

BACTERIOLOGICAL PROFILE AND ANTIBIOTIC SUSCEPTIBILITY PATTERNS OF BURN WOUND ISOLATES AT A TERTIARY CARE HOSPITAL IN PESHAWAR

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Abstract

Burn wound can easily infected by bacteria, and increasing resistance to of these bacteria to antibiotics is becoming a serious problem, especially in developing countries such as Pakistan. There is limited information about the bacteria found in burn wounds and their response to antibiotics in Peshawar. This Study was carried out at Burns and Plastic Surgery Centre Hayatabad, over a period of six months. The main aim of the study was to identify the bacteria found in burn wound and see how well they responded to different antibiotics. A total of 100 burn patients were included in using consecutive samples. The data was collected prospectively for one month from hospital microbiology laboratory until the required number of patients was reached. The bacterial identification and antibiotic susceptibility results were recorded on a structured proforma. Ethical approval was obtained before the data collected. Out of 100 isolates, Gram-negative bacteria were more common, making 76% of the total isolates. Gram-positive bacteria made up 24%. within Gram-negative bacteria, *Pseudomonas aeruginosa* and *Klebsiella* species were the most common. Among Gram-positive bacteria, *staphylococcus aureus* and MRSA were the main isolates. Colistin, meropenem and doripenem showed good activity against most Gram-negative isolates, on other hand higher resistance was seen against ampicillin, Augmentin, and sulfamethoxazole/trimethoprim. For Gram-positive bacteria, vancomycin, tigecycline, and linezolid showed high susceptibility. Most MRSA isolates were resistant to other antibiotics tested. The findings show Gram-negative bacteria was the main cause in burn wound infection is this study.

Introduction

Burns remains one of the most unevenly spread injuries in world health. The greatest number of serious burn cases happens in low to middle income countries and although burn rates are slowly decreasing in world, the total count of serious cases is expected to keep increasing to the mid of the century (1). The effect can be seen obviously in patient outcomes, patients treated in places with less medical resource often have high burns and die more than ten than patients treated in richer health system (2). A burn wound does not always become infected, in the beginning bacteria may only live on the surface of the wound without causing a serious infection. With time however, some bacteria can increase in number and move to deeper into damaged tissue which may lead to a true wound infection. The change of infection can become greater when the wound remains open for a long time or when the patient need prolong hospital care. Therefore, understanding how bacteria enters and grow in burn wound is important for preventing infection and supporting proper wound healing (3).

What makes a burn different from other injuries is that it removes the body natural protection against infection at the same time that the immune system become disturbed (4). A recent review of hospital infections in adult burn patients shows that changes in how medicine work in the body, long use of antibiotics, and changes in the type of bacteria during hospital stay all help resistant bacteria develop in burn unit (5). This problem is not limited to burn injuries. Gram negative are already more common

than gram positive bacteria in general wound samples for major hospitals showing a wider range in the types of bacteria that grow in the injured tissues (6). A study of burn wound samples in India found that *pseudomonas* and *staphylococcus* were the main bacteria while the colistin and vancomycin were the most effective antibiotics (7).

One reason bacteria become difficult to treat is due to their ability to form the biofilm which is a protective layer that bacteria make around him which can helps the bacteria to protect form antibiotics. In India more than half of gram-negative bacteria collected from burn wounds were able to form biofilm (8). Burn patients in Asia also shows that antibiotic resistant bacteria are very common in the region with *Acinetobacter*, *pseudomonas*, and *staphylococcus* and *Klebsiella* are among the main bacteria causing infection (9).

Pseudomonas Aeruginosa needs specific attentions because it can quickly develop resistance to antibiotics. A ten years study in United Kingdom found this bacterium in more than half of burn patients and its presence was linked with longer hospital stays and need of surgery (10). A study of Burn patients in Iran found that antibiotics resistance in this bacterium was related to biofilm formation and other harmful features of this bacterium. In Ethiopia about one third of *pseudomonas aeruginosa* bacteria collected from burn patients were resistant to several antibiotics (11). Similar problems have been also reported in countries closer to the area of present study. In Balochistan researchers found that *pseudomonas aeruginosa* can slow the healing of burn wounds (12).

Gram positive bacteria specially *Staphylococcus aureus* also show serious antibiotic resistance. A study of MRSA bacteria from burn wound in China found different genetic groups. These groups showed different responses to antibiotics (13). During patients' hospital stays the types of bacteria and their resistance also changes. One study found the change from easier to treat Gram positive at the beginning of the hospital stays to more difficult to treat gram negative bacteria later (14).

Correct identification of bacteria causing a burn wound infection is an important part of patient care. A wound may contain more than one type of bacteria and bacteria present may change during treatment (3). Clinical signs alone may not always show which bacteria is responsible for the infection so lab testing of wound sample can provide useful information. Bacterial culture and antibiotic susceptibility testing can help identify the causative organism and show which antibiotic is likely to work. This information is especially important in tertiary care hospital where many seriously injured patients receive prolonged treatment (15).

The present study was therefore conducted to determine the bacteriological profile of burn wound isolates and describe their antibiotic susceptibility patterns at a tertiary care hospital in Peshawar.

Methodology

The study was descriptive cross-sectional study conducted at the Burns and Plastic Surgery Centre, Hayatabad, Peshawar, over a period of six months. The study population consisted of burn patients whose burn wound samples were processed by the

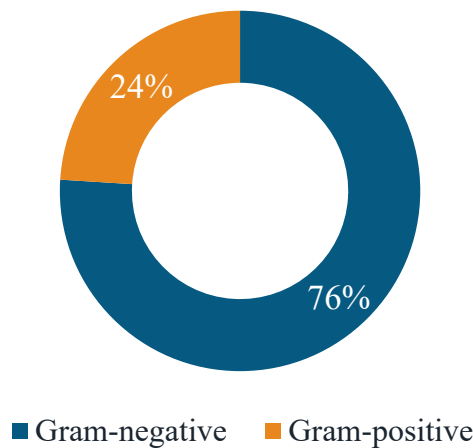
hospital microbiology laboratory, and 100 burn patients were included using consecutive sampling. Real-time data were collected prospectively from the hospital microbiology laboratory for one month during study period, with eligible burn wound culture results recorded as they become available. Patients were included if they had a burn wound, were admitted to or received treatment at the selected hospital during the study period, and had a wound sample sent to the hospital laboratory for bacterial culture and antibiotics susceptibility testing. Patients without a burn wound, without an available wound sample or with incomplete or missing required information were excluded. The study used results generated by the hospital microbiology laboratory as a part of routine diagnostic testing, and bacterial identification and antibiotic susceptibility results were recorded from laboratory reports using a structured proforma containing patient age, gender, culture results, identified bacterial organism, and antibiotic susceptibility results. Ethical approval was obtained from the relevant institutional ethical committee, permission was obtained from the concerned laboratory authority, and patient confidentiality was maintained by excluding the names and directly identifying information.

Results

Distribution of Bacterial Isolates

The study included a total of 100 patients with burn wound isolates. The provided laboratory results included in both Gram-negative and Gram-positive bacteria. Gram-negative bacteria were predominant with

76% and Gram-positive bacteria were 24% isolated.



Among the Gram-negative isolates, *Pseudomonas Aeruginosa* was the most frequently isolated organism, with 29 isolates, followed by *Klebsiella Species* with 25 isolates, *Staphylococcus aureus* with 16 isolates, *Acinetobacter spp.* with 12 isolates and MRSA with 7 isolates. *Enterobacter spp.* with 2 isolates and *proteus spp.* represented by 3 isolates. *Citrobacter species* with 4 isolates, *E. coli* and *Staphylococcus Epidermidis* were each represented by 1 isolate.

Table 1: Distribution of Bacterial Isolates

Organism	No. of Isolates	Percentage%	Gram Classification
<i>Pseudomonas Aeruginosa</i>	29	29%	Gram-negative
<i>Klebsiella spp.</i>	25	25%	Gram-negative
<i>Staphylococcus aureus</i>	16	16%	Gram-positive
<i>Acinetobacter spp.</i>	12	12%	Gram-negative
MRSA	7	7%	Gram-positive
<i>Citrobacter spp.</i>	4	4%	Gram-negative
<i>Proteus spp.</i>	3	3%	Gram-negative
<i>Enterobacter spp.</i>	2	2%	Gram-negative
<i>Staphylococcus Epidermidis</i>	1	1%	Gram-positive
<i>Escherichia Coli</i>	1	1%	Gram-negative
Total	100	100%	-

Table 2: Antibiotic Susceptibility Pattern of Gram-Negative Bacteria

Antibiotics tested	<i>Citrobacter</i> <i>spp.</i> N = 04	<i>Proteus</i> <i>spp.</i> N = 03	<i>Enterobacter</i> <i>spp.</i> N = 02	<i>Acinetobacter</i> <i>spp.</i> N = 12	<i>Klebsiella</i> <i>spp.</i> N = 25	<i>P. aeruginosa</i> N = 29	<i>E. Coli</i> N = 01
Ampicillin	R	R	R	R	R	NT	01 (100%)
Augmentin	R	R	R	R	02 (8%)	NT	00 (0%)
Tazobactam/ Piperacillin	02 (50%)	02 (67%)	01 (50%)	08 (67%)	18 (72%)	10 (34%)	00 (0%)
Imipenem	03 (75%)	02 (67%)	02 (100%)	07 (58%)	17 (68%)	10 (34%)	00 (0%)
Meropenem	04 (100%)	03 (100%)	02 (100%)	11 (92%)	25 (100%)	20 (69%)	00 (0%)
Doripenem	04 (100%)	03 (100%)	02 (100%)	11 (92%)	25 (100%)	20 (69%)	01 (100%)
Gentamicin	01 (25%)	02 (67%)	00 (0%)	07 (58%)	17 (68%)	05 (17%)	01 (100%)
Amikacin	03 (75%)	03 (100%)	02 (100%)	10 (83%)	21 (84%)	19 (66%)	00 (0%)
Tobramycin	01 (25%)	02 (67%)	00 (0%)	07 (58%)	17 (68%)	05 (17%)	00 (0%)
Cefoperazone/ Sulbactam	01 (25%)	00 (0%)	00 (0%)	07 (58%)	17 (68%)	10 (34%)	01 (100%)
Colistin	04 (100%)	R	02 (100%)	12 (100%)	25 (100%)	29 (100%)	01 (100%)
Ciprofloxacin	02 (50%)	00 (0%)	00 (0%)	06 (50%)	15 (60%)	13 (45%)	00 (0%)
Tetracycline	00 (0%)	R	00 (0%)	07 (58%)	14 (56%)	NT	01 (100%)
Tigecycline	02 (50%)	R	02 (100%)	10 (83%)	23 (92%)	NT	01 (100%)
Sulfamethoxazole/ Trimethoprim	00 (0%)	00 (0%)	00 (0%)	02 (17%)	02 (8%)	NT	00 (0%)
Cefotaxime	00 (0%)	00 (0%)	00 (0%)	05 (42%)	10 (40%)	NT	00 (0%)

N= Number of isolates, R= Resistant (naturally resistant), NT= Not Tested, Spp.= Species.

The Gram-negative susceptibility results showed high sensitivity to meropenem, doripenem and colistin among several organisms. Colistin showed 100% sensitivity to for *Citrobacter*, *Acinetobacter*, *Klebsiella*, *Pseudomonas aeruginosa* and single *E. coli* isolate, while meropenem and doripenem showed 100% susceptibility to *Citrobacter*, *Proteus*, *Enterobacter* and *Klebsiella* isolates and

92% susceptibility for *Acinetobacter*. *Pseudomonas aeruginosa* showed 69% susceptibility to both meropenem and doripenem and 66% to amikacin. Lower susceptibility was observed for several common tested antibiotics, including sulfamethoxazole/trimethoprim and cefotaxime among *Klebsiella* and *Acinetobacter* isolates.

Table 3: Antibiotic Susceptibility Pattern of Gram-Positive Bacteria

Antibiotics Tested	<i>Staphylococcus Aureus</i> N = 16	MRSA N = 07	<i>Staphylococcus Epidermidis</i> N = 01
Ampicillin	00 (0%)	00 (0%)	00 (0%)
Augmentin	10 (63%)	00 (0%)	01 (100%)
Tazobactam/Piperacillin	16 (100%)	00 (0%)	01 (100%)
Gentamicin	14 (88%)	00 (0%)	01 (100%)
Amikacin	14 (88%)	00 (0%)	01 (100%)
Imipenem	16 (100%)	00 (0%)	01 (100%)
Cefoxitin	16 (100%)	00 (0%)	01 (100%)
Vancomycin	16 (100%)	07 (100%)	01 (100%)
Tetracycline	12 (75%)	00 (0%)	01 (100%)
Tigecycline	16 (100%)	07 (100%)	01 (100%)
Cefotaxime	09 (56%)	00 (0%)	00 (0%)
Cefoperazone/Sulbactam	12 (75%)	00 (0%)	01 (100%)
Erythromycin	09 (56%)	00 (0%)	01 (100%)
Linezolid	16 (100%)	07 (100%)	01 (100%)
Ciprofloxacin	10 (63%)	00 (0%)	01 (100%)
Penicillin	00 (0%)	00 (0%)	00 (0%)

N= Number of isolates, R= Resistant (naturally resistant), NT= Not Tested.

Among the Gram-Positive isolates, *Staphylococcus aureus* was the most common organism with 16 isolates, followed by MRSA with 7 isolates and *Staphylococcus*

epidermidis with one isolate. *Staphylococcus aureus* showed 100% susceptibility to piperacillin/tazobactam, imipenem, cefoxitin, vancomycin, tigecycline and linezolid, while gentamicin and amikacin

showed 88% susceptibility. All seven MRSA isolates were susceptible to vancomycin, tigecycline, and linezolid, whereas no susceptibility was reported for other listed antibiotics. The Single *S. epidermidis* isolates showed susceptibility to most tested antibiotics except ampicillin, cefotaxime and penicillin.

Discussion

The study Examined the types of bacteria found in burn wound samples and their response to different antibiotics different antibiotics. A total of 100 burns wounds isolated were studied. The findings are discussed below by looking at the common bacteria. Antibiotics resistance between Gram-negative and Gram-positive bacteria an what these findings means for choosing antibiotics in a tertiary care burn unit.

Gram-negative bacteria made up the most isolates (76%). In both Gram-negative and Gram-positive bacteria, *pseudomonas aeruginosa* was most common bacteria, with 29 isolates followed by *Klebsiella species* with 25 isolates. Other important organisms were *Staphylococcus Aureus* with 16 isolates. *Acinetobacter spp.* with 12 isolates and MRSA with 7 isolates and remaining was found in smaller number. The high number of Gram-negative bacteria in this study is similar to findings from other burn wound studies. (16), is a study from a tertiary care hospital in Peshawar, also reported Gram-negative Bacteria as important cause of burn wound infections. With *Klebsiella Pneumoniae* and *Pseudomonas Aeruginosa* among the common organisms.(17) A study from Lahore also reported *Pseudomonas Species* as a major

organism in burn wound infections. Th differences in the number of bacteria between studies may be due to difference in patients, hospital environment, infection control practices wound care and antibiotic use.

The Gram-negative bacteria showed different levels of sensitivity to the tested antibiotics. Overall, the Meropenem, Doripenem and colistin showed the highest activity against several Gram-negative organisms. The highest sensitivity to colistin is an important finding because colistin is often use against bacteria that are resistant to many other antibiotics. In the present study all 29 *pseudomonas aeruginosa* isolates were sensitive to colistin. however, Colistin should not be used unnecessarily because resistant to this antibiotic can be develop. (18) reported increasing concern about colistin resistance in Gram-negative bacteria, including *Klebsiella Pneumoniae* and *pseudomonas aeruginosa*.

The commonly used antibiotics showed lower activity against many Gram-negative bacteria. Ampicillin showed no sensitivity in the tested Gram-negative groups except for the *E. Coli* isolates, which showed poor activity against the tested organisms. cefotaxime and sulfamethoxazole/trimethoprim showed particularly low sensitivity against *Klebsiella spp.* and *Acinetobacter species*. These findings are similar to a recent Pakistani study by (19), which reported high resistance to commonly used antibiotics among Gram-negative bacteria for wound infections. The poorer response to these antibiotics may be related

to their frequent use and the ability of bacteria to develop resistance.

Among the Gram-positive bacteria *Staphylococcus aureus* was the most common organism, with 16 isolates followed by MRSA with 7 isolates and *Staphylococcus epidermidis* with one isolate. Within 16 *Staphylococcus Aureus* isolates in the present study, piperacillin/tazobactam, imipenem cefoxitin, vancomycin, tigecycline and linezolid showed (100%) susceptibility. Gentamicin and amikacin each showed 88% susceptibility, while tetracycline and cefoperazone/sulbactam showed 75% susceptibility. No Susceptibility was found to ampicillin and penicillin. The complete lack of susceptibility to penicillin and in the present study it similar finding from recent study burn wound studies. (20) reported high resistance to penicillin among burn wound isolates, showing that older antibiotics may no longer be reliable against *Staphylococcus Aureus* in burn patient.(21) also reported very high resistance to penicillin while vancomycin and linezolid was remained active against all tested isolates.

MRSA was founded in 7% the total isolates in this study. All seven MRSA showed susceptibility to vancomycin, tigecycline and linezolid. However, no susceptibility to Augmentin, Gentamicin, amikacin, imipenem, cefoxitin, tetracycline, ciprofloxacin and penicillin. This shows that MRSA in the present study had a much wider resistance than Ordinary *Staphylococcus aureus*. The findings are generally similar to recent study done in Pakistan.(22) studied 751 *S. aureus* isolates from burn patients and

found 56% were MRSA. Their study reported 99.6% susceptibility to linezolid while very high resistance to several antibiotics used in routine like ciprofloxacin and trimethoprim-sulfamethoxazole.

Conclusion

The present study showed that burn wound infections were caused by a variety of bacteria, with Gram-negative bacteria were being more common than Gram-positive bacteria. Among the 100 patients included in this study, Gram-negative bacteria accounted for 76% of the most of isolates. The Antibiotics susceptibility results showed that the isolates had different levels of sensitivity and resistance to the tested antibiotics. Resistance to commonly used antibiotics is an important concern because it can make the treatment of burn wound infections more difficult. The findings also showed that knowing the types of bacteria causing an infection is important before selecting an antibiotic whenever testing is possible. Overall, the study gives a better understanding of the bacteria commonly found in burn wounds and their antibiotic susceptibility patterns at the study hospital.

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