

MICROBIAL ETIOLOGY AND ANTIBIOTIC RESISTANCE IN SURGICAL SITE INFECTIONS: IMPLICATIONS FOR OBSERVATIONAL TREATMENT

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ABSTRACT

Background: Surgical site infections (SSIs) remain a major contributor to postoperative morbidity, prolonged hospitalization, and increased healthcare costs. Rising antimicrobial resistance (AMR) among causative pathogens further complicates treatment, underscoring the need for local microbiological surveillance to guide empirical therapy. **Objective:** To determine the microbial etiology and antimicrobial susceptibility patterns of pathogens isolated from SSIs in postoperative patients. **Study Design:** Observational cross-sectional study. **Setting:** Department of Pathology, Allied Hospital, Faisalabad, Pakistan. **Duration of Study:** July 2022 to December 2022. **Methods:** A total of 100 wound swabs and pus specimens were collected from adult patients in surgical and orthopedic wards with clinically suspected SSIs. Samples were processed using standard microbiological protocols, and isolates were identified through biochemical testing. Antimicrobial susceptibility was evaluated using the Kirby–Bauer disk diffusion method, following CLSI guidelines. Data were analyzed with SPSS version 17.0. **Results:** Of the 100 bacterial isolates, 45% were Gram-positive and 55% were Gram-negative organisms. *Staphylococcus aureus* was the most frequent pathogen (43%), followed by *Escherichia coli* (36%), *Pseudomonas* spp. (9%), *Proteus* spp. (4%), and *Klebsiella* spp. (3%). *S. aureus* demonstrated high resistance to penicillin (88%) and ampicillin (82%) but remained fully susceptible to vancomycin (100%) and highly sensitive to linezolid (98%) and amikacin (90%). Gram-negative isolates showed high resistance to ampicillin (96.7%) and cotrimoxazole (93.3%), while colistin (100%), amikacin (88.3%), and meropenem (81%) were the most effective agents. **Conclusion:** *S. aureus* and *E. coli* were the predominant pathogens in SSIs, both exhibiting high resistance to first-line antibiotics. Culture-guided therapy, reinforced infection control practices, and continuous antimicrobial surveillance are critical to improving SSI management and combating AMR in surgical settings.

Keywords: Surgical Site Infection, *Staphylococcus Aureus*, *Escherichia Coli*, Antimicrobial Resistance, Kirby–Bauer Method

INTRODUCTION

Surgical site infections (SSIs) represent a significant burden on healthcare systems worldwide, complicating surgical procedures and leading to increased morbidity, prolonged hospital stays, and higher healthcare costs (1). The prevalence of SSIs has been closely linked to the rise of antimicrobial resistance (AMR) among pathogens, complicating management strategies as traditional antibiotic therapies become less effective. This necessitates an urgent reevaluation of prophylactic and therapeutic approaches in surgical settings (2).

The microbial etiology of SSIs is diverse, with both gram-positive and gram-negative organisms commonly implicated. Notably, the prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) and other multidrug-resistant organisms continues to rise, complicating treatment options and prompting healthcare professionals to adopt more rigorous infection control measures (3, 4). Furthermore, the role of endogenous flora is critical, as it contributes significantly to the microbial landscape at surgical sites, leading to infections that are often difficult to predict and manage (5).

In response to the rising rates of SSIs and the subsequent increase in AMR, several studies emphasize the importance of antimicrobial prophylaxis tailored to local microbiological patterns. The specificity of antibiotic therapy, informed by microbial studies, has been shown

to improve treatment efficacy and minimize the development of resistance (2, 6, 7). Enhanced surgical techniques, including the use of novel antiseptic agents and materials, aim to suppress bacterial colonization (8).

While understanding microbial pathogens and their resistance patterns is vital for shaping effective treatment strategies, it needs to be contextualized within specific populations. For instance, studies have indicated unique challenges in regions like Pakistan, driven by local healthcare delivery systems, socioeconomic conditions, and public health infrastructure (9, 10). Recent research has shown that the incidence of SSIs following procedures such as cesarean sections remains notably high, often exacerbated by inadequate postoperative care and limited resources for infection control (11, 12). Cultural and systemic factors significantly influence the management of SSIs and adherence to guidelines for antibiotic prophylaxis (13).

The landscape of surgical site infections underscores the critical need for targeted surveillance and research in specific contexts. By harnessing local epidemiological data, evidence-based interventions can be developed to address the particular microbial threats faced by healthcare facilities, enhancing patient outcomes and contributing to efforts against the growing crisis of antimicrobial resistance.

METHODOLOGY

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This observational cross-sectional study was conducted in the Department of Pathology at Allied Hospital Faisalabad (AHF), Pakistan, over six months from July 2022 to December 2022. AHF is a tertiary care referral center serving both urban and rural populations from across Punjab province. The study involved postoperative patients admitted to adult surgical and orthopedic wards who presented with clinical evidence of surgical site infection (SSI). Surgical site infections were defined according to the Centers for Disease Control and Prevention (CDC) criteria as infections occurring within 30 days of surgery, involving either the incision or the deep tissues at the operation site. Patients were included if they had purulent drainage from the incision or drain site before wound cleansing. Pediatric patients from the Child ICU, patients from non-surgical wards, and outpatient postoperative cases were excluded from the study.

A total of 100 specimens, including wound swabs and pus samples, were collected using a convenience sampling technique. New patients meeting the inclusion criteria were enrolled daily until the desired sample size was achieved. Demographic and clinical information, such as gender, age, ward, and relevant history, was recorded on a pre-designed structured proforma. Specimens were collected aseptically during surgical wound dressing before the wound was cleaned with an antiseptic solution. Using sterile cotton swabs, samples were obtained directly from the infected site to minimize contamination with skin commensals. The samples were then placed in Amies transport medium and transported to the laboratory within 30 minutes.

Microbiological processing followed standard laboratory protocols. Smears were air-dried, heat-fixed, and Gram-stained to allow preliminary classification of organisms. Samples were inoculated on blood agar, MacConkey agar, nutrient agar, and fresh blood agar, and incubated aerobically at 37 °C for 24–48 hours. Colony morphology was noted, and representative colonies were subcultured for further testing. The identification of bacteria was confirmed through standard biochemical tests, including catalase, coagulase, oxidase, indole, citrate utilization, and triple sugar iron agar reactions. For suspected anaerobic bacteria, inoculation onto fresh blood agar with a metronidazole disc was performed, and the plate was incubated in an anaerobic environment at 37 °C for up to five days.

Antimicrobial susceptibility testing was performed using the Kirby–Bauer disk diffusion method in accordance with guidelines from the Clinical and Laboratory Standards Institute (CLSI). Bacterial suspensions were prepared in sterile peptone water and adjusted to match the 0.5 McFarland turbidity standard. Mueller–Hinton agar plates were uniformly inoculated, and appropriate antibiotic discs were applied. After incubation at 37 °C for 18–24 hours, inhibition zones were measured in millimeters. Methicillin resistance in *Staphylococcus aureus* was determined using cefoxitin (30 µg) discs. Results were interpreted as sensitive, intermediate, or resistant based on CLSI breakpoints.

Data were entered and analyzed using SPSS version 17.0 (SPSS Inc., Chicago, IL, USA). Categorical variables were summarized as frequencies and percentages, with results presented in tables and figures illustrating the distribution of isolates and their antimicrobial susceptibility patterns. As the study utilized routinely collected diagnostic samples without patient identifiers and did not involve experimental interventions, formal ethical approval was not required. All procedures adhered to institutional biosafety and infection control standards.

RESULTS

A total of 100 postoperative wound swabs and pus specimens were analyzed, comprising 57 (57%) male and 43 (43%) female patients. Among the bacterial isolates, 45% were Gram-positive and 55% were Gram-negative organisms.

The predominant pathogen was *Staphylococcus aureus* (43%), followed by *Escherichia coli* (36%), *Pseudomonas* spp. (9%), *Proteus* spp. (4%), *Klebsiella* spp. (3%), and miscellaneous species (15%). (Table 1)

Table 1. Distribution of Microorganisms Isolated from Surgical Site Infections (n = 100)

Microorganism	Frequency (n)	Percentage (%)
Gram-Positive (n=45)		
<i>Staphylococcus aureus</i>	43	43.0
Other Gram-positive spp.	2	2.0
Gram-Negative (n=55)		
<i>Escherichia coli</i>	36	36.0
<i>Pseudomonas</i> spp.	9	9.0
<i>Proteus</i> spp.	4	4.0
<i>Klebsiella</i> spp.	3	3.0
Miscellaneous species	15	15.0
Total	100	100.0

Antimicrobial susceptibility testing revealed distinct resistance patterns between Gram-positive and Gram-negative isolates. Gram-positive isolates (*S. aureus*) exhibited high resistance to penicillin (88%) and ampicillin (82%), with methicillin resistance present in a notable proportion of isolates. High susceptibility was observed for vancomycin (100%), linezolid (98%), and amikacin (90%). Gram-negative isolates showed high resistance to ampicillin (96.7%), cotrimoxazole (93.3%), and first-generation cephalosporins (>85%). The highest sensitivities were to colistin (100%), amikacin (88.3%), and meropenem (81%). (Table 2)

Table 2. Antimicrobial Susceptibility Pattern of Major Isolates from Surgical Site Infections (n = 100)

Antibiotic	<i>Staphylococcus aureus</i> (n=43) Sensitive %	Gram-Negative (n=55) Sensitive % ¹
Penicillin	12.0	5.0
Ampicillin	18.0	3.3
Methicillin/Cefoxitin	62.0	12.0
Vancomycin	100.0	0.0
Linezolid	98.0	0.0
Clindamycin	84.0	25.0
Ciprofloxacin	65.0	55.0
Amikacin	90.0	88.3
Meropenem	95.0	81.0
Colistin	100.0	100.0
Cotrimoxazole	22.0	6.7

¹Gram-negative percentage represents the mean sensitivity of *Escherichia coli*, *Pseudomonas* spp., and *Klebsiella* spp.

S. aureus was the predominant isolate, representing 43% of cases. Gram-negative organisms constituted a slightly higher proportion of the isolates (55%) than did Gram-positive organisms (45%). Colistin, vancomycin, and amikacin were the most effective antimicrobials across all isolates. Resistance to first-line empirical antibiotics (penicillin, ampicillin, cotrimoxazole) was high, underscoring the need for culture-guided therapy.

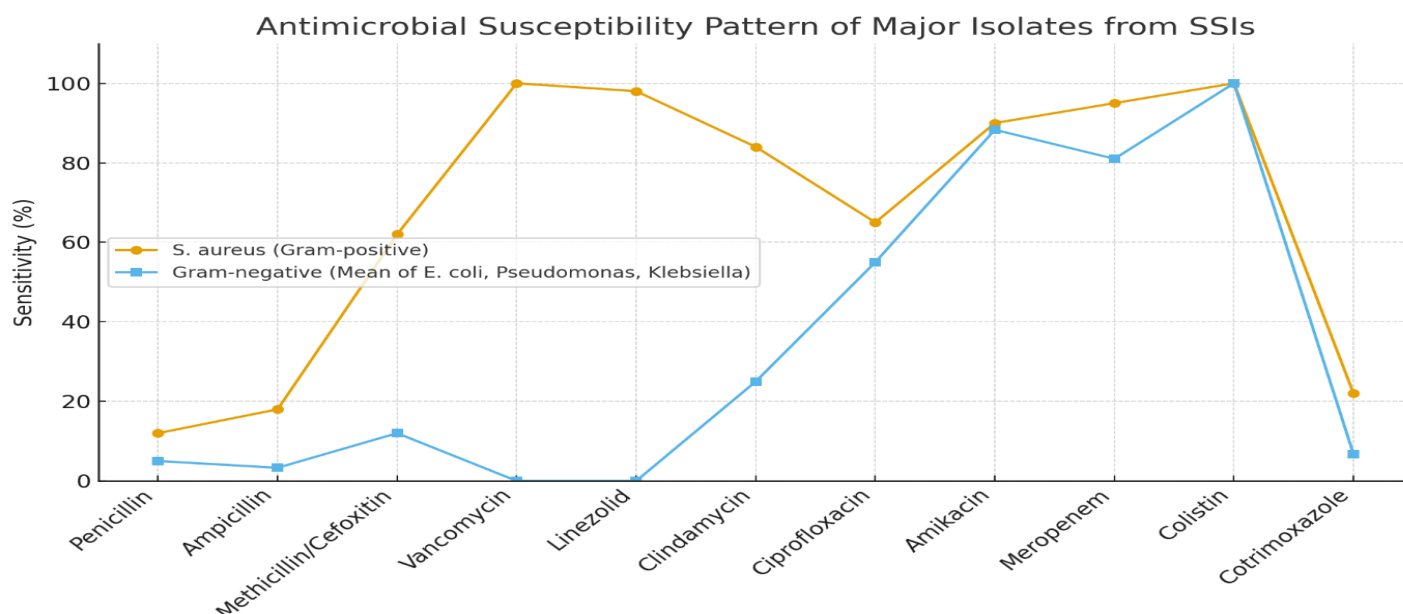


Figure 2. Antimicrobial susceptibility pattern of primary isolates.

DISCUSSION

The findings of this study provide valuable insights into the microbial etiology and antibiotic resistance patterns associated with surgical site infections (SSIs) in a diverse patient population. A total of 100 postoperative wound swabs and pus specimens were evaluated, revealing a higher prevalence of gram-negative organisms (55%) compared to gram-positive organisms (45%). *Staphylococcus aureus* was identified as the predominant pathogen (43%), followed by *Escherichia coli* (36%). This aligns with previous research conducted by Oba and Nuhu, which identified *S. aureus* as a leading cause of SSIs in a similar setting, affirming its clinical significance in postoperative complications (14).

In our study, we observed that the predominance of *S. aureus* aligns with findings from Amulio et al., who reported that *S. aureus* was the most prevalent organism in postoperative infections (15). Furthermore, our results align with those of Begum et al., who reported similar trends in pathogen prevalence in surgical site infections, underscoring the importance of *S. aureus* as a significant contributor to morbidity in surgical patients (16).

Moving to the resistance profiles, our study's finding regarding the high resistance rates of *S. aureus* to penicillin (88%) and ampicillin (82%) echoes the data presented by Nguyen et al., which highlighted similar resistance trends among isolates from clinical samples (17). The noteworthy methicillin resistance in our isolates, with 62% showing resistance, aligns with the alarming trend noted in other studies, including that of Troeman et al., where methicillin-resistant strains were implicated in SSIs, further complicating clinical management (18).

Additionally, the high susceptibility to vancomycin (100%) and linezolid (98%) in *S. aureus* isolates is consistent with results shown by Akhter et al., who reported comparable susceptibility patterns, suggesting that these antibiotics remain effective options for treating infections caused by *S. aureus* in our context (19). The high resistance of gram-negative isolates (96.7% to ampicillin and 93.3% to cotrimoxazole) found in our study is corroborated by Bassey et al., demonstrating similar resistance levels among gram-negative organisms in surgical settings (20). *Escherichia coli*'s prevalence in our study (36%) as a significant isolate is consistent with the findings

of a recent survey by Noor et al., which highlighted *E. coli* as a prominent pathogen in surgical site infections following laparotomy procedures (21). The gram-negative organisms in our results, including *Pseudomonas* spp. and *Klebsiella* spp., further underscore ongoing public health concerns, specifically due to their increasing resistance profiles and their emerging roles in complicated infections post-surgery.

In conclusion, our study highlights critical insights into the microbial and resistance patterns of pathogens responsible for surgical site infections in our cohort. The notable prevalence of *S. aureus* and *E. coli*, along with their respective resistance patterns to commonly used antibiotics, emphasizes the urgent need for continuous surveillance, tailoring of empirical treatment choices, and careful consideration of antibiotic stewardship to mitigate the rising tide of antimicrobial resistance.

In Pakistan, the limited resources and inadequate infection control measures in healthcare facilities may exacerbate the problem of SSIs, with the emergence of antibiotic resistance posing significant challenges (23). Given the high incidence of surgical site infections documented in this study, our findings underscore the urgent need for enhanced clinical protocols regarding antibiotic prophylaxis and treatment, as well as targeted educational programs to educate healthcare workers about infection prevention practices. This knowledge would not only inform better management strategies but also strategically guide antibiotic use to curtail the escalating issue of resistance prevalent in the local context.

CONCLUSION

This study highlights that *Staphylococcus aureus* and *Escherichia coli* remain the leading pathogens responsible for SSIs in our setting, with alarmingly high rates of resistance to first-line empirical antibiotics. The persistence of multidrug-resistant organisms, particularly MRSA and resistant Gram-negative bacilli, necessitates the integration of culture-based diagnosis into routine postoperative care. Vancomycin, linezolid, colistin, and amikacin demonstrated the highest efficacy and should be prioritized in empirical regimens only when strongly indicated to avoid the development of resistance. Implementation of robust infection prevention protocols, adherence to antibiotic

stewardship programs, and ongoing local epidemiological surveillance are essential to reducing SSI-related morbidity and combating the growing threat of AMR.

DECLARATIONS

Data Availability Statement

All data generated or analysed during the study are included in the manuscript.

Ethics approval and consent to participate

Approved by the department Concerned. (IRBEC)

Consent for publication

Approved

Funding

Not applicable

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTION

MUHAMMAD NOMAN

Conceived the study, collected data, performed initial analysis and prepared the first draft of the manuscript

LAIBA TARIQ

Assisted in data collection, literature review and manuscript editing

WARDA KHALID

Helped in data acquisition, statistical analysis and manuscript formatting

NAJID ALI

Contributed to methodology development, data organization and interpretation of results

SHAHZAIB

Contributed to literature search, referencing and manuscript proofreading

ALI HASSAN

Assisted in patient recruitment, data entry and results interpretation

MUHAMMAD HAROON

Supervised the study, validated results and approved the final version of the manuscript

All authors read and approved the final version of the manuscript.

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