Original Research Article

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Bacteriological study of post-operative wound infections with special reference to MRSA and ESBL in a tertiary care hospital

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ABSTRACT

Background: Surgical site infections are known to be one of the most common causes of nosocomial infections worldwide and raises an important public health concern.

Methods: A prospective observational study was conducted across 12 months (July 2018-June 2019) in a tertiary care hospital. The present study includes 50 pus samples from clinically suspected cases of post-operative wound infections from various surgical wards which were inoculated in Department of Microbiology. The phenotypic identification of different bacterial isolates especially MRSA and ESBL producers along with their antimicrobial susceptibility testing was interpreted according to CLSI guidelines.

Results: Out of 50 samples, 44 showed positive culture. The infections were more common in emergency situations, age group of 20-30 years and in females. The most common bacterial isolates were *Staphylococcus aureus* (43.18%) followed by *E. coli* (22.72%), *Klebsiella* (15.9%), *CONS* (13.63%) and *Pseudomonas* (4.54%). Out of 19 *Staphylococcus aureus* 9 were MRSA and these gram-positive bacteria were highly sensitive to Linejolid and Clindamycin. In gram negative group 22.72% of *E. coli* and 15.9% of *Klebsiella* were ESBL producers who were susceptible mainly to Aztreonam, Linezolid or Cefoxitine.

Conclusions: Post-operative wound or surgical site infections is the most important factor responsible for significant morbidity, mortality, prolonged hospitalization and additional cost to treatment in surgical patients. Marked resistance of isolates to commonly used antibiotics indicates the need of judicious use of these drugs to prevent the emergence of multi-drug resistant strains.

Keywords: Antibiotic susceptibility, Extended spectrum beta lactamase, Infection, Methicillin resistant *staphylococcus aureus*, Post-operative wounds, Surgery

INTRODUCTION

Surgical Site Infections (SSIs) are known to be one of the most common cause of nosocomial infections worldwide and accounts for nearly 20-25%. As defined by Centers for Disease Control and Prevention (CDC), infection at or near surgical incision or within 1 year of an operation, if a foreign body is implanted as part of the surgery is defined as SSI. In other words, surgical site infection is

an infection that occurs after surgery in the part of the body where the surgery took place. Surgical site infections can sometimes be superficial infections involving the skin only. Other surgical site infections are more serious and can involve tissues under the skin, organs or implanted material.

Surgical Site Infections contributes to substantial rate of mortality, significant morbidity, considerable

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prolongation in length of hospitalization and added treatment expenses. Despite the technical advancements that have been practiced over the past few years in surgical and wound management system, wound infections are still viewed as the most widely recognized nosocomial infections, particularly in patients experiencing surgery. SSIs were estimated approx. 31% of all Healthcare Associated Infections (HAI) which contributed 20% postsurgical readmissions as well. The situation is more severe in developing countries like India where resources are scarce, and staff is always in short supply.² The most common organisms encountered in surgical site infections are Staphylococcus aureus, negative Staphylococcus Coagulase (CONS). Pseudomonas, E. coli, Klebsiella, Enterobacter, Proteus, Acinetobacter and Enterococcus etc.3

The emergence of high antimicrobial resistance among bacterial pathogens has made the management and treatment of post-operative wound infections difficult. *Staphylococcus aureus* was susceptible to Penicillin but due to misuse of antibiotics, it is resistant to most of the Penicillin group worldwide due to Methicillin resistance. Most Methicillin Resistant *Staphylococcus aureus* (MRSA) infections occur in health care settings and are called Hospital acquired MRSA. Such infections are generally associated with invasive procedures, I.V catheters, surgical wounds, etc.⁴ The emergence of MRSA and Extended Spectrum Beta Lactamase (ESBL) posed serious therapeutic challenge.

The present study was conducted with aims

- To study the organisms and their antibiotic susceptibility in post-operative wound infections.
- To detect the prevalence of MRSA.
- To detect ESBL production in germ negative bacterial isolates.

METHODS

A prospective observational study was conducted from July 2018 to June 2019 in the Department of Microbiology at J.J.M. Medical College, Davanagere. In this study 50 pus samples from clinically suspected cases of post-operative wound infections from various surgical wards were included.

Inclusion criteria

All patients age >18 years admitted to different surgical wards (including General surgery, Orthopedics, Urosurgery, Obstetrics and Gynecology) having exudates from their surgical site were included.

Exclusion criteria

Patients with age <18 years having infected burn wounds, stitch abscess and those with episiotomy wound infection were excluded from the study.

The sample was collected from depth of wound with strict aseptic precautions with the help of dry sterile cotton swab sticks for bacteriological examination. Two culture swabs from each sample were obtained, one for the direct smear study and other for aerobic bacterial culture and sent immediately to the laboratory for investigation.

The culture swab was used to inoculate onto the blood agar and MacConkey agar media and incubate at 37°C for 24 hours. After incubation identification of bacteria from positive culture was done with standard microbiological technique.⁵ Strains of *Staphylococcus aureus* was identified by gram's staining, colony morphology, catalase production test, slide and tube coagulase test and mannitol fermentation test.⁶

Antimicrobial susceptibility testing was performed according to Clinical and Laboratory Standard Institute (CLSI) guidelines, by Kirby-Bauer disc diffusion technique on muller Hinton agar.⁷

Detection of MRSA was done by Cefoxitin disc diffusion method by Cefoxitin disc 30 µg (according to CLSI guidelines) and ESBL detection was done by Combined Disc Diffusion method by using disc Ceftazidime + Ceftazidime-Clavulanic acid and Cefotaxime + Cefotaxime-Clavulanic acid.8

RESULTS

The present study was conducted at J. J. M. Medical College, Davanagere. A total 50 samples collected from clinically suspected post-operative wounds were studied after inoculation. Out of 50 samples studied 44(88%) showed bacterial growth while 6(12%) did not show any growth after inoculation.

Table 1: Age wise distribution in post-operative wound infections.

| Age group (years) | No. of post-operative wound infections |
|-------------------|--|
| 20-30 | 23 |
| 30-40 | 7 |
| 40-50 | 9 |
| 50-60 | 5 |
| > 60 | 6 |

As regards to age distribution, the post-operative wound infection was maximum (52.27%) i.e. 23 out of 44 in age group of 20-30 years followed by 20.45% i.e. 9 out of 44 in middle age group of 40-50 years and was minimum 11.36% i.e. 5 out of 44 in age group of 50-60 years (Table 1).

Further with gender distribution out of 50 samples, 29 samples were collected from females and 21 from males. Out of these 19 out of 21(90.47%) of males and 25 out of 29(86%) of females were culture positive (Table 2).

Bacterial isolates in post-operative wound infections

Out of 44 isolates 56.88% (25 out of 44) were gram positive while 43.2% (19 out of 44) belongs to gram negative group.

Table 2: Sex distribution in post-operative wound infections.

| Sex | Total no. of samples | Culture positive samples |
|--------|----------------------|--------------------------|
| Male | 21 | 19(90.47%) |
| Female | 29 | 25(86%) |

Amongst gram positive cocci *staphylococcus aureus* 19(43.2%) was the most common isolate followed by *E. coli* 10(22.7%), *Klebsiella* 7(15.9%), coagulase negative *staphylococcus* (CONS) 6(13.6%) and the least prevalent was *Pseudomonas* 2(4.5%). (Table 3)

Table 3: Total Number of Isolates.

| Gram positive bacteria | Gram negative bacteria |
|--|------------------------|
| Staphylococcus aureus: 19(43.2%) | E. coli: 10(22.7%) |
| Coagulase negative | Klebsiella: 7(15.9%) |
| staphylococcus aureus (cons): 6(13.6%) | Pseudomonas: 2(4.5%) |
| Total: 25(56.8%) | Total: 19(43.2) |

In the study out of 19 *staphylococcus aureus* 9(47.36%) were MRSA by cefoxitin disc diffusion method and the rest were methicillin sensitive *staphylococcus aureus* (MSSA) (Figure 1).



Figure 1: MSSA and MRSA on muller Hinton's agar by using cefoxitin disc.

Out of total 19-gram negative isolates, 5 were found to be ESBL producers by Ceftazidime and Ceftazidime-Clavulanic Acid and Cefotaxime and Cefotaxime-Clavulanic acid by double disc diffusion method. Amongst them 3 out of 10(22.72%) *E. coli* were found to be ESBL producers and 2 out of 7(15.9%) *Klebsiella* were found to be

ESBL producers while none of the pseudomonas was found to be ESBL producer (Table 4).

Table 4: ESBL Producer Isolates.

| Organisms | Total no of isolates | ESBL producers | Percentage (%) |
|------------|----------------------|----------------|----------------|
| E. coli | 10 | 3 | 22.72% |
| Klebsiella | 7 | 2 | 15.9% |
| Total | 17 | 5 | 38.62% |

The findings of ELBS screen test is very well (Figure 2 and 3).

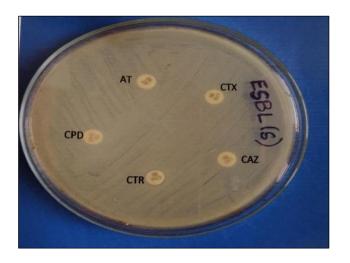


Figure 2: ESBL screening test-positive (disc diffusion method).

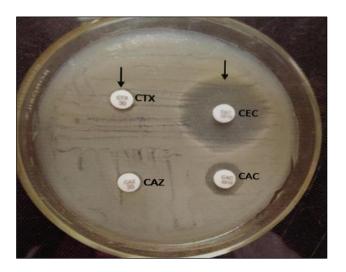


Figure 3: ESBL confirmatory test-positive (combined disc test).

Antibiotic susceptibility testing by Kirby Bauer disc diffusion method

Antibiotic susceptibility pattern of isolated organism found by Kirby Bauer disc diffusion method showed that *staphylococcus aureus* (both coagulase positive and

negative) were susceptible mainly to Linezolid, Doxycycline, Clindamycin while among gram negative bacteria both *E.coli* and *Klebsiella* were susceptible to predominantly Aztreonam, Linezolid or Cefoxitine (Figure 4 and 5).



Figure 4: Pattern of *staphylococcus aureus* susceptibility to antibiotics.

The different antibiotic susceptible pattern of both gram positive and gram-negative isolates (Table 5).



Figure 5: Pattern of gram-negative bacterial susceptibility to antibiotics.

DISCUSSION

Post-operative wound infection still remain one of the most important causes of morbidity and is one of the most common nosocomial infection in surgically treated patients. In present study, an attempt has been made to know the various bacterial flora responsible for surgical site infections and their antibacterial susceptibility pattern. The rate of SSI varies greatly worldwide and from hospital to hospital.

| Antibiotics (total) | Staph. Aureus (19) | E. coli (10) | Klebsiella (7) | Cons (6) | Pseudomonas (2) |
|---------------------|-----------------------|--------------|-------------------|-----------|-----------------|
| Aztreonam | 6(31.57%) | 4(40%) | 3(42.85%) | 1(16.66%) | 0 |
| Cefpodoxime | 9(47.36%) | 2(20%) | 3(42.85%) | 3(50%) | 2(100%) |
| Ceftazidime | 6(31.57%) | 2(20%) | 2(28.57%) | 2(33.33%) | 2(100%) |
| Cefotaxime | 8(42.10%) | 3(30%) | 2(28.57%) | 2(33.33%) | 2(100%) |
| Doxycycline | 9(47.36%) | 3(30%) | 2(28.57%) | 5(83.33%) | 2(100%) |
| Linezolid | 14(73.68%) | 4(40%) | 2(28.57%) | 5(83.33%) | 0 |
| Cefoxitine | 9(47.36%) | 3(30%) | 3(42.85%) | 3(50%) | 0 |
| Clindamycin | 9(47.36%) | 2(20%) | 2(28.57%) | 4(66.66%) | 0 |
| Amoxyclav | 6(31.57%) | 3(30%) | 2(28.57%) | 2(33.33%) | 2(100%) |
| Ceftriaxone | 5(26.31%) | 2(20%) | 2(28.57%) | 1(16.66%) | 0 |

Table 5: Antibiotic susceptibility pattern of organism.

In this study the incidence of positive growth pus culture was 88% (i.e. 44 out of 50) from clinically suspected infected cases from different surgical departments which is comparable with the study performed by Krishna et al, (90%). The study also revealed higher number of post-operative wound infections in younger age group as compared to the elderly one. In this study there was female preponderance in culture positive patients because a greater number of infections was seen in obstetrics and gynecological surgeries. This is comparable with the study performed by Ward et al. Higher incidence of

would infection as seen in emergency operations (80%), then the elective operation (20%) in our study was statistically significant. Swathi et al, also observed increased incidence of SSI in emergency cases (84%). ¹² The effect of emergency surgery on the rate of SSI is likely to be due the fact that emergency procedures lack routine pre-op preparations which reduce the rate of SSI. (e.g. control of diabetes) and most of emergency operations involve contaminated areas such as the bowel and the perianal region. ¹³

In this study among 44 sample isolates, Staphylococcus aureus (43.2%) was the most common isolate followed by E. coli (22.7%), Klebsiella (15.9%), CONS (13.63%), Pseudomonas (4.54%) etc. These observations were similar to findings by Krishnan et al, who reported Staph. Aureus as the most frequent species isolated followed by Pseudomonas, E. coli, Klebsiella, CONS, Proteus, Acinetobacter, citrobacter, providential species. In study by B Ananthi et al, similar reports of Staph.aureus being the most common isolate followed by E.coli, CONS, Klebsiella, Proteus, Enterococcus, Pseudomonas. Acinetobacter, Citrobacter was observed.¹⁴ In this study MRSA isolates rate is 47.36% which is also similar to the study by Badauli et al, (50%).15

In this study 25 isolates of gram-positive cocci were highly sensitive to Linezolid and Clindamycin similar to study of Krishnan et al. The high incidence of gramnegative organisms in this study (19 out of 44) can be attributed to be acquired from patients normal endogenous and microflora. Among 19 isolates of gram-Negative bacteria 22.72% of *E. coli* and 15.9% of *Klebsiella* were ESBL producers. This correlates with the observation by Kumar et al, (22.42% and 14.3% respectively).¹⁶

CONCLUSION

Postoperative wound infections are real risks associated with any surgical procedure and health care services around the world. It is one of the most important factors responsible for significantly morbidity, mortality, unwanted prolonged hospitalization and additional cost of treatment in surgical patients. As the control of postoperative complications is an essential component of total management, therefore periodic surveillance postoperative wound infections should be done in every hospital at regular intervals to evolve the control strategies and reduce the infection rate. Judicious and rational use of antibiotics in post-operative wound infection targeted towards the prevailing organisms in that community not only help in reducing antibiotic induced bacterial resistance but also the overall cost of treatment in developing countries including India.

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