with soap and water and treated with antibiotics. The high prevalence rate is due to indiscriminate use of antibiotics, lack of awareness and unethical treatment before coming to hospitals. (Wayne,1994)

Nowadays Vancomycin resistant *Staphylococcus aureus* isolates are emerging which indicates that the infections are spreading day by day and sensitive strains are slowly becoming resistant strains thus posing a threat to treat the infections. A study by Loomba and Juhi Taneja also indicates that Vancomycin resistant *Staphylococcus aureus* is emerging rapidly.

Since *Staphylococcus aureus* remains a major cause of skin infections due to the rise of highly virulent, drug resistant strains, the treatment becomes more difficult. As a result, burden of disease is rising with the hospital and community acquired settings, thus emerging the need for modified therapies. This study indicates regular surveillance of hospitals, proper hygiene control, and monitoring antibiotic sensitivity pattern, its prevalence and formulation of definite antibiotics to reduce the incidence of *Staphylococcus* infections and multidrug resistant *Staphylococcal* infections.

Conclusion

Staphylococcus aureus was the predominant pathogen isolated from various abscesses. These were resistant to many antibiotics and the incidence rate of infection is high in patients with skin ulcers, wound infections, diabetic foot comparatively to other site abscesses. The indiscriminate use of antibiotics for prophylactic as well as other therapeutic purposes may be the reason for increased prevalence of many antibiotic resistant *Staphylococcus aureus.* A relatively high number of strains are also resistant to the

commonly used antibiotics used against many human infections. Judicial use of antibiotics, better hygiene and good clinical practices should be followed to prevent the severity of these infections.

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