ResearchArticle

Frequency of causative organisms in surgical site infection in clean surgical cases: a cross-sectional study

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Abstract

Background: Surgical site infection (SSI) remains a clinical problem even after clean surgery, and the causative pathogens vary between populations and geographical areas. This study determined the frequency of causative organisms in SSI after clean surgical procedures. Methods: A descriptive cross-sectional study was conducted at the Department of Surgery, Abbass Institute of Medical Sciences, Muzaffarabad, from 27 February to 26 August 2021. A total of 160 patients aged 20–60 years with SSI after appendectomy, herniorraphy or cholecystectomy were enrolled by non-probability consecutive sampling. Wound swabs or pus, and deep tissue where possible, were Gram-stained and cultured on blood agar and MacConkey agar, and isolates were identified by colony morphology and biochemical tests. Data wereanalysed in SPSS 25.0; the chi-square test was applied after stratification ($p \leq 0.05$). Results: Mean age was 42.17 ± 9.45 years; 56.87% of patients were female and 61.25% lived in rural areas. Escherichia coli was the most frequent isolate (62; 38.75%), followed by Pseudomonas spp. (38; 23.75%), Staphylococcus aureus (29; 18.13%), coagulase-negative staphylococci (CoNS; 19; 11.87%) and Klebsiella spp. (12; 7.50%). CoNS differed significantly by age ($p = 0.032$) and type of procedure ($p = 0.0007$), S. aureus by gender ($p = 0.007$), Pseudomonas spp. by BMI ($p = 0.012$) and Klebsiella spp. by place of living ($p = 0.007$). Conclusion: E. coli was the most common pathogen causing SSI after clean surgery in this setting. Identifying the causative organism before starting antimicrobial therapy is recommended.

Keywords:surgical site infection; Escherichia coli; bacterial pathogens; clean surgery; Staphylococcus aureus; Pseudomonas

1. Introduction

surgical procedure (Shah et al., 2017). It may involve the skin and subcutaneous tissue, the musculofascial layers, or an organ or cavity opened during surgery (Bagnall et al., 2009). A number of risk factors increase the risk of SSI, including obesity, advanced age, diabetes mellitus, malnutrition, prolonged preoperative stay, infection at a remote site, duration and technique of surgery, presence of drains, inappropriate antimicrobial prophylaxis, perioperative temperature and poor postoperative glycaemic control (Curcio et al., 2019).

Despite efforts to control infection and a better understanding of sepsis, wound infection is still a clinical problem, and some infections in clean wounds remain unexplained (Kasukurthy and Bathala, 2020). In many SSIs the responsible pathogens originate from the patient's endogenous flora, and the causative pathogen depends on the type of surgery; the most commonly isolated organisms are *Staphylococcus aureus*, coagulase-negative staphylococci (CoNS), *Enterococcus* spp. and *Escherichia coli* (Kochhal et al., 2019; Narula et al., 2020). Reported frequencies differ widely between studies. An Indian study found CoNS (40.0%) to be the most common isolate, followed by *S. aureus* (37.50%), *Pseudomonas aeruginosa* (20.0%), *Klebsiella pneumoniae* (14.29%) and *E. coli* (9.09%) (Narula et al., 2020), whereas another study reported *S. aureus* (22.10%), CoNS (16.30%), *E. coli* (13.90%) and *P. aeruginosa* (10.5%) (Bhave et al., 2016), and a third found *P. aeruginosa* (29.31%) followed by *E. coli* (25.86%) (Kokate et al., 2017).

Because the prevalence of different pathogens in SSI varies from population to population and with geographical background, international results may not apply to our community, the majority of which lives in rural areas. Local data

on the causative organisms can support early identification and the selection of a proper antibiotic, and so reduce morbidity in the community. This study therefore aimed to determine the frequency of different causative organisms in SSI in clean surgical cases in the surgical ward.

2. Materials and methods

2.1. Study design, setting and participants

This descriptive cross-sectional study was carried out in the Department of Surgery, Abbass Institute of Medical Sciences, Muzaffarabad, AJK, from 27 February 2021 to 26 August 2021. The sample size of 160 was calculated for a 95% confidence level and 4.5% margin of error, taking the percentage of E. coli as 9.09% (Narula et al., 2020). Patients were enrolled by non-probability consecutive sampling. Patients of both genders aged 20–60 years who presented to the surgical outpatient department with SSI after a clean procedure (appendectomy, herniorraphy or cholecystectomy) were included. Patients already receiving antibiotics, patients with chronic renal failure (history and serum creatinine > 1.5 mg/dl) and those with non-surgical wounds were excluded.

2.2. Operational definitions

SSI was defined as infection at the site of surgery within two weeks of the operation, affecting the skin, flesh or deeper tissues, and was assessed by the presence of any two of: erythema (superficial redness of the skin, clinically assessed), pain not relieved by analgesics (self-reported) and ooze at the operation site (green, yellow or cloudy fluid). Body mass index (BMI) was calculated as weight (kg, weighing machine) divided by height squared (m^2, measuring tape); BMI > 30 kg/m^2 was classified as obese and ≤ 30 kg/m^2 as non-obese. Diabetes mellitus (DM) was defined as known diabetes (fasting blood sugar > 110 mg/dl on two consecutive occasions) for the previous two years, on medication with controlled disease.

2.3. Data collection and microbiological methods

After approval from the institutional ethical review committee [approval number to be added] and informed consent from every patient, swabs from the wound or pus were collected, and deep tissue specimens were collected wherever possible. A smear was prepared and Gram-stained, and specimens were cultured on blood agar and MacConkey agar. Isolates were identified on the basis of colony morphology, Gram staining and biochemical reactions (coagulase, catalase and oxidase tests and API 20). Each sample was examined by a consultant pathologist with at least three years of post-fellowship experience. The organisms recorded were CoNS, S. aureus, Pseudomonas, Klebsiella and E. coli. Age, gender, type of procedure, BMI, place of living (rural/urban), DM and the isolated organism were recorded on a predesigned proforma.

2.4. Statistical analysis

Data wereanalysed with SPSS version 25.0. Mean and standard deviation were calculated for age and BMI; frequencies and percentages for gender, type of procedure, place of living, DM and each organism (present/absent). Results were stratified by age, gender, type of procedure, BMI, place of living and DM, and the chi-square test was applied post-stratification. A p-value ≤ 0.05 was considered significant.

3. Results and discussion

3.1. Characteristics of the patients

A total of 160 patients with SSI were studied. Age ranged from 20 to 60 years (mean 42.17 ± 9.45 years); 110 (68.75%) patients were aged 41–60 years. There were 69 (43.13%) males and 91 (56.87%) females (male-to-female ratio 1:1.3), and 98 (61.25%) patients were from rural areas. Mean BMI was 28.93 ± 3.07 kg/m^2, and 66 (41.25%) patients had a BMI > 30 kg/m^2. Cholecystectomy (77; 48.12%) and appendectomy (63; 39.38%) were the most frequent procedures, and 65 (40.63%) patients were diabetic (**Table 1**).

Table 1. Baseline characteristics of the patients (n = 160).

Variable	Category	No. of patients	%
Age (years)	20–40	50	31.25
	41–60	110	68.75
Gender	Male	69	43.13

	Female	91	56.87
Place of living	Rural	98	61.25
	Urban	62	38.75
BMI (kg/m²)	≤ 30	94	58.75
	> 30	66	41.25
Type of procedure	Appendectomy	63	39.38
	Herniorraphy	20	12.50
	Cholecystectomy	77	48.12
Diabetes mellitus	Yes	65	40.63
	No	95	59.37

Mean ± SD: age 42.17 ± 9.45 years; BMI 28.93 ± 3.07 kg/m².

3.2. Frequency of causative organisms

Gram-negative bacilli accounted for 112 (70.0%) and Gram-positive cocci for 48 (30.0%) of the isolates. *E. coli* was the most frequent organism (62; 38.75%), followed by *Pseudomonas* spp. (38; 23.75%), *S. aureus* (29; 18.13%), CoNS (19; 11.87%) and *Klebsiella* spp. (12; 7.50%) (**Table 2**, **Figure 1**).

Table 2. Frequency of causative organisms in surgical site infection after clean surgery (n = 160).

Causative organism	No. of patients	%
Escherichia coli	62	38.75
Pseudomonas spp.	38	23.75
Staphylococcus aureus	29	18.13
Coagulase-negative staphylococci	19	11.87
Klebsiella spp.	12	7.50
Total	160	100.0

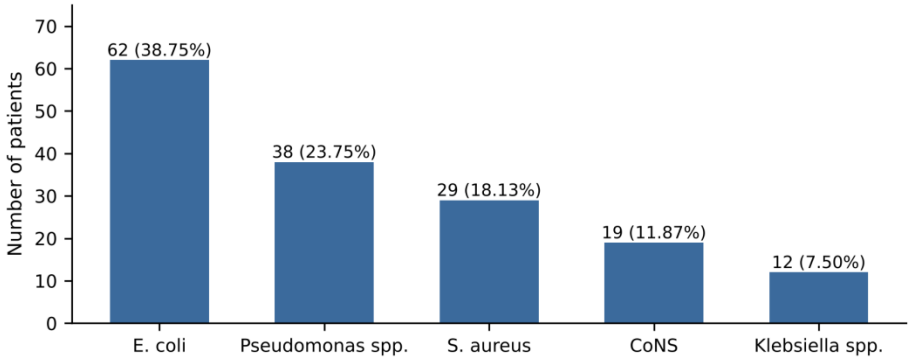


Figure 1. Frequency of causative organisms in surgical site infection after clean surgery (n = 160). CoNS, coagulase-negative staphylococci.

3.3. Stratification of organisms by patient characteristics

Stratified results are given in **Tables 3–8**. Most associations were not statistically significant, but five were: CoNS was more frequent in patients aged 20–40 years than in those aged 41–60 years (20.0% vs 8.18%; p = 0.032); *S. aureus* was more frequent in males than in females (27.54% vs 10.99%; p = 0.007); *Pseudomonas* spp. was more frequent in patients with BMI ≤ 30 kg/m² (30.85% vs 13.64%; p = 0.012); *Klebsiella* spp. was more frequent in urban than in rural patients (14.52% vs 3.06%; p = 0.007); and CoNS differed across procedures (herniorraphy 30.0%, appendectomy 17.46%, cholecystectomy 2.60%; p = 0.0007). Diabetes mellitus was not significantly associated with any organism (p = 0.869–0.951).

Table 3. Causative organisms (present) stratified by age, n (%).

Organism	20–40 y (n = 50)	41–60 y (n = 110)	p-value
E. coli	24 (48.0)	38 (34.55)	0.105
Pseudomonas spp.	15 (30.0)	23 (20.91)	0.210
S. aureus	8 (16.0)	21 (19.09)	0.638
Klebsiella spp.	4 (8.0)	8 (7.27)	0.871
CoNS	10 (20.0)	9 (8.18)	**0.032**

Table 4. Causative organisms (present) stratified by gender, n (%).

Organism	Male (n = 69)	Female (n = 91)	p-value
E. coli	25 (36.23)	37 (40.66)	0.569
Pseudomonas spp.	15 (21.74)	23 (25.27)	0.422
S. aureus	19 (27.54)	10 (10.99)	**0.007**
Klebsiella spp.	3 (4.35)	9 (11.11)	0.187
CoNS	7 (10.14)	12 (13.19)	0.556

Table 5. Causative organisms (present) stratified by BMI, n (%).

Organism	≤ 30 kg/m² (n = 94)	> 30 kg/m² (n = 66)	p-value
E. coli	33 (35.11)	29 (43.94)	0.259
Pseudomonas spp.	29 (30.85)	9 (13.64)	**0.012**
S. aureus	13 (13.83)	16 (24.24)	0.092
Klebsiella spp.	10 (10.64)	2 (3.03)	0.072
CoNS	9 (9.57)	10 (15.15)	0.283

Table 6. Causative organisms (present) stratified by place of living, n (%).

Organism	Rural (n = 98)	Urban (n = 62)	p-value
E. coli	41 (41.84)	21 (33.87)	0.314
Pseudomonas spp.	20 (20.41)	18 (29.03)	0.212
S. aureus	22 (22.45)	7 (11.29)	0.074
Klebsiella spp.	3 (3.06)	9 (14.52)	**0.007**
CoNS	12 (12.24)	7 (11.29)	0.856

Table 7. Causative organisms (present) stratified by type of procedure, n (%).

Organism	Appendectomy (n = 63)	Herniorraphy (n = 20)	Cholecystectomy (n = 77)	p-value
E. coli	28 (44.44)	6 (30.0)	28 (36.36)	0.429
Pseudomonas spp.	16 (25.40)	5 (25.0)	17 (22.08)	0.891
S. aureus	5 (7.94)	3 (15.0)	21 (27.27)	0.427
Klebsiella spp.	3 (4.76)	0 (0.0)	9 (11.69)	0.119
CoNS	11 (17.46)	6 (30.0)	2 (2.60)	**0.0007**

Table 8. Causative organisms (present) stratified by diabetes mellitus, n (%).

Organism	Diabetic (n = 65)	Non-diabetic (n = 95)	p-value
E. coli	25 (38.46)	37 (38.95)	0.951
Pseudomonas spp.	15 (23.08)	23 (24.21)	0.869
S. aureus	12 (18.46)	17 (17.89)	0.927
Klebsiella spp.	5 (7.69)	7 (7.37)	0.939
CoNS	8 (12.31)	11 (11.58)	0.889

CoNS, coagulase-negative staphylococci. Chi-square test; significant p-values (≤ 0.05) in bold.

3.4. Discussion

In this study *E. coli* was the leading cause of SSI after clean surgery (38.75%), followed by *Pseudomonas* spp. (23.75%). This differs from studies in which Gram-positive organisms predominated, such as the Indian reports of CoNS (40.0%) and *S. aureus* (37.50%) (Narula et al., 2020) or *S. aureus* (22.10%) (Bhave et al., 2016), and from the Pakistani study in which *S. aureus* (37.5%) was followed by *E. coli* (30.4%) (Malik, 2015). Our ranking is closer to that of Qaisar (2015), who reported *E. coli* (40.7%) followed by *P. aeruginosa* (26.31%) and *S. aureus* (19.73%), and to the series of Kokate et al. (2017), in which *P. aeruginosa* (29.31%) and *E. coli* (25.86%) led. In the Maharashtra study of Mundhada and Tenpe (2015), *S. aureus* (29%) was predominant, followed by *E. coli* (21%).

Differences in the distribution of isolates between settings may reflect diversity of the study population and local antimicrobial use, which favour the emergence of pathogens able to resist the antibiotics in current use. Another possible reason for the predominance of Gram-negative organisms is the type of surgery: appendectomy and cholecystectomy accounted for 140 (87.5%) of our patients, and Gram-negative organisms are predominantly reported in intra-abdominal procedures (Bibi et al., 2012). Other studies have likewise found E. coli to be the most common isolate from intestinal surgery (Chada et al., 2017). In the series of Akhi et al. (2015), Enterobacteriaceae (34.03%) were the predominant isolates, followed by P. aeruginosa (13.4%) and S. aureus (12.37%).

Several earlier studies from other countries reported a higher proportion of Gram-positive organisms, especially S. aureus, in SSI (Onche and Adedeji, 2010; Shriyan et al., 2010; Sani et al., 2012; Wassef et al., 2012), and S. aureus, CoNS and E. coli were also the organisms most prevalent in surgical wound infections according to the Centers for Disease Control. The pattern in our patients, in whom Gram-negative bacilli caused 70.0% of infections, therefore differs from part of the published literature and underlines that the distribution of pathogens depends on the population and setting studied.

From a clinical point of view, the identification of the causative microbe is beneficial to the patient and assists the selection of appropriate chemotherapy, which helps to avoid the emergence of multidrug-resistant organisms in hospital (Gelaw et al., 2013; Amare et al., 2011; Thu et al., 2006). The local frequencies reported here provide baseline data from a predominantly rural population (61.25% of our patients) that can inform routine practice guidelines for the early identification of the pathogen and the choice of antibiotic in SSI after clean surgery.

The stratified analysis indicates that the distribution of organisms was largely independent of age, gender, BMI, place of living, procedure and diabetes status, with the exceptions noted above. These significant findings arise from several subgroup comparisons, some with small numbers (for example 20 herniorraphy patients), and should be interpreted cautiously. This study has further limitations: it was conducted at a single centre with non-probability consecutive sampling, and antimicrobial susceptibility was not assessed, so resistance patterns among the isolates could not be described.

4. Conclusion

E. coli was the most common bacterial pathogen causing SSI after clean surgery in this setting (38.75%), followed by *Pseudomonas* spp. (23.75%), *S. aureus* (18.13%), CoNS (11.87%) and *Klebsiella* spp. (7.50%). We recommend that the causative organism be identified in these patients before any antimicrobial therapy is started, in order to reduce

resistance of pathogens to the most commonly used antimicrobial agents. Future multicentre studies including antimicrobial susceptibility testing are needed.

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