High Frequency of Methicillin-Resistant \textit{Staphylococcus aureus} in Intensive Care Unit in Karaj, Iran

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Abstract

\textbf{Objectives:} The current study aimed at finding the frequency of MRSA infections, contamination, and colonization in the teaching hospitals of Karaj city, Iran for the first time.

\textbf{Methods:} The current cross sectional study was conducted in Karaj on three teaching hospitals from July 2013 to July 2014. Sample collection from personnel and surfaces was conducted twice and monthly, respectively, during the study period. Also, all \textit{Staphylococcus aureus} species isolated from patients were included in the study. Antimicrobial susceptibility test was performed by the standard disk diffusion method. All isolates were subjected to \textit{mupA} and \textit{mecA}-specific polymerase chain reaction (PCR) to identify high-level mupirocin-resistant and MRSA isolates, respectively. Chi-square test was employed for data analysis.

\textbf{Results:} The majority of \textit{S. aureus} species were isolated from personnel and surfaces of the hospitals. One hundred sixty-eight \textit{S. aureus} and 49 MRSA species were isolated from Karaj teaching hospitals. The main frequency of MRSA was isolated from intensive care unit (ICU) (75%) and high rate of resistance to rifampicin (53%) was observed in MRSA isolates. Although 10 \textit{S. aureus} species were resistant to mupirocin by disk diffusion, no \textit{mupA} gene was detected in the isolates.

\textbf{Conclusions:} In conclusion, in comparison with the other studies from Iran, low frequency of MRSA was observed in the investigated hospitals. However high frequency (75%) of MRSA in ICU indicated that antibiotic policy is urgently needed to prevent the resistance development. Moreover, antibiotic susceptibility monitoring and regular screening surfaces and personnel of hospitals in terms of MRSA colonization, especially ICU, are indispensable.

\textbf{Keywords:} Methicillin-Resistant \textit{Staphylococcus aureus}, MRSA, Intensive Care Unit, ICU, Iran

1. Background

Nosocomial infection rate in ICUs is one of the most common hospital-acquired infections. High mortality, increased healthcare costs, and prolonged hospital stays are the results of infections caused by methicillin-resistant \textit{Staphylococcus aureus} (MRSA) (1-3). The intensive care unit (ICU) is more frequently colonized with MRSA rather than other departments in hospitals. Patients admitted to ICUs are at high risk for MRSA infection due to following reasons: more colonization of antimicrobial-agent resistant microorganisms such as MRSA in ICU, length of stay, severity of illness, and application of intravascular devices. Moreover, patients are admitted to ICU from different wards and discharged to many other wards or hospitals and can easily transmit MRSA inter or intra-hospitals (4). Prevalence of MRSA infection in ICUs varies in different regions, from 55% in the US and Iran to about 1% in the Netherlands (3, 5).

A programmed surveillance on screening of personnel, surfaces of departments of hospitals and patients in terms of nosocomial microorganisms such as MRSA is one the strategies to control infection (4). The first step to control the MRSA infection is having suitable and enough knowledge about the rate of infections, contamination, and colonization of the bacteria in the healthcare systems (4). Al-
2. Methods

2.1. Collection and Identification of Bacterial Isolates

The current cross sectional study was conducted in Karaj on three teaching hospitals from July 2013 to July 2014. The majority of S. aureus isolates were isolated from personnel and surfaces of hospitals. Sample collection from personnel was performed twice during the study. Specimens were collected by swabbing both nostrils of the personnel and subsequently culturing on the brain-heart infusion (BHI) agar. Sample collection from the surfaces of the three hospitals was performed monthly and cultured on the same media. The following hospital departments were included in the study: hospital No.1, emergency, burns, ICU, and internal medicine; hospital No. 2, females’ surgery, neonates, children, emergency, and ICU; and hospital No. 3, dialysis, angiosis, ICU, emergency, and males’ internal medicine. The isolates were cultured on the sheep blood agar and mannitol salt agar, and identified by conventional biochemical tests including catalase, tube coagulase, mannitol fermentation, and DNase (6). All S. aureus species isolated from patients were included in the study.

2.2. Antimicrobial Susceptibility Testing

The clinical and laboratory standards institute (CLSI) guideline was used for antimicrobial susceptibility tests by the standard disk diffusion method. The tested antibiotics included cefotaxime (30 µg), tigecycline (15 µg), vancomycin (30 µg), linezolid (30µg), synercid (quinupristin/dalfopristin) (15 µg), mupirocin (20 µg), teicoplanin (30 µg), and rifampicin (5 µg) (Mast, UK). S. aureus ATCC 25923 was used as the control strain.

2.3. DNA Extraction and Identification of High-Level Mupirocin Resistant and MRSA Isolates

DNA was extracted from S. aureus colonies by boiling method described previously (7). The polymerase chain reaction (PCR) was the amplification of S. aureus-specific nuclease (nuca) gene as described previously (8). Moreover, all isolates were subjected to mupA and mecA-specific PCR to identify high-level mupirocin resistant and MRSA isolates, respectively (7, 9).

2.4. Statistical Analysis

SPSS version 16 was used to analyze data (SPSS, Chicago, IL, USA). Differences of S. aureus and MRSA isolates frequencies among hospitals and departments were assessed using chi-square test. A P-value of < 0.05 was considered statistically significant.

3. Results

One hundred sixty-eight S. aureus and 49 MRSA species were isolated from Karaj teaching hospitals. The number and frequency of the isolates are shown in Table 1. More S. aureus and MRSA species were isolated from hospital No.1 and the P-value was significant (< 0.05). The number and frequency of S. aureus and MRSA species isolated from surfaces of different units of hospitals are shown in Table 2. Almost 75% of MRSA species were isolated from ICU departments and the P-value was significant. Table 3 shows the No. and frequency of S. aureus and MRSA isolates in personnel. Finally, antibiotic susceptibility pattern of MRSA and methicillin-sensitive S. aureus (MSSA) isolates is shown in Table 4. Although 10 S. aureus isolates were resistant to mupirocin based on disk diffusion findings; mupA was not observed in the isolates and accordingly it is concluded that the isolates had low resistance to mupirocin.

5. Discussion

Antibiotic pressure and indiscriminate administration of broad-spectrum antibiotics lead to high prevalence of resistant S. aureus especially MRSA in the world (1, 10). The contamination of hospital environments, especially ICUs, with MRSA is the main concern in recent years. Nevertheless, there are limited reports on the incidence rates of MRSA infection in ICUs in Iran (11-13). Furthermore, ICU-acquired infections are reported with high morbidity and mortality in Iran (14, 15). In the current study, the prevalence of MRSA varied greatly between investigated hospitals, from 16% to 40% (Table 1). As compared with the recent studies findings, the data showed that the rate of MRSA was lower than those of the other studies in Iran (52.7% - 93.3%), India (78%), Oman (52%), Australia (30.3%), and Turkey (35% - 43%) (1, 5, 16-18). According to Table 1, the frequency rate of MRSA in hospital No. 1 was higher than the other two hospitals. Some important reasons for the higher rate in this hospital included the high number of admitted patients, poor hygiene, and lack of infection control strategies. Despite low rate of MRSA colonization in Karaj hospitals, out
Table 1. Number and Frequency of *Staphylococcus aureus* and MRSA Species Isolated From Karaj Teaching Hospitals

| Hospital | Surface | MRSA (%) | Personnel | MRSA (%) | Patient | MRSA (%) | S. aureus | MRSA (%) |
|----------|---------|----------|-----------|----------|---------|----------|-----------|----------|
| No. 1    | 51      | 23 (45)  | 35        | 10 (28.5)| 4       | 3 (75)   | 90        | 36 (40)  |
| No. 2    | 25      | 3 (12)   | 11        | 4 (36)   | 6       | 0        | 42        | 6 (16.5) |
| No. 3    | 22      | 2 (9)    | 6         | 0        | 8       | 4 (50)   | 36        | 7 (16.5) |
| Total    | 98      | 28 (28.5)| 52        | 13 (25)  | 18      | 7 (39)   | 168       | 49 (29)  |

Table 2. Number and Frequency of *Staphylococcus aureus* Species Isolated From Surfaces of Karaj Teaching Hospitals

| Department | ICU | Emergency | Internal Medicine | Surgery | Neonates | Burns | Angiography | Dialysis | Total |
|------------|-----|-----------|-------------------|---------|-----------|-------|-------------|----------|-------|
| No. of S. aureus (%) | 43 (44.5) | 19 (19) | 14 (14.5) | 4 (4) | 1 (1) | 2 (2) | 2 (2) | 98 (100) |
| No. of MRSA (%) | 21 (75) | 2 (7.25) | 2 (7.25) | 1 (3.5) | 1 (3.5) | 1 (3.5) | 0 | 28 (100) |

Table 3. Number and Frequency of *Staphylococcus aureus* and MRSA Species isolated From Personnel of Karaj Teaching Hospitals

| Hospital | No. 1 | No. 2 | No. 3 | Total (%) |
|----------|-------|-------|-------|-----------|
| No. of samples (each nostril) | 98    | 98    | 23    | 219       |
| No. and frequency of S. aureus isolates (%) | 25 (25.5) | 7 (7) | 4 (17) | 36 (16.5) |
| No. and frequency of personnel carrying MRSA (%) | 6 (6) | 2 (2) | 0 | 8 (3.5) |

Table 4. Antibiotic Susceptibility Pattern of MRSA and MSSA Isolates

| Antibiotic (Disk) | MRSA (N = 49) | MSSA (N = 119) | Total (N = 168) |
|-------------------|---------------|----------------|-----------------|
|                   | No. of Resistant Isolates (%) | No. of Resistant Isolates (%) | No. of Resistant Isolates (%) |
| Rifampicin        | 26 (53)       | 1 (0.8)        | 27 (16)         |
| Mupirocin         | 4 (8)         | 6 (5)          | 10 (6)          |
| Teicoplanin       | 0             | 0              | 0               |
| Vancomycin        | 0             | 0              | 0               |
| Synercid          | 0             | 0              | 0               |
| Linezolide        | 0             | 0              | 0               |
| Tigecyclin        | 0             | 0              | 0               |

of 98 and 28 *S. aureus* and MRSA isolates, 43 (44.5%) and 21 (75%) species were isolated from ICU surfaces, respectively (Table 2) which showed an alarm in the hospitals. High frequency of MRSA in ICU was in agreement with those of previous studies (19). Also in the studies conducted in Tehran, *S. aureus* were the most frequent bacteria isolated from ICU surfaces (13, 20). Factors such as prolonged hospital stay, infection sites, invasive procedures, underlying disease conditions, and exposure to multidrug-resistant bacteria mostly cause higher rates of infection among patients in ICUs (11).

Although health care workers are the main reservoirs for MRSA, they may be the victims in the health-care settings. The nasal carriage rate of *S. aureus* and MRSA in the personnel in the current study was 16.5% and 3.5%, respectively, which was lower than those of other studies conducted in other regions of Iran (1, 21, 22). However, another study from Ghana reported low nasal carriage among inpatients and health staff (23). Some factors such as quality and size of samples, application of different techniques, and different interpretation guidelines may affect the prevalence of nasal carriage of *S. aureus* strains (21). Similar to high contamination with *S. aureus* and MRSA in hospital No. 1, Table 3 also shows the highest colonization of personnel with those strains in this hospital. These data also approved the probable poor hygiene and lack of infection control strategies in the mentioned hospital. However, the number of studied personnel in hospital No. 3 was less than those of other hospitals and it was a limitation in the current study.

The ICUs are the regions with remarkable drug usage and high frequency of drug-resistant pathogenic bacte-
Mupirocin can be the cause of such high resistance in current study isolates. Since mupirocin was not used to eradicate MRSA. In spite of low resistance to mupirocin in the isolates, an antibiotic policy is urgently needed to prevent the re-emergence of MRSA. In contrast with a previous study (1), there was no significant resistance to methicillin and mupirocin in the current study S. aureus isolates. In spite of low resistance to mupirocin in the isolates, high susceptibility to mupirocin was not observed in the current study isolates. Since mupirocin was not used to eradicate MRSA in the personnel of the mentioned hospitals, in agreement with other studies, previous exposure to mupirocin can be the cause of such high resistance in S. aureus isolates. Moreover, low rate of resistance to mupirocin was more common among MRSA than MSSA isolates (8% versus 5%). The prevalence of mupirocin-resistant MRSA was different in studies from Jordan (2.6%), Greek (1.6%), Korea (5%), China (6.6%), and Iran (70%) (1). In conclusion, in comparison with the other studies from Iran, there was low frequency of MRSA in the investigated hospitals. However, high frequency of MRSA (75%) in ICU indicated that an antibiotic policy is urgently needed to prevent the resistance development. Moreover, antibiotic susceptibility monitoring and regular screening of the hospital surfaces and personnel in terms of MRSA colonization, especially ICUs, are indispensable.

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Mupirocin is one of the most effective antibiotics currently used to eradicate MRSA. In contrast with a previous study (1), there was no simultaneous resistance to methicillin and mupirocin in the current study S. aureus isolates. In spite of low resistance to mupirocin in the isolates, high susceptibility to mupirocin was not observed in the current study isolates. Since mupirocin was not used to eradicate MRSA in the personnel of the mentioned hospitals, in agreement with other studies, previous exposure to mupirocin can be the cause of such high resistance in S. aureus isolates. Moreover, low rate of resistance to mupirocin was more common among MRSA than MSSA isolates (8% versus 5%). The prevalence of mupirocin-resistant MRSA was different in studies from Jordan (2.6%), Greek (1.6%), Korea (5%), China (6.6%), and Iran (70%) (1). In conclusion, in comparison with the other studies from Iran, there was low frequency of MRSA in the investigated hospitals. However, high frequency of MRSA (75%) in ICU indicated that an antibiotic policy is urgently needed to prevent the resistance development. Moreover, antibiotic susceptibility monitoring and regular screening of the hospital surfaces and personnel in terms of MRSA colonization, especially ICUs, are indispensable.

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