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Antibiotic Susceptibility Pattern of Bacterial Isolates from Surgical Site Infections in a Teaching Hospital, Tirupati, Andhra Pradesh

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Abstract: Surgical site infection (SSI) is a severe problem in the surgical specialties, which can cause mortality, morbidity, delays incision healing and economic burden. Advances in control of infections have not completely eradicated this problem because of development of drug resistance. Aims and objectives are isolation and identification of the bacterial pathogens from the infection site and determine the antibiotic susceptibility pattern of the isolated pathogens. A total number of 210 clinically suspected cases of surgical site infections in various surgical departments during July 2009 to April 2010 were included. Standard bacteriological methods were used for bacterial isolation and Susceptibiliity testing. Among enrolled cases the age of the patients ranged from ≤20 yrs to ≥ 60 yrs. Out of 210 wound swabs 167/210 were culture positive aerobically yielding 235 bacterial isolates, while 43 were sterile. Of this 112/131 were emergency surgery patients. The maximum rate of isolates was seen after EmLSCS. Of all risk factors pre-operative stay of >7 days was considered as the most significant. All Gram positive cocci isolated were sensitive to Vancomycin (100%), followed by Amikacin and Clindamycin. 81.16% of S.aureus were sensitive to Oxacillin. All Gram negative bacilli isolate showed 100% sensitivity to imipenem. Klebsiella pneumoniae showed 95.35% sensitivity to Amikacin. Escherichia coli showed 91.18% sensitivity to Amikacin, followed by Piperacillin/tazobactam (70.58%). Proteus species showed 100% sensitivity to Amikacin & Piperacillin/tazobactam followed by 80% Ceftrioxone & Ticarcilin/ clavulanate. Pseudomonas aeruginosa showed 100% sensitivity to Imipenem, 82.61% to Amikacin & 80.43% to Piperacillin-tazobactam. Citrobacter species showed 85.71% sensitivity to Amikacin, 71.43% to Piperacillin-tazobactam and 57.14% to Ticarcillin/clavulanate. The present bacteriological study has determined the commonest bacteria and antibiotic susceptibility patterns of the surgical site infections. There is a need to develop national surveillance of antibiotic – resistant organisms.

Keywords: Surgical Site Infection, Common Pathogens, Emergency surgeries, Antimicrobial Susceptibility pattern, Antibiotic policy.

INTRODUCTION

Surgical Site Infection (SSI) can occur anytime from 0 to 30 days after a procedure in which no implant is used and up to 1 year if foreign material like prosthetic heart valve or hip prosthesis is implanted. About 80% to 90% of all Postoperative infections (SSI) occur within 30 days after the operative procedure. Most of the wound infection manifest within a week of surgery. Streptococcus pyogenes infections appear within a day or two; while Staphylococcal infections typically take four or five days and Gram negative bacillary infections take six or seven days to appear. Staphylococcus aureus is the most commonly isolated human bacterial pathogen and it is an important cause of skin and soft-tissue infections [1].

Wound infections have been a problem in the field of infections has not completely eradicated the problem because of development of resistance [2]. Antimicrobial resistance can increase complications and costs associated with procedures and treatment. An infected wound complicates the postoperative course and results in prolonged stay in the hospital and delayed recovery [3].

Postoperative SSI is among the most common problems for patients who undergo operative procedures and the third most frequently reported nosocomial infection in the hospital population [4].

The wide spread use of antibiotics, together with the length of time over which they have been available have led to major problems of resistant organisms, contributing to morbidity and mortality. Pathogens that infect surgical wounds can be part of the patients normal flora (endogenous source) or acquired from the hospital environment or other infected patients (exogenous source). The skin bacteria comprise commensals, transients and pathogens. The transient organisms include Staphylococcus aureus and Coliforms. Identification of a microbe that has been recovered from a clinical specimen is beneficial to the patient and assists in selection of chemotherapy [5]. Gram positive cocci and Gram negative bacilli with unusual patterns of resistance have also been frequently isolated. Since bacteria resistant to multiple antibiotics are commonly found, there is possibility of extensive outbreak which may be difficult to control. Therefore, it is imperative to establish an early diagnosis and formulate an effective antibiotic policy [6].

This study will assess the prevalence of Surgical Site Infections and current anti-microbial susceptibility patterns of bacterial isolates in post-operative wound infections in among the patients admitted to S.V. Government Medical College, Teaching Hospital, Tirupati, Andhrapradesh. It assists the clinicians in appropriate selection of antibiotics for prophylaxis and treatment.

AIMS AND OBJECTIVES

1. To isolate and identify the bacterial pathogens from the infected surgical site.
2. To determine the antibiotic susceptibility pattern of the isolated pathogens.

MATERALS AND METHODS

The present study was conducted on patients admitted for surgery in various surgical units of S.V. Medical College; teaching hospital includes S.V.R.R. Government General Hospital and Government Maternity Hospital, Tirupati, Andhrapradesh.

SAMPLE SIZE

A total number of 210 clinically suspected cases of surgical site infections during July 2009 to April 2010 were included in this study.

SAMPLING PROCEDURE

A questionnaire was used to obtain data from the patient after obtaining an informed consent from the patient/ or care taker. Double wound swabs were aseptically obtained from each surgical site before the wound was cleaned using an antiseptic solution. The specimen was collected on sterile cotton swab without contaminating them with skin commensals. The samples were transported soon after being obtained. In the laboratory, the specimens were registered and macroscopically examined for color, odor and consistency. One of the swabs collected was used for preparation of direct smear and stained by Gram's stain. The smears were screened under oil immersion objective. Second swab was used for culture by streaking on blood agar and MacConkey agar and nutrient agar; plates were incubated at 37^{o}c over night. After incubation the plates were examined for bacterial growth. Further identification and confirmation of organisms was done by the standard identification tests.

ANTIBACTERIAL SUSCEPTIBILITY TESTING

Susceptibility testing was performed by Kirby-Bauer disk diffusion technique according to criteria set by CLSI 2009. The inoculums was prepared by picking parts of similar test organisms with a sterile wire loop and suspended in sterile normal saline. The density of suspension to be inoculated was determined by comparison with opacity standard on McFarland 0.5 Barium sulphate solution. The test organism was uniformly seeded over the Muller-Hinton agar (oxoid) surface and exposed to a concentration gradient of antibiotic diffusing from antibiotic-impregnated paper disk into the agar medium, and then incubated at 37^{o}c for 16-18 hours. Diameters of the zone of inhibition around disks were measured to the nearest millimeter using a ruler and classified as sensitive, intermediate and resistant according to the standardized table supplied by CLSI 2009.

The drugs tested for gram positive cocci were Penicillin G (10U), Ampicillin (10 µg), Oxacillin (1µg), Ciprofloxacin (5µg), Gentamicin (10µg), Amikacin (30µg), Clindamycin (2µg), Erythromycin (5µg), Novobiocin (30µg), Co-trimoxazole (1.25/23.75 µg) and Vancomycin (30µg). The drugs tested for Enterobacteriacea were Gentamicin (10µg,) Amikacin (30µg), Ampicillin (10 µg), Cefotaxime (30µg), Ceftriaxone (30µg), Ceftazidime (30µg), Ciprofloxacin (5µg), Co-trimoxazole (1.25/23.75 µg), Amoxycillin/clavulanate (20/10µg), Imipenam (10µg), Piperacillin/tazobactam (100/10µg) and Ticarcillin/clavulanic acid (75/10µg). The drugs tested for Pseudomonas aeruginosa were Gentamicin (10µg), Amikacin (30µg), Cefotaxime (30µg), Ceftriaxone (30µg), Ceftazidime (30µg), Ciprofloxacin (5µg), Co-trimoxazole (1.25/23.75 µg), Amoxycillin/clavulanate (20/10µg), Imipenam (10µg), Piperacillin/tazobactam (100/10µg), and Ticarcillin (75 µg).

STATISTICAL ANALYSIS

All the study data were enterd into the computer database using standard format, checked for errors and verified. Data maintained in the computer sheets were organised by SPSS version 17.0 software for Windows. Data will be peresented in aprropriate Table, Charts by caliculating of percentage, rate etc.

ETHICAL ISSUES

The study was approved and ethically cleared by the Research and Ethical Review Committee of Sri Venkateswara Medical College, Tirupati, Andhrapradesh. Written consent was obtained from each study participants and parents or care takers. All patient information was kept confidential.

RESULTS

A total number of 210 clinically suspected cases of post-operative wound infections were included in this study between July 2009 to April 2010. The ages of study groups ranged from ≤20 yrs to ≥ 60 yrs.

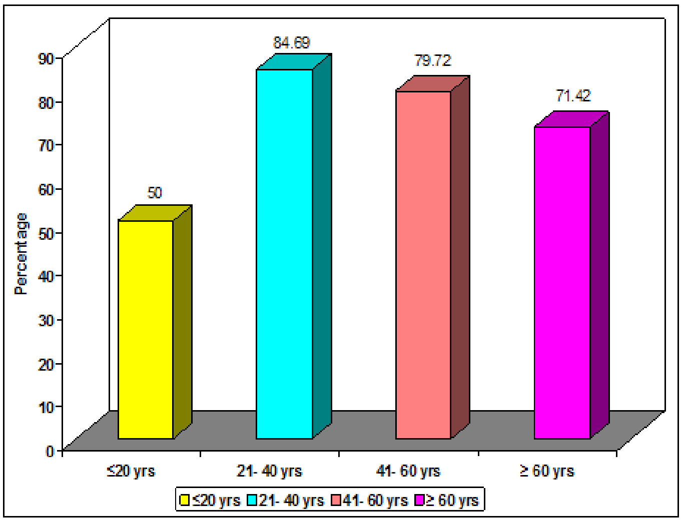

Chart-1: Age wise distribution of cases

In the present study, samples were collected from age group ranging from ≤20 yrs to ≥ 60 yrs and categorized in to four main age groups, <20 years, 21-40 years, 41-60 years and >60 years. Among age wise distribution of cases the maximum rate of isolation of pathogens was from 21-40 years (84.69%) followed by 41-60 years (79.72%), >60 years (71.42%) and <20 years (50%) (Chart-1).

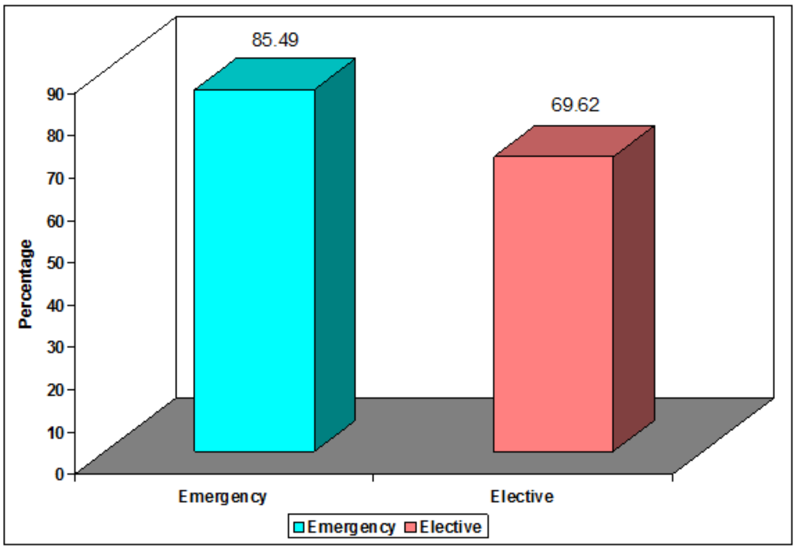

Chart-2: Effect of type of surgery on SSI

The maximum rate of isolation of pathogens was from emergency surgery (85.49%) than elective surgery (69.62%).

Table-1: Distributions of isolates in various surgeries

S. No	Surgery	Cases	Number of Positive Cases (167)	Rate of Isolation
1.	Emergency Lower Segment Caesarian Section (Em LSCS)	43	33	15.71%
2.	Hysterectomy	17	12	5.71%
3.	Family planning sterilization	11	6	2.85%
4.	Colorectal surgery	10	10	4.76%
5.	Herniorraphy	9	5	2.38%
6.	Appendicectomy	9	6	2.85%
7.	Small bowel surgery	32	26	12.38%
8.	Gastro duodenal surgery	19	15	7.14%
9.	Limb amputation	7	7	3.33%
10.	Open Reduction Internal Fixation (ORIF)	25	25	11.90%
11.	Arthroplasty	8	6	2.85%
12.	Split skin graft	10	4	1.9%
13.	I &D abscess drainage	5	5	2.38%
14.	Oncological surgeries	4	4	1.9%
15.	Tracheostomy	1	1	0.4%

Highest infection rate was observed in EmLSCS (15.71%) and lowest rate was in Tracheostomy surgeries (0.4%) (Table-1).

Total Number of Specimens - 210
Total Number of Isolates - 235

Table-2: Aerobic bacterial pathogens isolated from SSI cases

Organisms	Monomicrobial	Polymicrobial	Number of Isolates	Percent Among the Isolates
Staphylococcus aureus	52	17	69	29.36%
Staphylococcus epidermidis	6	9	15	6.3%
Enterococcus faecalis	2	4	6	2.55%
Klebsiella pneumoniae	9	34	43	18.2%
Escherichia coli	13	21	34	14.4%
Proteus vulgaris	0	1	1	0.42%
Proteus mirabilis	0	4	4	1.7%
Citrobacter koseri	1	5	6	2.55%
Citrobacter freundii	1	7	8	3.4%
Pseudomonas spp.	14	32	46	19.75%
Providencia rettgerii	0	1	1	0.42%
Serratia marcescens	0	1	1	0.42%
Enterobacter aerogenes	0	1	1	0.42%

A total of 235 bacterial isolates were obtained, 145 (61.7%) were aerobic gram negative bacilli. While 90 (39.29%) were aerobic gram positive cocci. Staphylococcus aureus was the predominant organism isolated 69 (29.36%), followed by Pseudomonas aeruginosa (19.75%), Klebsiella pneumoniae (18.2%), Eschericia coli (14.4%) (Table– 2).

Total Number of Isolates – 134

Table-3: Distributions of organisms in polymicrobial growth

S. No	Polymicrobial Growth	Cases	Percentage
1	Klebsiella pneumonie and Pseudomonas aeruginosa	17	25.3%
2	Klebsiella pneumoniae and Escherichia coli	11	16.4%
3	Staphylococcus aureus and Pseudomonas aeruginosa	9	13.4%
4	Staphylococcus aureus and Escherichia coli	5	7.4%
5	Staphylococcus epidermidis and Citrobactar koseri	5	7.4%
6	Citrobactar freundii and Pseudomonas aeruginosa	4	5.9%
7	K.pneumoniae and Proteus mirabilis	3	4.4%
8	Escherichia coli and Klebsiella pneumoniae and Staphylococcus epidermidis	2	2.9%
9	Escherichia coli and Pseudomonas aeruginosa	2	2.9%
10	Staphylococcus epidermidis and Serratia marcescens	1	1.4%
11	Staphylococcus epidermidis and Providencia rettgerii	1	1.4%
12	Staphylococcus aureus and Proteus vulgaris	1	1.4%
13	Enterobacter aerogens and Pseudomonas aeruginosa	1	1.4%
14	Enterococcus faecalis, Staphylococcus aureus and Staphylococcus epidermidis	1	1.4%
15	Enterococcus faecalis and Citrobactar freundii	1	1.4%
16	Enterococcus faecalis and Klebsiella pneumoniae	1	1.4
17	Enterococcus faecalis and Escherichia coli	1	1.4%
18	Staphylococcus aureus and Proteus mirabilis	1	1.4%

Among distribution of organisms in polymicrobial growth total number of isolates were 134 while Klecsiella pneumonia (25.3%) was commonest polymicrobial growth pattern seen followed by Eschericia coli and Klebsiella pneumonia (16.4%) (Table-3).

Table-4: Antibiotic sensitivity pattern of aerobic Gram positive cocci isolated from SSI cases

Antibiotic	Staphylococcus aureus		Staphylococcus epidermidis		Enterococcus faecalis	
	S %	R %	S %	R %	S %	R %
Penicillin	0	100	13.33	86.66	-	100
Erythromycin	76.81	23.19	73.33	26.67	66.66	33.34
Clindamycin	81.16	18.84	73.33	26.67	-	-
Amikacin	85.51	14.49	78.09	21.91	-	-
Gentamicin	63.77	36.23	76.64	23.36	71.85	28.15
Ciprofloxacin	31.88	68.12	19.91	80.09	-	-
Co-trimoxazole	50.72	49.27	18.89	81.11	-	-
Oxacillin	81.16	18.84	79.63	20.37	-	-
Vancomycin	100	0	-	-	100	-
Novobiocin	-	-	100	-	-	-

In our study all Gram positive cocci showed 100% susceptibility to Vancomycin. Followed by Amikacin (85.51%), Clindamycin and Oxacillin (81.16%), Erythromycin (76.81%), Gentamicin (63.77%) and the least effective drug was Ciprofloxacin. And 18.84% of the isolates of Staphylococcus aureus were resistant to Oxacillin. The Gram positive bacterial isolates were found to be 100% resistant towards Penicillin (Table-4).

Table-5: Antibiotic sensitivity pattern of aerobic Gram negative bacilli isolated from SSI cases.

Antibiotic	Klebsiella pneumoniae		Escherichia coli		Pseudomonas aeruginosa		Proteus spp.		Citrobacter spp.		Providencia rettgerii		Serratia narcescens	
	S %	R %	S %	R %	S %	R %	S %	R %	S %	R %	S %	R %	S %	R %
Ampicillin	9.3	90.7	5.88	94.12	-	-	0	100	14.29	85.71	0	100	0	100
Amikacin	95.35	4.65	91.18	8.82	82.61	17.39	100	0	85.71	14.29	0	100	100	0
Gentamicin	81.39	18.61	91.18	8.82	76.09	23.91	60	40	71.43	28.57	0	100	100	0
Cefotaxime	25.58	74.42	17.65	82.35	23.91	76.09	20	80	14.29	85.71	0	100	0	100
Ceftriaxone	32.56	67.44	26.47	73.52	26.09	73.91	80	20	21.43	78.57	100	0	0	100
Ceftazidime	23.25	76.75	23.53	76.47	67.39	32.61	40	60	21.43	78.57	0	100	0	100
Ciprofloxacin	46.51	53.49	20.59	79.41	28.26	71.74	0	100	21.43	78.57	0	100	0	100
Co-rimoxazole	46.51	53.49	32.35	67.65	13.04	86.96	0	100	28.57	71.43	100	0	100	0
Amoxyclav	9.3	90.7	20.59	79.41	4.35	95.65	0	100	14.29	85.71	0	100	0	100
Piperacillin/ tazobactam	76.74	23.26	70.58	29.42	80.43	19.57	100	0	71.43	28.57	100	0	100	0
Ticarcillin/ clavulanate	20.93	79.07	29.41	70.59	-	-	-	-	57.14	42,86	100	0	0	100
Ticarcillin	-	-	-	-	76.09	23.91	80	20	-	-	-	-	-	-
Imipenam	100	0	100	0	100	100	100	0	100	0	100	0	100	0

Among all Gram negative isolates showed 100% susceptibility to Imipenam. Klebsiella pneumoniae showed 95.35% Sensitivity to Amikacin, Escherichia coli showed 91.18% sensitivity to Amikacin, Pseudomonas aeruginosa Showed 100% sensitivity to Imipenem & (82.61%) sensitivity to Amikacin and Citrobacter species showed 85.71% sensitivity to Amikacin.

DISCUSSION

Surgical site infections or post-operative wound infections have been found to pose a major problem in the field of surgery for a long time. The surveillance of nosocomial infections with an emphasis on antimicrobial audit will reduce the risk of surgical site infections [7]. Despite the introduction of the meticulous antiseptic regimen in surgical practice, surgical site infections do occur in the patients and a number of exogenous and endogenous factors play an important role in the occurrence of the infections [8].

Data from the past several years show an increasing resistance for drugs that were considered as the first line of treatment for surgical site infections [9]. There is a need to reduce the cost which is potentially associated with antibiotic misuse viz. use of high-end antibiotics, multiple antibiotic / irrational combinations and the prolonged duration of treatment. Optimal antimicrobial prophylaxis in the appropriate dose, time and duration, which has been selected on the basis of the antimicrobial susceptibility pattern of the most common isolates in the hospital, would ensure a decreased rate of surgical site infections [10-12].

Similarly, there was statistically high infection rate in emergency surgeries as compared to the elective ones. Similar observations have been made by others Renvall *et al.*, [13] and Gil Egea *et al.*, [14] who reported a higher incidence in patients requiring emergency operations.in emergency surgeries, a combination of various factors like physical condition of the patient, short time interval for preparing the case for operation and lacking of rigorous aseptic measures due to urgency may predispose the individual to the infection.

The preoperative prophylactic antibiotic significantly prevents the post-operative wound infections or SSI. However, the use of antibiotics in the preoperative period may destroy susceptible organisms and permit colonization with resistant virulent organisms [15]. To be more effective a manner that ensures substantial tissue level at the time of incision and should target the pathogens commonly associated with specific operation undertaken [16].

Amongst different kinds of surgery, the maximum rate of isolation of pathogens was seen in EmLSCS (15.71%) and small bowel surgery (12.38%). Among gastrointestinal surgeries highest incidence of

SSI was seen in small bowel surgeries (12.38%), followed by gastro duodenal surgeries (7.14%).This finding corresponded with the study of Suchitra and Lakshmidevi *et al*., [17] who reported an increased incidence in EmLSCS, hysterectomy, gastrointestinal surgeries.

The association of risk factors and medical illness in the causation of SSI was also studied. A higher incidence of SSI was seen in surgeries with pre-operative stay >7days (31.9%), followed by the time of local preparation of >12 hours (26.19%), the presence of drains (21.9%), smoking (21.42%), alcohol (19.72%), presence of septic focus(10%) and diabetes mellitus (7.6%). This finding was correlated with the work carried out by others. Kowli *et al*., [18] reported an infection rate of 17.4% and 71.4% with a pre-operative stay of 0-7 days and >21 days respectively. Suchitra and Lakshmidevi *et al*., [17] also reported an increased incidence of SSI in patients with pre-operative stay of >7days and in diabetic patients. Lilani *et al*., [19] reported an SSI rate of 22.41% in cases where drain was used than in non-drained wounds (3.03%). Nagachinta *et al*., [20], De SA LA, Sathe LJ, Bapat RD *et al*., [21] reported an infection of 36.6% in patients with septic focus. This is due to impaired host defenses in these patients and longer hospitalization for correction of underlying disease leads to increased risk of colonization by hospital strains of bacteria.

As indicated in many previous studies, Staphylococcus aureus was the most frequently isolated pathogenic bacteria from surgical site infections. Vancomycin (100%) can be the antibiotic of choice for the surgical prophylaxis of this organism. 81.16% of the isolates of Staphylococcus aureus was sensitive to oxacillin. This finding of sensitivity pattern was comparable with the studies of Lilani *et al*., [19] and Jyoti Sonawane *et al*., [22].

All Gram negative bacilli isolate showed 100% sensitivity to imipenem. *Klebsiella pneumoniae* showed 95.35% sensitivity to Amikacin followed by Gentamicin (81.39%) and Piperacillin / tazobactam (76.74%). *Escherichia coli* showed 91.18% sensitivity to Amikacin and Gentamicin, followed by Piperacillin / tazobactam (70.58%), Co-trimoxazole (32.35%), Ceftrioxone (26.47%) and Ceftazidime (23.53%).

Proteus species showed 100% sensitivity to Amikacin and Piperacillin / tazobactam followed by 80% sensitivity to Ceftrioxone and Ticarcilin / clavulanate (80%). *Citrobacter species* showed 85.71% sensitivity to Amikacin followed by Gentamicin and Piperacillin / tazobactam (71.43%) and Ticarcillin / clavulanate (57.14%). *Pseudomonas aeruginosa* showed 100% sensitivity to Imipenem, 82.61% sensitivity to Amikacin and 80.43% sensitivity to Piperacillin / tazobactam sensitivity.

Citrobacter spe cies showed 85.71% sensitivity to Amikacin followed by Gentamicin& Piperacillin-tazobactam (71.43%) and Ticarcillin/clavulanate (57.14%).

This pattern of antibiotic sensitivity was comparable with the studies of Giacometti *et al*., [23], Kiran Ruhil and Bharti Arora *et al*., [24].

CONCLUSION

There is an alarming increase of infections caused by antibiotic - resistant bacteria. Lack of uniform antibiotic policy and their indiscriminative use of antibiotics may have led to emergence of resistant bacterial strains. The knowledge of the susceptibility patterns of the bacterial strains in a hospital will guide the clinicians to choose appropriate and judicious antibiotics for surgical prophylaxis. Updating the antibiogram periodically will further reduce the rate of surgical site infections to a considerable extent.

In addition, regular antimicrobial susceptibility surveillance is essential for area wise monitoring of the resistance patterns. An effective national and state level antibiotic policy and draft guidelines should be introduced to preserve the effectiveness of antibiotics and for better patient management.

In conclusion extensive and exhaustive studies are needed to explore the various problems in the area of nosocomial infections. The use of antibiotics must be confirmed with antibiotic sensitivity testing of isolates to prevent the emergence of drug resistant strains. The battle for elimination of surgical site infections will continue and with adequate surveillance and with proper coordination of microbiologists, this battle would definitely be won.

COMPETING INTERESTS

The author(s) declare that they have no competing interests.

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