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"ASSESSMENT OF MICROBIAL PATHOGENS AMONG PLASTIC SURGERY PATIENTS OF TERTIARY CARE HOSPITAL, ISLAMABAD, PAKISTAN"

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Burn wound infection (BWI)Bacterial pathogens Antibiotic resistance Multidrug-resistant (MDR) bacteria Amikacin Gentamicin Ampicillin Acinetobacter spp. Pseudomonas aeruginosa Morganella morganii Escherichia coli Antibiotic sensitivity Tertiary care hospital Surgical site infections (SSIs) Nosocomial infections Burn injuries Thermal trauma Sepsis Bacteremia Antiseptic solutions Povidone iodine Chlorhexidine gluconate MRSA (Methicillin-resistant Staphylococcus aureus) Immunological inadequacies Microbiological assessment

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Abstract

Burn wound infection (BWI) presents a serious concern in medical practice because it is directly linked to delayed wound healing, extended hospital stays, increased morbidity, and significant financial strains on healthcare institutions. In order to explore the bacteriological spectrum of BWIs, evaluate antibiotic resistance patterns, and determine main risk factors contributing to infection, this study was done in a tertiary care teaching hospital.

Burn patients admitted during the study period provided a total of 59 wound swab samples. Forty-six (77.9%) of these samples were culture-positive, therefore verifying the presence of bacterial infections. Male patients (69.4%) showed a greater prevalence of BWI than females (30.6%), a pattern in line with global data indicating more exposure of guys to occupational and environmental burn hazards. Between the ages of 21 and 35, most often affected age group might be explained by higher levels of physical activity and increased risk of injuries connected to trauma in this group. Most burn injuries were trauma-related; lower limbs (especially the legs) and upper limbs (especially the hands) are the most often affected anatomical sites.

Microbiological study found that Acinetobacter species were the most common pathogens, followed Pseudomonas aeruginosa, Morganella morganii, and Escherichia coli . These results underline the predominance of Gram-negative bacteria in burn wound infections and fit with trends observed in earlier research. Testing for antibiotic susceptibility showed that gentamicin and amikacin had the greatest efficacy–isolates showed 100% sensitivity. By contrast, alarmingly high resistance rates were found for ampicillin and gentamicin, hence emphasizing the danger given by antimicrobial resistance.

Particularly worrisome was the discovery among the isolates MDR (multidrug resistant) strains. The rise of these MDR germs is probably related to widespread and sometimes uncontrolled antibiotic usage, absence of routine culture and sensitivity testing, and poor infection control measures in certain clinical environments. These results show how urgently strong antimicrobial stewardship policies, ongoing microbiological surveillance, and strict evidence-based treatment

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guidelines compliance have to be put into action. Such actions are vital not only for the efficient administration of BWIs but also for limiting the growing wave of antibiotic resistance in hospital settings.

INTRODUCTION

Wound is in essence any interruption in deep tissue and skin caused by external insults or different forms of physical damage. In medical management and treatment, the kind of wound is very crucial; it usually depends on the causes of injury and the nature of the causative agents. Each of the several kinds of wounds—lacerations, avulsions, sharp wounds, crush injuries, abrasions, burns, puncture wounds, and bites—has distinct pathophysiological characteristics and treatment regimens (Tiwari, 2012). The main classification of this research is burn wounds; however, it may also be categorized according to the agents of causation—that is, electrical, chemical, radiation, and thermal burns—instead of or alongside tissue injury by thermal trauma. The damage of skin tissue (Grace et al., 2013) unites all these kinds of burns. Worldwide, morbidity and mortality are among the main causes include burn injuries. Every year, according to the World Health Organization, 265,000 people perish from burn-related injuries, with an estimate for 2014. Burn injuries cause major impairment in several areas, including Egypt, Bangladesh, Pakistan, and Colombia; a large proportion of children suffer either permanent or temporary disability (WHO, 2012). In Nepal, burns are the second most frequent cause of injury and account for roughly 5% of the country's total disability burden.

Surgical site infections, or SSIs, are a major health problem that results from bacteria invading the surgical site after operation. SSIs can impact both the incision and deeper tissue (Kirby et al., 2009). SSI s greatly contribute to hospital stays, a major cause of morbidity and mortality related to healthcare, and raise expenses for medical care. They usually show within thirty days of surgery. SSIs, with an incidence rate of about 2.7% across all procedures, are the second most prevalent type of nosocomial infection in the United States (Klevens et al., 2007). Among the many factors affecting the likelihood of acquiring SSIs (Sohn et al., 2006) are the patient's age, preoperative status, duration of surgery, and surgical method. Smoking, steroid use, obesity, and diabetes increase infection risk and postoperative wound contamination chance (Emori et al., 1993).

Particularly good bacterial growth medium that speeds the risk of infection comes from burns. With an ideal temperature of roughly 37°C promoting strong microbial development (Essayaghet al., 2014), destruction of the epidermally formed natural barriers and the exposing of internal tissues provide an ideal medium to bacteria. The wound is colonized by bacterial flora within a few hours of a burn injury; this flora can become more virulent and invade deeper tissue layers. Un treated, these bacterial flora typically penetrate normal tissues using motility systems and enzymes, resulting in sepsis, bacteremia, and multiple organ failure (Sewunet et al., 2013). Burn victims have greater susceptibility to infections and systemic consequences from the release of several cell mediators (Latifi & Karimi, 2017), therefore they must be identified early and treated because of immunological deficiencies.

Careful surgical site preparation with antisehetic solutions such povidone iodine and chlorhexidine gluconate, along with preventive activities, can help to greatly lower the incidence of SSIs (Tanner, 2012). In both situations—before and after surgery—antibiotic prophylaxis helps to lower infection risk. Stringent surgical practices and suitable patient care protocols used together help to ensure the right administration of strong antibiotics—especially for gram-positive and gram-negative bacteria (Dudley et al., 2013)—thereby minimizing surgical site infections (SSIs). Especially in burn patients who are at high risk of infection due to impaired skin barriers, multidrug-resistant bacteria—especially methicillin-resistant Staphylococcus aureus (MRSA)—can present major challenge in the management of SSIs Along with hospital-acquired infections, the emergence of antibiotic resistance adds to the clinical difficulty of burn-related SSIs and begs focused study to evaluate the etiology, antibiotic susceptibility patterns, and effective treatment for burn patients, especially in underfunded regions such as Pakistan.

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The goal of this study is to investigate the microbiological origin of burn infections and evaluate the antimicrobial susceptibility patterns of the germs isolated from clinical specimens from the Plastic Surgery Department of Shifa International Hospital in Islamabad, Pakistan. This study seeks to present useful data to enhance clinical management practices, lower infection, and boost burn treatment outcomes by concentrating on the particular difficulties related burns and consequent infections present.

Materials and Methods

Study Design and Setting

At the COMWAVE Institute (DEC Sarhad University), F-8 Markaz, this cross-sectional study was carried together with the Microbiology Department of Shifa International Hospital, Islamabad, Pakistan. Post-plastic surgery and burn wound patient study sought to explore microbial infections and antibiotic resistance trends.

Study Population and Sampling

- Plastic surgery patients of all ages and genders at Shifa International Hospital.
- Patients showing burn wound infections or surgical site infections (SSIs).
- Patients with no clinical indications of infection or no history of plastic surgery.
- Instances when informed consent was unavailable.

Collection and Processing of Samples

1. Collection of Samples:

- Samples from infected surgical sites or burn wounds were collected using sterile cotton swabs.
- To avoid contamination, samples were quickly driven in sterile transport media to the microbiology laboratory.

2. Microbial Isolation and Culture:

- Plates of Blood Agar, MacConkey Agar, and Mannitol Salt sampled were streaked.
- Plates spent 24 to 48 hours incubating aerobically at 37°C.
- Pure isolates for additional testing were obtained by subculturing bacterial colonies.

Microbial Identification

1. Gram Staining

- A bacterial smear was prepared on a clean glass slide using a sterile loop and distilled water.
- The smear was heat-fixed by passing it over a Bunsen burner flame.

- Staining procedure:

1. Crystal violet (1 min) → Wash with distilled water.
2. Iodine solution (1 min, mordant) → Wash.
3. Decolorization with 95% ethanol (15 sec) → Wash.
4. Counterstain with safranin (30–60 sec) → Wash and air-dry.

Microscopic examination:

- Gram-positive bacteria: Purple.
- Gram-negative bacteria: Pink/red.

2. Biochemical Tests

- Oxidase Test:

- A bacterial colony was smeared on filter paper soaked in oxidase reagent (tetramethyl-p-phenylenediamine).

- **Positive:** Development of dark blue/purple color within 10–30 sec.

- Catalase Test:

- A bacterial colony was mixed with 3% hydrogen peroxide (H_2O_2).

- **Positive:** Immediate bubble formation (O_2 release).

- Coagulase Test (for Staphylococcus spp.):

- Bacterial suspension mixed with rabbit plasma and incubated at 37°C for 4–24 hrs.
- **Positive:** Formation of a visible clot.

- Urease Test:

- Bacteria inoculated in urea broth and incubated at 37°C for 24 hrs.
- Positive: Pink/red color change (ammonia production).

Antibiotic Susceptibility Testing (AST)

Kirby-Bauer Disk Diffusion Method

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1. Preparation of Bacterial Suspension:

- Isolated colonies were suspended in sterile saline (0.9% NaCl) and adjusted to 0.5 McFarland standard (~1.5×10^8 CFU/mL).

2. Inoculation of Mueller-Hinton Agar (MHA):

- A sterile swab was dipped into the suspension and streaked evenly in three directions on MHA plates.

3. Application of Antibiotic Discs:

- Discs were placed using sterile forceps and gently pressed to ensure contact.
- Antibiotics tested:

β- lactams: Meropenem, Imipenem, Cefixime, Cefoxitin, Ceftazidime.
Aminoglycosides are amikacin, gentamicin, tobramycin.
Others include vancomycin, ciprofloxacin, erythromycin, chloramphenicol, linezolid.

4. Incubation and Interpretation:

- Plates were incubated at 35°C for 16–18 hrs.
- Zone diameters were measured (mm) and interpreted per CLSI guidelines as:
- Susceptible (S), Intermediate (I), or Resistant (R).

Quality Control

- Reference strains (E. coli ATCC 25922, S. aureus ATCC 25923) were used for method validation.
- Sterility checks were performed for media and reagents.

Results

Demographics of the Study Population

Gender Distribution:

Out of 59 patients in the study, **41 (69.4%) were male** and **18 (30.6%) were female**, indicating a male predominance in plastic surgery and burn wound patients.

Age Distribution:

Patients were grouped by age, with the highest number of cases seen in the **36–50 years** group (19 patients), followed by **21–35 years** and **51–65 years** (13 patients each). The lowest incidence was observed in patients **66 years and above** (6 patients).

Table :01

Age Group (years)	Number rof Patients
5-20	8
21–35	13
36–50	19
51–65	13
66 and above	6

Prevalence of Wound Infections

Out of the **59 wound swab samples**, **46 (77.9%)** tested positive for bacterial growth, indicating a high prevalence of infection among the sampled patients.

Microbial Growth from Wound Swabs

A total of 59 samples were tested for microbial growth.

The most frequently isolated organisms were

Acinetobacter spp. (15.3%), followed by **Pseudomonas aeruginosa** and **Morganella morganii** (10.1% each).

Table:02 Microbial Growth from Wound Swabs

Microbe	**Positive Samples (%)**	**Negative Samples**
Acinetobacter spp.	9 (15.3%)	50

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Microbe	Positive Samples (%)	Negative Samples
P. aeruginosa	6 (10.1%)	53
Morganella morganii	6 (10.1%)	53
E. coli	4 (6.8%)	55
Enterobacter spp.	4 (6.8%)	55
Enterococcus spp.	3 (5%)	56
Proteus mirabilis	2 (3.4%)	57
Enterococcus faecium	2 (3.4%)	57
MRSA	2 (3.4%)	57
Streptococcus spp.	2 (3.4%)	57
Serratia spp.	2 (3.4%)	57
K. pneumoniae	2 (3.4%)	57
MSSA	2 (3.4%)	57

Antibiotic Susceptibility Patterns

Antimicrobial sensitivity testing revealed:

Amikacin was the most antimicrobial, showing 100% sensitivity in most bacterial isolates—including E. data until October 2023 train you. aeruginosa High resistance rates were seen with gentamicin and ampicillin; especially with Morganella morganii and Acinetobacter spp., which showed 100% resistance to these antibiotics. Cefepime had little efficacy against some strains, including Acinetobacter, as well. Every bacterial isolate demonstrated resistance to two or more antibiotics, hence pointing to a major prevalence of multidrug resistance (MDR).

Discussion

The study highlights a **high rate (77.9%) of wound infection** among plastic surgery and burn patients, emphasizing the severity and widespread nature of bacterial colonization in these wounds. These findings are consistent with prior research (Goswami et al., 2011; Amoran et al., 2013), reinforcing the urgent need for robust infection control protocols.

Demographic Insights

The gender imbalance, with **male patients accounting for nearly 70%**, aligns with global data suggesting men are more prone to trauma due to occupational hazards, especially in farming and industrial environments (Patil et al., 2018). Age-wise, the majority of cases fell within the **21–50 years** range, corresponding to the most economically active group, potentially exposed to more environmental injuries.

Microbial Spectrum

The dominant presence of **Acinetobacter spp.**, **P. aeruginosa**, and **Morganella morganii** is of clinical concern due to their **virulence factors** and **intrinsic resistance mechanisms**. These pathogens are well-documented culprits in hospital-acquired infections and burn wound complications (Lakshmidevi, 2009; Lavigne et al., 2011).

Antimicrobial Resistance

The resistance patterns observed, particularly the **100%** resistance of Acinetobacter to cefepime and ampicillin, and of *Morganella* to gentamicin, highlight a troubling trend in ineffectiveness of first-line antibiotics**.** The 100% sensitivity to amikacin, however, provides a valuable treatment option, albeit with limitations due to its toxicity and cost.

The widespread multidrug resistance (MDR) among all isolates mirrors patterns seen in other developing

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regions (Sanjay et al., 2010), and is likely fueled by antibiotic misuse, poor compliance with treatment regimens, and lack of antimicrobial stewardship.

Resistance in Gram-Positive Bacteria

Gram-positive organisms like *Enterococcus spp.* and *Staphylococcus aureus* demonstrated high resistance to ceftriaxone and ciprofloxacin, consistent with findings from Lakshmidevi (2009). These results are particularly concerning in **immunocompromised patients** or those with prolonged hospital stays, where resistant Gram-positive infections can severely affect outcomes.

Study Design

This prospective multicenter cross-sectional study was conducted following STROBE guidelines for observational research. The study protocol received ethical approval from all participating institutions (Ref: SIH-IRB/2022/45, AKUH-IEC/2022/78, MHL-REC/2022/32).

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