Microbial Etiology and Antibiogram of SSIs following LSCS in a Tertiary Care Hospital in Amritsar.

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ABSTRACT

Background: Surgical Site Infections (SSIs) are the second most commonly reported nosocomial infections after Urinary tract infection following a delivery by caesarean section. They add significantly to the morbidity, mortality and socioeconomic consequences in both patients as well as health care systems. **Aim:** To study the microbial etiology and antibiogram of post caesarean SSIs. **Methods:** This prospective study was conducted on pregnant women undergoing elective/emergency caesarean section irrespective of the indication during a study period from Jan 2017 to June 2018. 250 pregnant women undergoing LSCS developing signs and symptoms suggestive of SSI till the 30th post-operative day were included as cases. Under all aseptic precautions, two pus swabs were collected from every post caesarean women enrolled in the study. The first swab was used for gram staining while the other was inoculated on 5% blood agar and Macconkey agar. The isolates were identified by their colony characters and their standard biochemical tests. Antimicrobial susceptibility pattern was performed on Mueller Hinton agar plates by Kirby Bauer disc diffusion methods and interpreted as per latest CLSI guidelines. **Results:** Among 250 cases suspected of having SSIs, 93 aerobic isolates were obtained. Staphylococcus aureus (41.9%) was the most common organism isolated followed by Acinetobacter baumanii (15.1%), Escherichia coli(11.8%) Klebsiella pneumoniae(9.7%). 15/39(38.4%) of Staphylococcus aureus were observed to be Methicillin resistant. 15/45 (33.3%) of Gram negative isolates were Extended Spectrum Beta- Lactamase producers. **Conclusion:** Since MRSA is the leading cause of post caesarean SSI and the incidence of multidrug resistant pathogens as a cause of post-operative wound infections is rising with the increased prevalence of ESBL, therefore rational and judicious use of antimicrobials, active surveillance and reporting of SSI, enforcement of aseptic measures, implementation of infection control strategies and their periodic review is the need of an hour to reduce the rate of post caesarean SSIs.

Keywords: SSIs, Post caesarean, Bacterial pathogens, Antimicrobial resistance, MRSA, ESBL.

INTRODUCTION

Surgical site infections(SSIs) are the second most commonly reported nosocomial infections leading to lengthening of the hospital stay by an average of 7.4 days with reported incidence from as low of 2.5% to as high as of 41.9%.[1] SSI continues to be a major public health problem even in hospitals with most modern facilities despite of the advancement in infection control and surgical practices.[2] In Obstetrics, Emergency Lower Segment Caesarean Section (LSCS) is one such procedure whose incidence has increased substantially worldwide due to emphasis on improving the maternal and perinatal outcome. The incidence of SSI has also increased with the increase in the numbers of LSCS being performed.[3] The rate of post caesarean SSI ranges from 3 to 15% depending upon the surveillance methods used to identify infections, the patient population and the use of antibiotic prophylaxis. Determinants of infection may be related to the host, microbe, environment, procedure adopted or perioperative antibiotic prophylaxis. In clean-contaminated procedures like caesarean section the polymicrobial aerobic–anaerobic flora closely resembling the normal endogenous microflora of the operated organ constitutes the most frequently isolated pathogens. Among the aerobic bacterial agents causing post- caesarean surgical site infections, Gram negative bacilli includes Escherichia coli, Klebsiella, Pseudomonas, Acinetobacter, Proteus etc. Most frequently isolated

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Gram positive bacteria includes Staphylococcus aureus, Coagulase negative Staphylococcus, Streptococcus faecalis, group B Streptococcus.

Since, SSI is a major cause of sepsis after caesarean section and in spite of the availability of antibiotics, they are still responsible for much morbidity and far reaching socioeconomic consequences for both patients as well as health care system. The use of prophylactic antibiotic is a common practice to avoid SSIs; however indiscriminate use of antibiotics can lead to problems including an increase in costs and the emergence of resistant micro-organisms. Studies have shown an increase in the trend of SSIs attributable to antimicrobial resistant pathogens such as Methicillin Resistant Staphylococcus aureus and Extended spectrum beta lactamases producing microorganisms. Hence, a working knowledge of the microbial etiology and antibiogram will help in rationalizing the use of appropriate antibiotics with proper timing and dosage for surgical prophylaxis thereby contributing to the reduction in rate of SSIs.

MATERIALS AND METHODS

The present prospective study was conducted in the Department of Microbiology, Government Medical College and Department of Obstetrics & Gynaecology, Guru Nanak Dev Hospital, Amritsar over a period of one and a half year from January 2017 to June 2018. 250 pregnant women undergoing elective/ emergency caesarean section developing signs and symptoms suggestive of SSI till the 30th post-operative day were included as cases after taking written informed consent from every patient enrolled in the study. Due approval of Institutional Ethics Committee, Government Medical College, Amritsar was taken before the commencement of the study. Under all aseptic precautions, two pus swabs were collected from every post caesarean women enrolled in the study. The first swab was used for gram staining4 while the other was inoculated on 5% blood agar and Macconkey agar and incubated at 37◦C for 48 hours before being reported as sterile. The isolates were identified by their colony characters and their battery of standard biochemical tests.[5,6] Antimicrobial susceptibility testing was performed on Mueller Hinton agar plates by Kirby Bauer disc diffusion methods and interpreted as per latest CLSI guidelines.[7] All Staphylococcus strains isolated from SSIs were tested for Methicillin resistance and all Gram negative strains isolated from surgical site infections were further tested for Extended spectrum beta lactamase (ESBL) production as per the recommended CLSI guidelines.[7]

Stastical Analysis

The data was entered in MS EXCEL worksheet and analysed using statistical software SPSS version 17.0. The percentage of various aerobic bacteria causing SSI and antimicrobial susceptibility profile were summarized by frequency (percentage) by using Frequency tables.

RESULTS

A total of 250 cases out of 1225 women who underwent elective/emergency LSCS during the study period satisfied the inclusion criteria for SSIs. Hence the rate of SSI was 250/1225(20.4%) with aerobic culture positivity obtained in 89/250 (35.6%) cases while 161/250(64.4%) showed no aerobic bacterial growth. Most of the SSIs in this study were found to be superficial and limited to the stitch line. 58/89(65.2%) cases showed maximum culture positivity at 6th to 8th day postoperatively followed by 14(15.7%) on 10th -12th day, 11(12.3%) on 4thpost operative day and the remaining 6 (6.74%)cases developed SSI during their post discharge follow up . Out of 89 culture positive cases, 85(95.5%) showed monomicrobial growth while 4(4.5%) showed mixed infection with two bacterial isolates thereby accounting to total of 93 bacterial isolates. Gram positive bacterial growth was obtained in 48/93(51.6%) of the cases and Gram negative bacteria contributed 45/93(48.4%) of all the bacterial isolates. Among the Gram positive isolates, Staphylococcus aureus 39/93(41.9%) was the commonest followed by Coagulase negative Staphylococcus 9/93(9.8%). Among the Gram negative bacteria, Acinetobacter baumanni was the most common isolate 14/93(15.1%) followed by Escherichia coli 11/93(11.8%), Klebsiella pneumoniae 9/93 (9.7%), Citrobacter species 7/93(7.5%) and Pseudomonas aeruginosa 4/93(4.3%). [Table 1]

Table 1: Isolation of Various Pathogens Causing Post Caesarean Surgical Site Infections

Organisms isolated	No.of isolates (n=93)	Percentage (%)
Staphylococcus aureus MSSA MRSA	39 24 15	41.9 61.5 38.5
Acinetobacter baumanii	14	15.1
Escherichia coli	11	11.8
Klebsiella pneumoniae	9	9.7
Coagulase negative Staph	9	9.7
Citrobacter species Citrobacter freudii Citrobacter koseri	7 6 1	7.5 85.7 14.2
Pseudomonas aeruginosa	4	4.3

Among the Gram positive isolates ie S.aureus and CoNS, maximum resistance was seen to ampicillin (71.8% and 77.7%) with less resistance to amikacin(12.8% and 33.3%) and no resistance was observed to linezolid and vancomycin respectively. Both gram positive isolates were moderately sensitive to amoxyclav (51.2% and 44.4% respectively). (Table 2) Methicillin resistance was seen in 38.4% of the S.aureus isolates and 44.4% of Coagulase negative Staphylococcus isolates.[Figure 1] D test positivity was seen in 6 out of 39 (15.3%) of Staphylococcus aureus isolates. [Figure 2]

Table 2: antibiotic resistance pattern among gram positive isolates.

	No of resistant isolates (%)										
Organism	**Amp**	**Amc**	**Ak**	**G**	**Cs**	**Cx**	**Cot**	**Cf**	**E**	**Lz**	**Va**
S.aureus n=39 %age	28 71.8	20 51.2	5 12.8	10 25.6	24 61.5	15 38.4	12 46.2	14 35.8	23 58.9	0 0	0 0
CoNS n=9 %age	7 77.7	4 44.4	3 33.3	5 55.5	7 77.7	4 44.4	3 33.3	4 44.4	6 66.6	0 0	0 0

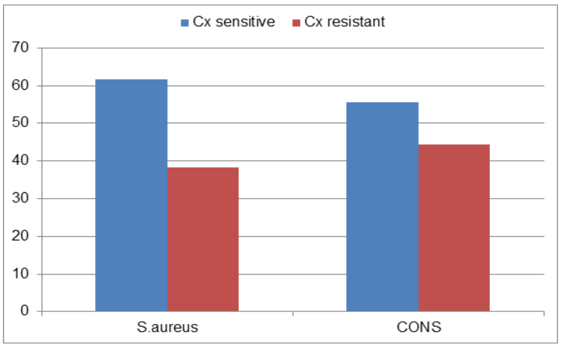


Figure 1: Methicillin Resistance in Staphylococcus aureus and Coagulase negative Staphylococcus

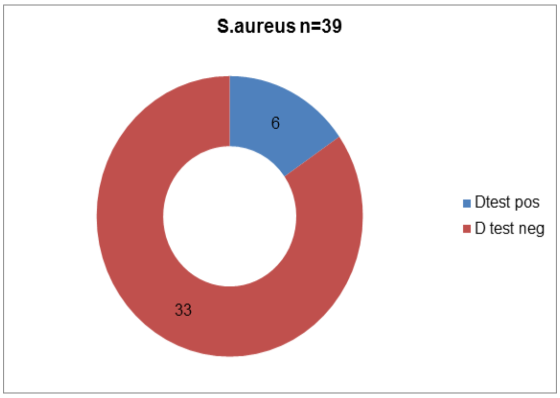
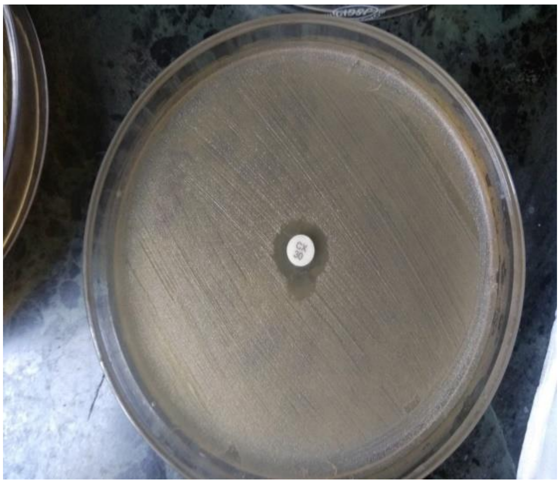


Figure 2: Inducible Clindamycin Resistance with the help of D-test

D Test Positive Showing Inducible Clindamycin Resistance.



Methicillin Resistant Staphylococcus Aureus

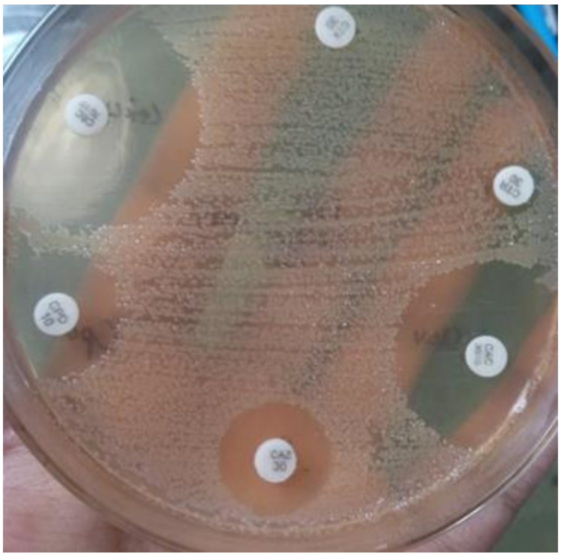
The Gram negative isolates showed minimum resistance to piperacillin-tazobactam seen in 11.1% of K.pneumoniae, 21.4% of A.baumanii, 28.5% of Citrobacter species and 27.2% of E.coli isolates. No resistance to piperacillin-tazobactam was seen in Pseudomonas aeruginosa . Gram negative isolates showed higher rate of resistance to ceftazidime and cefotaxime in addition to ciprofloxacin. Acinetobacter baumanii was one of the most resistant organism with maximum resistance to ciprofloxacin (85.7%) followed by ceftazidime (78.5%), cefotaxime (71.4%) and amikacin (71.4%). 2/4(50%) of P.aeruginosa were sensitive to all the antimicrobials whereas 1/4(25%) strain was an ESBL producer. Escherichia coli showed maximum sensitivity to piperacillin tazobactam (72.8%) and cefotaxime clavulanate (72.8%) followed by aminoglycosides (63.7%) while maximum resistance was seen to ciprofloxacin(81.8%). Imipenem resistance was seen in 2 /11(18.1%) strains of E.coli. (Table -3) All the Gram negative isolates resistant to cefotaxime were further tested for ESBL production by phenotypic confirmatory method i.e Combined disc method. ESBL production was observed in 33.3% of all the Gram negative isolates. 5/9 (55.5%) K.pneumoniae, 4/11(36.3%) E.coli, 3/14 (21.4%) A.baumanii, 2/7 (28.5%) Citrobacter species and 1/4 (25%) P.aeruginosa strains were ESBL producers. (Table -4)

Table 3: Antibiotic Resistance Pattern Among The Gram Negative Isolates.

	No of resistant isolates (%)								
Org	**Ak**	**G**	**Cf**	**Pc**	**Pctz**	**Cz**	**Ctx**	**Ctxclav**	**Imp**
Ac. bau n=14 %	10 71.4	8 57.1	12 85.7	10 71.4	3 21.4	11 78.5	10 71.4	7 50	3 21.4
Kleb pn n=9 %	2 22.2	3 33.3	4 44.4	5 55.5	1 11.1	7 77.7	7 77.7	2 22.2	2 22.2
E.coli n=11 %	4 36.3	4 36.3	9 81.8	8 72.7	3 27.2	7 63.6	6 54.5	3 27.2	2 18.1
Citro spp n=7 %	3 42.8	3 42.8	5 71.4	3 42.8	2 28.5	5 71.4	4 57.1	2 28.5	1 14.2
Ps aer n=4 %	1 25	1 25	1 25	1 25	0 0	1 25	1 25	0 0	0 0

Table 4: ESBL Production Among The Gram Negative Isolates

S.No	Organisms	No of ESBL producers	ESBL (%age)
1	Klebsiella pneumoniae	5/9	55.5
2.	E.coli	4/11	36.3
3.	Acinetobacter baumanii	3/14	21.4
4.	Citrobacter species	2/7	28.5
5.	Pseudomonas aeruginosa	1/4	25.0



Esbl phenotypic confirmatory test.

DISCUSSION

Healthcare-associated infections constitute an essential health challenge worldwide and pose a major threat to patient safety.[8] Among the Health care associated infections (HAI), Surgical site infections (SSI) previously termed as post-operative wound infections are one of the most common HAI in low and middle income countries. Despite the advancement in operative techniques and a better understanding of the pathogenesis of the wound infections, SSIs still contribute a significant burden due to the emergence of antimicrobial resistant pathogens. The increasing incidence of caesarean deliveries worldwide has contributed to greater wound morbidity.[9] The rates of SSI are considered as an indicator of the quality of surgical and postoperative care provided by the hospitals.

- The overall rate of SSI in the present study was found to be 250/1225 (20.4%) and compared favourably with other reported rates ranging from 2.5 to 41.9%.[10,11] Similar results were obtained in study conducted by Sonal Saxena at Lady Hardinge Medical College, New Delhi in 2013,[12] that reported the incidence to be 24.4% including the post discharge surveillance. A prolonged preoperative stay with exposure to hospital environment, instrumentation, duration of surgery and presence of co-morbid illness like diabetes, hypertension may contribute to increased rate of SSI.[13]
- In our study a total of 89/ 250 (35.6%) showed aerobic bacterial growth as compared to the study conducted by Preethishree et al,[14] in Karnataka, India that reported a higher culture positivity of 59.76% . In the present study, this can be attributed to the fact that preoperative antibiotics were given to 37.2% cases even before the sample was sent for culture or the presence of anaerobes which could not have grown on aerobic culture.
- In our study, the most commonly isolated organism was Staphylococcus aureus (41.9 %) followed by Acinetobacter baumanii(15.9%).CONS was isolated in 9.7% of the culture positive cases. This was similar to National Nosocomial Infection Surveillance (NNIS) service survey (1997–2001) that reported Staphylococcus aureus (47%) including MRSA and Staphylococcus epidermidis (CONS) as the most common organism causing SSI.[15] In another study by S P Lilani et al,[16] Staph. aureus was found as the most common organism isolated from the SSI sites. Staphylococcus aureus is a predominant cause of SSIs worldwide with a prevalence rate ranging from 4.6% to 54.4%.[17] Infection with S. aureus is most likely associated with endogenous source as it is a part of the skin and nasal flora and also with exogenous contamination

from environment, surgical instruments or from hands of health care workers.[18,19] More than 70% isolates of Staphylococcus aureus and CoNS were resistant to penicillin. Ineffectiveness of penicillin in Staphylococcus aureus has been reported in other studies as well.[16,20,21] In our study methicillin resistance was seen in 38.4% of S.aureus isolates and 44.4% of CoNS isolates. In study done by Saxena et al.,[12] methicillin resistance was observed in 23.8% strains of S.aureus. Another study by Malik S1 reported MRSA in 55.7% of the Staphylococcus isolates. MRSA infections are of great concern due to high morbidity and mortality rates.

- Among the Gram negative isolates, Acinetobacter baumanii (15.1%) was the most common organism followed by Escherichia coli (11.8%), Klebsiella pneumoniae (9.7%), Citrobacter species (7.5%) and Pseudomonas aeruginosa (4.3%) which was the least common among the isolated organisms. As reported in various other studies as in Saxena et al,12 Acinetobacter species is frequently isolated from the operation theatre environment and this could have been the reason for contamination of the wound edges at the time of surgery. Among the Gram negative isolates , maximum resistance was seen to ciprofloxacin followed by third generation cephalosporins like ceftazidime and cefotaxime. This is in concordance with the study conducted by Mpogoro et.al.,22 where a total of 10-22% of gram-negative enteric bacteria were found to be resistant to third generation cephalosporins. Acinetobacter baumanii was among the most resistant organism due to its predominant role in nosocomial infections. The high levels of antibiotic resistance seen in this study could be attributed, at least in part, to inappropriate use of antimicrobial agents that affected both the cost and efficacy, thus favouring the emergence of resistant bacteria.

- The frequency of ESBL producers of 33.3percent in our study is comparable to previous Indian studies.1,23 The most frequent ESBL producing isolates in our study were Klebsiella(55.5%) and E.coli(36.3%) species. The higher rate of ESBL production was observed in a study conducted by Aniruddha S24 where all the cefotaxime resistant strains of E.coli and K.pneumoniae were ESBL producers. ESBL producers show cross- resistance to other antimicrobial agents also, thus limiting the therapeutic choice. Thus screening for ESBL production as a routine procedure in clinical laboratories gives valuable information to the clinician in appropriate selection of antimicrobial agents. Based on the prevalence rate of the ESBL producers in a healthcare facility, antibiotic policy of the institution can be tailored to achieve superior therapeutic outcome and bring about a reduction in healthcare costs. It also eliminates the misuse of conventional cephalosporins in a significant proportion of patients.

CONCLUSION

Since MRSA is the leading cause of post caesarean SSI and the incidence of multidrug resistant pathogens as a cause of post-operative wound infections is rising with the increased prevalence of ESBL, therefore it becomes imperative to understand the local antibiotic susceptibility patterns existing in a community so that an institutional antimicrobial policy based on local microbiological data may be formulated.

In addition, reducing the rate of SSI will help to reduce the unnecessary morbidity and mortality associated with it as well as reduce the wastage of healthcare resources, treatment cost and economic burden on the patient. The recommendations include addressing the modifiable risk factors in the preconception period, ensuring a sterile environment, aseptic surgeries, meticulous haemostatic techniques, and a variety of preventive measures aimed at neutralizing the threat of microbial contamination posed by operative staff, the operating room environment, and the patient's endogenous skin flora to reduce the incidence of infection. Additionally, rational and judicious use of antimicrobials, timely administration of antibiotic prophylaxis, suitable antibiotic policy development, active surveillance and reporting of SSI, enforcement of aseptic measures, implementation of infection control strategies and their periodic review is the need of the hour.

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