ORIGINAL ARTICLE

# Bacterial and Fungal Isolates from Surgical Site Infections in a Point Prevalence Survey for Healthcare-Associated Infections in Nasarawa State, Nigeria

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**Abstract**

**Background:** Surgical site infections (SSIs) are a group of healthcare-associated infections (HAIs) that pose a major public health challenge due to their contribution to increased morbidity, mortality, and increased cost of healthcare. Although bacteria are the usual agents associated with SSIs, fungi are now increasingly associated as well. Over 70% of bacteria associated with healthcare-associated infections are reported to be antimicrobial-resistant. Identifying these pathogens and their antimicrobial susceptibility is important in guiding therapy. This study aimed to determine the prevalence of surgical site infections, identify the associated bacterial and fungal pathogens, and determine their antimicrobial susceptibility patterns.

**Methodology:** The study was a point prevalence survey conducted among hospitalised patients in public health facilities across the State. The ECDC point prevalence survey protocol for HAIs was adopted for data collection. Data were collected from 208 eligible patients between February and March 2025. Wound swab samples were collected from six patients who met the case definition for SSIs. Samples were processed to isolate and identify bacterial and fungal pathogens and to determine their antimicrobial susceptibility patterns.

**Results:** The study found an SSI prevalence of 2.9%, constituting 33.3% of all HAIs among hospitalised patients in this study. Both bacteria and fungi were isolated from surgical sites, with *Escherichia coli* and *Aspergillus niger* being the most common bacteria and fungi, respectively. All the bacterial isolates were found to be resistant to several antimicrobial agents, while the fungal isolates had low zones of inhibition to the antifungal agents tested.

**Conclusion**: The co-occurrence of bacterial and fungal infections in SSI and their resistance to several antibiotics demonstrates the need to have a high index of suspicion for polymicrobial infections in surgical sites to effect appropriate treatment early and reduce the risks associated with HAIs in hospitalised patients

**Keywords**: Surgical Site Infections, Isolates, Antimicrobial Susceptibility.

## INTRODUCTION:

Surgical site infections (SSIs) are a group of healthcare-associated infections (HAIs) that pose a major public health challenge due to their contribution to increased morbidity, mortality, and increased cost of healthcare.[1] Surgical site infections refer to infections occurring in patients within 30 days of receiving healthcare or within a year of an implant being inserted.[2] Most reports show that SSIs are the most common HAIs globally, with the burden higher in developing countries. [3]Surgical site infections are estimated to constitute about 20% of all HAIs in the United States (US) and Europe and cost the US about $900 million and 400,000 extra hospital days annually. [4]Between 2 to 36% of patients who undergo surgery are reported to develop SSIs, depending on the type of procedure, with the highest rates in patients who have undergone orthopaedic procedures. [1] The impact of SSIs mirrors that of HAIs, as the effects are both on the patient and the health system. Surgical site infections,

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like other HAIs, increase morbidity, healthcare costs, the risk of hospital readmission, disability, and mortality.[2] They also increase healthcare expenditure, workforce demands, staff workload and burnout, antimicrobial use, and the risk of antimicrobial resistance.[1,5,6]

Although bacteria are the usual agents associated with SSIs, fungi are now increasingly associated as well.[7,8] A variety of pathogens have been associated with SSIs, the majority of them being of bacterial and fungal origin. The common bacteria associated with SSIs include Gram-positive organisms, especially members of the genus Staphylococcus: both methicillin-resistant (MRSA) and methicillin-sensitive (MSSA), as well as coagulase-negative Staphylococci. A review report indicates that Staphylococci are the second most common organisms associated with SSIs in Africa. [9] However, most reports suggest that Gram-negative bacilli are the most common cause, with *Escherichia coli* reported as the most common Gram-negative organism associated with SSIs. [10,11] Other common Gram-negative organisms associated with SSIs are *Pseudomonas aeruginosa*, *Proteus mirabilis*, and *Acinetobacter spp*. [11]

Over 70% of all bacteria associated with healthcare-associated infections are reported to be antimicrobial-resistant.[11] Antimicrobial resistance increases the chance of HAIs and other co-morbidities getting worse, and the worsening of the patient's condition leads to prolonged treatment and the need for more antimicrobial agents, predisposing to additional risk of development of resistance. [6]

The commonest fungal pathogens associated with SSIs are Candida spp.[12] Candida infection may occur alone or, in most cases, as a mixed infection with bacteria. [13] Candida is reported to have the ability to form biofilms, hence an important cause of device-associated SSI in patients with implants. [14]

Identifying these pathogens and their antimicrobial susceptibility patterns will guide antibiotic therapy and the implementation of infection prevention and control measures. This study, therefore, aimed to determine the prevalence of healthcare-associated surgical site infections, the associated bacterial and fungal pathogens, and their antimicrobial susceptibility pattern in Nasarawa State, Nigeria.

**MATERIALS AND METHODS:**

Study location and population: The study was conducted in Nasarawa State, North-central Nigeria, among hospitalised patients in public secondary and tertiary health facilities across the State's three senatorial zones.

Study design: The study was a cross-sectional study, using the point prevalence survey (PPS) design. The European Centre for Disease Control (ECDC) protocol for HAIs and Antibiotic Use was adopted for data collection.[15]

Sample size: The sample size was determined using the formula below for cross-sectional studies, where the variables are qualitative.[16]

$n = z^2 p (1 - p)/d^2$

Where:

n = sample size

z = standard normal variation at 5% ($p < 0.05$), which is 1.96

p = expected prevalence of HAIs in the population based on previous studies is 14.3%.[17,18]

d = precision level determined to be 0.05.

Therefore,

$n = 1.96^2 \times 0.143(1 - 0.143) / 0.05^2$

$n = 3.84 \times 0.12255 / 0.0025$

n = 188.4. If an attrition rate of 10%, (18.84), is added, this was 207.24, giving a minimum sample size of 207. A total of 208 patients from one tertiary and four secondary facilities were included, drawn from one of the two tertiary health facilities and four secondary health facilities across the three senatorial zones of the state.

Sampling method: Stratified random sampling method was used. Of the two tertiary facilities in the state, one was randomly selected. The secondary facilities were stratified into the three senatorial zones in the state. One facility was selected from each zone. When the sample size was not met, an additional facility was randomly selected from the remaining secondary facilities to make up the sample size.

The sampled secondary facilities were General Hospitals Doma, Nassarawa Eggon, Keffi and Obi. While the sampled tertiary facility was Dalhatu Araf Specialist Hospital (DASH), now Federal University Teaching Hospital (FUTH), Lafia.

Ethical consideration: Ethical approval was obtained from the State Health Research Ethics Committee (NHREC Protocol No: 18/06/2017) and the Health Research Ethics Committee of the tertiary health facility (FUTHREC/534). All data was handled confidentially. Consent was waived for data collection from patient's medical records. However, individual consent was sought and obtained from all patients who met the case definition for SSIs before wound swabs were collected.

Data were collected from Patients' medical records into a proforma. All data from the secondary facilities and each inpatient ward of the tertiary facility were collected on the same day.

Survey data collection:

Data was collected over two weeks between February and March 2025. All data from each secondary facility were collected on the same day, and in the tertiary facility, data from each ward were collected on the same day. In all selected health facilities, all hospitalised patients were included, except those admitted after 8 am on the survey date, those in Emergency wards, and those receiving same-day care. Data collection was carried out on working days in all the facilities. Patient data were extracted from patients' medical records (electronic and physical). The case definition for the different types of HAIs was the presence of signs and/or symptoms of the HAI type on the survey date, or a documented diagnosis by a physician, and/or microbiological confirmation. The case definition for surgical site infections (SSI) was based on diagnosis by a clinician, and/or pain, redness, swelling or warmth to touch, and/or purulent discharge or abscess, or spreading cellulitis at the surgical site, or Microbiological (MCB) confirmation.[19,20]

Sample collection and processing:

After obtaining consent from patients who met the case definition for SSIs, wound swab samples were collected from them. The wounds were cleaned with sterile saline, and a sterile cotton swab was gently rotated on the wound. Two swabs were collected from each patient and labelled

appropriately. The samples were transported to the Medical Microbiology Laboratory of the DASH (now FUTH), Lafia. One of the wound swab pairs was inoculated into Blood Agar and MacConkey Agar and incubated at 35-37°C for 24 hours. The second swab pairs were inoculated into Sabouraud Dextrose Agar and incubated at room temperature for up to 2 weeks. The bacterial isolates were identified using colonial morphology, Gram stain, and appropriate biochemical tests in line with standard protocols. [21] The fungal isolates were identified by their growth characteristics, colonial morphology, Germ tube test, and microscopic features in line with standard protocols.[21]

Antimicrobial susceptibility testing:
The Kirby-Bauer method of antimicrobial susceptibility testing was used for both the bacterial and fungal isolates. Mueller-Hinton Agar and modified Mueller-Hinton Agar were used for bacterial and fungal isolates, respectively, in line with standard methods.[22] The antibiotic discs used were Ceftazidime, Cefuroxime, Erythromycin, and Ceftriaxone at 30 µg, Levofloxacin, Amoxicillin, Augmentin, and Rifampicin at 20 µg, and Streptomycin, Gentamicin, Pefloxacin, and Azithromycin at 10 µg. The zones of inhibition were measured and interpreted in accordance with the Clinical and Laboratory Standards Institute (CLSI) interpretative charts.[23] Staphylococcus aureus CB008 and Escherichia coli NCTC CB002 were used as controls. The Antifungal discs used for fungal isolate susceptibility testing were Fluconazole (25 µg), Itraconazole (10 µg), Griseofulvin, and Nystatin. While the susceptibility tests against bacterial pathogens were interpreted in line with the CLSI guidelines, the antifungal susceptibility tests are only descriptive.

Statistical analysis
Data were entered and analysed using SPSS software. A descriptive statistical analysis was conducted for all variables. Categorical variables, i.e., age, sex, facility types, bacterial and fungal isolates, were presented as frequencies and percentages, while continuous variables were summarised using measures of central tendency and dispersion. The age of study participants was reported as mean and standard deviation (SD). Age was further categorised into age groups for analysis of SSI distribution. The overall prevalence of surgical site infection (SSI) was calculated as the number of patients with SSI divided by the total number of hospitalised patients surveyed. Antimicrobial susceptibility patterns for both bacterial and fungal isolates were reported as the number and percentage of isolates that were resistant, intermediate, or susceptible to each tested agent, based on CLSI/EUCAST breakpoints.

**RESULTS:** The study participants were aged from less than one to over 70 years with a mean age of 32 ± 19.8 years. The majority of them were from the tertiary health facility (73.1%) and female (59.6%) as shown in Table 1.

Five of the six patients who had SSI were males. Two patients with SSI each were aged between 21 and 30 years and 51 years and older, respectively, while the remaining two were less than 20 years old.

The overall prevalence of surgical site infection (SSI) among the 208 hospitalised patients in this point prevalence survey for healthcare-associated infections was 2.9%. Surgical site infections were the most common type of HAI, constituting 33.3% of all the HAIs. (Table 2)

Bacterial growth was detected in five (83.3%) of the six SSI specimens. *Escherichia coli* was the predominant isolate (3/6, 50.0%), followed by *Klebsiella pneumoniae* and *Staphylococcus aureus* (1/6, 16.7% each), Table 3. In addition, fungal growth was detected in four (66.7%) specimens. *Aspergillus niger* was the most frequently isolated fungus (2/6, 33.3%), while *Aspergillus fumigatus* and *Candida albicans* were each isolated from one specimen (16.7%). No fungal growth was observed in two specimens (33.3%).

**Antimicrobial susceptibility testing results**
Results of susceptibility testing of both the bacterial and fungal isolates to conventional antibacterial and antifungal agents showed a high level of resistance by all the isolates.

Antibacterial susceptibility testing
As shown in Table 4, all bacterial isolates exhibited multidrug resistance. All *Escherichia coli* isolates were resistant to Azithromycin, Erythromycin, Amoxicillin, Cefotaxime, Rifampicin, Ciprofloxacin, Cefuroxime, Gentamicin, Ceftazidime, Ceftriaxone, and Augmentin, while two were also resistant to Pefloxacin. *Klebsiella pneumoniae* demonstrated resistance to a similar range of antibiotics. The *Staphylococcus aureus* isolate was resistant to penicillins, aminoglycosides, and macrolides. However, all bacterial isolates remained susceptible to streptomycin. In addition, all isolates from the tertiary facility were susceptible to levofloxacin, whereas both isolates from the secondary facility were resistant.

Antifungal susceptibility testing
All fungal isolates exhibited inhibition zones ranging from 0 to 18 mm against the antifungal agents tested (Table 5). *Candida albicans* was resistant to all antifungal agents tested. *Aspergillus niger* showed inhibition zones of 10–18 mm with ketoconazole, Fluconazole, and Griseofulvin, whereas *Aspergillus fumigatus* was resistant to all agents except nystatin.

Table 1. Demographic and facility characteristics of study participants

| Characteristic | | Frequency | Percentage (%) |
|---|---|---|---|
| **Age (years)** | | | |
| **Mean age 32.3 ± 19.8 years** | | | |
| | 0 – 10 | 33 | 15.9 |
| | 11 – 20 | 25 | 12.0 |
| | 21 – 30 | 48 | 23.1 |
| | 31 – 40 | 38 | 18.3 |
| | 41 – 50 | 26 | 12.5 |
| | > 51 | 38 | 18.3 |
| **Gender** | | | |
| | Male | 84 | 40.4 |
| | Female | 124 | 59.6 |
| **Facility type** | | | |
| | Tertiary | 152 | 73.1 |
| | Secondary | 56 | 26.9 |

Table 2: Types of Healthcare -associated infections among study participants

| Characteristics | Frequency n (N=208) | Percentage (%) |
|---|---|---|
| Bloodstream Infections | 1 | 0.5 |
| Gastrointestinal infections (Diarrhoea) | 1 | 0.5 |
| Neonatal sepsis | 5 | 2.4 |
| **Surgical site infection** | **6** | **2.9** |
| Skin and soft tissue infections | 4 | 1.9 |
| Urinary Tract Infection | 1 | 0.5 |

Table 3: Bacterial and fungal isolates from Surgical Site Infections among the participants

| Isolates | n | Percentage |
|---|---|---|
| **Bacterial** | | |
| Escherichia coli | 3 | 50.0 |
| Klebsiella pneumoniae | 1 | 16.7 |
| Staphylococcus aureus | 1 | 16.7 |
| No bacterial growth | 1 | 16.7 |
| **Fungi** | | |
| Aspergillus niger | 2 | 33.3 |
| Aspergillus fumigatus | 1 | 16.7 |
| Candida albicans | 1 | 16.7 |
| No fungal growth | 2 | 33.3 |

Table 4: Antibacterial susceptibility patterns of bacterial isolates from SSI in Nasarawa state.

| Patient ID | Levofloxacin | Azithromycin | Erythromycin | Amoxicillin | Ciprofloxacin | Ceftazidime | Cefuroxime | Rifampicin | Streptomycin | Gentamicin | Pefloxacin | Ceftriaxone | Cefoxitin | Augmentin |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Zone of inhibition in millimeters | | | | | | | | | | | | | |
| **F1** | **26** | 0 | 0 | 0 | **28** | 0 | 0 | 0 | 18 | 0 | 0 | 0 | 9 | 0 |
| **F2** | **25** | 0 | 0 | 0 | **25** | 0 | 0 | 0 | 20 | 0 | **18** | 0 | 0 | 0 |
| **F3** | **24** | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 22 | 0 | 0 | 0 | 8 | 0 |
| **K1** | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 12 | 0 | 7 | 0 | 0 | 0 |
| **K2** | 0 | 12 | 0 | 0 | 0 | 0 | 0 | 0 | 23 | 0 | 19 | 0 | 0 | 0 |
| **CLSI (E)** | ≥17 | * | ≥23 | ≥19 | ≥21 | ≥21 | ≥16 | NA | ≥15 | ≥16 | ≥15 | ≥13 | * | ≥19 |
| **CLSI (S)** | ≥21 | * | ≥15 | ≥19 | ≥21 | ≥17 | * | ≥20 | NA | ≥16 | * | * | ≥21 | ≥25 |

*No official breakpoint F: Tertiary facility; K- Secondary facility

Keys: F1- *Escherichia coli*; F2- *Escherichia coli*; F3- *Klebsiella spp*; K1- *Escherichia coli*; K2- *Staphylococcus aureus*; S- Susceptible; I- Intermediate; R- Resistant; NA-Not applicable. CLSI (E)- standard zone for Enterobacteriaceae; CLSI (S)- standard zone for Staphylococci

Table 5: Antifungal susceptibility of fungal isolates from SSI in Nasarawa state.

| Patient ID | Isolate | Itraconazole | Nystatin | Ketoconazole | Fluconazole 25 µg |
|---|---|---|---|---|---|
| **F1** | *Aspergillus niger* | 12 | 18 | 0 | 0 |
| **F2** | *Aspergillus niger* | 15 | 0 | 0 | 0 |
| **F3** | *Aspergillus fumigatus* | 0 | 10 | 0 | 0 |
| **F4** | *Candida albicans* | 0 | 0 | 0 | 0 |
| **CLSI standard** | | ** | | | *≥19 |

*CLSI breakpoints for Candida albicans ** No CLSI breakpoints for disk diffusion for *Candida spp* and *Aspergillus spp*.[22]

## DISCUSSION

This study found the prevalence of surgical site infections to be 2.9% among hospitalised patients, representing 33.3% of all healthcare-associated infections. This estimate is comparable to the 3.7% previously reported by Ikrama *et al*.[24] and is consistent with the finding from another study that used similar methodology.[18] However, it is substantially lower than estimates reported in many other studies, including the 14.5% prevalence reported by Olowo-Okere *et al*. in a 2019 systematic review and meta-analysis.[25]

The study found the SSIs were polymicrobial, with both bacteria and fungi isolated from the surgical sites. *Escherichia coli* was the most prevalent bacterium, followed by *Staphylococcus aureus* and *Klebsiella pneumoniae.* This finding is consistent with a similar hospital-based study conducted in North-Central Nigeria.[24] However, the finding contrasts with other reports, which found the predominant SSI-associated bacteria to be *Staphylococcus aureus.*[26,27] Although Aspergillus species were the most common fungi isolated in this study, this differs from other reports in which Candida species were the predominant SSI-associated fungi. However, the small sample size limits the generalizability of this finding and may not accurately represent the true fungal profile of SSIs.[14,27]

All the bacterial and fungal isolates in this study displayed resistance to multiple antimicrobial agents. The bacterial isolates, including *Escherichia coli, Staphylococcus aureus,* and *Klebsiella pneumoniae,* all showed resistance to more

than three classes of antibiotics. This finding aligns with other reports in Nigeria and globally, where healthcare-associated pathogens are increasingly becoming multidrug-resistant.[28,29] However, our findings are limited by the small sample size, and the interpretation of the findings may not be a true representation of the antimicrobial susceptibility pattern of all pathogens associated with surgical site infections.

Although the fungal isolates in this study had low sensitivity to the antifungal agents tested, including the Imidazole, Triazoles, Polyenes, and Heterocyclic Benzofurans, this result does not necessarily indicate resistance because the method used for the antifungal susceptibility did not have validated CLSI disc diffusion breakpoints for the organisms and the antifungal agents tested.

Other reports, such as those by Ejembi *et al*[5] and Abban *et al*,[7] who previously reported increasing resistance by healthcare-associated fungal isolates. However, the antifungal susceptibility findings in this study should be interpreted with caution because of the small sample size and the limited range of antifungal agents tested. Although the high level of multidrug resistance observed among the bacterial isolates is concerning because of its potential impact on patient outcomes and healthcare costs, these findings require confirmation in larger studies to better characterize antimicrobial resistance patterns and inform strategies for improving the management of surgical site infections.

**Conclusion:** This study found an SSI prevalence of 2.9% in Nasarawa state, Nigeria from a point prevalence study. The co-occurrence of both bacteria and fungi in surgical sites is a finding that should influence decisions regarding the care and treatment of SSIs. While *Escherichia coli* and *Aspergillus species* were the most common bacterial and fungal pathogens in this study, all the bacterial isolates demonstrated multidrug resistance. There is a need for regular microbiological testing and guidance, and the need to strengthen antimicrobial stewardship programs in all public health facilities. Furthermore, the strengthening of infection prevention and control programs in public health facilities will reduce the burden of surgical site infections with their associated consequences.

**Limitations**: This study used a point prevalence design and may not fully represent pathogens associated with SSIs, as it excluded post-discharge infections and patients with short hospital stays. The small sample size for pathogen identification also limits the generalizability of the findings to Nasarawa State.**Conflict of interest**: The authors declare no conflict of interest.

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**Author's contributions:**
ESA conceived the study, developed the theoretical framework, implemented the research, and drafted the manuscript. AC, AAA, GAO, and ZA contributed to the study design and implementation. SOB, ZA, LYW, AAA, and MH contributed to the design, implementation, and data analysis. All authors discussed the findings and contributed to the final manuscript.

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