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Original Article

Antimicrobial resistance pattern of bacterial isolates from burn wounds in an Iranian University Hospital

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ABSTRACT

Objective: About 73% of death cases in the first 5 days after burning are due to infection complications. The aim of this study was to identify the causing agents of infections in burn patients and the sensitivity pattern of them to the commonly used antimicrobials in an Iranian Burn center University Hospital.

Methods: In this cross-sectional study, patients who were admitted to one of the Iranian Burn center University hospitals in 2009 and had nosocomial infection due to burn wound, whom received antimicrobial agents for therapeutic reasons, with a hospitalization period of more than 48 hours were enrolled. Gram stain analyses were performed to help identifying growing colonies. Differential tests for identification of pathogenic bacteria species were performed following primary tests. E-test strips of each antimicrobial were placed on the culture medium plate in order to determine the minimum inhibitory concentration Studied antimicrobials for isolated Gram-negative bacteria were meropenem, piperacillin/tazobactam, ceftriaxone, cotrimoxazole, and for *Staphylococcus aureus*, vancomycin, piperacillin/tazobactam, cotrimoxazole, and cephalothin.

Findings: Only 16% of *Pseudomonas aeruginosa* species were sensitive to meropenem, and 13% were sensitive to piperacillin/tazobactam. Ten out of 29 *Klebsiella* species (34%) were sensitive to meropenem and piperacillin/tazobactam. All isolated strains of *Staphylococcus aureus* were sensitive to vancomycine while they were all resistant to cotrimoxazole.

Conclusion: *Pseudomona, Klebsiella* and *Staphylococcoci* are the most common species causing burn infection in this medical center. Results showed the importance of limiting irrational use of wide-spectrum antimicrobials and recommends strict management of infections in burn injury centers.

Keywords: Antimicrobial drug resistance; burns; wound infection

INTRODUCTION

Burn patients are among the most critically ill patients a physician can ever visit in his vocational lifetime. These patients are at the risk of failure of different organs, and in those who survive the acute phase; infections are the most common cause of mortality.[1] In fact more than 75% of deaths due to burn in severe burned patients are attributed to sepsis, infection complications and inhalation injury.[2,3]

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Burn wounds have a better prognosis if they are not infected, but the condition of such wounds along with other factors predisposes them to infection. About 73% of death cases in the first five days after burning are said to be due to infection complications.[4] Skin destruction due to heat and simultaneous suppression of cellular and humoral immune system leads to burn infection.[5-7] Although burn wounds are sterile at first, finally microorganisms grow there,[8,9] which can lead to infection based on the nature and

extension of the wound, and the species and number of microorganisms. Paying enough attention to the infection of burn wounds is of utmost importance because this contamination may delay the healing process[10] and may lead to bacteremia, sepsis and finally multiple organ dysfunction.[11,12] On the other side, this will increase the risk of microbial resistance especially for nosocomial infections. Therefore, identifying causing bacteria and their resistance pattern to antimicrobials is essential in burn management centers. Despite the different patterns of antimicrobial sensitivity, Methicillin-resistant *Staphylococcus aureus* (MSRA), methicillin-resistant coagulase-negative *Staphylococci*, vancomycin-resistant enterococci (VRE) and multiple drug-resistant gram-negative bacilli including *Pseudomonas aeruginosa* and Acinetobacter are considered as the most important nosocomial pathogens.[1] As a result, regular audits and evaluations for the pattern of antimicrobial resistance especially for major pathogens seem to be essential for empiric therapy in each medical center.

In this study, we have identified the causing agents of infections in burn patients and the sensitivity pattern of them to the most commonly used antimicrobials in an Iranian Burn center University Hospital.

METHODS

In this cross-sectional study, which was conducted in Imam Mousa Kazem [PBUH] Burn center University Hospital (affiliated to Isfahan University of Medical Sciences, Iran) in 2009, burn wound samples of 81 burn patients who had nosocomial infection due to burn wound[13] with a hospitalization period of more than 48 hours were studied. Infection was diagnosed by the attending physician according to the standard definition of nosocomial infection of the wound.[13]

A sterile swab was rubbed on the wound for sampling, and then it was transferred to transport medium of Stuart. Samples were sent to the microbiology lab of medicine faculty for diagnostic tests. In order to isolate Gram-positive and Gram-negative bacteria, swab samples were removed from Stuart transport medium and were used to inoculate various culture media including McConkey agar, Blood agar, Eosin Methylene Blue (EMB) by streaking method. Cultures were incubated for 18-24 hours at 37°C and Gram stain analyses were then performed to help identifying growing colonies. Differential tests for identification of bacterial species [including catalase test, culture on mannitol salt agar and coagulase test for *Staphylococci* and Triple Sugar Iron (TSI), citrate consumption test, urea degradation test and oxidase spot test for differentiating gram negative bacteria] were performed following primary tests. After grown bacteria were indentified, pure colonies were isolated in 24 hours, and several colonies were prepared in saline 0.85% suspension with standard turbidity of 0.5 McFarland under aseptic conditions. Then, a plate containing Mueller-Hinton Agar culture medium was smeared with the microbial suspension using a sterile swab. E-test strips of each antimicrobial were placed on the culture medium in order to determine the minimum inhibitory concentration (MIC). After 24 hours of incubation at 37°C, an oval-shaped inhibitory zone was seen around E-test strips. The crossing point of this halo with the strip indicates MIC in μg/ml. After reading MIC data, they were compared with tables of clinical and laboratory standard institute (CLSI) and the results were reported as sensitive, resistant and intermediately sensitive.[14]

Studied antimicrobials for isolated Gram-negative bacteria were meropenem, piperacillin/tazobactam, ceftriaxone, cotrimoxazole, and for *Staphylococcus*, vancomycin, piperacillin/tazobactam, cotrimoxazole and cephalothin.

RESULTS

In this study, 43 (53.1%) out of 81 hospitalized patients were men. Their age ranged from 1 to 83 years old. Multiple burns comprised most of the burns (68 cases) while eight people had lower limb burn and four people had upper limb burns, and one person had body and abdomen burns.

In 37 patients, only one bacterial specie, in 38 patients, two bacteria species, and in 6 patients, 3 bacterial species (in sum, 131 species) were isolated. *P. aeruginosa* was the most commonly isolated bacteria with 62 cases (47.3%). Other bacterial species isolated were *Klebsiella* (31 cases, 23.7%), *Staphylococcus aureus* (25 cases, 19.1%), *Acinetobacter* (7 cases, 5.3%), *Enterobacter* (4 cases, 3.1%), *Proteus mirabilis* and *Escherichia coli* (1 case for each, 0.8%).

Table 1 shows the resistance of isolated bacteria to studied antimicrobials. As can be seen, only 16% of *P. aeruginosa* species were sensitive to meropenem, and 13% were sensitive to piperacillin/tazobactam. Of 29 *Klebsiella* species, 10 species (34%) were sensitive to meropenem and piperacillin/tazobactam. All isolated species of *S. aureus* were sensitive to vancomycine while they were all resistant to cotrimoxazole.

DISCUSSION

In the order of frequency, the studied bacteria in the present research were *P. aeruginosa, S. aureus, Acinetobacter, Enterobacter, E.coli,* and *P. mirabilis*.

Table 1: Rate of bacterial resistance to the commonly used antimicrobials in the studied burn wound samples

Bacterium	Antimicrobials					
	Meropenem	Ceftriaxone	Cotrimoxazole	Tazocin	Cefalotin	Vancomycin
Pseudomona aeruginosa	84%	95.5%	100%	87%	-	-
Klebsiella spp.	66%	76%	100%	63%	-	-
Staphylococcus aureus		-	100%	16%	36%	0
Acinetobacter	57%	71%	100%	57%	-	-
Enterobacter	50%	75%	75%	25%	-	-
Escherichia coli	0	0	0	0	-	-
Proteus mirabilis	0	0	0	0	-	-

Similar results were reported as per major infecting microorganisms of burns in some other studies done in Iran.[4,15,16] In one study in 2003 on 170 burn patients in a teaching University Hospital located in Shiraz (Iran), *P. aeruginosa* was the most common infecting agent of burns (54.4%), followed by coagulase-negative *Staphylococcus, S. aureus, Klebsiella,* and *E.coli* with 5%, 3.26%, 1.75%, and 1.25%, respectively.[15] In another study by Ekrami and Kalantari, which was accomplished in a burn injury hospital in Iran, it was revealed that the principal pathogens of nosocomial infections (76.9% of which were wounds) included *Pseudomonas aerogonosa* (37.5%) *S. aureus* (20.2%) and *acinetobacter* (10.4%) (4). Rastegar *et al* also introduced *Staphylococci* as main cause of hospital-acquired infections in burn-injured patients.[16]

In some other studies, *P. aeruginosa* was introduced as the most common infecting agent in burns.[17,18] A few studies have introduced another bacterium (except *P. aeruginosa*) as the infecting agent of burns. Santucci *et al*. found *S. aureus* as the major cause of infection,[19] while two other studies found *Staphylococcus epidermidis* as the main pathogen.[20,21]

In the present study, isolated pseudomona species were all highly resistant to all antimicrobials. A study in Ghotbodin Hospital in Shiraz, aiming to study the sensitivity pattern of isolated *P. aeruginosa* from burn patients, showed the sensitivity of isolated *P. aeruginosa* species to imipenem, meropenem, and ciprofloxacin. Compared to our study, more isolated species were sensitive to meropenem in this study (36.7%).[18] The resistance of *P. aeruginosa* to new antimicrobials is a serious and major problem in burn patients in hospitals. High resistance of this bacterium to the antimicrobial agents has complicated the treatment of infections caused by this bacterium, and made it one of the major medical predicaments. Despite applying anti-*Pseudomonas* antibiotics, this pathogen is one of the most difficult causing agents related to morbidity and mortality of burn patients in the world.[19,20]

In the present study, all *Staphylococcus* species were sensitive to vancomycin which is consistent with the results of Ekrami and Kalantar.[4] Furthermore, another study found some *Staphylococcus* species highly resistant to cephalothin, which is a first-generation cephalosporin (36%). Therefore, it cannot be trusted for treating severe infections due to burn.

Considering the high resistance of all isolated species (*Staphylococcus* and Gram-negative bacteria) to cotrimoxazle, it can be inferred that this antimicrobial may have no place in treatment of burn infection in this University burn management center. These results, shows the importance of limiting irrational use of wide-spectrum antimicrobials and strict management of infections in burn injury centers. The most important undertakings to control and prevent infections include (but not limited to): The isolation of patients in individual rooms and avoid transportation of medical and surgical instruments to multiple rooms, persistent disinfection of instruments, wearing gloves by medical and nursing staff, sterile dressings and face masks and hand washing before and after visiting patients.[1] Moreover, preventive use of topical antimicrobials (e.g., silver sulfadiazine and mafenide) may lead to a decrease in microbial burden and causative infections.[22-23] These, of course would not obviate the need for determination of antimicrobial sensitivity of pathogens in separate studies.

Finally in this study we have showed that *Pseudomona, Klebsiella* and *Staphylococcus* were the most common species causing burn infection in Imam Mousa Kazem [PBUH] burn center University hospital. Antimicrobial therapy for burn patients should cover these pathogens although the resistance of bacteria (especially gram negative species) to the studied wide spectrum antimicrobials was too high. Regarding to the high resistance of these prevalent pathogens to commonly used antimicrobials in burn wounds, preventive measures to essentially avoid infections in burn wounds should be considered by the infection control committee of hospitals. This may consequently limit the unreasonable application of antimicrobials.

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AUTHORS' CONTRIBUTION

All authors contributed the idea of research, design of study, data analysis and manuscript preparation.

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