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Research Article Isolation, Identification and Characterization of Bacteria Isolated from Wound Sepsis from Patients in a Secondary Medical Care Facility in Nigeria

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Abstract

Background and Objective: A wound is an injury to living tissue caused by a cut or other impact, typically one in which skin is cut or broken. The study aimed to isolate, identify and characterize bacteria associated with wound sepsis in this locality. **Materials and Methods:** The isolation and identification of bacteria from wounds was carried out on sixty specimens of wound swabs collected from General Hospital, Minna. This was achieved by using standard bacteriological procedures and techniques. **Results:** The results showed that the majority of the wounds were contaminated with various bacteria isolates of which *Staphylococcus aureus* was the most prevalent with 21 (35.0%) followed by *Pseudomonas aeruginosa*, 16 (26.7%), *Proteus* sp., 10 (16.7%), Coagulase-negative *Streptococcus* sp., 8 (13.3%) while *Klebsiella* spp. and *Escherichia coli* has the least occurrence of 3 (5.0%) and 2 (3.3%), respectively. The presence of these bacteria in the various wound samples investigated could have been as a result of exposure to the dirty environment, contaminated water or materials used for treatment and even from the hospital (nosocomial infections). **Conclusion:** Avoiding these factors could help to prevent mortality, amputation and debridement of the infected part of the patients or individual. In conclusion, the presence and multiplication of the above bacterial in the wound may delay the healing process of the wound. Therefore, it is concluded that wound healing is predicated on good hygiene, proper care of wound infection as well as the use of effective antimicrobial drugs.

Key words: Isolation, characterization, bacteria, wound sepsis, antimicrobial drugs

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

A wound is a bodily injury that usually involves a laceration or cut on the skin's membrane as well as damage to the underlying tissues and is caused by an assault, accident or surgery¹. When virulence factors produced by one or more bacteria in a wound overcome the host's natural immune system, the germs can penetrate and grow in live tissue, causing a cascade of local and systemic host reactions². Local reactions include pus discharge and painful spreading erythema due to vasodilation, both of which are symptoms of cellulite is around a wound². The majority of acute and chronic wound infections are caused by a combination of aerobic and anaerobic bacteria. Polymicrobial colonization of the environment is characterized by the presence of numerous potentially harmful bacteria.

Any wound has the potential to become contaminated^{2,3}. Even though microorganisms cause wound infection, there is still substantial controversy about the particular mechanisms by which they do so as well as their function in non-healing injuries that show no indications of infection². Infection can occur when bacteria begin to proliferate and attach to the wound site without causing tissue damage³. The wound healing process is not hindered solely by colonization and in some cases, colonization might speed up the healing process⁴. This can also happen when the number of bacteria in the environment increases substantially and the immune system of the host becomes overwhelmed⁵. During this stage, the wound's granulation bed seems unhealthy, with atrophied, deep red or grey colouring and increased secretion but no evidence of invasion of the surrounding tissue⁴⁻⁶. Infection of a wound slows healing, causes wound breakdown, increases treatment time and increases trauma and medical costs7. According to studies, wound infection is ubiquitous and the types of bacteria vary by geographical region, host skin bacteria, clothing at the wound site and time between wound occurrence and evaluation⁸. Due to widespread antibiotic resistance and an increase in infections caused by methicillinresistant Staphylococcus aureus (MRSA) and polymicrobial flora, controlling wound infections has become more difficult⁸. Staphylococcus aureus, Proteus mirabilis, Pseudomonas aeruginosa, Klebsiella aerogenes, Escherichia coli. Staphylococcus epidermidis, Streptococcus pyogenes and Streptococcus faecalis are among the bacteria commonly associated with wound infections. Candida albicans and Candida tropicalis have also been linked to the spread of the disease². This study is therefore designed to isolate, identify and characterize bacteria associated with wound sepsis in General Hospital in Minna Niger State.

MATERIAL AND METHODS

Study area: The study was carried out from April to July, 2021 in General Hospital, Minna.

Participants/subjects: The subjects of this investigation were patients attending General Hospital Minna, Niger State for treatment of their wounds.

Sample collection: Sixty wound swap samples were collected aseptically, using sterile swab sticks from both male and female patients with wounds by rotating the sterile cotton swab stick over the wound and the swap stick is inserted into a sterile transport tube containing saline and labeled⁹.

Media used: The media used were Blood agar, MacConkey agar and Nutrient agar. The media were prepared according to the manufacturer's specifications labelled on the media containers. The agars were autoclaved for 15 min at 121°C and dispensed aseptically into a sterile petri dish.

Isolation methods: All specimens collected were immediately applied to freshly prepared blood agar, MacConkey agar and Nutrient agar, streaked and incubated overnight at 37°C for 24 hrs. After incubation, the bacterial colonies were observed and discrete colonies were picked and purified by sub-culturing onto freshly prepared Nutrient agar, MacConkey agar and blood agar using a streak plate technique. Isolated colonies that grew on the plates were then transferred onto Nutrient agar slants with a proper label. These agar slants were stored in the refrigerator at 4°C and were used for further characterization.

Characterization and identification of bacterial isolates:

Characterization and identification of bacterial isolates were based on standard microbiological methods including gram staining, morphological and cultural characteristics on Nutrient agar media, catalase test, coagulase test, indole production test, oxidase test, triple sugar iron test, citrate test and urease test.

RESULTS

The colonial characteristics of bacterial isolates on different media (MacConkey agar, Chocolate agar and Nutrient agar) and colonial morphological were shown in Table 1. *Staphylococcus aureus* and *Streptococcus* species have round colonies while *Pseudomonas aeruginosa, Klebsiella* species and *Escherichia coli* exhibited different colonial morphology.

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| Table 1: Colonial | characteristics of bacterial isolates |
|-------------------|---------------------------------------|
| | |

| Organisms | MacConkey agar | Chocolate agar | Nutrient agar | Colony morphology |
|------------------------|----------------|----------------|---------------|---------------------|
| Staphylococcus aureus | Golden yellow | Golden yellow | White | Round |
| Pseudomonas aeruginosa | White | White | Green | Round and irregular |
| Proteus sp. | Pink | Pink | White | Swampy and flooded |
| Streptococcus sp. | Pink | Pink | White | Round |
| <i>Klebsiella</i> sp. | White | White | White | Raised and round |
| Escherichia coli | Pink | White | White | Convex and round |

Table 2: Biochemical characteristics of bacterial isolates

| | | | Test | | | | | | |
|------------------------|---------------|--------------------------|----------|-----|--------|---------|---------|--------|-----------|
| Suspected organism | Gram reaction | Shapes | Catalase | TSI | Urease | Oxidase | Citrate | Indole | Coagulase |
| Staphylococcus aureus | Gram-positive | Singly, clusters | + | - | - | - | - | - | + |
| Pseudomonas aeruginosa | Gram-negative | Singly, rods | + | + | - | + | - | - | - |
| <i>Proteus</i> sp. | Gram-negative | Rods | + | + | + | - | - | - | - |
| Streptococcus sp. | Gram-positive | Cocci in chains | - | - | - | - | - | - | - |
| <i>Klebsiella</i> sp. | Gram-negative | Singly, stout | + | + | - | - | + | - | - |
| Escherichia coli | Gram-negative | Singly, rods in clusters | + | + | - | - | - | + | - |

TSI: Triple sugar iron, +: Positive and -: Negative

Table 3: Occurrence of bacterial isolates

| Organisms suspected | Total number isolated | Occurrence (%) |
|--------------------------|-----------------------|----------------|
| Staphylococcus aureus | 21 | 35.0 |
| Pseudomonas aeruginosa | 16 | 26.7 |
| Proteus sp. | 10 | 16.7 |
| <i>Streptococcus</i> sp. | 8 | 13.3 |
| <i>Klebsiella</i> sp. | 3 | 5.0 |
| Escherichia coli | 2 | 3.3 |
| Total | 60 | 100 |

Biochemical characteristics of bacterial isolates confirmed the presence of *Staphylococcus aureus*, *Streptococcus* species, *Pseudomonas aeruginosa*, *Klebsiella* species and *Escherichia coli* in the wound sepsis as indicated in Table 2. Out of the 60 samples cultured, *Staphylococcus aureus* has the highest number of occurrence, 21 (35.0%) followed by *Pseudomonas aeruginosa*, 16 (26.7%) while *Escherichia coli* has the least occurrence, 2 (3.3%) in Table 3.

DISCUSSION

This study was carried out to isolate and characterize the bacterial pathogens associated with wound sepsis. The findings show that *Staphylococcus aureus* was the commonest isolates accounting for 21 (35.0%) followed by *Pseudomonas aeruginosa*, 16 (26.7%), *Proteus* sp., 10 (16.7%), Coagulase-negative *Streptococcus* sp., 8 (13.3%) while *Klebsiella* sp. and *Escherichia coli* has the least occurrence of 3 (5.0%) and 2 (3.3%), respectively (Table 3). These results were in agreement with previous studies carried out globally and in different parts of the country such as Umuahia², Wukari³, Ibadan¹⁰, Benin city¹¹ and Ekpoma¹². Findings from the study carried out at a University hospital in Nigeria showed that the commonly isolated bacteria were *Staphylococcus* aureus (25%) and Pseudomonas aeruginosa (20%)¹. A similar study carried out at a University teaching hospital in Iran, also reported Staphylococcus aureus to be the commonest bacteria isolated (43%)¹³. The high prevalence of Staphylococcus aureus infection maybe because it's an endogenous infection source. Contamination from the environment, such as contamination of surgical instruments, can also cause infection with this organism. Staphylococcus aureus, the prevalent bacteria on surfaces such as human skin, can easily enter wounds due to the disruption of the natural skin barrier. Also, Anthanasopoulos et al.¹⁴ proposed that the extracellular adherence protein (Eap) of Staphylococcus *aureus* played a key role in delayed wound healing by blocking angiogenesis in the proliferative state and slowing the inflammatory response. Inflammation plays a crucial role in wound healing and is responsible for the elimination of germs¹⁵. The presence of bacterial components in chronic wounds, on the other hand, may trigger an excessive inflammatory response and chronic wounds may not heal unless the excessive inflammation is decreased¹⁶.

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The results show that most of the wounds used in this study were contaminated with these bacteria and are therefore at risk of infection if proper care and treatment modalities are not adhered to.

CONCLUSION

Staphylococcus aureus was confirmed to be the most prevalent bacteria in wounds accounting for 21 (35.0%) from this study. Wound infection is an ongoing burden when an injury is sustained and increased the rate of infection due to contamination of wound when risk factors are present such as diabetes. The presence of these bacteria in the various wound samples investigated could have been as a result of exposure to the dirty environment, contaminated water or materials used for treatment and even from the hospital (nosocomial infections). To avoid wound infection, good personal hygiene should be maintained and the wound should regularly be cleaned with antibiotics to prevent bacteria colonization around the wound. Finally, the presence and multiplication of the above bacterial in the wound may delay the healing process of the wound. Therefore, whenever there is a wound, especially one which delays healing, routine culture should be carried out to determine bacterial associated with such wound and its susceptibility to various antibiotics should also be carried out to determine the choice of antibiotic for treatment. Hence good hygiene and proper care of wound infection plus the cooperation of antimicrobial drugs during treatment is advised.

SIGNIFICANCE STATEMENT

This study discovered the implication of wound infection and its solution that can be beneficial for the medical caregiver and the patients. This study will help the researchers to uncover the critical areas of causes of delays in the hospital that many researchers were not able to explore.

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