



Prevalence and Antibiotic Resistance Pattern of Gram-Positive Isolates from Burn Patients in Velayat Burn Center in Rasht, North of Iran

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ABSTRACT

Background and objectives: Bacterial contamination of wounds is a serious problem, particularly in burn patients. Gram-positive bacteria are the predominant cause of infection in newly hospitalized burn cases. This study aimed to survey the prevalence and antibiotic resistance pattern of gram-positive bacterial isolates among burn patients in Rasht, North of Iran.

Methods: This cross-sectional study was conducted on burn patients with a positive culture for gram-positive isolates who were hospitalized in the Velayat Burn Center in Rasht, North of Iran, during 2017-2020. The isolates were identified using standard microbiological methods. Moreover, the antibiotic resistance pattern was determined by the disk diffusion method.

Results: During the study period, 671 bacterial cultures were obtained, of which a total of 16 gram-positive isolates were taken from the patients. The frequency of coagulase-negative staphylococci (CoNS), *Staphylococcus aureus*, and *Enterococcus* spp. was 68.7%, 18.8%, and 12.5%, respectively. In addition, the highest rate of resistance in CoNS isolates was against trimethoprim/sulfamethoxazole. The highest rate of resistant among *S. aureus* isolates was recorded against penicillin. Moreover, *Enterococcus faecalis* isolates showed a high level of resistance to ampicillin, erythromycin, tetracycline, gentamicin, and ciprofloxacin. All isolates were susceptible to teicoplanin. Moreover, the frequency of methicillin-resistant *S. aureus* isolates was 66.7%.

Conclusion: Given the increasing prevalence of drug-resistant strains, especially in susceptible burn patients, it is imperative to analyze the bacterial etiology of nosocomial infections periodically and epidemiologically.

Keywords: [Staphylococcus aureus](#), [Enterococcus](#), [Burns](#), [Gram-positive bacterial infections](#).

INTRODUCTION

One of the main problems in trauma wards of hospitals is burn wound infections, which are mainly caused by bacteria (1, 2). Pneumonia and blood infections such as septicemia and wound infections are the leading cause of mortality in burn patients (2). Wounds are naturally suitable for colonization of bacteria; therefore, burn patients are at great risk of life-threatening complications (3). Emergence of multidrug-resistant (MDR) strains that carry transferable plasmids, integrons, or other transferring elements is another health problem (4, 5).

Bacterial contamination of wounds could have either an endogenous (microorganisms in nasopharynx, gastrointestinal tract, and the surrounding skin) or exogenous (healthcare settings) origin (6). The prevalence of nosocomial infections caused by gram-positive bacteria, particularly MDR strains, is on the rise (7). The most commonly isolated gram-positive bacteria are *Staphylococcus aureus*, coagulase-negative staphylococci (CoNS), and *Enterococcus spp.*, which are recognized as important causes of nosocomial infections in the world (8).

Enterococci, especially vancomycin-resistant enterococci (VRE) infections, are becoming common and challenging to treat. In addition, the increased incidence of methicillin-resistant *S. aureus* (MRSA) and contingency of vancomycin resistance highlights the need for rapid and reliable characterization of isolates and control of MRSA and VRE spread in hospitals (9-11). Due to the importance and increased rate of antibiotic resistance, it is essential to prevent or control pathogens in clinical settings (12-14). Therefore, the present study aimed to investigate the prevalence and antibiotic resistance pattern of gram-positive bacteria isolates from burn patients in the North of Iran.

MATERIALS AND METHODS

This retrospective cross-sectional study was performed on burn patients admitted to the Velayat burn injuries hospital in Rasht, North of Iran, from March 2017 to September 2020. The study was approved by the Ethics Committee of the Guilan University of Medical Sciences (ethical code: IR.GUMS.REC.1399.276) and was performed according to the Helsinki's declaration. Demographic and clinical information of

patients who had a positive culture for a gram-positive isolate were collected.

Superficial cleaning of the wounds was performed using normal saline. Then, each sample was taken by rotating a sterile, pre-moistened swab across the wound surface of a 1 cm area in a zig-zag motion, from the center to the outside of the wound. Next, the swab was placed in an enrichment tube and transferred to the Microbiology Laboratory for testing. Briefly, each swab was plated into blood agar, mannitol salt agar, and bile esculin agar and incubated overnight at 37 °C. All culture media were purchased from Merck, Germany. Grown isolates were identified by morphological examination, followed by biochemical testing using the conventional microbiological tests (15, 16).

Antibiotic susceptibility testing: The antibiotic susceptibility pattern of gram-positive isolates was determined by the disk diffusion method according to the Clinical and Laboratory Standards Institute (CLSI) recommendations (17). The antibiotics used were ampicillin, penicillin, cefoxitin, erythromycin, tetracycline, teicoplanin, gentamicin, ciprofloxacin, clindamycin, trimethoprim/sulfamethoxazole, and rifampin. All antibiotic disks were purchased from Oxoid, UK. The *S. aureus* strain ATCC 25923 was used for quality control. In addition, MDR was defined as acquired non-susceptibility to at least one agent in three or more antimicrobial categories (18).

Data analysis was performed using SPSS™ software (version 21, IBM Corp., USA). The results are presented using descriptive statistics in terms of relative frequency.

RESULTS

During the study period, 671 bacterial cultures were obtained, of which a total of 16 gram-positive isolates were taken from burn patients. Overall, 68.7 % (11/16), 18.8% (3/16), and 12.5 % (2/16) of the isolates were CoNS, *S. aureus*, and *Enterococcus spp.*, respectively. Out of the 16 culture-positive samples, 56.2% (9/16) belonged to males and 43.8% (7/16) to females.

The patients' median age was 35.3 years (age range: 2 to 76 years). The isolates were taken from the surgery ward and intensive care unit (ICU, 81.3%). Most of the isolates (93.8%) were taken from wounds. Of 16 culture

positive cases, two (12.5%) patients died (Table 1).

The antimicrobial-susceptibility patterns of the gram-positive isolates are shown in table 2. Overall, 87.5% (14/16) of isolates were MDR. The resistance pattern of CoNS showed that the highest resistance rate was against trimethoprim/sulfamethoxazole (81.8%), followed by clindamycin (72.7%), and penicillin (72.7%), whereas the lowest

resistance was to gentamicin (45.4%). Enterococci isolates were almost resistant to all tested antibiotics, except ampicillin and rifampin. Moreover, 66.7% of isolates were MRSA based on the ceftoxitin disk diffusion method.

In *S. aureus* isolates, the resistance rate was highest against penicillin (100%) and lowest against gentamicin (33.3%) and rifampin (33.3%).

Table 1- The demographic and clinical characterization of burn patients' with bacterial infections

Variable	Number	Percent
Wards		
Surgery	3	18.7
ICU	13	81.3
Outcome		
Death	2	12.5
Discharge	14	87.5
Type of sample		
Wound	15	93.8
Tissue	1	6.2
Median of age (range)	35.3 (2-76 years)	-
Hospitalization duration	31.8 ± 91.5 days	-
Hospitalization until infection diagnosis	2.37 ± 2.5 days	-

Table 2- The antimicrobial-susceptibility patterns of isolates taken from burn patients

Antimicrobial category	Antimicrobial agent	Enterococci (N=3)	CoNS	<i>S. aureus</i>
		No. (%)	(N=11) No. (%)	(N=3) No. (%)
Penicillins	Ampicillin	1 (33.3)	-	-
	Penicillins	-	8 (72.7)	3 (100)
	Ceftoxitin	-	6 (54.5)	2 (66.7)
Macrolide	Erythromycin	2 (66.7)	7 (63.6)	2 (66.7)
Tetracyclines	Tetracycline	2 (66.7)	6 (54.5)	2 (66.7)
Lipoglycopeptide	Teicoplanin	0	-	-
Aminoglycosides	Gentamicin	2 (66.7)	5 (45.5)	1 (33.3)
Fluoroquinolones	Ciprofloxacin	2 (66.7)	6 (54.5)	2 (66.7)
Lincosamide	Clindamycin	-	8 (72.7)	2 (66.7)
Sulfonamides	Trimethoprim/sulfamethoxazole	-	9 (81.8)	2 (66.7)
Ansamycins	Rifampin	1 (33.3)	3 (27.3)	1 (33.3)

DISCUSSION

Bacterial contamination of wounds is a serious problem, particularly in burn patients. This often has poly-microbial nature, and the presence of MDR microorganisms frequently collaborates with vigorous clinical appearance and insignificant response to antimicrobial therapy (19). Nevertheless, proper identification of pathogenic microorganisms in burn hospitals' and determining their susceptibility to commonly used antibiotics will help clinicians manage wound infection more efficiently. In the present study, CoNS and *S. aureus* were the most common gram-positive isolates, which is similar to findings

of a study (20) but inconsistent with others (21, 22). Generally, difference in the frequency of isolates could be related to geographical location, hygiene measures, and the number of isolations. Other studies in Iran also showed that the number of nosocomial infections caused by enterococci, particularly resistant strains, is growing (23, 24).

In 2001, the National Nosocomial Infection Surveillance System stated that the incidence of burn wound infections caused by *Enterococcus* was 11% (5). In this regard, our country experienced a dramatic increase in rate of antibiotic resistance for enterococci (21).

This is an alarming issue for the healthcare system (25). In the present study, we found no VRE isolate, which is similar to results of a similar study in Tehran (26). However, other studies have isolated VRE strains from burn wound infections (27, 28). That is an alarming issue for the hygiene system and medical healthcare (26). What found no VRE isolate in our study, which is similar to findings of Heidari et al. (25) and in contrast with some previous studies (27, 28). These discrepancies could be due to the difference in geographical location and antibiotic prescriptions in the study area.

In a study conducted by Chirife et al., *S. aureus* was the most common gram-positive bacteria isolated from wound infections (29). In some countries such as Iraq, *S. aureus* has been considered as a significant cause of nosocomial infection in burn patients (30). Our results demonstrated that CoNS was the most prevalent gram-positive isolate taken from burn patients.

The emergence of worldwide antibiotic resistance, particularly among nosocomial pathogens, limits the efficiency of available antibiotics for treatment of burn wound infections (20, 21, 31). The frequency of MRSA and MDR isolates in our study was significantly higher than that in other studies (32-34). This might be related to the lack of awareness in taking care of burn wounds, horizontal transmission of MDR by the healthcare staff, transferring resistance genes by mobile genetic elements, and non-adherence to the hospital guidelines. The prevalence of MDR and MRSA strains among burn patients in hospitals is alarming. However, periodic monitoring of commonly used antibiotics and susceptibility information can help to overcome the emergence of drug-resistant strains.

Retrospective studies are not without limitation. In the present study, we only studied a single referral hospital, which makes results difficult to generalize. Second, because of the study design, additional clinical information could not be provided. Despite these limitations, this study provides useful information regarding microbiological aspects of gram-positive bacterial infections among burn patients.

CONCLUSION

According to the results, the prevalence of MDR and MRSA isolates is alarmingly high in hospitalized burn patients. Given that a high proportion of mortalities in burn patients is because of nosocomial infections, particularly those caused by resistant strains, it is essential to conduct periodical and epidemiological studies of nosocomial diseases.

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Ethics approvals and consent to participate

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Conflict of interest

The authors declare that there is no conflict of interest regarding publication of this article.

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