

Characterization of Bacterial isolates from Wound Infections

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ABSTRACT

INTRODUCTION: Wound infection is common across all sex and age groups which are responsible for significant human mortality and morbidity worldwide. Therefore, the objectives of this study were designed towards isolation and identification of bacteria involved in wound sepsis at Provincial hospital of Madhesh Pradesh, Nepal. **MATERIALS AND METHODS:** A total of 45 samples were collected from patients of different age groups of both male and female at provincial hospital, Madhesh Pradesh, Nepal. The samples were transferred immediately and processed in the microbiology laboratory at Model Multiple College, Janakpur, Nepal. Characterization and identification of the obtained bacteria was based on standard microbiological methods. **RESULTS:** Out of 45 samples 40 (88.89%) showed bacterial growth. Among the isolates *Staphylococcus aureus* were present in 15 (37.5%), *Pseudomonas aureogenosa* 7 (17.5%), *E. coli* in 9 (22.50%), and *Streptococcus pyogenes* in 9 (22.5%) of the total isolated samples. **CONCLUSIONS:** *Staphylococcus aureus* in wound infections was found to be dominant.

Keywords: Pus, Sepsis, Wound

INTRODUCTION

Wound infections have been regarded as the most common nosocomial infections and are associated with increased morbidity and mortality [1,2]. The intensity of wound infections may range from a simple self-healing to a severe and life threatening [3]. Tissue invasion by bacterial pathogens is determined by the location of wound [4]. Wound infections can be monomicrobial or polymicrobial [5]. The wound infection depends on a complex interaction between host factors like immunity, nutritional status and age, wound related factors like magnitude of trauma, dead space, devitalization presence of hematoma and microbial factors like toxins, invasion and resistance to antibiotics [6]. Most wound infections can be classified into two major categories, skin and soft tissue infections, although they often overlap as a

consequence of disease progression. Exogenous wound infection includes those associated with traumatic injury or decubitus pressure ulcer, animal or human bites, burns or foreign bodies in skin or mucous membrane. Endogenous wounds and abscess may be associated with appendicitis, cholecystitis, cellulitis, dental infection, septic arthritis, osteomyelitis, empyema, and sinusitis. Most of these processes are nosocomial contracted after invasive procedures, surgical manipulation, and placement of prosthesis [7]. Infections in a wound delays healing, causes wound breakdown, prolonged hospital stay, increased trauma care and treatment costs [8]. Many studies have shown that the common bacterial pathogens isolated from wound infections are *Staphylococcus aureus*, *S. epidermidis*, *S. pyogenes*, coagulase negative

staphylococci (CoNS), *Acinetobacter* spp., *Escherichia coli*, *Klebsiella* Spp., *Proteus* spp., *Enterobacter* spp., *Citrobacter* spp., and anaerobes such as *Clostridium* spp. and *peptostreptococcus* spp. [9,10]. Among all the species, *Staphylococcus aureus* has been found in a wide range of wounds dominant position. Patients with wound infections face additional exposure to microbial populations circulating in a hospital set up as the hospital environment is always charged with microbial pathogens. The control of wound infections has become more challenging due to widespread bacterial resistance to antibiotics and to a greater incidence of infections caused by methicillin-resistant *Staphylococcus aureus* (MRSA) and polymicrobial flora [10].

The importance of wound infections, in both economic and human terms should not be underestimated. Less availability of literatures reported on wound infections from Terai region is a topic of debate and key concern these days in the scientific community of Nepal. Therefore, the objective of this study was designed to isolate and identify bacteria from wound infection patients attending Provincial hospital of Madhesh Province, Nepal.

MATERIALS AND METHODS

Study design and setting

A hospital based descriptive cross sectional study was conducted in the month of April and May, 2021 from the sample collected from wound infection patients visiting at Provincial Hospital, Madhesh Province, Nepal.

Patients and procedures

A total of 45 samples (post-operative, abscess, bruise, burns) were collected from patients of different age groups of both male and female at Provincial hospital, Madhesh Province, Janakpurdham with the aid of sterile cotton swab stick which was rubbed in the area having wound for five seconds and kept in peptone water which was immediately transferred in the Microbiology laboratory of Model Multiple College,

Janakpurdham for the further processing and culture. The samples were streaked on plates of Nutrient agar, Blood agar, Mannitol salt agar and Mac-conkey agar and incubated for 24 to 48 hours at 35-37 degree celcius. After incubation, the bacterial colonies were observed and discrete colonies were picked and purified by sub-culturing on different agar plates and kept by proper labeling and were used for further characterization. Characterization and identification of the obtained bacteria was based on standard microbiological methods including Gram's staining, morphological and cultural characteristics on Nutrient agar, Macconkey agar and Manitol salt agar media, different biochemical tests like Catalase test, Coagulase test, Indole test, Motility test, Sub culturing on selective and differential media etc as per standard protocol following Monica Cheesebrough, 2002 [11].

Statistical analysis and data management

Data were entered into Microsoft excel for analysis. Frequencies and percentages were used to present the findings. Pie-chart was used to present distribution of samples showing growth pattern.

Ethical considerations

A written letter of study approval was obtained from Model Multiple College, Janakpurdham prior to study commencement. Informed consent was obtained from the patients about their participation explaining the objectives of the study.

RESULTS

Out of 45 samples, 40 (89.0%) samples showed growth of bacteria while 05 (11.0%) samples showed no growth on agar plates (Figure 1).

Bacteria identified were *Staphylococcus aureus* 15(37.5%), *Pseudomonas aeruginosa* 07(17.5%), *Escherichia coli* as 09(22.5%) and *Streptococcus* sps. as 09(22.5%). The dominance of *Staphylococcus aureus* in the wound samples was frequent which are the bacteria responsible for wound sepsis as shown in Table 1.

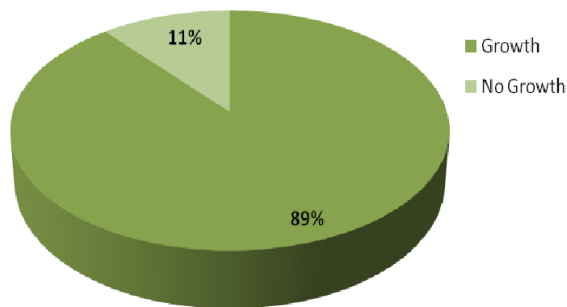


Figure 1 Distribution of samples showing growth

Bacteria	Number of isolates	Percentage
S. aureus	15	37.5
Streptococcus	09	22.5
Pseudomonas aeruginosa	07	17.5
E. coli	09	22.5
Total	40	100

Colony character	Catalase	Oxidase	Indole	Coagulase	Motility	Gram's Staining	Remarks
Yellow, white, smooth, spherical	Positive	Negative	Negative	Positive	Non-motile	Positive cocci in bunch	<i>Staphylococcus aureus</i>
Creamy white, mucoid,	Negative	Negative	Negative	Negative	Non-motile	Positive cocci in chain	<i>Streptococcus sps.</i>
Blue, green, smooth	Positive	Negative	Negative	Negative	Motile	Negative rod	<i>Pseudomonas aeruginosa</i>
Pinkish smooth, circular	Positive	Negative	Positive	Positive	Motile	Negative rod	<i>Escherichia coli</i>

Table 2 describes the characterization and identification of isolates from wound infections. Catalase test was positive for *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli* but for *Streptococcus* species it was found to be negative.

DISCUSSION

Wound infection plays an important role in the development of chronicity, delaying wound healing. The intensity of wound infections may range from a simple self-healing to a severe and life threatening [3]. Wound infection is a burning public health issue especially in developing countries. Severe wound infection can cause great loss including higher rate of morbidity and mortality; longer hospital stays, delay in wound healing, increase economic burden and increase discomfort which in turn increases disease burden significantly. Wound infection is being a common nosocomial infection which accounts for 0–80% of patient's mortality [12, 13]. Modernization in control and prevention of infections has not completely controlled wound infection due to increasing problem of antimicrobial resistance. So,

isolation and identification of bacteria in wound infections is a significant concern for treatment in a health care facility [14]. In present study in 40 (88.89%) samples showed growth of bacteria while 05 (11.11%) of samples showed no growth at all, this might be due to the therapeutic status of patients, condition of microbial growth, nature of growth media, type of species involved, insufficient incubation period. This study is comparable to the study by Shrestha P [15]. 72.4% showed bacterial growth. This might be due to similar culture medium used and growth environment condition for isolation of organisms. The present study concludes that *Staphylococcus aureus* in 15(37.5%), *Pseudomonas aeruginosa* in 07(17.5%) samples, *Escherichia coli* as 09(22.5%) and *Streptococcus sps.* in 09(22.5%). In similar type of study conducted on bacterial isolates from infected wounds the most common bacterial species detected was *Staphylococcus aureus* (37%), followed by *Pseudomonas aeruginosa* (17%), *Escherichia coli* (6%) [16] almost comparable to our study. Also, Upreti et al. [17] reported *Staphylococcus aureus* as most predominant bacteria followed by *Escherichia*

coli likely to our results and other bacterial isolates were coagulase negative staphylococci (7.8%), Acinetobacter spp. (5.2%), Klebsiella pneumoniae (5.2%), Pseudomonas aeruginosa (4.3%), Enterobacter spp. (4.3%), Citrobacter freundii (2.6%), Proteus vulgaris (1.6%) and P. mirabilis (0.9%) in pus samples of patients with wound infection visiting KIST Medical College Teaching Hospital, Kathmandu, Nepal. The predominance of S. aureus in wound infection is supported by different studies conducted by Parikh et al., Rajput et al., Gelaw et al., Goswami et al., [18-21]. As being a normal flora of human skin, it can get access into the wound easily. Another similar study reported that 82.5% of bacterial growth in pus samples and 13 different bacterial species were isolated where S. aureus was predominant (57.7%) species followed by E. coli (11%) and CoNS (3%) [22]. S. aureus was the most common bacteria (49%) found in wound infections followed by E. coli (25.9%), Klebsiella spp. (9.5%), P. aeruginosa (8.6%), Proteus spp. (4%) and Acinetobacter (2.7%) spp. S. aureus is the most common strain (25%) as a commensal organism of human skin and nasal passage in another similar type of study [23]. The previous results are about liable to our study although the sample size varies among different studies. Hence, most frequent isolation of S. aureus from pus specimens might also be due to contamination of collected specimens with skin normal flora [24]. The present study shows Staphylococcus aureus was the most predominant organism isolated from wound infection followed by Streptococcus spp., E. coli and Pseudomonas spp. In line with this study, similar

type of results was reported by Maharjan and Mahawal [25]. Nazeer et al. [26] found similar results in which the most predominant isolate from a wound infection was Staphylococcus aureus (37.12%) followed by Klebsiella species (20.45%). The study reported by Karkee P [27] also supported the finding of this study in which the most common bacterial isolates were S. aureus. E. coli (12.38%) emerged as the next common organism causing wound infection followed by CONS (11.40%) and P. aeruginosa (7.49%) likely similar to our results. However, the least common bacteria isolated were C. freundii (0.65%) not included in our study. In Saudi Arabia, Abussaud MJ [28] isolated S. aureus (35%), P. aeruginosa (25%) and Klebsiella spp. (10%) as the major causative agents nearly analogous to our study. The high prevalence of Staphylococcus aureus infection may be because it is an endogenous source of infection. Infection with this organism may also be due to contamination from the environment. With the disruption of natural skin barrier, Staphylococcus aureus which is a common bacterium on surfaces such as the human skin easily find their way [4].

CONCLUSIONS

The most predominant bacteria detected were *Staphylococcus aureus*. An incessant monitoring and update studies on the microbial isolates are crucial and compulsory prerequisite for an enhanced management and treatment of wound sepsis, which would result in better patient care, safety and health care outcomes.

ADDITIONAL INFORMATION AND DECLARATIONS

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Author Contributions: Study design, reviewed literatures, inscription of 1st and final draft of manuscript- NPY; Sample collection, data analysis, involved in writing first draft-RKJ; revision and amended 2nd draft of manuscript-RKY; intellectual critics in scripting, editing and final decisive revision of final draft of manuscript-OPY. All authors have read and agreed with the contents of the final manuscript towards publication.

Data Availability: Data will be available upon request to corresponding authors after valid reason.

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