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Research Article

# Nasal carriage of methicillin-resistant Staphylococci among healthcare workers of a university teaching hospital, Iran

## Nasale Trägerrate Methicillin-resistenter Staphylokokken bei medizinischem Personal eines iranischen Universitätslehrkrankenhauses

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## Abstract

**Background:** The opportunistic pathogens, methicillin-resistant-*Staphylococcus aureus* (MRSA) and *Staphylococcus epidermidis*, are associated with severe nosocomial infections and high levels of mortality. Healthcare workers colonized with either MRSA or methicillin-resistant *S. epidermidis* (MRSE) in the nasal cavity are high risk groups for transmitting the agent to hospitalized patients.
**Objective:** This study was carried out to investigate the prevalence of nasal carriage of MRSA and methicillin-resistant *S. epidermidis* among healthcare providers of Imam Reza University Teaching Hospital, Tabriz, Iran.
**Methods:** A total of one hundred two nasal swabs were obtained from participants working on different wards of the hospitals. The antibiotic resistance pattern was investigated using disk diffusion methods, which were subsequently evaluated by polymerase chain reaction (PCR) for the *mecA* gene.
**Results:** In the screened population, 22 isolates of *S. aureus* and 72 of *S. epidermidis* were detected. Of these, 7 isolates of *S. aureus* and 36 of *S. epidermidis* were cefoxitin resistant. Three isolates of *S. aureus* isolates and 35 of *S. epidermidis* were MRSE and positive for *mecA* amplification. Moreover, all isolates were penicillin G resistant but vancomycin and linezolid sensitive. High resistance was observed to clindamycin (74%).
**Conclusions:** The present study indicates that healthcare workers are at high risk of acquisition and transmission of methicillin-resistant Staphylococci. Early screening and decolonization of hospital staff, as well as education on standard sanitation measures, especially hand hygiene practice, remain the most effective strategies for controlling transmission of infectious agents.

**Keywords:** MRSA, methicillin-resistant S. epidermidis, nasal carrier, healthcare worker, infection control

## Zusammenfassung

**Hintergrund:** Die opportunistischen Erreger Methicillin-resistenter *Staphylococcus aureus* und Methicillin-resistenter *Staphylococcus epidermidis* (MRSE) können schwer verlaufende nosokomiale Infektionen mit hoher Mortalität verursachen. Medizinisches Personal, das entweder mit MRSA oder mit MRSE in der Nasenhöhle kolonisiert ist, kann die Erreger auf hospitalisierte Patienten übertragen.
**Zielsetzung:** Die Studie wurde durchgeführt, um die Prävalenz der nasalen Vorkommens von MRSA und MRSE bei medizinischem Personal des Imam Reza-Universitätslehrkrankenhauses in Tabris, Iran, zu untersuchen.

**Methoden:** Es wurden 102 Abstriche im Vestibulum nasi von medizinischem Personal auf verschiedenen Stationen der Krankenhäuser entnommen. Das Antibiotikaresistenzmuster wurde im Plättchendiffusionstest untersucht; anschließend wurde das mecA-Gen mittels Polymerase-Kettenreaktion (PCR) nachgewiesen.
**Ergebnisse:** Bei 22 Personen (21,6%) wurde *S. aureus*, bei 72 Personen (70,6%) *S. epidermidis* nachgewiesen. Davon waren 7 *S. aureus*- und 36 *S. epidermidis*-Isolate Cefoxitin resistent. Drei *S. aureus*- und 35 *S. epidermidis*-Isolate enthielten das mecA-Gen. Darüber hinaus waren alle Isolate resistent gegen Penicillin G, aber sensitiv gegen Vancomycin- und Linezolid. Es wurde eine hohe Resistenz gegenüber Clindamycin beobachtet (74%).
**Schlussfolgerungen:** Die Studie weist darauf hin, dass Beschäftigte im Gesundheitswesen dem Risiko des Erwerbs und der Übertragung Methicillin resistenter Staphylokokken ausgesetzt sind. Früherkennung und Dekolonisierung des Krankenhauspersonals und die Aufklärung über die Einhaltung der Basishygienemaßnahmen, insbesondere der Händedesinfektion, sind nach wie vor die wichtigste Strategie zur Prävention nosokomialer Infektionen.

**Schlüsselwörter:** MRSA, Methicillin-resistanter S. epidermidis, nasaler Träger, medizinisches Personal, Infektionskontrolle

# Introduction

As a normal inhabitant of the body, *Staphylococcus aureus* is frequently found in the respiratory tract, nose, and skin. Approximately 20% to 60% of the healthy population are asymptomatic nasal carriers of *S. aureus* [1], [2]. However, nasal colonization of methicillin-resistant *S. aureus* (MRSA), whether transient or persistent, poses a hazard in nosocomial environments [3]. Complicated MRSA infections result in an additional burden for both patients and healthcare settings, due to prolonging the hospital stay and increasing the mortality rate [4], [5]. *Staphylococcus epidermidis* is prevalent in skin and mucosa microflora, and is also associated with nosocomial infections [6]. Immunocompromised and hospitalized patients with medical devices, such as catheters and medical implants, are in serious danger of *S. epidermidis* colonization [7], [8]. Thus, the incidence of *S. epidermidis* – particularly with a resistance profile – is a great concern on many hospital wards [9]. Healthcare workers may carry either MRSA or methicillin-resistant *S. epidermidis* in the nasal cavity, without manifesting disease. Since MRSA transmission generally occurs by direct physical contact, carriers with inadequate hygiene may inadvertently endanger hospitalized patients [10]. Moreover, self-infection of carriers and introduction of pathogens to colleagues and community are further consequences of carriage status [2]. Findings indicate that screening and identification of asymptomatic carriers is a key measure to reduce intra-hospital transmission and subsequent risks [4]. The aim of this study was to evaluate the prevalence of nasal mucosa colonization of MRSA and methicillin-resistant *S. epidermidis* (MRSE) among healthcare providers.

# Methods and materials

## Study design

This cross-sectional study was conducted from June to November 2017 and December 2019 at Imam Reza University Teaching Hospital in Tabriz, Iran. The study was approved by the National ethics committee (registration number IR.TBZMED.VCR.REC.1398.401).
Healthcare workers voluntarily participated in the study, and written informed consent with individual variables was obtained from each participant. Volunteers who had received antibiotic treatment for the previous 10 days were excluded from the study. A total of 102 nasal swabs were collected from participants and inoculated into tryptic soy broth (TSB) medium (Merck, Germany), then transferred to the microbiology laboratory of Imam Reza Hospital. Samples were inoculated on blood agar containing 5% sheep blood and incubated at 35 °C for 24 h. The isolates were identified by conventional microbiological methods such as gram-staining, catalase test, coagulase test, and DNase test.

## Antibiotic susceptibility testing

*Staphylococcus* spp. isolates were examined for antibiotic susceptibility using a modified Kirby-Bauer disk diffusion method, according to Clinical and Laboratory Standard Institute (CLSI) guidelines [11]. The antibiotic disks (Mast, UK) used for susceptibility testing comprised penicillin G (10 units), vancomycin (30 µg), gentamicin (10 µg), amikacin (30 µg), ciprofloxacin (5 µg), clindamycin (2 µg), chloramphenicol (30 µg), linezolid (30 µg), cefoxitin

(30 µg), novobiocin (30 µg), and erythromycin (15 µg) [12].

## Polymerase chain reaction (PCR)

Isolates with a cefoxitin-resistant profile were further subjected to PCR assay to confirm the presence of the *mecA* gene. DNA extraction was performed using the tissue buffer boiling method [13]. The assay was conducted in 25 µL reaction mixture with the following thermal program: initial denaturation at 95°C for 6 min, followed by 35 cycles of denaturation at 95°C for 1 min, annealing at 52°C for 30 sec, extension at 72°C for 45 sec, and a final extension of 7 min at 72°C in a thermal cycler (BIO-RAD, T100™ Thermal Cycler). *S. aureus* ATCC® 49476™ was used as the positive control. Primers (F-AAAATCGATGGTAAAGGTTGGC and R-AGTTCTGCAGTACCGGATTTGC) were used for *mecA* amplification.

# Results

## Demographic information

A total of 102 healthcare workers were screened for *Staphylococcus* spp. nasal colonization, of which 78 (76.47%) were female and 26 (25.49%) were male. Participants' age ranged from 20 to 48 years. Participants were classified by their position: physician, nurse, and paramedic from intensive care units (ICUs), surgical wards and infectious wards. Of the screened population, 22 (23.4%) samples were gram-positive, catalase-, coagulase-, and DNase-positive, which were classified as *S. aureus*; 72 (76.6%) were coagulase-negative and novobiocin susceptible, and identified as *S. epidermidis*. Neither gram-negative bacteria nor Enterococci were detected.

## Antimicrobial susceptibility profiles of isolates

Inhibition zone diameter was interpreted according to CLSI guidelines. Isolates were classified as resistant (R), intermediate (I), and sensitive (S). All isolates were resistant to penicillin G, and sensitive to vancomycin and linezolid. Of 22 *S. aureus* isolates, 7 were cefoxitin resistant; of the 72 *S. epidermidis* isolates, 36 presented resistance to cefoxitin. The highest resistance was observed for clindamycin, with 74.46% being resistant and 3.19% intermediate. For the remaining antimicrobials, the resistance profile was as follows: cefoxitin, 56.38% R; erythromycin, 53.2% R, 8.51% I; ciprofloxacin, 26.6% R, 5.32% I; gentamicin, 21.27% R; amikacin, 10.63% R, 1.06% I; and chloramphenicol, 2.12% R, 1.06% I.

## PCR results

Of the 22 *S. aureus* isolates, 7 were cefoxitin resistant according to the disk diffusion method. Of these, 3 isolates were positive for the *mecA* gene, as shown by the PCR method (Figure 1). For *S. epidermidis*, PCR molecular test results were positive for 35 of the 36 cefoxitin-resistant isolates, and 35 isolates harbored the *mecA* gene (Figure 2).

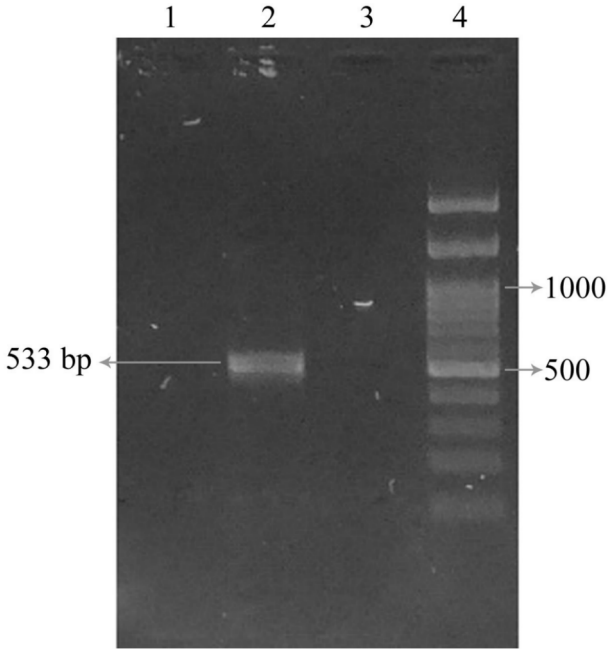


**Figure 1: PCR product of *mecA* gene amplified from *S. aureus* isolate. Lane 1: negative control; Lane 2: *mecA* positive *S. aureus* isolate; Lane 3: *mecA* negative *S. aureus* isolate; Lane 4: 100–1,200 bp DNA ladder**

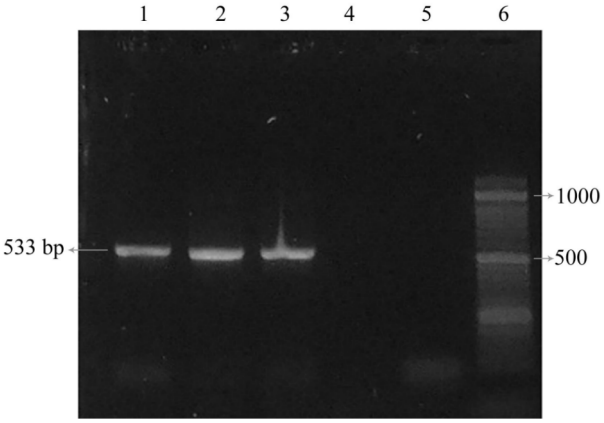


**Figure 2: PCR products of *mecA* gene amplified from *S. epidermidis* isolates. Lane 1: *S. aureus* ATCC® 49476™ as positive control; Lanes 2 and 3: *mecA* positive *S. epi*dermidis isolates; Lane 4: *mecA* negative *S. epidermidis* isolate; Lane 5: negative control; Lane 6:100–1,200 bp DNA ladder**

## Discussion

Staphylococci account for almost 30% of nosocomial infections and 50% of nosocomial septicemia [14]. Hospital staff colonized with methicillin-resistant staphylococci are high risk groups for transmitting the agent to patients. In the present study, we attempted to evaluate the nasal carriage of staphylococci among healthcare workers. Our results indicate that 23.4% of the screened population was recognized as *S. aureus* carriers. This prevalence is comparable to previously conducted studies from various countries, 29% in the US [15], 19.1% in Spain [16], 22.6% in Egypt [14], and 27% in Iran [17]. Higher colonization rates were reported from Nigeria, Brazil, and Saudi Arabia with 32%, 40.8%, and 43.2%, respectively [18], [19], [20]. However, the overall carriage rate of MRSA was low in this study (2 out of 75 isolates), similar to the observations from Italy [21] and Saudi Arabia [22]. In contrast, another study from Iran reported the prevalence of MRSA nasal colonization to be 32% [17], considerably higher than that of our study. On the other hand, *S. epidermidis* was the predominant bacterium in this study (76.6%), similar to rates reported elsewhere. The methicillin-resistant gene distribution among *S. epidermidis* isolates was lower than in previous studies (49.12% vs. 70.1%, 87.1%, and 96.25%) [7], [9], [23]. Our data show that the overall resistance in *S. epidermidis* was considerably higher compared to *S. aureus* isolates. This supports Najar-Peeraye et al.'s speculation that methicillin-resistant coagulase-negative Staphyloccoci show higher resistance rates than does *S. aureus* [6].

Of the *S. aureus* isolates, 7 showed cefoxitin-resistant profiles in the disk-diffusion test; however, only two isolates were positive for the *mecA* gene. This finding is similar to previous observations and provides evidence for the greater efficacy of PCR [24].

Analysis of antibiotic susceptibility data reveals that all isolates were susceptible to vancomycin and linezolid, which is in accordance with previous findings [6], whereas all isolates were penicillin G resistant. In a study conducted by Sadeghi et al., similarly high resistance was observed to clindamycin and erythromycin [25]. We hypothesize that this could be due to elevated consumption of these antibiotics in our region. Since these antibiotics, particularly clindamycin, are alternatives for *S. aureus* infections, high resistance may be a problem in healthcare settings.

It is important to point out that these organisms, particularly *S. epidermidis*, have significant potential to act as a reservoir and horizontally transfer antimicrobial and virulence genes [26]. For instance, *S. epidermidis* is known as a putative reservoir of cefoxitin- and mupirocin-resistance genes [27]. Acquisition of resistance to ß-lactams, quinolones, aminoglycosides, and glycopeptides by *S. aureus* contributes to further dissemination of resistant isolates and increased burden of nosocomial infections [28], [29]. Results from Bloemendaal et al.'s [30] study strongly support horizontal interspecies gene transmission. In that study, the authors indicate that *S. epidermidis* transferred a resistance gene to *S. aureus*, resulting in MRSA [30].

Self-infection of carriers is the other issue of concern. Studies indicate a correlation of skin and soft tissue infections with *S. aureus* nasal carriage [31], [32]. On average, 80% of patients with skin wounds were also *S. aureus* nasal carriers, and 65% of *S. aureus* strains isolated from nose and lesion had same phage type [33]. Moreover, von Eiff el al. [34] reported that the incidence of bacteremia was significantly greater in *S. aureus* carriers, in which 80% of bacterimic strains were identical to *S. aureus* isolates from nurses.

Since hospital settings and community are linked environments, it is necessary to implement control strategies for antimicrobially resistant bacteria. Screening and decolonization of healthcare providers with nasal antiseptics could be an effective strategy for controlling the transmission of infectious agents and probable subsequent infections [33].

This study has possible limitations. Many staff members did not participate in sample collection because of pain involved in sample collection, and only staff that completed the consent form were tested in this study.

## Conclusion

The present study indicates that health professionals are at high risk of acquisition and transmission of methicillin-resistant *Staphylococci*. Education on standard sanitation measures, especially hand hygiene practices, remains the most important factor in controlling the spread of MRSA. Early screening and identification of colonized healthcare workers may help reduce the transfer of resistance to sensitive strains and decrease the incidence of MRSA.

## Notes

### Competing interests

The authors declare that they have no competing interests.

### Acknowledgment

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