Bacterial diversity and antimicrobial resistance in Surgical site infections: A challenge to be tackled at the earliest!

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ABSTRACT

Background and Objectives: *SSI's are ranked among the most common health care associated infections. They cause significant morbidity, increased cost of care and prolonged hospital stay. A spectrum of microorganisms with varied antimicrobial susceptibility patterns have been identified as causative agents of SSI's, which vary with time, hospital location and with the type of surgical procedure performed. We conducted this study with an objective to assess the burden of SSI, its causative aerobic bacteria and their in vitro antibiotic susceptibility patterns.*

Materials and Methods: *All samples from post operative surgical site were processed and identified by gram staining, colonial morphology and standard biochemical tests. Antibiotic susceptibility testing was performed as per CLSI guidelines.*

Results: *A total of 110 samples were processed out of which 40 (36.36%) were positive for SSI's. Male to female ratio was 1.53:1. Most common age group was 20-40 years(41.86%). E.coli(44.19%) was the predominant organism isolated, followed by S.aureus(13.95%). Most of the gram negative isolates were found to be sensitive to Polymyxin B (86.66%), Imipenem (80%), and Amikacin (73.33%). Gram positive isolates showed 100% sensitivity to Vancomycin and Linezolid.*

Conclusion: *Our study highlights an increasing incidence of SSI's, which is an alarming situation to clinical microbiologists and surgeons of our country. Hence good empirical pre operative antibiotic therapy, with post operative prophylaxis will help in the long run to decrease the incidence of SSI and improve the cost of living.*

Keywords: *SSI, E.coli, Polymyxin B, Imipenem, S.aureus, Vancomycin, Linezolid.*

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Quick Response Code:	Website: www.innovativepublication.com
	DOI:

INTRODUCTION

Surgery has made great advances in last three quarters of this century and post operative wound infection is the most common complication faced by surgeon since the advent of surgery. Results of infection cause delayed recovery and may leave permanent disability.[1]

In 1992, U.S Centre for disease Control (CDC) revised the definition of "wound infection" by creating the definition, 'Surgical site infection'(SSI).[2] SSI's are ranked among the most common health care associated infections, along with pneumonia, urinary tract infections and blood stream infections. They cause significant morbidity, increased cost of care and prolonged hospital stay.[3]

SSI's are defined as infections which occur within 30 days after the operative procedure (except in case of added implants, when the duration extends to one year from operation). CDC, USA classifies SSI's into:

i. Superficial incisional SSI which involves only skin and subcutaneous tissue of incision.
ii. Deep incisional SSI which involves deep soft tissues of the incision. (eg: fascia and muscles layer).
iii. Organ/ Space SSI includes infection apparently related to the operative procedure and infection involves any part of the body, excluding skin incision, fascia, muscle layer that is operated or manipulated during operative procedure.[4]

A surgical wound may get infected by exogenous bacterial flora which may be present in the environment air of operation theatre, or by endogenous flora.[5] A summation of several factors contribute to the development of SSI such as inoculum of bacteria introduced into the wound during the procedure, the virulence of the contaminants, the micro environment of each wound and the integrity of the patients host defense mechanism.[5] Some types of surgery carry a greater risk of surgical wound infection than others, for example: operations on the bowel are at higher risk, because of the large number of microorganisms present in the operation site at the time of operation.[6] A spectrum of microorganisms with varied antimicrobial susceptibility patterns have been identified as causative agents of SSI's, which vary with time, hospital location and with the type of surgical procedure performed.[5]

Hence, as clinical microbiologists, we conducted this study with an objective to assess the burden of SSI, its causative aerobic bacteria and their in vitro antibiotic susceptibility patterns.

MATERIALS AND METHODS

I. **Study design and site:** The present prospective study was conducted in the department of Microbiology, at Shimoga Institute of Medical Sciences, Shimoga. The study period was from January 2015 to July 2015. Institutional ethics committee approval was obtained.

II. **Specimen collection and culture:** All samples from post operative surgical site sent to Microbiology laboratory, were included in this study. The samples were inoculated on both Blood agar and MacConkey's agar plates and incubated at 37^0C for 24 hours aerobically. Gram staining was also carried out for direct examination of the sample. The isolates were identified initially by colonial morphology and Gram staining. The isolates were further confirmed by standard biochemical tests as per CLSI guidelines 2014. Escherichia coli ATCC 25922 and Staphylococcus aureus ATCC 25923 were used as controls.[7]

III. **Antimicrobial susceptibility testing:** In vitro antibiotic susceptibility of the identified organisms was determined using Kirby Bauer disc diffusion technique using CLSI guidelines. Commercially available antibiotic discs from Himedia labs, Mumbai were used. Zones of inhibition after incubation were observed and measured. The interpretation of the measurements as sensitive, intermediate sensitive and resistant was made as per CLSI guidelines 2014 (M100-S24).[8]

RESULTS:

A total number of 110 samples were collected during the period of the study out of which 40 (36.36%) were positive for surgical site infections. Most of the samples were obtained from individuals between the age of 20-40 years. Infection rate was was more in males (60.47%) compared to females (39.53%). Male to female ratio in our study was 1.53:1. The details of distribution of samples by age and sex are depicted in table no 1.

Table 1: Distribution of samples by age and sex.

Age in years	Number (Percent)	No of males (Percent)	No of females (Percent)
0-20	12 (27.91%)	8 (18.60%)	4 (9.30%)
21-40	18 (41.86%)	9 (20.93%)	9 (20.93%)
41-60	12 (27.91%)	8 (18.60%)	4 (9.30%)
61-80	01 (2.32%)	1 (2.32%)	0 (0.00%)
Total	43 (100%)	26 (60.47%)	17 (39.53%)

The most common organism isolated in our study was Escherichia coli (44.19%). This was followed by Staphylococcus aureus (13.95%). Our findings correlate with other studies[1,5] and are depicted in Table 2.

Table 2: Distribution of isolates in SSIs

Gram reaction	Organism	Number (Percent)
Gram negative bacteria n=30 (69.76%)	Escherichia coli	19 (44.19%)
	Gram negative non fermenters	4 (9.30%)
	Pseudomonas species	3 (6.98%)
	Proteus species	2 (4.65%)
	Klebsiella species	2 (4.65%)
Gram positive bacteria n=13 (30.24%)	Staphylococcus aureus	6 (13.95%)
	Staphylococcus epidermidis	4 (9.30%)
	Enterococcus species	3 (6.98%)
Total= 43 (100%)	TOTAL	43 (100%)

In our study, most of the gram negative isolates were found to be sensitive to Polymyxin B (86.66%), Imipenem (80%), and Amikacin (73.33%). Most of the isolates showed resistance to other antibiotics. The antibiotic susceptibility pattern in gram negative isolates has been documented in Table 3.

Table 3: Antibiotic susceptibility pattern of Gram negative isolates

Antibiotic	E.coli n=19	GNNF n=4	Pseudomonas spp; n=3	Proteus spp n=2	Klebsiella spp n=2
Imipenem (IPM)	16(84.21%)	2 (50%)	2 (66.66%)	2 (100%)	2 (100%)
Polymyxin B (PB)	19(100%)	2 (50%)	3 (100%)	0 (0%)	2 (100%)
Ampicillin (AMP)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Gentamycin (GEN)	9(47.37%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Ciprofloxacin (CIP)	2(10.53%)	1 (25%)	1 (33.33%)	1 (50%)	0 (0%)
Amikacin (AK)	16(84.21%)	1 (25%)	2 (66.66%)	1 (50%)	2 (100%)
Cefotaxime (CTX)	5(26.32%)	0 (0%)	0 (0%)	0 (0%)	1 (50%)
Cefotaxime Clavulunate(CEC)	8(42.11%)	1 (25%)	0 (0%)	0 (0%)	1 (50%)

All the Gram positive bacteria in our study were senstive to Linezolid and Vancomycin. However, most of the isolates showed resistance to Penicillin, Cefoxitin, Ampicillin, Ciprofloxacin and Clindamycin. This finding correlates with findings of other studies.[5]

Table 4: Antibiotic susceptibility pattern of Gram positive isolates

Antibiotic	Staphylococcus aureus n=6	Staphylococcus epidermidis n=4	Enterococcus species n=3
Penicillin (P)	0 (0%)	0 (0%)	0 (0%)
Erythromycin (E)	2 (33.33%)	2 (50%)	0 (0%)
Clindamycin (CD)	2 (33.33%)	2 (50%)	0 (0%)
Gentamycin (GEN)	2 (33.33%)	2 (50%)	1 (33.33%)
Ciprofloxacin (CIP)	2 (33.33%)	2 (50%)	0 (0%)
Cefoxitin (CX)	3 (50%)	2 (50%)	-
Vancomycin (VA)	6 (100%)	4 (100%)	2 (66.66%)
Linezolid (LZ)	6 (100%)	4 (100%)	2 (66.66%)

DISCUSSION

The problem of post operative wound infection is seen in both developed and developing countries, despite the introduction of meticulous antiseptic regime in surgical practice, sterilization and operative techniques. In our study, we obtained 40 positive samples from January 2015 to July 2015. 43 organisms were isolated from 40 patients. Out of 43, 30 (69.76%) were Gram negative bacteria and 13 (30.24%) were Gram positive. Makanjuola et al., have also observed higher incidence of Gram negative bacteria (62.8%) compared to Gram positive bacteria (37.2%).

We observed a higher rate of infection in age group of 20-40 years (39.53%). This correlates with the findings of Makanjuola et al. The reason for obtaining a higher rate of post operative wound infection in young adults could be due to greater number of young adults getting operated for exploratory laparotomy for intestinal perforation and appendicular perforation or appendicitis.[1]

The spectrum of bacteria most frequently involved in surgical infections has changed over a period of time. Streptococcus being the most frequent and feared pathogen nearly a century ago was replaced by Staphylococcus about eight decades later and by Gram negative isolates as principal offenders in recent years.[5]

The predominant organism isolated in our study was Escherichia coli, as it had the highest prevalence with 44.19%. Kakati et al., have also documented E.coli as the predominant cause of SSI.

Staphylococcus aureus was the second predominant bacteria isolated, and the most commonly isolated Gram positive organism in our study. Kakati et al., also have observed similar findings in their study. Aniruddha S et al., have isolated Staphylococcus aureus as the most common pathogen in SSI.

The increased infection rate with prolonged pre operative hospital stay may be due to lowered immunity of patients due to age and other diseases which are responsible for pre operative stay and during the pre operative stay, the patient becomes increasingly exposed to bacteria to which he has not developed immunity and these bacteria may be antibiotic resistant.

While deciding pre operative antibiotic therapy, many factors must be considered including previous antibiotic therapy, knowledge of the usual causative organisms and their antibiotic susceptibilities. SSI's can be polymicrobial, hence empirical therapy should include relatively broad spectrum antibiotics, especially for patients with severe infections and those who are immunocompromised.[2]

There is a gradual increase in the emergence of antibiotic resistant organisms in surgical patients.[9] Most sensitive antibiotics in our study were Polymyxin B, Imipenem, Amikacin and to some extent Fluroquinolones in Gram negative bacteria and to Linezolid, Vancomycin and partly to fluroquinolones in Gram positive bacteria. Similar findings have been observed by Ramesh et al. Hence these drug combinations should be used for empirical therapy, but the prophylaxis should be continued with lower drugs according to available surgical prophylaxis guidelines to prevent spread of resistance.

CONCLUSION

The incidence of infection varies from surgeon to surgeon, from hospital to hospital, from one surgical procedure to another, and most importantly from one patient to another.[10] Our study highlights an increasing incidence of SSI's, which is an alarming situation to clinical microbiologists and surgeons of our country. A plethora of risk factors contribute to post operative SSI's. It is evident that SSI increases with an increase in degree of contamination of wounds operated upon. Prevention, in the form of adequate infection surveillance, ensuring standard aseptic techniques as well as proper preparation and maintenance of operating room are very important in addition to pre operative antibiotics, in decreasing the incidence of SSI's.

As our findings are similar to findings of other studies, it is important to emphasize the fact that antibiotic resistance in SSI's, is a global phenomenon. Hence good empirical pre operative antibiotic therapy, with post operative prophylaxis will help in the long run to decrease the incidence of SSI and improve the cost of living.

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How to cite this article: Misbah N, Prakash. N, Halesh L.H, Siddesh K.C, Bacterial diversity and antimicrobial resistance in Surgical site infections: A challenge to be tackled at the earliest! Indian J Microbiol Res 2015;2(4):206-209.