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Original Research Article

Aerobic bacterial pathogens in burn wound infections: experience in a teaching institution

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ABSTRACT

Background: Infections are a serious cause of burn mortality and morbidity. Post initial resuscitation burn wound infections account for 75% of burn mortality. With rising number of multidrug resistant pathogens and changing microbiological colonizers of the burn wound, the insight provided by documenting the pathogens will help streamline the management. The aim of our study was to isolate, identify and study the profiles of aerobic bacteria in patients with burn wound infections.
Methods: This was a hospital based observational study carried out in the department of microbiology and department of burns and plastics, Safdarjung hospital after obtaining hospital ethical committee approval. Day 1, day 3, day 5 and day 10 swabs from burn sites were taken and processed after taking proper aseptic precautions from a total of 100 patients.
Results: Most of the aerobic isolates were gram negative in nature with *K.pneumoniae* (27.43%) being the most common followed by *P. aeruginosa* (22.12%), *S. aureus* (14.15%), *P. mirabilis* (10.61%), *Acinetobacter spp* (10.17%), *CoNS* (8.40%), *E. coli* (3.53%), *P. stuartii* (1.32%), *Enterobacter spp* (1.32%), *K. oxytoca* (0.44%) and *C. koseri* (0.44%).
Conclusions: We concluded the study recognizing *K. pneumoniae* as the most common isolate that cause burn wound infections. Further studies which include anaerobic isolates are required for identifying full range of organism profile of burn wound infections.

Keywords: Burn wound infections, bacterial pathogens, Aerobic

INTRODUCTION

Burn can be categorized as one of the most common cause of trauma which needs specialized medical attention. India with the second largest population in the world, have a large proportion of total incidence of burns around the world. With round 11 million burn victims seeking medical help worldwide in the year of 2004 according to a WHO report, and 6 to 7 million annual burn incidences in India according to Gupta et al it can be stated that burn incidence is much high in India compared to rest of the world.[1-3] Historically over the years, burn management strategies have changed from dressings impregnated with pig fat and resins as proposed by Hippocrates to fluid resuscitation, antibiotics, excision, skin graft, management of metabolic derangements and nutrition.[4] But overuse of antibiotics, lack of organized burn care and inability to prevent nosocomial infections are the major challenges faced today. With high incidences of burn per annum nationwide, measures to prevent and treat burn infections have great clinical significance.[2] Furthermore, post initial resuscitation, burn

wound infections account for 75% of burn mortality.[5]Among several isolates *Klebsiella spp, Pseudomonas spp, Staphylococcus aureus, Acinetobacter spp, Proteus spp* and *Citrobacter spp* are the major pathogens along with *Candida spp.*[6] There are wide variations in types and frequency of these isolates.[7] Burn injury causes coagulative necrosis of the skin and the underlying tissue. The amount of damage caused by burn is directly proportional to the energy that the causative agent (like fire, hot liquid, and other offending substance) imparts on the skin.[8] Skin to an extent decreases the transfer of heat to internal structures, but the damage to underlying tissue occur anyway due to local tissue responses. The risk of subsequent wound infection and systemic infection in a patient of burn correlates with the amount of skin involved.[9] This is usually represented as total body surface area (TBSA).[9] Burn of significant TBSA can lead to immunosuppression that predisposes the patient to wound and systemic infections. Even though local inflammation is a necessary factor for wound healing, burn of significant TBSA can lead to a systemic inflammatory response that at first is proinflammatory but later become anti-inflammatory to preserve homeostasis.[9] The usual pattern of burn wound infection and colonization is from gram positive (commensals) to gram negative organism which are thought to arise from the endogenous enteric flora.[10] In hospital setting, exogenous microorganism also colonize burn wounds via direct contamination from the hands of health care workers and other sources like untreated water, soil, and other articles of daily use and worsen the wounds.[11] Considering all this factors the aim of our study was to isolate, identify and study the profiles of aerobic bacteria in patients with burn wound infections.

METHODS

This was a hospital based observational study,carried out in department of microbiology and department of burns and plastics, Vardhman Mahavir medical college and Safdarjung hospital after obtaining hospital ethical committee approval. Duration of the study was from January 2019 to June 2020. Burn wound infections are polymicrobial in nature and prevalence of bacteria in burn wound infections in different Indian studies are in the range of 50 - 70%.[6,12,13] Taking prevalence as 60% (allowable error of 10%) and applying the formula;

$$N = 4PQ/E2$$

Where P is the prevalence, Q=1-P and E is the allowable error), the number of sample size was calculated as 96, therefore a total of 100 patients were recruited in the study using non-probability sampling method (convenience sampling).[14] For this study, patients admitted in burn emergency, burn intensive care unit (BICU) and burn wards were recruited and informed consent was taken. Following inclusion criteria was utilized for the selecting the study subjects.

Adult patients with burns ranging from 20% to 60% total body surface area (TBSA). Pediatric patients with burns ranging from 10% to 60% total body surface area (TBSA).

History was taken from patient or their reliable attendant using predesigned questionnaires to determine socio-demographic data, type of burn injury, TBSA, length of hospital stay, antibiotics given etc. Wound swabs of the burn patients were taken with a sterile cotton swab moistened with normal saline and transported to lab in less than 30 minutes as such or in Brain Heart Infusion (BHI) broth (if pus is scanty) within 24 hours of patient admission. Swabs were taken on day 1 of admission and then on 3rd, 5th and10th day of hospital stay. Only patients, whose day 1, day 3 and day 5 samples were processed was included in the study. Patients who were lost to follow up after day 5 sample collection, were also included in the study. Swab collected from burn patients were inoculated on to blood and MacConkey agar. Colonies thus isolated from the culture was further processed using standard microbiologic techniques. Atypical findings were further analyzed and confirmed using Biomérieux VITEK 2® using manufactures instructions. Statistical analysis was done, and categorical variables were presented in number and percentage (%) and continuous variables were presented as mean±SD and median. The data was entered in MS Excel spreadsheet and analysis was done using statistical package for social sciences (SPSS) version 21.0.

RESULTS

A total of 100 patients were recruited for the study of which 55 were male and 45 were female. Seventy-eight (78) patients were from burn ward and 22 were from burns intensive care unit (BICU). During the study, data was tabulated and later compiled as tables, graphs, diagrams and tabulated. Age and sex of study subjects were recorded along with other relevant history. Majority of patients in this study were of the age group of 21 to 30 years (32%) followed by 31 to 40 years (27%). The mean age group was 33.42 with a standard deviation of 14.7 (33.41±14.7). Median age of patients was 31.5 and the Inter Quartile Range (IGR) were 24 to 40.75 (Table 1). Total Body Surface Area (TBSA) of the study subjects were also recorded and the mean TBSA was 39.59±11.61 with a median of 40 and IQR between 30-47.75 (Table 1). The total range of TBSA in patients were 15%-60%.

During the study period, samples were collected on 1st day of admission, 3rd day, 5th day and 10th day of admission. A total of 375 swabs were processed for the study. Of these, 105 swabs were sterile (28%) while 270 swabs (72%) gave positive culture. Single isolates were grown in 103 samples (38.15%) and 167 swabs (61.85%) grew multiple isolates (Table 2). Results were analyzed based on the day of hospital admission for better understanding of the colonization pattern during hospital stay. Total number of bacterial isolates identified during

this study were 226. Most common isolate was *Klebsiella pneumoniae* (62) followed by *Pseudomonas aeruginosa* (50). The rest of the isolates were *Proteus mirabilis* (24), *Staphylococcus aureus* (32), *Acinetobacter spp.* (23), CoNS (19), *Escherichia coli* (8), *Providencia stuartii* (3), *Enterobacter spp.* (3), *Klebsiella oxytoca* (1) and *Citrobacter koseri* (1) in that order. Total number of isolates and their distributions are represented in the chart below (Table 3).

Table 1: Age and gender distribution with location of study subjects.

Gender	Frequency	Age (years)	Frequency	(%)
Male	55	≤20	16	16.00
Female	45	21-30	32	32.00
Total	100	31-40	27	27.00
Patient location		41-50	13	13.00
Ward 22	24	51-60	6	6.00
Ward 22A	54	>60	6	6.00
BICU	22	Mean± SD	33.41±14.7	
Total	100	Median (IQR)	31.5 (24-40.75)	
		Range	2-80	

Table 2: Swab culture reports of the study participants.

Culture of Swabs	Frequency
Sterile	105
Non-sterile	270
Single isolates/multiple isolates	103/167
Total	375

DISCUSSION

Burn patients are at increased risk of acquiring burn wound infection caused by highly drug resistant nosocomial bacterial strains. In this hospital based observational study, 100 patients admitted in burn ICU or burn ward were studied for their burn wound isolates and their frequency of isolation. Majority of the patients of this study were in the age group of 21 to 30 years (32%) which is similar to the study findings by Mundhada et al and Priyadarshini et al having 70% and 42% respectively.[6,15] Patients were mostly males (55%) which is similar to the observation made in the study conducted by Jauhari et al with 60.7% patient population as males.[16] But other Indian studies such as Mundhada et al, Priyadarshini et al and Gupta et al had female predominance.[6,15,17] The mean Total Body Surface Area (TBSA) involvement of the study subjects were 39.59±11.61% with a median of 40 and range of 15 to 60%.In our study adult patient with TBSA 20 to 60% and pediatric patient with TBSA from 10 to 60% were included. Mundhada et al had recruited adult patients with TBSA 20 to 40%, Lunawat et al had excluded all the patients with TBSA above 70%, and Priyadarshini et al had excluded all patients with less than 10% TBSA.[6,7,15] Another study by Erol et al done at University hospital, Erzurum, Turkey had a mean the mean TBSA of 22.9% and range 5-75%.[18] Our study was done in both - burn ward and burn ICU, with 78% of patients coming from burn wards. A total of 375 wound swabs cultured, 28% had no isolates, while remaining swabs had bacterial growth indicating an isolation rate of 72%. This observation is corroborated by the finding of Gupta et al, with isolation rate of 61.87% and Jauhari et al, with 70.6% isolationrate.[16,17] There were other studies that show higher isolation rate such as Mundhada et al and Priyadarshini et al with 89.69% and 96% respectively.[6,15] Of the 270 swabs with bacterial isolates, single isolates were grown in 38.15% swabs and 61.85% had multiple isolates. A similar study done by Mundhada et al, which had a much smaller sample size showed a different result.[6] In that study, swabs cultured on day 4, day 10 and day 16 of admission in burn ward were analyzed, and single isolates were seen in 71.28% swabs and multiple isolates were seen in 18.31%.[6] Most of the bacterial isolates in the study were found gram-negative in nature. Of the 226 isolates, 175 bacterial isolates were gram-negative (77%) and 51 isolates were gram positive (23%). Similar findings are reported in studies of Mundhada et al, Priyadarshini et al and Jauhari et al with gram-negative bacteria constituting 72.4%, 59.26 and 60.2%respectively.[6,15,16] *Klebsiellapneumoniae* constituting 62 (27.43%) of the 226 isolates were the most common organism followed by *Pseudomonas aeruginosa*as 50 (22.12%). Study by Mundhada et al showed *Klebsiella pneumoniae* (34.4%) as the most common isolate followed by *Pseudomonas aeruginosa* (23.94%).[6] However, in Chauhan et al and Gupta et al *Klebsiella pneumoniae* was the second most common organism isolated.[8,17] In case of Bhatt et al studies, *Staphylococcus aureus* was the second common isolate.[19]However, comparing this with western reviews and studies (Norbury et al and De Macedo et al) *Staphylococcus aureus* is quoted as the chief causative agent of burn wound infections.[10,20] Other isolates in our study, in the descending order of representation are *Staphylococcus aureus* (14.15%), *Proteus mirabilis* (10.61%), *Acinetobacter spp*. (10.17%), *CoNS* (8.40%), *Escherichia coli* (3.53%), *Providentiastuartii* (1.32%), *Enterobacter spp*. (1.32%), *Klebsiella oxytoca* (0.44%) and *Citrobacter koseri* (0.44%). This cluster of isolates are, more or less similar to other studies like Chauhan et al, Gupta et al and Bhatt et al, showing a common pattern of infections in burn patients.[8,17,19] After analyzing day wise swab culture results, day 1 sample of 100 patients had 87% sterile swabs and rest with isolates. This indicates that majority of the burn patients have sterile wounds at the time of presentation. Day 1 isolates mainly constituted gram-positive organisms (89%). The isolates that were obtained on day 1 consisted mainly of *CoNS* which is a known commensal of skin. Sample collected

on day 3 also had a significantly high number of sterile swabs (53%) with gram-positive organisms like *Staphylococcus aureus* and *CoNS* as the predominant isolates.

Table 3: Distribution of bacterial isolates from the study.

Bacterial isolates	Day 1	Day 3	Day 5	Day 10	Total	
					Frequency	%
Klebsiella pneumoniae	0	8	23	31	62	27.43
Pseudomonas aeruginosa	0	4	28	18	50	22.12
Proteus mirabilis	0	5	11	8	24	10.61
Staphylococcus aureus	2	15	8	7	32	14.15
***Acinetobacter*spp.**	1	7	9	6	23	10.17
CoNS	6	10	3	0	19	8.40
Escherichia coli	0	3	3	2	8	3.53
Enterobacter spp.	0	0	1	2	3	1.32
Providencia stuartii	0	0	2	1	3	1.32
Klebsiella oxytoca	0	0	0	1	1	0.44
Citerobacterkoseri	0	0	0	1	1	0.44
Total	9	52	88	77	Total = 226	

This finding is similar to the study of Mundhada et al, where day 4 swabs had *Staphylococcus aureus* as the most common isolate.[6] Samples of day 5 had only 27% sterile swabs with *Pseudomonas aeruginosa* (23%) as the most common isolate followed by *Klebsiella pneumoniae*. Day 5 samples indicated a shift in pattern, with Gram negative organisms (87%) representing the bulk of isolates. Samples collected on Day 10 had the least number of sterile swabs (22.78%) with *Klebsiella pneumoniae* (30.69%) predominating in the number of bacterial isolates. Gram negative (91%) was the predominant isolate in day 10 samples. *Acinetobacter spp*. have emerged as a significant nosocomial pathogen resulting in considerable morbidity in burns patients. In our study *Acinetobacter spp*. was the fifth (10.17%) most common isolate having more frequency than *Escherichia coli* (3.53%). This is similar toGupta *et al* and Bhatt *et al* who had an isolation rate of 14.83% and 17.27% for *Acinetobacter spp*. respectively.[17,19] Other studies like Mundhada et al, and De Macedo et al recorded a very low isolation rate 2.75% and 3.9% respectively.[6,20] Some of the limitations of the current study was that the anaerobic bacterial isolates in burn wound infection could not be included in the study. Also burn wound histopathologic evaluation for invasiveness of the infective agents could have shed more light on to the significance of all the bacterial isolates tested during our study. Fungal agents are also a known etiologic agent for burn wound infections, which were not covered in our study.

CONCLUSION

In the current study, we tried to shed light on the aerobic bacterial isolates which commonly cause wound infection in burn patients. The results of our study helped to determine the predominance of gram-negative bacterial isolates in burn wounds. *Klebsiella pneumoniae* came out to be the most common bacterial isolate in our study. Also, a surge in isolation rate of *Acinetobacter spp* was seen in our study, more research is required in this regard. The study revealed a time related increase in the colonization of burn wounds, with day 1 and day 3 samples predominating gram positive isolates and day 5 and day 10 samples were predominated by gram negative isolates. Sampling of the patient endogenous flora from various site and from environmental samples and comparison with the isolates from burn wound can give better idea regarding the source of infection and this information can further be used to streamline infection control programs of the institute.

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