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# Surgical Site Infection by Multidrug Resistant Organisms at a Tertiary Care Hospital in Dhaka City

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**Abstract**

**Background:** Surgical site infection (SSI) is one of the common causes of hospital borne infections. **Objective:** This study was performed to find out the proportion of multidrug resistant bacteria causing SSI. **Methodology:** This cross-sectional study was conducted among 99 hospitalized cases in the Department of Microbiology at, Bangladesh University of Health Sciences, Dhaka from July 2016 to June 2017. Ninety-nine bacteria were isolated from Pus samples collected aseptically from infected wounds from patients of BIHS general hospital. Isolation, identification and antibiotic sensitivity was done as per standard method. **Results:** Among the 99 organisms isolated, Gram negative bacteria were predominant (70.7%) than gram positive bacteria (29.3%). Among Gram positive bacteria (n=29), 31.0% were methicillin resistant. Among Gram negative bacteria (n=70), 7.1% isolates were ESBL, 78.6% AmpC β-lactamase and 12.3% were Carbapenemase producer. Higher generation antimicrobial agents like Meropenem and Imipenem are still effective against most of the organisms except carbapenemase producing ones. All Gram negative bacilli isolated were sensitive to colistin except *Proteus species.* **Conclusion:** In conclusion most of the isolates were multiply resistant to commonly prescribed antimicrobial agents. [*Bangladesh Journal of Infectious Diseases, December 2022;9(2):69-75*]

**Keywords:** Surgical site infection; antimicrobial susceptibility; multi-drug resistance.

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## 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 (e.g. prosthetic heart valve, hip prosthesis) is implanted[1]. The development of surgical site infection (SSI) is related to three factors. Firstly, the degree of microbial contamination of the wound during surgery; secondly, the duration of the procedure, and thirdly, host factors such as diabetes, malnutrition, obesity, immune suppression, advanced age and a number of underlying disease states[2].

The causative pathogen depends on the type of surgery; the most commonly isolated organisms are *Staphylococcus aureus (S. aureus)*, coagulase negative *Staphylococci* (CoNS), *Enterococcus species* and *Escherichia coli (E. coli)*[3]. The

incidence of surgical site infection (SSI) varies from hospital to hospital and also varies in different studies that have been reported from time to time[4]. The incidence of hospital-based postoperative infection varies from 10.0% cases to 25.0% cases in India[5]. According to a study conducted in Bangladesh, it was reported that among nosocomial infections, more than 50.0% were due to wound infection[6]. Postoperative wound infection delays recovery, increased hospital stays and may produce long lasting sequelae[7]. Despite efforts to control infection and better understanding of sepsis, wound infection is still a clinical problem and some infections in clean wounds still remain unexplained[8].

As a result of indiscriminate use of antimicrobial agents, significant changes occur in microbial genetic ecology, so spread of antimicrobial resistance is now a global problem[9]. Due to mutation and gaining plasmid, the bacteria are now becoming multidrug resistant. *Staphylococcus aureus,* due to gaining mec A or *mecC* gene, becoming resistant to Penicillinase resistant penicillin and other wide range of antibiotics[10]. Gram negative bacteria are producing different enzyme such as ESBL, AmpC β lactamase and carbapenemases[11].

The enzymes are responsible for resistance of the bacteria to a wide range of antibiotics. Studying the antibiotic susceptibility profile of SSI paves way to select the empirical antibiotic accordingly and thereby reducing the rate of SSI. The present study had been designed to find out the proportion of multidrug drug resistant bacteria isolated from surgical site infection in a tertiary care hospital. It would assist the clinicians in appropriate selection of antibiotics for prophylaxis and treatment.

**Methodology:**

**Study Settings and Population:** This cross-sectional study was conducted in the Departments of Microbiology of Bangladesh University of Health Sciences, Dhaka. This study was carried out during the period from July 2016 to June 2017 for duration of one year.

**Study Procedure:** Ninety-nine bacteria were isolated from Pus samples collected aseptically from infected wounds from patients of BIHS general hospital. Isolation, identification and antibiotic sensitivity was done as per standard method[12-17].

**Detection of ESBL:** For detection of ESBL positive organisms, double disk diffusion test (DDDT) was done along with routine sensitivity test[11,18]. AmpC β-lactamases was screened by decreased susceptibility to cefoxitin (30 µg) by disk diffusion test[11,19]. Carbapenemase was screened by disc diffusion test using decreased susceptibility to meropenem disc (10 µg)[11,20].

**Quality Control:** *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25922 was used as control organism[12].

**Statistical Analysis:** Statistical analyses was performed with SPSS software, versions 22.0 (IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp.). Continuous data that were normally distributed were summarized in terms of the mean, standard deviation, median, minimum, maximum and number of observations. Categorical or discrete data were summarized in terms of frequency counts and percentages. When values are missing, the denominator was stated. Chi-square test was used for comparison of categorical variables. Every effort was made to obtain missing data. A two-sided P value of less than 0.05 was considered to indicate statistical significance.

**Ethical Clearance**: All procedures of the present study were carried out in accordance with the principles for human investigations (i.e., Helsinki Declaration) and also with the ethical guidelines of the Institutional research ethics. Formal ethics approval was granted by the Bangladesh University of Health Sciences (BUHS). Participants in the study were informed about the procedure and purpose of the study and confidentiality of information provided. All participants consented willingly to be a part of the study during the data collection periods. All data were collected anonymously and analyzed using the coding system.

**Results**

Figure 1 revealed that among the 99 respondents, 34(34.3%) were male and 65(65.7%) were female where the mean age of them were 58.2(±10.9) and 50.7(±14.7) years respectively.

Out of 99 organisms studied, most of the bacteria were gram negative 70(70.7%) and about one third of the bacteria were gram positive 29(29.3%). Among the isolated gram positive bacteria (n=29), *Staphylococcus aureus* 21(72.4%) was predominant

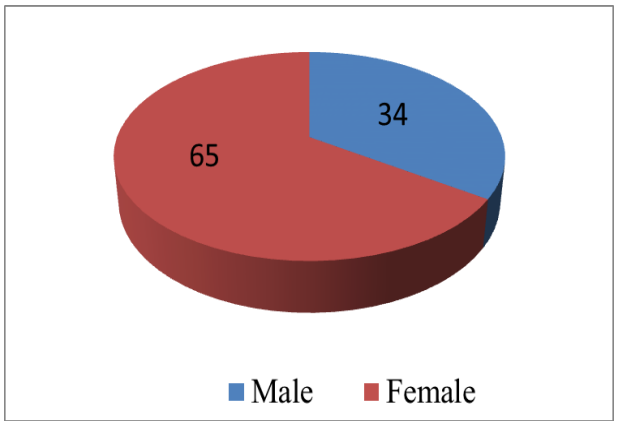


**Figure 1: Gender Distribution among the Study Population**

**Table 1: Distribution of Organisms in Study Population (n=99)**

| Name of Bacteria | Frequency | Percent |
|---|---|---|
| **Gram Positive Bacteria (n=29)** | | |
| *Staphylococcus aureus* | 21 | 72.4 |
| *Staphylococcus epidermidis* | 4 | 13.8 |
| *Enterococcus* species | 4 | 13.8 |
| **Gram Negative Bacteria (n=70)** | | |
| *Pseudomonas* species | 20 | 28.6 |
| *Proteus* species | 13 | 18.6 |
| *Klebsiella* species | 12 | 17.1 |
| *Escherichia coli* | 9 | 12.9 |
| *Enterobacter* species | 6 | 8.6 |
| *Citrobacter* species | 6 | 8.6 |
| *Acinetobacter* species | 4 | 5.7 |
| **Total** | **29** | **100** |

followed by *Staphylococcus epidermidis* in 4(13.8%) and *Enterococcus* species in 4(13.8%). Among the gram negative bacteria (n=70), *Pseudomonas* species was predominant which was 20(28.6%) followed by *Proteus* species in 13(18.6%), *Klebsiella* species in 12(17.1%), *Escherichia coli* in 9(12.9%), *Citrobacter* species in 6(8.6%), *Enterobacter* species in 6(8.6%) and *Acinetobacter* species in 4(5.7%) (Table 1).

In this study out of 29 Gram positive cocci 9(31%) isolates were multidrug resistant of which 7(33.3%) isolates were Methicillin Resistant (MRSA) *Staphylococcus aureus* and 2(50%) isolates were Methicillin Resistant *Staphylococcus epidermidis*. Neither VRSA (Vancomycin Resistant *Staphylococcus aureus*) nor VRE (Vancomycin Resistant Enterococci) was found (Table 2).

**Table 2: Proportion of Resistant Strain in Gram-Positive Organism**

| Gram-Positive Bacteria | Methicillin Resistant | Vancomycin Resistant |
|---|---|---|
| *S. aureus* (n=21) | 7(35.0%) | 0(0.0%) |
| *S. epidermidis* (n=4) | 2(50.0%) | 0(0.0%) |
| *Enterococcus* (n=4) | - | 0(0.0%) |
| **Total =29** | **9(31%)** | **0(0%)** |

Out of 70 gram negative Bacilli, 5(7.1%) isolates were ESBL positive, 55(78.6%) isolates were AmpC β-lactamase positive and 9(12.9%) were carbapenemases positive. Coproduction of two or three enzyme was also noted. ESBL was produced by *Escherichia coli* (33.3%), *Citrobacter* species (16.7%) and *Klebsiella pneumoniae* (8.3%). AMPC-BL was produced in varying degrees (54.0% to 100.0%) by all isolated organisms. Carbapenemases was produced by *Acinetobacter baumannii* (50.0%), *Klebsiella pneumoniae* (25.0%) and *Pseudomonas* species (20.0%). However, among these, only *Klebsiella* produced all the enzymes in varying degrees (Table 3).

**Table 3: Proportion of Multidrug Resistant Strains in Gram-Negative Bacteria**

| Gram-Negative Bacteria | ESBL (+ve) | AMPC-BL (+ve) | Carbapenemases (+ve) | Total |
|---|---|---|---|---|
| *Pseudomonas* species | 0(0.0%) | 19(95.0) | 4(20.0%) | 20 |
| *Proteus* species | 0(0.0%) | 7 (53.9%) | 0(0.0%) | 13 |
| *Klebsiella* species | 1(8.3%) | 9(75.0%) | 3(25.0%) | 12 |
| *Escherichia coli* | 3(33.3%) | 6(66.7%) | 0(0.0%) | 9 |
| *Enterobacter* species | 0(0.0%) | 6(100%) | 0(0.0%) | 6 |
| *Citrobacter* species | 1(16.7%) | 4(66.7%) | 0(0.0%) | 6 |
| *Acinetobacter* species | 0(0.0%) | 4(100%) | 2(50.0%) | 4 |
| **Total** | **5(7.1%)** | **55(78.6%)** | **9((12.9%)** | **70** |

**Table 4: Antibiogram of Multidrug Resistant Bacteria**

| Status of Antibiogram | Gram Positive Cocci | Gram negative bacilli | | |
|---|---|---|---|---|
| | **MRSA** | **ESBL +ve** | **AmpC BL+ve** | **Carbapenemase +ve** |
| 100% resistant to | Penicillin, Cephalosporin, Cefoxitin, tetracycline & Doxycycline, azithromycin, clindamycin, Penicillin | Penicillin, Cephalosporin (except Cefoxitin), Aztreonam | Ampicillins, Cephalosporin, Piperacillin / Tazobactam, Aztreonam | Ampicillin, Cephalosporin, Piperacillin/ Tazobactam. Aztreonam, quinolones, Carbapenem, |
| 100% sensitive to | Linezolid, Vancomycin, Carbapenem, Gentamicin & Netilmicin | Colistin, Carbapenem, Tigecycline, Cefoxitin, Gentamicin & Netilmicin, Quinolones. | Colistin, Carbapenem | Colistin |
| Partly sensitive to | Amikacin (80-100%); Quinolones (0-30%) | Amikacin (50 to 100%); cotrimoxazole, Piperacillin/ Tazobactam, Tetracycline & Doxycycline (0-33%), | Tigecycline (75-100%); Amikacin (50-100%); cotrimoxzole (14-25%); Quinolones (25-50%); Gentamicin & Netilmicin (25-100%), Tetracycline and Doxycycline (10-50%). | Tigecycline (33-100%); Amikacin, Cotrimoxazole, gentamicin and Netilmicin (25-50%); Tetracycline & Doxycycline (0-50%); |

Some bacteria were 100.0% isolates resistant to some antibiotics, 100.0% sensitive to some others and partially sensitive that means some percentage of bacteria are sensitive and others resistant to some antibiotics (Table 4).

## Discussion

Surgical site infection(SSI) is one of the common cause of hospital borne infections. This is responsible for delayed recovery, prolonged hospital stay, increased cost and may produce long lasting sequelae[6-7]. This study was done to find out the proportion of drug resistant bacteria in SSI's and thus to help clinicians to select the appropriate antibiotics. Ninety-nine patients were enrolled in the study. Among them 34(34.3%) were male and 65(65.7%) were female. Mean age of respondent was 58.2 (±10.9) and 50.7 (±14.7) years among male and females respectively. Other studies regarding SSI showed that the mean age of the respondents were between 20 to 30 years[21-23]. The higher mean age in this study might be due to the fact that mainly diabetic patients come to BIHS General Hospital for treatment. As in most cases, diabetes is type 2 variety which occurs in adults and elderly people. Predominance of female respondents was found in the study. This might be due to the fact that data were also collected from Gynecology and Obstetrics department where patient flow in this hospital is much more than surgery and orthopedics departments. This contrasts with study of Hope et al[24]. Out of 99 bacteria in this study, gram negative bacteria (70.7%), was predominant than gram positive bacteria which was 29(29.3%). This correlates with the study of Hope et al[24] where gram negative bacteria were 61(65.6%) isolates as compared to gram positive bacteria 32(34.4%). This also correlates with the study of Dessie et al[25] where gram negative and gram positive bacteria were

76(73.1%) and 28(26.9%) respectively. Among the isolated gram positive bacteria, *Staphylococcus aureus* (72.4%) was predominant followed by *Staphylococcus epidermidis* (13.8%) and *Enterococcus* species (13.8%). This correlates with the findings of Hope et al[24] where *Staphylococcus aureus*, CoNS and *Enterococci species* were 21.5%,7.5% and 5.4% respectively and Dessie et al[25] where proportion of *Staphylococcus aureus*, CoNS and Group B *Streptococci* was 18.3%, 3.8% and 4.8% respectively.

Among the gram negative organisms isolated, Pseudomonas species (28.6%) was predominant followed by *Proteus* species (18.6%), *Klebsiella* species *(*17.1%), *Escherichia coli* (12.9%), *Enterobacter* species (8.6%), *Citrobacter* species (8.6%) and *Acinetobacter* species (5.7%). Some of the organisms correlates with that of Hope et al[24] where the organisms were *Klebsiella* species (29.0%), *Proteus* species (11.8%), *Escherichia coli* (9.7%), *Enterobacter* species *(*3.2*%), Serratia* species (2.2%) and unidentified gram negative bacilli (9.7%). Dessie et al[25] also reported *Escherichia coli* 24 (23.1%), *Acinetobacter* species (22.1%), *Klebsiella pneumoniae* (9.6%), *Klebsiella ozaenae* (2.9%), *P. aeruginosa* (5.8%) and *Proteus vulgaris* (5.8%). At present Multidrug resistance is shown by organisms which undergo some mutation gaining new gene[10] providing with some structural change or gain plasmid due to which different enzymes[11] are produced rendering them multidrug resistant. In this study, multidrug resistant bacteria were found in gram positive cocci in the form of MRSA (Methicillin Resistant *Staphylococcus aureus*) or MRSS (Methicillin Resistant Staphylococcus species) and in gram negative bacilli in the form of ESBL (Extended Spectrum β-Lactamase) positive, AmpC β-Lactamase positive and carbapenemase positive in varying degrees. The present study found that 35.0% of *Staphylococcus aureus* were MRSA and 50.0% *Staphylococcus epidermidis* were MRSS. In study of Dessie et al[25] 10.5% (n=19) were MRSA and 100.0% (n=4) were MRSS. In the study of Hope et al[24], all (100.0%) 20 of *Staphylococcus* aureus were MRSA and 42.9% (n=7) of CoNS were methicillin resistant. Vancomycin *Staphylococcus aureus* (VRSA) and Vancomycin Resistant Enterococci (VRE) was not found in this study. Study of Dessle et al[25] and Hope et al[24] also did not find any VRSA or VRE. ESBL was produced by *Escherichia coli* (33.3%), *Citrobacter* species (16.7%) and *Klebsiella* species (8.3%) which was consistent with the result of Gajbhiye and Gajbhiye[28] where they showed that *Klebsiella sp*. (40.62%), *Escherichia coli* (35.89%), *Citrobacter* species (33.33*%), Proteus* species (26.08%) were ESBL producers.

AmpC β-lactamases (AmpC-BL) was produced in varying degrees (54.0 to 100.0%) by all isolated organisms in our study. Study of Gajbhiye & Gajbhiye[26] reported that *Klebsiella* species (17.18%), *Escherichia coli* (10.25%), *Proteus* species (11.11%) and *Citrobacter* species (8.69%) were AmpC producers. Kokate et al[27] found that *Citrobacter* species (33.33%), *Escherichia coli* (20.0%) and *Pseudomonas* species (17.64%) were AmpC producers. The results of present study found that carbapenemases was produced by *Acinetobacter* species (50.0%), *Klebsiella* species (25.0%) and *Pseudomonas* species (20%).

In the study of Kotb et al[28], out of 361 enterobacteriaceae (*Klebsiella* species, *Escherichia coli* and *Enterobacter* species) isolated from surgical wounds, 165(45.7%) were carbapenemases resistant. In this study some of the multidrug resistant bacteria are found 100% sensitive to some drugs, 100.0% resistant to others and still partially sensitive to some other drugs. Methicillin Resistant *Staphylococci* (MRSA /MRSS) are resistant to pencillinase resistant penicillin (e.g. methicillin) and also all extended spectrum penicillin, carbapenem and cephalosporin (except a new cephalosporin, ceftalorine)[12]. These bacteria can be treated with linezolid, vancomycin, daptom, Quinopristn-dalfopristin and ceftalorine[11,12]. This finding was also found in this study where MRSA/MRSS were 100.0% resistant to penicillin, cephalosporin, cefoxitin, tetracycline, doxycycline, azithromycin and clindamycin; 100.0% sensitive to linezolid, vancomycin, gentamicin and netilmicin; However, they were partially sensitive to amikacin (80.0% to 100.0%) and quinolones (0.0% to 30.0%). The ESBLs are able to hydrolyze the penicillin, narrow-spectrum and third-generation cephalosporin, and monobactams and are inhibited by β-lactamase inhibitors such as clavulanic acid, Tazobactam and so one[29-30] . This was also demonstrated in this study where ESBL positive Gram negative bacilli were 100% resistant to penicillin and cephalosporin (except cefoxitin) and were inhibited by clavulanic acid. In this study, ESBLs were 100.0% sensitive to Colistin, carbapenem, tigecycline, cefoxitin, gentamicin and netilmicin, quinolones and aztreonam. However, they are partially sensitive to amikacin (50.0% to 100.0%), cotrimoxazole, Piperacillin/Tazobactam, Tetracycline and doxycycline (0.0% to 33.0%).

AmpC β lactamases active on penicillin but even more active on cephalosporin and can hydrolyze

cephamycins such as cefoxitin and cefotetan; oxyimino-cephalosporin such as ceftazidime, cefotaxime, and ceftriaxone; and monobactams such as aztreonam and are not inhibited by β-lactamase inhibitors[29]. This was demonstrated in this study also where AmpC βL positive gram negative bacteria were 100% resistant to penicillin, Cephalosporin, piperacillin/tazobactam and aztreonam; 100% sensitive to colistin and carbapenem. But partially sensitive to Tigecycline (75.0% to 100.0%); amikacin (50.0% to 100.0%); cotrimoxzole (14.0% to 25.0%); quinolones (25.0 to 50.0%); Gentamicin & Netilmicin (25.0% to 100.0%), tetracycline and doxycycline (10.0% to 50.0%). Carbapenemases are diverse enzymes that vary in the ability to hydrolyze carbapenem and other β-lactams[31]. This was found in this study also. Carbapenemase producing gram negative bacteria were found 100% resistant to penicillin, cephalosporin, piperacillin/ tazobactam, aztreonam, quinolones and carbapenem; 100% sensitive to colistin. They are partially sensitive to Tigecycline (33-100%), amikacin, Cotrimoxazole, gentamicin, netilmicin (25.0% to 50.0%), tetracycline and doxycycline (0.0% to 50.0%); Antibiotic treatment should be started after culture sensitivity. This is urgently needed for proper treatment of the patients and also to prevent the spread of multidrug resistance to sensitive bacteria

## Conclusion

SSI's are being caused by both of gram positive and gram negative multidrug resistant bacteria. There is limited therapeutic option for these Multidrug resistant bacteria. Pathogens that produce carbapenemases along with an ESBL ± AmpC β-lactamases are particularly challenging for clinicians and are a major threat worldwide. Ceftriaxone, a third generation cephalosporin was found to be ineffective against most of the isolates. In contrast higher generation antimicrobial agents like Meropenem and Imipenem are still effective against most of the organisms except carbapenemases producing ones. All Gram negative bacilli isolated are sensitive to colistin except *Proteus* species. Antibiotic treatment should be started after culture sensitivity. This is urgently needed for proper treatment of the patients and also to prevent the spread of multidrug resistance to sensitive bacteria.

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**Conflict of Interest**
We declare that we have no conflict of interest.

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**Contribution to authors:**
Meherun Nesa, Shah Md. Zahurul Haque Asna : Conception and design, or design of the research; or the acquisition, analysis, or interpretation of data; conceptualized and designed the overall study. Meherun Nesa ,Una Jessica sarker: involved in data collection; Drafting the manuscript or revising it critically for important intellectual content. Meherun Nesa, Mousumi Karmaker: involved in data input and data cleaning. Meherun Nesa, Una Jessica sarker: conducted data analysis. Meherun Nesa, Shah Md.Zahrul Haque Asna: drafted the manuscript. All authors reviewed and approved the final manuscript.

**Data Availability**
Any questions regarding the availability of the study's supporti ng data should be addressed to the corresponding author, who can provide it upon justifiable request.

**Ethics Approval and Consent to Participate**
The Institutional Review Board granted the study ethical approval. Since this was a retrospective study, not every study participant provided formal informed consent. Each method followed the appropriate rules and regulations.

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