

Original Research Article

Isolation, identification and antibiotic sensitivity pattern of aerobic bacteria from burn wound patient admitted in tertiary care hospital

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ABSTRACT

Background: Significant burn injuries induce a state of immunosuppression that predisposes patients to infectious complications, thus the rate of nosocomial infections are higher. Rapidly merging multidrug resistant among the various isolate in indoor burn patients are depending on time-line becoming serious threat for managing therapeutically. Objective of this study is to determine the aetiological factor, prevalence, antimicrobial susceptibility pattern and emerging nosocomial pathogens.

Methods: A prospective study was carried in burn ward of K.L.E.'s Dr. Prabhakar Kore Hospital and Medical Research Centre, Belgaum for the period of 1 year. Pair of wound swab were collected from patient having burnt more than 30% (RULE OF NINE) on 3rd day of stay. Sample were collected aseptically from 30 patients and processed by convectional culture and biochemical identification procedures and tested against commonly used antibiotics.

Results: 30 patients that fall under inclusive criteria were enrolled in the study. The total burn surface area (TBSA) ranges from 30-82%. The ratio of female to male patient suffering burn wound in our study is 1.5:1. Aetiology of burn is heat (moist/dry) mostly. Depending upon degree of burn, most of patient suffered from 20 degree (superficial to deep) injury. From 30 swab cultures, 42 isolates were identified during the study in which mixed were 66.66% and one is fungi. The most commonly isolated is *Pseudomonas aeruginosa* (45.24%) then *Klebsiella pneumoniae* (19.04%), *Acinetobacter spp.* (14.28%), *Staphylococcus aureus* (11.90%). Among gram positive isolates, isolates are found to be most resistant to Erythromycin (100%) and Co-trimoxazole (100%) and sensitive to Vancomycin (71.42%). Among gram negative isolates are found to be most resistant to Gentamicin (91.65%), Ciprofloxacin (82.35%), Ceftazidime (82.35%) and sensitive to Meropenem (52.95%), Piperacillin (35.30%), Carbenicillin (29.41%).

Conclusions: *Pseudomonas aeruginosa* was found to be the most common isolate. The nature of microbial wound colonization and flora changes with time should be taken into consideration in empirical antimicrobial therapy.

Keywords: Antimicrobial resistant, Antimicrobial sensitivity, Burn, Nosocomial infection

INTRODUCTION

Burn patients are at risk of acquiring infection because of the loss of skin barrier and suppressed immune system, compounded by prolonged hospitalization and invasive therapeutic procedures. The major challenge for a burn

team is nosocomial infection in patient which is known as to cause over 50% of burnt deaths.¹ Based on National Nosocomial Infection Surveillance System (NNIS) criteria, all the burn patients are required to follow the distribution of bacterial species among burn isolates and the antimicrobial susceptibility of the pathogens in order to adopt empirical antibiotic strategies.¹ Infection is most

common and most serious complication of major burn injury is related to burn size.² Microbial colonization of the open burn wounds is established by the end of the first week, primarily from an endogenous and exogenous source from environment and healthcare associates. Due to overcrowding in burn ward, cross-infection may come into play. In various country including India, the importance of *Acinetobacter spp* is emerging as nosocomial pathogen rapidly. In addition, the problem of multi-drug resistance in gram negative bacilli due to extended spectrum of beta-lactamases (ESBL) production is becoming a serious threat to the healthcare worker, who are likely to contract the infection, as therapeutic option to these organism are limited.³ The common pathogens isolated from burn patients include *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella spp.* and various coliform bacilli.^{1,4} Fungi can also cause infection. The pattern of infection differ from hospital to hospital; the varies bacterial flora of infected wound may change considerably during the healing period.

With reference to above literature, the present study is carried to identify the commonest pathogen isolated from burn ward and to gain in-depth knowledge of the resistant organisms and their antibiotic sensitivity pattern so that infection related morbidity and mortality will improved.

METHODS

The present study was done in microbiology department, JNMC and K.L.E's Dr. Prabhakar Kore Hospital and Medical Research Centre, Belgaum. The clinical Specimens collected on sterile cotton swab from patient admitted in burn ward and plastic surgery ward following the inclusive criteria like more than two days of hospital

stay, more than 30% of burn injury, sample collecting before the antiseptic used. Exclusion criteria like sample are collected before two days of admission of patient, patients with less than thirty percentage of burn injury, sample collected after use of antiseptic, Sample collected on cotton swab are dried up.

The 30 clinical samples has been transferred immediately to the bacteriology laboratory where processed aseptically. Preliminary, Gram's stain was done with one swab and cultured on Blood agar, MacConkey agar. And further Gram's stain, Catalase, Oxidase, Coagulase, Motility test etc done from the growth. The isolate were characterized by battery of biochemical tests. The biochemical tests includes mostly indole production test, methyl red test, voges-proskauer test, citrate utilisation test, urea hydrolysis test, triple sugar iron test, amino acid deaminase test, mannitol motility medium, sugar medium, phenylpyruvic acid test etc. Antibiotic susceptibility testing is done by Kirby Bauer disc diffusion technique using Mueller Hilton agar. Sensitivity result were interpreted according to Clinical Laboratory Standard Institute (CLSI).

RESULTS

In this study 30 patients that fall under inclusive criteria were enrolled in the study. The total burn surface area (TBSA) ranges from 30-82%. The ratio of female to male patient suffering burn wound in our study is 1.5:1. Most of the patient's aetiology of burn is heat (moist/dry) 80%; Chemical 16.66% and Electrical 3.34%.

Depending upon degree of burn, most of patient suffered from 2^o degree (superficial to deep) injury is 50%; 1^o degree is 26.66% and 3^o degree is 23.34%.

Table 1: Antibiotic susceptibility pattern (%) of isolates in burn patients.

Antibiotics/ conc ⁿ	<i>P. aeruginosa</i> % (No.19)	<i>Acinetobacter</i> <i>spp.</i> % (No.6)	<i>K. pneumonia</i> % (No.8)	<i>Citrobacter</i> <i>freundii</i> % (No.1)	<i>Staph.</i> <i>Aureus</i> % (No5)	<i>Enterococcus</i> <i>spp.</i> % (No.2)
Amikacin(30)	26.31 (5)	50 (3)	25 (2)	100 (1)	N.D	N.D
Gentamicin(10)	10.52 (2)	16.67 (1)	0 (0)	0 (0)	N.D	N.D
Ciprofloxacin(5)	11.10 (1)	16.67 (1)	0 (0)	0 (0)	0(0)	50 (1)
Ceftazidime(13)	26.37 (5)	16.67 (1)	0 (0)	0 (0)	N.D	N.D
Meropenem(10)	26.37 (5)	83.33 (5)	100 (8)	0 (0)	N.D	N.D
Piperacillin(100)	47.37 (10)	33.33 (2)	0 (0)	0 (0)	N.D	N.D
Carbenicillin(100)	36.84 (7)	33.33 (2)	0 (0)	100 (1)	N.D	N.D
Ampicillin(10)	N.D	N.D	N.D	N.D	20 (1)	0 (0)
Cotrimoxazole (1.25/23.7)	N.D	N.D	N.D	N.D	0 (0)	0 (0)
Erythromycin(15)	N.D	N.D	N.D	N.D	20 (1)	0 (0)
Amoxycillin(20)	N.D	N.D	N.D	N.D	60 (3)	0 (0)
Vancomycin(30)	N.D	N.D	N.D	N.D	80 (4)	50 (1)
Cefoxitin(5)	N.D	N.D	N.D	N.D	20 (1)	N.D

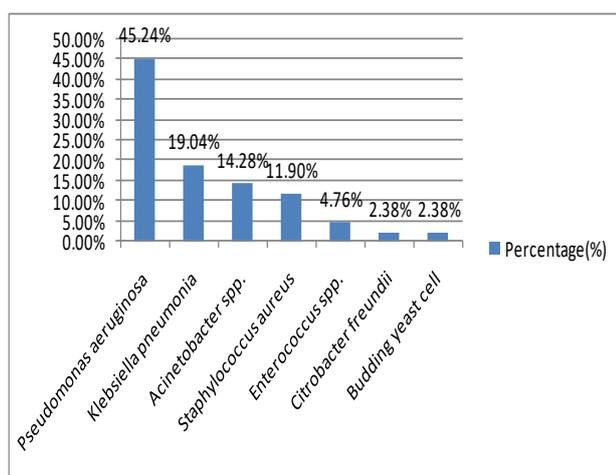


Figure 1: Incidence of isolates in burn patients.

It has been observed that polymicrobial infection in 66.66% patient. From 30 swab cultures, 42 isolates were identified during the study. And one of them is fungi. None of the swab show no growth petriplate.

Among 42 isolates, the most commonly isolated is *Pseudomonas aeruginosa* (45.24%) then *Klebsiella pneumonia* (19.04%), *Acinetobacter spp.* (14.28%), *Staphylococcus aureus* (11.90%) (Figure 1). Among gram positive isolates, isolates are found to be most resistant to Erythromycin (100%) and Co-trimoxazole (100%) and sensitive to Vancomycin (71.42%) (Table 1). Among gram negative isolates are found to be most resistant to Amikacin (67.65%), Gentamicin (91.65%), Ciprofloxacin (82.35%), Ceftazidime (82.35%) and sensitive to Meropenem (52.95%) and then with Piperacillin (35.30%), Carbenicillin (29.41%) (Table 1).

DISCUSSION

The burn wound is considered as the one of the major health issue in the world. The infection is frequent and complications are severe in patients who have sustained burn. The dysfunction of the immune System, a large cutaneous bacterial load, the possibility of gastrointestinal bacterial translocation, prolonged hospitalization, and invasive diagnostic and therapeutic procedures all contribute to sepsis.⁵ Thus, knowledge of the burn ward microbial flora and the current antibiotic sensitivities at any point of time is important for the better treatment of burn patients.

In this study, a total of 32 patient enrolled from burn ward. 2 (6.25%) of them died during the study period. One of the patients who died had TBSA of 58% and colonized by two different kinds of bacteria on his wound. The isolates were *MRSA*, *Pseudomonas aeruginosa*. Another patient who died had TSBA 50% and colonized by *Pseudomonas aeruginosa* on his

wound. This indicates that the organism could enter the bloodstream through the wound and is a potential threat for disseminated infection which can be life threatening.^{6,7}

It has been estimated that up to 75% of all deaths following burns are related to infection.⁶ On study there were 14% death in Nepal among a total of 50 patients whose mean TBSA was 33.9%.⁸ But in this study the low mortality rate (6.25%) was probably due to factors such as, continuous clinical and microbiological surveillance leading to quick detection of etiology, antibiotic therapy and care in our hospital. It is also stated that the death rates of burn patients have improved substantially in the past few decades due to advances in resuscitation, nutritional support, pulmonary care, wound care, and infection control practices in specialized burn units.⁸

The people are under poor socio-economic status and low health awareness patients may not seek treatment as early as possible so that their wound may get exposed for microbes from the environment. It is also stated that immediate colonization could be from the patient's endogenous origin, or via contact with contaminated external environmental surfaces, water, fomites, air, and from hands of health care workers.⁹

The most common isolate was *Pseudomonas aeruginosa*-55.0%, followed by *Staphylococcus aureus*-19.29%, *Klebsiella spp.*-11.43%, *Acinetobacter spp.*-7.14%. *Proteus spp* 4.29%, *Es-cherichia coli*-2.85%. Resistance of *S.aureus* was 40% observed with Oxacillin and 84% to Erythromycin whereas all strains were susceptible to Vancomycin. Authors analyzed that *Pseudomonas* which was the commonest isolate was most resistant to Ceftazidime (70%) followed by Cefotaxime. Ciprofloxacin (55.5%) and Amikacin (54.0%) were found to be most effective antimicrobial agent.^{10,11} Other Gram-negative organisms were highly resistant to Cefotaxime (66.0%) followed by Gentamycin (60.0%). Imipenem was found to be less resistant (26%) against *Pseudomonas*.¹²

In this study among 42 isolate, the most common isolate are *Pseudomonas aeruginosa*-45.24% followed by *Klebsiella pneumonia*-19.04%, *Acinetobacter spp.*-14.28%, *Staphylococcus aureus*-11.90%, *Enterococcus spp.*-4.76%, *Citrobacter freundii*-2.38%, fungal isolate-2.38% which is nearly similar to study done by Anuradha (Figure 1).¹²

It was seen that very high culture positivity of 96% in the samples from burn wound accounting single isolate of 62.5% of case and multiple isolates were noted in 37.5% cases.¹²

This study shows very high culture positivity of 100% in the samples from wound accounting single isolate of

were 33.34% and mixed were noted in 66.66%. This is not similar to study done by Anuradha.¹² This is because of sample exclusion criteria. There were 189 males (46.9%) and females (53.1%) with female to male ratio of 1.13 to 1. Thermal burns being most common type is also reported by most of the workers, Among the causes of thermal burns leading causes are Kerosene stove (32.3%), open flame (chulla etc.) (23.1%), Kerosene lamp (14.2%) and Gas stove (5.7%). Thus Kerosene was the main accelerant accounted for burns. This is probably because kerosene is cheap and easily accessible and more use of kerosene stove and kerosene lamp by the people of low socioeconomic strata in India, where obsolete and unsafe uses of fire for cooking and light are still prevalent.¹³

In this study shows that male is 12 (40%) and female 18 (60%). The ratio of female to male is 1.5:1. The most common type of burn is concluded as thermal burn which account for 80%. The severity of burn wound depending on the TBSA is 2⁰ degree. Most of patient had burnt are of superficial to deep. This result is analog with study Ushma.¹³ Five mixed growths of *Pseudomonas* and *Klebsiella* were observed from patients and three mixed growth of *Pseudomonas* and MRSA in this study. It was observed that a high level of drug resistance among gram negative isolates especially Gentamicin (91.18%). It was moderately resistant to meropenem (47.05%) whereas resistance was more marked for other antibiotics. The gram-negative bacteria isolated from burn patient showed considerable resistance to Carbenicillin (70.59%), Ceftazidime (82.35%), Ciprofloxacin (82.35%), Amikacin (67.65%).

In this study, the ratio of GNB and GPC is found to be 4.85:1. The gram-positive bacteria account for 16.67% whereas *S. aureus* infection in burn wound is by 11.90% and *Enterococcus spp.* by 4.76%. Among *S. aureus*, 80% were detected as methicillin resistant *S. aureus*. The Antibiotic susceptibility pattern showed that *S. aureus* mostly susceptible to Vancomycin (80%). which is similar to Alireza.¹

CONCLUSION

Pseudomonas aeruginosa was found to be the most common isolate. The commonest isolated among GPC is *S. aureus* was tested for methicillin and found to be 80% resistant. Fungal isolates are also prevalent in burn trauma that represents 2.38% in this study. The nature of microbial wound colonization and flora changes with time should be taken into consideration in empirical antimicrobial therapy of burnt patients. Indiscriminate and overuse of broad-spectrum antibiotics predisposes burn centres in to sites of multi drug resistant virulent micro flora. This should be control by making some judicial act to combat misuse of precious antibiotic. In

conclusion, present observations seem to be helpful in providing useful guidelines for choosing effective therapy against isolates from burn patients.

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Ethical approval: The study was approved by the Institutional Ethics Committee

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