Original Article

Antimicrobial Susceptibility Patterns of Bacterial Isolates from Surgical Site Infections: An Experience from Two Tertiary Level Hospitals in Dhaka City, Bangladesh

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Abstract

Surgical site infection (SSI) is an infection created by an invasive surgical procedure within 30 days if no prosthetic in situ or 1 year if a prosthetic is implanted in the patient. It is one of the most critical post-operative complications worldwide. A cross-sectional, observational study was conducted, between July 2022 and June 2023, at Dhaka Medical College Hospital and Mugda Medical College Hospital in Dhaka, Bangladesh, to identify causative bacteria of surgical site infections and their antimicrobial susceptibility patterns. Wound infection with cellulitis, no drainage, and suture abscesses were excluded. Demographic and laboratory data of the patients were recorded. Pus samples or wound swabs of clinically suspected SSI cases sent to the Department of Microbiology of the respective hospitals and were inoculated and interpreted. The mean age of the participants was 35.3±14.4 years. 88(47.31%) were males and 98(52.69%) were females (male to female ratio was 1: 1.1). Among 186 study subjects, E. coli (28.6%) was the most common isolated organism followed by Pseudomonas spp. (17.9%), Klebsiella (17%), Acinetobacter (15.2%), Staph. aureas (9.8%). Isolated Gram-positive bacteria were resistant to amoxiclav (100%), azithromycin (90%) and all were sensitive to linezolid and vancomycin (100%). Isolated Gram-negative bacteria were resistant to cefixime (100%) followed by cefuroxime and levofloxacin (97%). Appropriate prophylactic therapy for any major surgeries reduces incidence of SSI, and thereby reduces morbidity, mortality and cost burden in patients.

Keywords: *Surgical site infection, bacterial isolates;antimicrobial resistance*

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INTRODUCTION

Surgical site infection (SSI) is an important healthcare associated infection (HAI) globally that arises following surgery which has an impact on patient morbidity and mortality[1]. Appearance of infection in surgical wound was known as post-operative surgical wound infection. 'Surgical site infection' term is used instead of 'surgical wound infection' by the Surgical Wound Infection Task Force in 1992 to include infections of organ or spaces deep in the skin and soft tissues[2]. It can be a) primary infection which originates during operation or b) secondary infection which occurs after the operation from sources in the ward or as a result of some complications[3]. SSI is defined as an infection that happens within 30 days of the operation or within 1 year of operation if an implant is left in place[4]. At least 5% of patients develop SSI after their surgical procedures[5]. SSI is a critically serious problem in postoperative complications which constitutes approximately 20% of all HIAs[6]. There are many advance techniques to prevent and control of infection, but SSIs still remain a burden on the healthcare system. It is the commonest healthcare associated infection (HAI) in the low- and middle-income countries (LMIC) like Bangladesh and second most in Europe and USA[7]. Globally, the rate of SSI is 2.5% to 41.9% [8]. The rate of SSI is 2.6% in the United States, in Brazil ranges from 1.4% to 38.8%, 1.6% in Germany[9] and 10 to 25% in India[10]. SSI is responsible for up to 16% of nosocomial infections in all hospitalized patients and 38% of all surgical patients[11]. In the low- and middle-income countries, more than 1 in 10 surgical patients develop SSI. The risk of SSI in LMICs is 3 to 5 times more than in the high income countries[12]. The incidence of hospital based postoperative infection in India varies from 10 to 25%. Postoperative patients develop more wound infections (49%) than preoperative patients (15.9%)[10]. SSIs can be characterized by various sign symptoms, such as pain, tenderness, warmth, erythema, swelling, drainage etc.[13]. There are various factors that increase the risk of SSI which can be classify as intrinsic factors (host/patient related) and extrinsic (operation related) factors[14]. Host factors including age, obesity, body weight, anemia, DM, HTN and other chronic diseases, concurrent infections at other sides of the body are very important. Regarding operation, duration of surgery, site and depth of the wound, surgery techniques are remarkable[15]. Most surgical site infections are caused by the contamination with patient's normal microbial flora during surgery or from an external source after surgery[5]. Various studies showed that both gram-positive and gram-negative bacteria are responsible for SSI[2]. Most common identified organisms are *Staphylococcus aureus, Streptococcus pyogens, Enterococci, Escherichia coli, Klebsiella pneumonia, Proteus species and Pseudomonas aeruginosa*[7]. According to a study conducted by Ramesh et al., *E. coli* (20.8%) is the most common organism followed by *S. aureus* (16.1%) for SSI. Some other studies also found *Staphylococcus aureus* is the most common organism for SSI[16]. Antimicrobial resistance (AMR) is making SSIs more difficult to treat. Although bacteria naturally evolve and develop resistance to antimicrobial agents, but misuse and overuse of antibiotics have increased this process dramatically[17]. It is a major challenge for surgeons to face the SSI caused by multidrug resistant bacteria[18]. The microorganism's resistance to at least one antibiotic of 3 or more than 3 different classes of antibiotics are considered as multidrug resistance (MDR)[19]. Other common resistant organisms are *Methicillin resistant S. aureus,* extended spectrum â-lactamase producing *Enterobacteriaceae*, *Pseudomonas aeruginosa, coagulase negative staphylococci* and vancomycin resistant *Enterococcus* are important[20]. Hence, we proposed this study to identify causative bacteria of surgical site infections and their susceptibility patterns. Information obtained from this study is expected to facilitate preventive measures by the surgical teams at respective institutions.

METHODS

This cross-sectional, observational study was conducted among 186 patients visited at Departments of Obstetrics & Gynaecology, Surgery and Orthopedics at Dhaka Medical College Hospital and Mugda Medical College Hospital, between June 2022 and July 2023. Patients above 18 years and operation done within last 30 days were included in the study. Wound infections with cellulitis or suture abscesses were excluded. Patients satisfying the inclusion criteria were enrolled to the study after obtaining informed written consent from the patients. Data were collected through a face-to-face short interview by using a previously designed questionnaire. Wound/pus samples were collected from patient with the help of a sterile swab stick. Normal saline was used to wash the wound prior to collection of pus to remove contaminating materials like debris, dried exudates and dressing residue. In case of dry wound, then the swab was moistened with sterile saline. Care was taken that the swab only came into contact with the wound and not the surrounding skin. Tip of the swab was applied to wound with gentle pressure and rotated between fingers in a zig-zag pattern across the entire wound. In case of a large wound at least 1 to 2 cm area was sampled and materials from both the wound bed and wound margin were collected. It was then transferred to the specimen container. The specimen was transported to the Microbiology laboratory of the respective institute for processing as soon as possible. The pus samples were inoculated onto the media immediately and were incubated at 35°C aerobically for 24 hours. The isolated organisms were recognized by standard methods after incubation, preliminary detection of bacteria was based on colony characteristics such as hemolysis on blood agar, pink color production for lactose fermentation and other physical characteristics of the colony. Gram-negative rods were recognized by performing a sequence of biochemical tests. Namely: oxidase, indole production, urease production, Simon's citrate agar and motility. Gram-positive cocci were recognized based on their hemolytic properties, gram reaction, catalase and coagulase test results[21]. Antibiotic susceptibility testing was done by the Kirby Bauer disc diffusion method and necessary data were collected according to the objectives of the study. Statistical analysis was carried out by using MS-Excel and Statistical Package for Social Science (SPSS) version 26.0 for Windows.

The study was approved by the National Research Ethics Committee of Bangladesh Medical Research Council (BMRC), Dhaka, Bangladesh.

RESULTS

The sociodemographic profile of these patients showed that the highest number of participants with SSI were found within the age range of 18-27 years (36.0%) and the mean age of the participants was 35.3±14.4 years. 88(47.31%) were males and 98(52.69%) were females (male to female ratio was 1: 1.1) (Table -I). The overall culture positive SSI rate on these patients was 62.9%. The culture positive rate was higher in males (71.6%) than females (55.1%); the difference was statistically significant (P=0.020). Of these, 10.26% (n=12) were Gram-positive and 89.74% (n=105) Gram-negative. Out of all the culture positives, the findings showed that *E. coli* (28.6%) was the most common isolated bacteria among patients with SSI followed by *Pseudomonas spp*. (17.9%) and *Klebsiella* (17.0%), *Acinetobacter* (15.2%), *Staphylococcus* spp. (9.8%), *Enterobacter* (7.1%), *Proteus* (3.5%) and *Streptococcus* spp. (0.9%) (Fig. 1). Polymicrobial SSI was experienced by 5(4.3%) study subjects, 2(40%) had *Pseudomonas* and *Klebsiella* (Fig. 2). Table-II shows the comparative distribution of bacterial isolates between two hospitals (DMCH vs. MuMCH). Gram-positive bacteria resistant to amoxiclav was (100%), azithromycin was (90%) followed by clindamycin was (86%). All isolates were (100%) sensitive to linezolid and vancomycin (Table-III). Gram-negative bacteria were resistant to cefixime (100%) followed by cefuroxime and levofloxacin (97%). 87% of the isolates were sensitive to colistin (Table-IV).

Table-I: *Age and gender distribution of the study participants (n=186)*

Variables	Culture Positive		Culture Negative		Total	
	Frequency	Percentage	Frequency	Percentage	Frequency	Percentage
Age group (in years)						
18-27	42	62.69	25	37.31	67	36.02
28-37	28	53.85	24	46.15	52	27.96
38-47	16	53.33	14	46.67	30	16.13
48-57	17	80.95	4	19.05	21	11.29
>57	14	87.5	2	12.5	16	8.60
Mean±SD			35.3±14.4 years			
Gender						
Male	63	71.59	25	28.41	88	47.31
Female	54	55.1	44	44.9	98	52.69

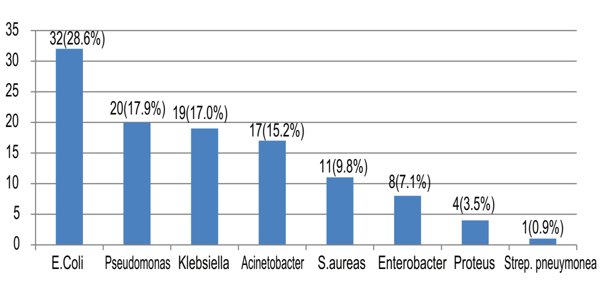

Fig. 1: *Monomicrobial distribution of samples according to isolated bacteria (n=112)*

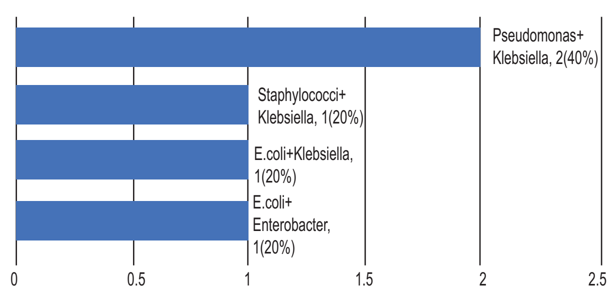

Fig. 2: *Polymicrobial distribution of samples according to isolated bacteria (n=5)*

Table-II: *Comparison of distribution of bacteria between two hospitals (DMCH vs. MuMCH)*

Bacteria	DMCH Frequency (Percentage)	MuMCH Frequency (Percentage)
S. aureas (n=11)	8 (73)	3 (27)
Strep. Pneumonea (n=1)	1 (100)	-
E. coli (n=32)	24 (75)	8 (25)
Pseudomonas (n=20)	15 (75)	5 (25)
Klebsiella (n=19)	14 (74)	5 (26)
Acinetobacter (n=17)	17 (100)	-
Enterobacter (n=8)	7 (88)	1 (12)
Proteus (n=4)	4 (100)	-

Table-III: *Antibiotic susceptibility pattern of Gram-positive isolated bacteria (n = 12)*

Antibiotics	Sensitive		Intermediate		Resistant	
	Frequency	Percentage	Frequency	Percentage	Frequency	Percentage
Amoxiclav	-	-	-	-	5/5	100
Azithromycin	1/10	10	-	-	9/10	90
Ciprofloxacin	3/8	37	-	-	5/8	63
Cloxacillin	2/5	40	-	-	3/5	60
Clindamycin	1/7	14	-	-	6/7	86
Doxycycline	6/10	60	-	-	4/10	40
Levofloxacin	1/2	50	-	-	1/2	50
Linezolid	12/12	100	-	-	-	-
Vancomycin	11/11	100	-	-	-	-

Table-IV: *Antibiotic susceptibility pattern of Gram-negative isolated bacteria (n= 105)*

Antibiotics	Sensitive		Intermediate		Resistant	
	Frequency	Percentage	Frequency	Percentage	Frequency	Percentage
Amikacin	51/94	54	1/94	1	42/94	45
Azithromycin	1/11	9	-	-	10/11	91
Aztreonam	4/35	11	1/35	3	30/35	86
Cefepime	3/31	10	-	-	28	31
Cefuroxime	1/33	3	-	-	32/33	97
Ceftazidime	9/98	9	4/98	4	85/98	87
Ceftriaxone	4/86	5	-	-	82/86	95
Cefixime	-	-	-	-	48/48	100
Ciprofloxacin	27/94	29	-	-	67/94	71
Colistin	75/86	87	3/86	4	8/86	9
Cotrimoxazole	15/74	20	1/74	1	58/74	79
Gentamicin	47/101	46	1/101	1	53/101	53
Imipenem	16/31	52	2/31	6	13/31	42
Levofloxacin	2/69	3	-	-	67/69	97
Meropenem	45/91	49	2/91	2	44/91	48
Piperacillin	33/86	38	4/86	4	49/86	57
Tigecycline	26/54	48	3/54	5	15/54	47

DISCUSSION

On evaluating the sociodemographic characteristics of the study subjects in this present study, 88 (47.3%) were males and 98 (52.6%) were females. Surgical site infection (SSI) rate is more in age group of 18 to 27 years. The culture positive rate was higher in males 71.6% than females 55.1%. The reasons behind these findings could involve factors like gender related physiological differences and potential variations in postoperative care or hygiene practices. A similar study reported that 52% were females and 63% were from the 15-64 yeers age group[22], which supports the finding of the present study.

In this study, the overall culture positive SSI rate was high. A similar study Gunne et al., revealed that 77.5% (62/80) patients had a positive culture[23]. This information contributes to understanding the microbial diversity and complexity of these surgical site infections[21]. The current study showed (10.3%) and (89.7%) of Gram-positive and Gram-negative bacteria respectively which is comparably similar with a study done on surgical site infections which reported (30.6%) Gram-positive and (69.4%) Gram-negative[20]. A higher rate of SSIs caused by Gram-negative bacteria, *E. coli* (28.6%) due to majority of specimens were collected from abdominal surgeries. During abdominal surgeries, that may cause spillage of gut flora, which is rich in Gram-negative bacteria[24]. Another study report showed that the most frequent causative organism for SSI was the *S. aureus* (72.6%) of all SSI isolates which dissimilar with this study[23].

Regarding the susceptibility patterns of Gram-positive bacteria, our study showed that the organisms varied in their susceptibility to all the antimicrobials used. Majority of them showed multi-drug resistant (resistance to two or more classes of antimicrobials). Rate of isolates resistant to amoxicillin, azithromycin followed by clindamycin and ciprofloxacin. All isolates were (100%) sensitive to linezolid and vancomycin. These findings might be because of bacteria naturally vary in their susceptibility to different antibiotics due to differences in their genetic makeup and biological properties. Some antibiotics might be effective against certain bacterial species while less effective against others. When bacteria are exposed to antibiotics over time, they can develop mechanisms to evade their effects. This can lead to the emergence of multi-drug resistant strains. Linezolid and vancomycin target specific mechanisms in bacterial cells that are less prone to resistance development. This could contribute to the higher sensitivity observed. The high resistance rates to certain antibiotics highlight the need for careful antibiotic stewardship to preserve the effectiveness of these crucial medications[25]. A study suggested that the susceptibility rates of *Streptococcus pneumoniae* and *Streptococcus pyogenes* for amoxicillin were (92%) and (100%), respectively. For *Hemophilus influenzae*, ampicillin susceptibility was 91% and amoxicillin/ clavulanic acid was 97%, respectively which is not in congruence with the present study and that dissimilarity might be due to race and geographical variation of the patients[26].

This study revealed that isolated Gram-negative bacteria are mostly resistant to cefixime, cefuroxime, levofloxacin followed by azithromycin. This resistance might be due to overuse and misuse of antibiotics, both in healthcare settings and in the environment, contribute significantly to the development of antibiotic resistance. The frequent exposure of bacteria to antibiotics creates selective pressure, favoring the survival and proliferation of resistant strains. Some bacteria naturally possess mechanisms that make them inherently resistant to specific antibiotics. This can be due to impermeable cell membranes, efflux pumps that expel antibiotics from the cell, or specific enzymes that neutralize the antibiotic's effects. Bacteria can acquire resistant genes from other bacteria through horizontal gene transfer. Resistance to one antibiotic can sometimes confer resistance to other antibiotics within the same class or with similar mechanisms of action due to cross resistance. Bacteria can undergo genetic mutations that provide them with a survival advantage against antibiotics. Over time, these mutations can accumulate, leading to higher levels of resistance[27]. A previous study showed that resistance rates against Gram negative bacteria were ampicillin/sulbactam, (92.6%); cefotaxime, (88.9%); ceftazidime, (74.1%); cefepime, (74.1%); gentamicin, (55.6%); piperacillin/ tazobactam, (48.1%); meropenem, (7.4%) and amikacin, (3.7%), which supports the findings of present study[28].

There are some limitations in our study. The sample size was relatively small, and data was collected from only two centers of Dhaka city. Due to unavailability, anaerobic culture could not be done.

CONCLUSION

Gram-negative bacteria were the most common pathogen isolated from infected surgical sites. Among them *E. Coli* was the predominant Gram-negative bacilli. A proper guideline regarding infection prevention practices and the selection of antibiotics for prophylaxis or empiric treatment may reduce the prevalence of SSI. Besides, continuous monitoring with fair use of antimicrobial agents is essential to prevent the multidrug resistance pattern.

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