ORIGINAL ARTICLE

Incidence, Clinical Evaluation and Antibiogram of Bacterial Isolates Obtained from Burn Patients

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ABSTRACT

Background: Microbiological contamination plays salient role in governing the outcome and time span in the hospital for burn victims in burn unit. Therefore, regular supervision of microbes and its resistance pattern is mandatory. The emergence of Multi Drug Resistant (MDR) bacteria provoked researchers to develop new plans to combat against the threat.

Objective: To assess the infectious organisms, to know the drug resistance of isolates and ascertain the effectiveness of antibiotics against microbes that are found in burn patients.

Methodology: Descriptive case series study performed and Quantitative data was collected in Jan 2019 to April 2019. Two hundred sample from burn patients admitted in Pak Italian Burn Unit Multan was collected, processed for culture, bacterial identifications and to test the antibiotics sensitivity in accordance with CLSI (clinical and laboratory standards institute) guidelines.

Results: Gram negative bacteria were 189 (94.5%) and gram positive were 11 (5.5%).Organisms isolated were Proteus vulgaris 53 (26.5%) followed by Escherichia coli 47 (23.5%), Pseudomonas aeruginosa 45 (22.5%), Enterobacter 44 (22%) and Staphylococcus aureus 11 (5.5%). Imipenem 156 (78%), Meropenem150 (75%), Moxifloxacin 17(8.5%), Levofloxacin 67 (33.5%), Piperacillin/Tazobactam 123 (61.5%), Oxifloxacin 22 (11%) and Amikacin 16 (8%) were used for antibiotic sensitivity.

Practical implication: Our study results can helpful for all health care personals to select new regime of antibiotics which significantly declined in all types of burn patients and also we are able to decrease the mortality rate in such patients. Assertive contagion limitations can helpful in confining the disease and proliferation of MDR pathogens.

Conclusion: This study proved to be successful because antibiotic associated complications were decreased significantly as a result mortality rate declination was also noticed in all the types of burn patients.

Keywords: Antibiogram, Burn, Clinical evaluations, Infection, Multi drug resistance (MDR), Mortality.

INTRODUCTION

Burn is defined as injury present in the epidermis, inner layer or the inner most layer of skin because of heat, electricity, oil, or other hot liquids etc. Historically, grading of burn was established by Hilden's in 1607. Burn constitutes one of the most important causes of morbidity and mortality worldwide.[1] Reported burn injuries are 2,500,000 out of which become 100,000 hospitalized and 12,000 die annually. Burn is the second leading cause of trauma deaths after motor vehicle accidents[2]. As stated by WHO in 2016, most of the burns are because of heat from fire, smoke and hot water. According to a more recent research, causes of burn include thermal or hot liquid (scald), 68.7%; hot material injury, 3.7%; flame burn, 1.5%; and chemical burn, 1.2%. In this regards burns are of different types caused by electricity, chemicals, radiations, heat and scald. All these types can make lethal impact on patient's body if left untreated[3]. Degree of burns is helpful in better assessment of patients. 1st and 2nd degree burns involve epidermis and dermis with intense pain and minimal scarring while 3rd and 4th degree burns involve subcutaneous tissue with minimal pain but severe scarring. In burn laboratory, diagnosis is very crucial to evaluate burns clinically and to prevent development of complications especially among burn patients. Blood count, coagulation profile, renal parameters, C-reactive protein (CRP) and serum electrolytes are important in detection and evaluation of contamination in burn wound leading to inflammation. Wounds of burn patients are ideal for growth of opportunistic organisms[4].

Normally, burn wounds remain sterile initially and after 24 hours, colonization of gram negative bacteria usually begins. Pseudomonas aeruginosa is the foremost and prominent microbe that is isolated from burn wounds; some anaerobic organisms are also involved including bacteria like Bacteroides fragilis, Pepto-streptococcus, Fusobacterium, Enterobacter and E. coli, and fungi like Zygomycetes and Aspergillus niger. There is a peculiarity about Staphylococcus aureus in this regard that it is a common colonizer that is invulnerable to the most of antimicrobial drugs. After years of research, it is found that pervasiveness of Pseudomonas aeruginosa is decreasing and antibiotic resistance has developed among those gram negative bacteria that were not previously considered as important or predominant microbes. Different antibiotics have been used in the past to treat burn and its related complications. Amikacin and Fluoroquinolones were the most prominent drugs used against gram negative bacteria especially in burn patients[5,6].

This study was performed to evaluate bacterial prevalence and antibiotic sensitivity pattern in adult burn patients, because decline in antibiotic associated complications may also lead to significant mortality rate declination in all types of burn patients.

MATERIALS AND METHODS

Samples were taken from Pak Italian Burn Unit (PIBU) Multan from January 2019 to March 2019 for this descriptive case series study using non-probability purposive sampling. Patients of both genders with the history of burns having TBSA 20 % admitted to PIBU belonging to age group 15 to 60 years were included after taking their informed consent. Patients with some kind of terminal illness or other chronic conditions like hypertension and diabetes were excluded from this study. Clinical evaluation of burn patients was also integral part of our research. Base line test e.g. LFT, RPM, S/E, Hb and coagulation profile were performed to evaluate patients clinically as early diagnosis.

Samples were collected using sterile swab and transported in Amie's transport media. Samples consisted of pus from wounds, wound debris with complete aseptic precautions and were transported immediately to microbiology lab where these samples were inoculated onto Blood and MacConkey agar and incubated at 37°C for 24 hours. Slides were prepared for gram staining and then proceed it for microscopy to differentiate between gram positive and gram negative bacteria. Conventional methods were used for identification of disease causing bacteria outlying in pure cultures. Morphology emergence of colonies on culture medium, appearance under the microscope accustomed biochemical were used to attribute bacteria and identify them. Catalase and coagulase tests were performed to differentiate between Staphylococcus aureus and other gram positive bacteria. Triple

Sugar Iron, citrate and indole tests were performed to identify Escherichia coli and other gram negative bacteria (Table 1).

To check antimicrobial sensitivity by disk diffusion method, antibiotics like Moxifloxacin, Piperacillin/Tazobactam, Amikacin, Imipenem, Meropenem and Levofloxacin were used.

Collected data was analyzed by SPSS – 22 to calculate mean and standard deviation for numerical values like age, sex, type of burn and socioeconomic status. SProportions in terms of frequencies and percentages were recorded for gender, residential status, socioeconomic status, type of burn, age groups, causative organisms and their antibiotic sensitivity pattern.

Table 1: Biochemical Tests

Microbes	Enterobacter	S. aureus	E. coli	P. vulgaris	P. aeruginosa
Catalase	-	+	-	+	+
Indole	-	-	+	+	-
Oxidase	-	-	-	-	+
TSI	A/A	-	A/A, gas production	AK/A with H2S production	AK/AK
Citrate	+		-	+	+
H2S	-		-	+	+
Coagulase	-	+	-	-	-
Motility	-	-	+	+	+

RESULTS AND DISCUSSION

Our study included 200 burn patients with 53.5 % male patients and 46.5% female patients with age ranging between 15 to 60 years. 90% were from rural areas and 10% were from urban areas[7]. Burns included in this study included flame (68%), electric (19%) and scald (13%). Bacterial isolates were Proteus vulgaris (26.5%), Escherichia coli (23.5%), Pseudomonas aeruginosa (22.5%), Enterobacter (22%) and Staphylococcus aureus (5.5%).[8] (Table 2)

Table 2: Clinical Evaluation of Burn Patients

Lab reports	Pathology			
	Flame burn n=136 pathological data	Electric burn n=38 pathological data	Scald burn n=26 pathological data	Normal
Liver function test	94	26	18	62
Serum electrolyte	110	30	21	39
Hemoglobin	21	12	08	159
Coagulation profile	23	11	05	161
Renal function test	30	10	06	154

Antibiotic sensitivity performed using Müller Hinton agar for all gram positive and negative bacteria. Results were interpreted according to clinical laboratory and standard institute (CLSI) 2019 guidelines.

Mortality rate of burn patients after use of new regime of antibiotics significantly declined in all types of burn patients like in flame burn 43/136 (32%), electric burn 07/38 (18%), scald burn 03/26 (12%). Overall mortality rate in all type of burns is 31%[9] (Table 4). All data were taken from PIBU.

Table 3: Antibiotics Sensitivity Patterns of Isolated Bacteria

Antibiotics	Imipenem	meropenem	Moxifloxacin	Levofloxacin	Piperacillin/Tazobactam	Oxifloxacin	Amikacin
Bacteria	Sensitivity (%)	Sensitivity (%)	Sensitivity (%)	Sensitivity (%)	Sensitivity (%)	Sensitivity (%)	Sensitivity (%)
P. vulgaris n=53	79.21	75.40	9.40	20.72	62.22	11.32	7.51
E. coli n=47	80.85	78.72	14.81	12.76	65.91	0.00	12.76
P. aeruginosa n= 45	71.11	66.66	15.55	20	60	11.11	20
Enterobacter n=44	86.36	79.54	0.00	25	68.18	11.36	9.09
S. aureus n=11	90.90	72.72	27.27	36.36	54.54	0.00	18.18

Table 4: Comparison Between new and old Antibiotics Applied on Burn Patients

Antibiotics (old class)	Mortality rates Sept-Dec 2018	Antibiotics (new class)	Mortality rates March-July	Types of burn
Co-amoxiclave Amikacin Tetracycline Ciprofloxacin Netlimycin	47%	Carbapenem Cotrimazole Cefotaxime Levofloxacin Piperacillin/ Tazobactam	32%	Flame
	26%		18%	Electric
	19%		12%	Scald

A three year retroactive study was performed in China in order to evaluate and check the antibiotic resistance and bacterial distribution (especially disease causing bacteria) among burn patients. EMR system performed to get data on 1449 hospitalized patients from Fujian Medical University Union Hospital[10].

Out of 3835 samples, 1891 strains of disease causing bacteria were found and the total detection rate was 49.3% (1891/3835).[11] The active microbes were gram-positive bacteria (689 strains; 36.5%) and gram negative bacteria (1089 strains; 57.7%), Predominant bacteria Staphylococcus aureus (19.0%), fungi (113 strains; 6.1%) Acinetobacter baumanni (17.5%), Pseudomonas aeruginosa (16.6%), Enterococcus faecalis (4.4%), and Klebsiella pneumoniae (7.5%). MRSA accounted for Staphylococcus epidermidis accounted for 40.7% (69/170), coagulase negative staphylococcus species (S. aureus 74.1% (265/359), 72.5% (50/69) of which were MRSE. Unfortunately, MRSE and MRSA were resistant to ampicillin and penicillin.[12]

One of the gram negative bacteria Acinetobacter baumanni is the most prevalent bacteria which completely resistant to aztreonam, amoxicillin, ampicillin and co-amoxiclave. Numerous amount of K. pneumoniae were resistant to ampicillin, cefazolin and amoxicillin. Mainly Escherichia coli isolates were unaffected with sulfamethoxazole, piperacillin, ampicillin, cefazolin and tetracycline. Discernment rates of ESBL among K. pneumoniae and E. coli and isolates were 67.3% (41/61) and 44.7% (62/139). Antibiotics having more efficacy included vancomycin, Linezolid, tigecycline and teicoplanin.[13]

In our study, clinical evaluation, antibiogram and incidence of bacteria isolated from burn patients are evaluated. Clinically isolated bacteria obtained from this research are mostly resistant to most of the antibiotics. These bacteria are proteus vulgaris, Escherichia coli, Enterobacter, Pseudomonas aeruginosa and Staphylococcus aureus. We have found Proteus vulgaris is the most pervasive gram negative bacteria in burn patients. The increase prevalence of Proteus vulgaris in hospital is contrasting from United State, Europe and South America[14].

A total of 200 burn patients tested of both genders between age of 15 to 60 years presenting with flame, electric and scald burn will be included in the study and admitted in the inpatient department of plastic surgery Pak Italian Modern Burn Center, Nishtar Hospital Multan, a tertiary care govt. hospital in South Punjab. Out of 200 samples, proteus vulgaris is the most pervasive microorganism 53 (26.5%) followed by Escherichia coli 47 (23.5%), Pseudomonas aeruginosa 45 (22.5%), Enterobacter, P. vulgaris 44 (22%) and Staphylococcus aureus 11 (5.5%). Only one Gram

positive bacteria isolated out of five gram negatives. Attributed morphological emergence of colonies on gram staining and on media and accustomed biochemical tests including catalase, Coagulase, urease, hydrogen sulfide and sugar utilization and citrate, Oxidase and indole were used to attribute bacteria and identify them because of the high antibiotic resistance, complications and mortality rate increases day by day. Despite of the advance technology and researches, pathogenesis and antibiotic.[15]

Resistance mechanisms, Enterobacter and P. aeruginosa were found to be MDR. All isolates, in our study were found to be imipenem and meropenem sensitive. A strict and periodic vigilance is required to know the pattern of the distribution of the organisms among burn patients[16].

Our medical unit is in South Punjab where the climate is warm sometimes moist and hard, this hypothesis could be an explanation for our results also and should be further studied. In nutshell, the results of this study exhibited that, patients' restored clinically because of better/more stable lab reports including liver function tests, serum electrolytes, hemoglobin, renal and coagulation profiles due to the use of new regime of antibiotics. This study proved to be successful because mortality rate declination in all the types of burn patients especially when we compare old and new regime of antibiotics and antibiotic culture and antibiogram shows that drugs (antibiotics) associated complications were decreased significantly. MDR bacteria are serious threat to burn patients. Assertive contagion limitations can helpful in confining the disease and Proliferation of MDR pathogens.

CONCLUSION

- The result of our study showed that Proteus vulgaris followed by Enterobacter, E. coli and P. aeruginosa are the leading cause of contamination in burn patients.
- Only imipenem and meropenem drugs are ray of hope for managing burn patients because they are effective against gram negative organisms especially in patients effected with electric, scald and flame burn injuries. .

- The results from our study spotlight about the incidence, clinical management and antibiogram of bacterial isolates in burn patients.

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