A Study on surgical Site Infections, their bacteriological profile and antimicrobial susceptibility pattern

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Abstract
Introduction: Surgical site infection (SSI) is second most common hospital acquired infection. The rate of SSI ranges from 2.5% to 41.9% globally. The identification of bacterial pathogens and their antibiotic susceptibility testing is required for successful treatment of SSI.

Objective: To study the bacterial pathogens in patients with SSI and their antibiotic susceptibility pattern.

Results: Out of 107 samples collected from post operative cases with symptomatic wound infection, 60 (56.07%) samples showed single isolates where as 13 (12.14%) samples showed multiple isolates and 34 (31.77%) did not show any bacterial growth. Out of 86 isolates, 29 (33.7%) isolates are Gram positive organisms and 57 (66.3%) isolates are Gram Negative organisms, among Gram positive isolates Staphylococcus aureus (MSSA) (44.8%) is predominant pathogen and in Gram negative isolates Escherichia coli (42.1%) is the predominant pathogen. In the present study Gram positive organisms showed high susceptibility to vancomycin and linezolid (100%) followed by gentamicin (79.5%) and Gram negative organisms showed high susceptibility to polymyxin B (94.7%) followed by imipenem (75.4%).

Conclusion: The study gives an insight into bacterial pathogens and their antibiotic sensitivity patterns isolated from SSI and there should be surveillance of SSI which helps to reduce the rate of SSI as well appropriate use of antibiotics.

Keywords: Hospital acquired infections, Surgical site infections (SSI), Antibiotic sensitivity testing, Gram positive bacteria, Gram negative bacteria.

Introduction
As a part of innate immunity the main function of intact skin in humans is to control the microbes that are resident on the skin surface and also it prevents the underlying tissues from colonization or invasion by pathogens.

If due to any condition (wounds) where there is exposure of subcutaneous tissue due to loss of integrity of skin it provides good environment for colonization and proliferation of microorganisms and so any wound is at risk of developing infection.

Infections occurring in the wound are major barriers for healing which shows impact on patients, which may prolong the hospital stay and effects the quality of life and wound healing requires a healthy environment which will result in normal healing process and also with minimal scar formation.

SSI was previously termed as post operative wound infections was termed by US center for disease control in order to prevent the confusion between infection at site of surgical incision and infection at the site of traumatic wound and SSI can be defined as proliferation of pathogenic microorganisms at the site of surgical incision which may involve skin and subcutaneous fat (superficial), Musculofacial layers (deep) in an organ/cavity.

Hospital acquired infections are common type of nosocomial infections in surgical patients and SSI is the second most common hospital acquired infection. Generally SSI occur within 30 days after the procedure but in cases of any added implants the duration of SSI may also extend upto one year from the operation procedure.

Despite efforts to prevent these SSI, the data of National Centre for Health Statistics and National Healthcare Safety Network suggests that 2.50,000 to 1 million SSI complicate 26.6 million inpatient surgical procedures performed annually in USA and this impact of SSI have been estimated to be 3.7 million hospital days and also excess cost of 1.6 billion dollars.

The rate of SSI ranges from 2.5% to 41.9% globally and the risk of acquiring hospital acquired infection is high in patients undergoing surgery and also 77% of death of patients with hospital acquired infections are related to SSI.

Hospital acquired infections are complicated by the increasing prevalence of some multi drug resistant organisms like Methicillin resistant Staphylococcus aureus (MRSA), Coagulase Negative Staphylococci, Vancomycin Resistant Enterococci (VRE), Escherichia coli, Acinetobacter baumannii, Pseudomonas aeruginosa which increase the mortality and morbidity.

Materials and Methods
This is a prospective observational study carried out after approval by institutional ethics committee for a period of one year (from August 2017 to August 2018) and a total of 107 samples were collected from either gender and different age groups after taking intra operative and post operative details. Samples were collected from SSI from patients with complaint of pain, swelling, discharge, delayed or non healing wound at surgical site.

Two swabs were collected from each subject from the surgical site following standard procedure. One swab was kept in a sterile test tube and the other in sterile nutrient broth (in order to maintain the viability of organisms) and they were immediately transported to microbiology.
laboratory and these specimens were inoculated onto Nutrient agar, Blood agar, Chocolate agar, MacConkey agar within 30 minutes to 1 hour after collection and these are incubated at 35°C-37°C aerobically and are observed for growth after 24 hrs and the plates which did not show growth after 24 hrs are re-incubated for other 48 hrs.

The isolates were then identified by colony morphology, Gram’s stain and conventional biochemical tests used for Gram positive and Gram negative bacteria. Antibiotic susceptibility pattern of the isolates was studied by Kirby bauer disc diffusion technique following Clinical Laboratory Standards Institute (CLSI) guidelines and the diameter of zone of inhibition was measured and interpreted as sensitive(S), intermediate (I) and resistant (R) after incubation at 35°C-37°C for 18-24 hrs using antibiotic discs (HiMedia Labs) Control strains like Escherichia coli (ATCC 25922), Staphylococcus aureus (ATCC 25923), Pseudomonas aeruginosa (ATCC 27853) were included in the study and the study findings are being explained in words, percentages and tables. ESBL detection in gram negative isolates was performed after screening by checking for presence of resistance against ceftriaxone, cefotaxime, cefazidime, cefpodoxime and confirmed by combined disc diffusion test using cefotaxime (30mcg) and cefazidime (30mcg) antibiotic disc (HiMedia labs) with and without clavulanic acid (10mcg) and MRSA detection was done by E-test using MHA with 2% NaCl with 0.5McFarland density according to CLSI guidelines.

Results

In present study a total number of 4642 (100%) surgical procedures were done in our institute during the study period out of which 107(2.30%) (Table 1) samples were collected from symptomatic SSI cases and out of the samples collected from post operative cases, 60(56.07%) samples showed growth of single isolates whereas 13(12.14%) samples showed multiple isolates and 34(31.77%) did not show any bacterial growth after 48 hrs of aerobic incubation. (Table 2).

In the present study out of 86 isolates, 29(33.7%) isolates were gram positive organisms and 57(66.3%) isolates were gram negative organisms (Table 3). Gram negative organisms were more frequently isolated than gram positive organisms.

Out of 29 Gram positive isolates, Staphylococcus aureus (MSSA) (44.8%) is the predominant pathogen followed by MRSA (31.03%), Coagulase Negative Staphylococci and Enterococcus species (10.3%), Streptococcus pyogenes (3.44%) respectively. (Table 4). Out of 57 Gram negative isolates, 24(42.1%) showed Escherichia coli followed by Klebsiella species 17(29.8%), Pseudomonas aeruginosa 15(26.3%), Acinetobacter baumannii 1(1.8%) respectively (Table 5). In the present study out of 86 isolates, 57(66.3%) were isolated from superficial SSI and 29(33.7%) from Deep or organ/space SSI and in superficial SSI among gram positives MSSA (15.78%) is predominant pathogen and among gram negatives Escherichia coli (26.31%) is predominant pathogen and from deep or organ /space SSI also MSSA (13.78%) is predominant pathogen among gram positives and Escherichia coli (31.03%) is predominant pathogen among gram negatives (Table 6).

In the present study, Gram positive organisms showed high susceptibility to vancomycin and linezolid (100%) followed by gentamicin (79.3%), tetracycline (75.9%), neomycin (55.2%), co-amoxiclav (48.3%), erythromycin, oxacillin, ceftriaxone and cephalothin (44.8%), cotrimoxazole (34.5%), ciprofloxacin (27.6%), penicillin (3.4%) (Table 7) whereas Gram negative organisms showed high susceptibility to polymyxin B (94.7%) followed by imipenem (75.4%), amikacin (73.7%), meropenem (71.96%), gentamicin (64.9%), piperacillin-tazobactam (52.6%), neomycin (49.1%), cefapirazone-sulbactam (43.8%), ciprofloxacin (40.3%), co-amoxiclav (33.3%), cotrimoxazole (26.3%), ceftazidime (10.5%), cephalothin (7%) respectively (Table 8). In the present study out of 86 isolates, 23(26.7%) are multidrug resistant and 8(9.3%) are carbapenem resistant (Table 9) and among 57 gram negative isolates 25(43.9%) were ESBL producers (Table 10) and overall rate of HAI in our institute is 1.9%.

Table 1: Shows total number of surgical procedures done during study period, no. of symptomatic SSI and rate of Hospital Acquired Infection (HAI) in our institute.

| No. of surgical procedures | 4642(100%) |
|----------------------------|------------|
| No. of symptomatic SSI      | 107(2.30%) |
| HAI                        | 86(1.9%)   |

Table 2: Shows number of isolates from clinical specimens.

| Single isolates | 60(56.07%) |
|----------------|------------|
| Multiple isolates | 13(12.14%) |
| No bacterial growth | 34(31.77%) |
| Total            | 107(100%)  |

Table 3: Shows number of Gram positive and Gram negative organisms isolated from clinical specimens.

| Total no. of isolates | 86(100%) |
|-----------------------|----------|
| Gram positive organisms | 29(33.7%) |
| Gram negative organisms | 57(66.3%) |

Table 4: Shows different types of Gram positive organisms.

| Organisms                        | No. of isolates |
|----------------------------------|-----------------|
| Staphylococcus aureus (MSSA)     | 13(44.8%)       |
| Staphylococcus aureus (MRSA)     | 9(31.03%)       |
| Coagulase Negative Staphylococci | 3(10.3%)        |
| Enterococcus species             | 3(10.3%)        |
| Streptococcus pyogenes           | 1(3.44%)        |
| Total                            | 29(100%)        |
Table 5: Shows different types of Gram negative organisms.

| Organisms                     | No. of isolates |
|-------------------------------|-----------------|
| Escherichia coli              | 24(42.1%)       |
| Klebsiella species            | 17(29.8%)       |
| Pseudomonas aeruginosa        | 15(26.3%)       |
| Acinetobacter baumanii        | 1(1.8%)         |
| Total                         | 57(100%)        |

Table 6: Shows distribution of bacterial isolates based on SSI class.

| Bacterial isolate                      | Superficial SSI   | Deep tissue or organ.Space SSI | Total |
|----------------------------------------|-------------------|-------------------------------|-------|
| Staphylococcus aureus (MSSA)           | 9(15.78%)         | 4(13.78%)                     | 13    |
| Staphylococcus aureus (MRSA)           | 7(12.28%)         | 2(6.89%)                      | 9     |
| Coagulase Negative Staphylococci       | 3(5.26%)          | 0(0%)                         | 3     |
| Enterococcus species                   | 2(355%)           | 1(3.44%)                      | 3     |
| Streptococcus species                  | 1(1.75%)          | 0(0%)                         | 1     |
| Escherichia coli                       | 15(26.31%)        | 9(31.03%)                     | 24    |
| Klebsiella species                     | 10(17.54%)        | 7(24.10%)                     | 17    |
| Pseudomonas aeruginosa                 | 9(15.78%)         | 6(20.68%)                     | 15    |
| Acinetobacter baumanii                 | 1(1.75%)          | 0(0%)                         | 1     |
| Total                                  | 57                | 29                            | 86    |

Table 7: Showing the antibiotic susceptibility pattern of gram positive organisms to various antibiotics.

| S. No | Name of the Antibiotic       | Sensitive | Resistant | Intermediate |
|-------|------------------------------|-----------|-----------|--------------|
| 1     | Vancomycin                   | 29(100%)  | -         | -            |
| 2     | Linezolid                    | 29(100%)  | -         | -            |
| 3     | Erythromycin                 | 13(44.8%) | 14(48.3%) | 2(6.9%)      |
| 4     | Tetracycline                 | 22(75.9%) | 7(24.1%)  | -            |
| 5     | Oxacillin                    | 13(44.8%) | 9(31.03%) | -            |
| 6     | Penicillin                   | 1(3.4%)   | 28(96.5%) | -            |
| 7     | Co-Amoxyclav                 | 14(48.3%) | 15(51.7%) | -            |
| 8     | Cotrimoxazole                | 10(34.5%) | 19(65.5%) | -            |
| 9     | Ceftriaxone                  | 13(44.8%) | 9(31.03%) | -            |
| 10    | Cephalothin                  | 13(44.8%) | 9(31.03%) | -            |
| 11    | Gentamicin                   | 23(79.3%) | 6(20.9%)  | -            |
| 12    | Ciprofloxacin                | 8(27.6%)  | 16(55.2%) | 5(17.2%)     |
| 13    | Neomycin                     | 16(55.2%) | 13(44.8%) | -            |

Table 8: Showing the antibiotic susceptibility pattern of gram negative organisms to various antibiotics.

| S. No | Name of the Antibiotic       | Sensitive | Resistant | Intermediate |
|-------|------------------------------|-----------|-----------|--------------|
| 1     | Polymyxin B                  | 54(94.7%) | 3(5.3%)   | -            |
| 2     | Imipenem                     | 43(75.4%) | 13(22.8%) | 1(1.7%)      |
| 3     | Meropenem                    | 41(71.9%) | 14(24.6%) | 2(3.5%)      |
| 4     | Piperacillin-tazobactam      | 30(52.6%) | 23(40.3%) | 4(7%)        |
| 5     | Cefepirazeo-sulbactam        | 25(43.8%) | 28(49.1%) | 4(7%)        |
| 6     | Ciprofloxacin                | 23(40.3%) | 34(59.6%) | -            |
| 7     | Amikacin                     | 42(73.7%) | 14(24.6%) | 1(1.7%)      |
| 8     | Gentamicin                   | 37(64.9%) | 18(31.6%) | 2(3.5%)      |
| 9     | Co-Amoxyclav                 | 19(33.3%) | 38(66.7%) | -            |
| 10    | Ceftriaxone                  | 6(10.5%)  | 36(63.1%) | -            |
| 11    | Cephalothin                  | 4(7%)     | 38(66.7%) | -            |
| 12    | Cotrimoxazole                | 15(26.3%) | 27(47.4%) | -            |
| 13    | Neomycin                     | 28(49.1%) | 22(38.6%) | 7(12.3%)     |
| 14    | Ceftazidime                  | 6(10.5%)  | 9(15.8%)  | -            |
Table 9: Shows no. of Multi Drug Resistant (MDR) and carbapenem resistant strains.

|                        | Total no of isolates | MDR strains | Carbapenem resistant strains |
|------------------------|----------------------|-------------|-----------------------------|
| Total no of isolates   | 86(100%)             | 23(26.7%)   | 8(9.3%)                     |
| MDR strains            |                      |             |                             |
| Carbapenem resistant   |                      |             |                             |

Table 10: Shows no.of ESBL producers among Gram negative isolates.

|                       | Total no of isolates | ESBL producers |
|-----------------------|----------------------|---------------|
| Total no of isolates  | 57(100%)             | 25(43.9%)     |
| ESBL producers        |                      |               |

Discussion

SSI is a problem in both developing countries as well as in developed countries inspite of introduction of various infection control practices and antibiotic regimens into surgical practice. Management of patients with SSI either with gram positive organisms or gram negative organisms depend on selection of effective and appropriate antibiotic or regimen against the organisms as antibiotics play an important role in both prophylaxis and treatment of infectious diseases.

According to Nandita pal et al, 23.3% showed single isolates whereas 36.7% showed multiple isolates. Mamma et al reported single isolates in 91.6% whereas multiple isolates in 8.4% and also reported that 87.4% samples were culture positive and 12.6% samples did not show any bacterial growth. In the present study the growth of single isolates are most frequent (56.07%) than multiple isolates (12.14%).

In the present study, 33.7% were Gram positive organisms and 66.3% were Gram negative organisms where Escherichia coli is the predominant isolate in gram negatives (42.1%) and Staphylococcus aureus (MSSA) (44.8%) in gram positives. According to Goswami et al, 31.15% were gram positives and 68.85% were gram negatives where Staphylococcus aureus (26.23%) is predominant pathogen whereas Escherichia coli showed 15.85%. According to Mamma et al, 47% were gram positives and 53% were gram negative organisms. Among these isolates, Staphylococcus aureus is the predominant isolate (32.4%) followed by Escherichia coli (20%). According to Amare et al, 44.1% were gram positive and 55.9% were gram negative organisms respectively and also reported that Escherichia coli was the most common isolate in gram negatives (24.3%) followed by Staphylococcus aureus (23.4%) in gram positives.

In the present study, gram positives showed 100% susceptibility to vancomycin and linezolid followed by gentamicin (79.3%), tetracycline (75.9%) whereas erythromycin, cephalothin, ceftriaxone (44.8%), ciprofloxacin (27.6%), penicillin (3.4%). According to Mamma et al, gram positives showed 100% susceptibility to vancomycin followed by gentamicin (91.2%), ceftriaxone (80.9%), ciprofloxacin (89.7%), erythromycin (77.9%), cephalothin (57.3%), tetracycline (48.5%), penicillin (13.2%). According to Goswami et al, vancomycin showed 61.4% susceptibility followed by ciprofloxacin (47.4%), tetracycline (42.1%), erythromycin (38.6%), penicillin (29.8%), gentamicin (29.8%) respectively.

In the present study gram negatives showed 94.7% susceptibility to polymyxin –B followed by imipenem (75.4%), meropenem (71.9%), amikacin (73.7%), piperacillin-tazobactam (52.6%), gentamicin (64.9%), ciprofloxacin (40.3%), ceftazidime-sulbactam (43.8%), cefoxitin (33.3%), cotrimoxazole (26.3%) whereas ceftriaxone and ceftazidime (10.5%). According to Nandita pal et al, 100% susceptibility was showed for imipenem, meropenem, piperacillin -tazobactam, ceferaerzone-sulbactam, amikacin followed by ciprofloxacin (85.7%), gentamicin (71.4%), ceftriaxone (30%), ceftazidime and cotrimoxazole (28.6%), co-amoxiclav (14.3%) respectively. According to Goswami et al, ciprofloxacin showed 67.5% susceptibility followed by meropenem (48.4%), cotrimoxazole (19.8%).

Conclusion

The study gives insight into bacterial pathogens and their antibiotic susceptibility patterns isolated from SSI in a tertiary care hospital. Surveillance of SSI along with feedback from surgeons will help to reduce the SSI rate and this surveillance system should be developed in all hospitals and also guidelines for antibiotic use among surgical patients should also be developed and strictly followed which may provide the estimate of incidence of SSI.

From the present study it was observed that microorganisms, both gram positive and gram negatives became resistant to more commonly used drugs like penicillin, cephalosporins and even quinolones which are cost effective. We are now left with few reserve drugs like carbapenems which should be used judiciously.

New technological advances (eg: Minimally invasive procedures) and emergence of antibiotic resistant organisms (eg: MRSA) led to additional challenges in prevention, identification and treatment of SSI.

Although there are many programmes centered to basic key principles of surgical care and antibiotic prophylaxis, there are still some unresolved issues regarding some aspects in antibiotic prophylaxis in surgical care patients like drug dose in obese patients, specific timings of antibiotic administration, role of anti MRSA prophylaxis etc. To conclude there is still much to learn about pathophysiology, prevention and surveillance of SSI even after 150 years of discoveries of Louis Pasteur and Joseph lister.

Conflicts of Interest: None.

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