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Clinical and Demographic Correlates of MRSA and mecA-Positive Isolates in a Tertiary Hospital, Tehran: A Cross-Sectional Study

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ARTICLE INFO	ABSTRACT
Article type: Research Article	***Background*:** Methicillin-resistant *Staphylococcus aureus* (MRSA) is a major cause of healthcare-associated infections, with the mecA gene serving as a key marker of resistance. This study investigates the clinical and demographic factors associated with MRSA and mecA carriage in a tertiary hospital in Tehran to inform targeted infection control strategies..
	***Methods*:** This cross-sectional study included 125 hospitalized patients with *Staphylococcus aureus* infections from a tertiary hospital in Tehran. Clinical and demographic data were collected via structured questionnaires and medical records, and MRSA/mecA status was determined using phenotypic methods and real-time PCR, with associations analyzed using chi-square and Fisher's exact tests in SPSS v18..
	***Results*:** MRSA was identified in 40.8% of *Staphylococcus aureus* isolates, with no significant association found with age or sex. Prolonged hospitalization, immunocompromised status, and frequent antibiotic use were significantly associated with MRSA infection ($p < 0.05$), while a lower prevalence was observed among patients with infected wounds.
*Corresponding Authors: Haniyeh Bashi Zadeh Fakhar: Department of Genetics, Islamic Azad University, Science and Research Branch, Tehran, Iran. *Tel*: +98-21-22749213, *E-mail*: haniyehfakhar@yahoo.com.	***Conclusion*:** This study highlights a high prevalence of MRSA in a Tehran tertiary hospital, with prolonged hospitalization, immunocompromised status, and antibiotic overuse identified as key risk factors. Targeted infection control strategies integrating clinical risk assessment and microbiological surveillance are recommended to mitigate MRSA transmission.

Introduction

Methicillin-resistant *Staphylococcus aureus* (MRSA) is a major Gram-positive pathogen associated with both healthcare- and community-acquired infections, ranging from skin and soft tissue infections to life-threatening conditions such as sepsis and pneumonia (1). The global rise of multidrug-resistant MRSA, particularly healthcare-associated (HA-MRSA) strains, has made it a leading cause of nosocomial infections and a significant public health challenge (2). The World Health Organization (WHO) has warned that antimicrobial resistance could lead to 10 million annual deaths by 2050 if effective interventions are not implemented (3).

The primary mechanism of methicillin resistance is the acquisition of the mecA gene, which encodes penicillin-binding protein 2a (PBP2a), a variant with low affinity for β-lactam antibiotics (4). This gene is carried on the mobile genetic element SCCmec and serves as a definitive molecular marker for MRSA (5). While mecA detection via real-time PCR offers high sensitivity and specificity for rapid diagnosis (6), its presence alone does not fully explain the clinical and epidemiological patterns of MRSA transmission.

Epidemiological studies in Iran have demonstrated a high and variable prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA), with reported rates ranging from 36% to 47.5% in Tehran hospitals (7). A national meta-analysis estimated the overall prevalence of mecA-positive isolates at 52.7%, although regional variation is substantial, with rates spanning from 20.48% to 90% across different cities (8). Higher MRSA burdens are consistently observed in high-risk units such as intensive care units and emergency departments (9). Molecular characterization reveals that SCCmec type III is the predominant genotype in Iranian healthcare settings, accounting for 87% of isolates, followed by types IIIA and IV (10).

MRSA isolates in Iran exhibit high levels of multidrug resistance, particularly to macrolides (e.g., erythromycin: 90.9%), clindamycin (85.4%), and fluoroquinolones, yet remain universally susceptible to glycopeptides (vancomycin), oxazolidinones (linezolid), tigecycline, and daptomycin (9-10). Prior hospitalization is a significant risk factor for MRSA acquisition, although community-associated cases are also documented (11). Despite this body of evidence, data on the clinical and demographic correlates of MRSA and mecA carriage in hospitalized patients remain fragmented, particularly in relation to immune status, infection history, and antibiotic exposure patterns.

A meta-analysis comparing molecular and culture-based methods for MRSA screening in surgical patients revealed that PCR-based assays exhibit superior sensitivity (99.2%) and specificity (82.2%) compared to traditional culture techniques (12). Furthermore, rapid molecular screening in surgical units has been linked to lower MRSA acquisition rates, whereas patients screened using conventional culture methods were 1.49 times more likely to acquire MRSA (13).

Therefore, this cross-sectional study aims to evaluate the prevalence of MRSA and mecA-positive isolates among hospitalized patients in a tertiary care center in Tehran, with a specific focus on identifying clinical and demographic factors—including age, sex, hospitalization duration, immune status, and infection history—associated with MRSA and mecA carriage. By linking microbiological findings with patient characteristics, this study seeks to inform targeted infection control strategies and improve risk assessment in high-burden healthcare settings.

Materials and Methods

Study Design and Setting

This cross-sectional study was conducted at a tertiary-care hospital in Tehran, Iran, from

February 2023 to August 2024. The primary objective was to evaluate the clinical and demographic factors associated with methicillin-resistant *Staphylococcus aureus* (MRSA) and mecA-positive isolates among hospitalized patients. A total of 125 clinical specimens (blood, urine, and sputum) were collected from patients with clinically suspected or microbiologically confirmed infections.

Hospitalized patients with positive S. aureus cultures were included. Participants were recruited using a convenience sampling method. Exclusion criteria included polymicrobial infections, concurrent non-S. aureus infections, or underlying immunodeficiencies that could confound infection outcomes, ensuring a more homogeneous population for analysis. Written informed consent was obtained from all participants prior to enrollment.

Data Collection

Demographic and clinical data were collected through a structured, pre-tested questionnaire administered by trained research personnel during face-to-face interviews or via medical record review, ensuring consistency and accuracy. The questionnaire was designed based on standardized epidemiological tools and included socio-demographic variables such as age, sex, educational level, and hospital ward of admission.

Clinical variables encompassed duration of hospitalization (categorized as <7 days or ≥7 days), underlying medical conditions (e.g., diabetes mellitus, malignancy, chronic kidney disease), immunocompromised status (defined by conditions such as HIV, organ transplantation, or long-term corticosteroid use), and history of recurrent antibiotic use within the past six months. Additional variables included prior history of *Staphylococcus aureus* infection, presence of indwelling medical devices (e.g., catheters, ventilators), surgical procedures within the last year, and existence of chronic wounds or infected ulcers—recognized risk factors for MRSA acquisition.

All data were anonymized and entered into a secure database using double-entry verification to minimize transcription errors. The collected information was then linked to microbiological results (MRSA status) and molecular findings (mecA positivity) for subsequent correlation and statistical analysis. This integrated approach enabled the assessment of potential associations between host-related factors and the presence of resistant S. aureus isolates, supporting a comprehensive evaluation of risk profiles in the hospital setting.

Microbiological Identification and Phenotypic Confirmation of MRSA

S. aureus isolates were identified using standard microbiological methods, including Gram staining, catalase and coagulase tests (slide and tube), DNase production, and mannitol fermentation on mannitol salt agar (MSA). Species identity was further confirmed by amplification of the 16S rRNA gene.

Methicillin resistance was phenotypically assessed using oxacillin screening agar (Mueller-Hinton agar supplemented with 4% NaCl and 6 µg/mL oxacillin), incubated at 35°C for 24 hours. Isolates showing growth were classified as MRSA.

Molecular Detection of mecA Gene

Genomic DNA was extracted from bacterial pellets using the QIAamp DNA Mini Kit (Qiagen, Germany) following cell lysis with lysostaphin (30 mg/mL, 37 °C, 4 h). DNA concentration and purity were assessed via spectrophotometry (NanoDrop), with samples of acceptable A260/A280 (1.8–2.0) stored at −20 °C.

The presence of the mecA gene was confirmed by real-time PCR using SYBR Green Master Mix in a 20 µL reaction containing template DNA, gene-specific primers, and nuclease-free water.

Amplification was performed on an ABI 7300 Real-Time PCR system with an annealing temperature of 52 °C. A 214 bp fragment was targeted, and results were analyzed based on amplification curves and quantification cycle (Ct) values. Negative (nuclease-free water) and positive (known MRSA strain) controls were included in all runs.

Statistical Analysis

Data analysis was performed using IBM SPSS Statistics (version 18.0). Descriptive statistics were used to summarize the study variables. Continuous variables, such as age and hospitalization duration, were presented as mean ± standard deviation (SD), while categorical variables—including sex, immune status, antibiotic exposure, prior infection history, and MRSA/mecA status—were reported as frequencies and percentages.

The primary objective of inferential analysis was to identify clinical and demographic factors associated with MRSA infection and mecA gene carriage. The Chi-square test (χ^2) was employed to assess the association between categorical variables. When expected cell counts were less than 5, Fisher's exact test was used as an alternative to ensure validity. Crude associations were expressed as proportions with p-values, and a two-sided p-value < 0.05 was considered statistically significant.
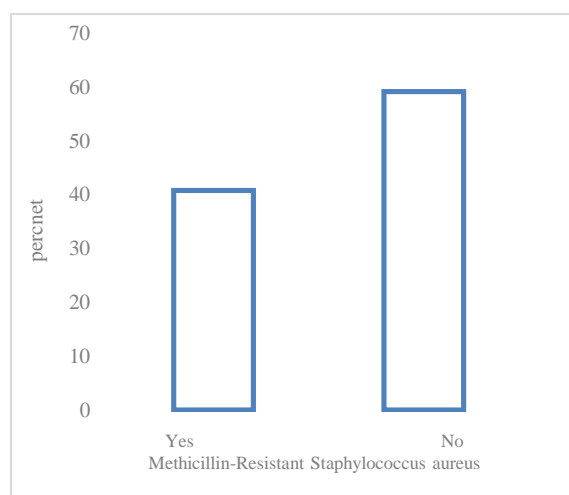
To evaluate the strength of associations, odds ratios (OR) with 95% confidence intervals (CI) were calculated for significant variables in univariate analysis. This approach enabled a more interpretable assessment of risk factors for MRSA and mecA-positive isolates, supporting evidence-based conclusions for infection control planning.

Results

Microbiological and molecular procedures were performed as part of a broader diagnostic project; however, the clinical correlation and epidemiological analysis presented here are original and not previously reported. These findings highlight specific patient subgroups at higher risk for MRSA, particularly those with prolonged hospitalization, immunosuppression, and repeated antibiotic exposure.

The mean age of the 125 patients with *Staphylococcus aureus* infection was 55.68 ± 17.65 years, with a nearly balanced distribution between individuals under 60 years (50.40%) and those aged 60 years or older (49.60%), indicating a representative sample across adult age groups. The study population included slightly more females (52%) than males (48%), reflecting the demographic composition of hospitalized patients in the study setting.

Figure 1. Frequency of MRSA in the studied samples.

MRSA was identified in 40.80% (51 out of 125) of S. aureus isolates, confirming a high burden of methicillin resistance in this tertiary hospital (figure 1). However, no statistically significant association was observed between MRSA infection and age group (38.71% in patients ≥60 years vs. 42.86% in those <60 years, p = 0.637),

Table 1.   Frequency of methicillin-resistant *Staphylococcus aureus* according to study variables.

Variable		Yes (Resistant) (N:51)*	No (N:74)**	P-value
		Percent (100 %)		
Age	Less than 60 yrs	27 (42.86)	36 (57.14)	0.637
	More than 60 Yrs	24 (38.17)	38 (61.29)	
Sex	Male	22 (36.67)	38 (63.33)	0.366
	Female	29 (44.62)	36 (55.38)	
Patients with a history of hospitalization	Yes	28 (79.41)	39 (21.58)	0.001
	No	23 (66.39)	35 (34.60)	
Frequent use of antibiotics	Yes	44 (38.6)	70 (61.4)	0.001
	No	7 (63.64)	4 (36.36)	
Immunodeficiency disease	Yes	19 (47.5)	21 (52.5)	0.001
	No	32 (37.65)	56 (62.35)	
History of *Staphylococcus* infection	Yes	21 (38.18)	34 (61.82)	0.598
	No	30 (42.86)	40 (57.14)	
History of infected wound	Yes	33 (38.37)	53 (61.63)	0.001
	No	18 (46.15)	21 (53.85)	
Sensitive to antibiotics	Vancomycin and rifampin	39 (45.35)	47 (54.65)	0.124
	Others	12 (30.77)	27 (69.23)	
Resistance to antibiotics	P,TE,CN,CP,CZ,GN,CF,E	36 (47.37)	40 (52.63)	0.063
	Others	15 (30.61)	34 (69.39)	

suggesting that age alone may not be a dominant risk factor for MRSA acquisition in this cohort. Similarly, although a higher proportion of female patients were infected with MRSA (44.62% vs. 36.67% in males), this difference did not reach statistical significance (p = 0.366), possibly due to balanced distribution or insufficient power to detect gender-based differences.

In contrast, several clinical factors showed strong and significant associations with MRSA. Prolonged hospitalization (≥7 days) was significantly linked to MRSA infection (41.79% vs. 39.66% in shorter stays, p = 0.001), highlighting the role of healthcare exposure in the development of resistant infections. Immunocompromised status was also a significant predictor, with MRSA detected in 47.50% of immunocompromised patients compared to 37.65% of immunocompetent individuals (p = 0.001), underscoring the vulnerability of this subgroup.

Furthermore, patients with a history of frequent antibiotic use had a markedly higher prevalence of MRSA (63.64%) compared to those without such history (38.60%, p = 0.001), reinforcing the impact of antimicrobial pressure on resistance selection.

Interestingly, MRSA was less prevalent among patients with infected wounds (38.37%) compared to those without such history (46.15%, p = 0.001), a counterintuitive finding that may reflect differences in infection source (e.g., community vs. hospital), prior treatment, or wound management practices. No significant associations were found between MRSA and prior history of S. aureus infection or multidrug resistance patterns, suggesting that these factors may not independently drive MRSA acquisition in this population (Table 1).

Discussion

In this study , Microbiological and molecular procedures were performed as part of a broader

diagnostic project; however, the clinical correlation and epidemiological analysis presented here are original and not previously reported

MRSA prevalence in Iran demonstrates significant variation across different populations and time periods. A comprehensive meta-analysis by Dadashi et al. (2018)(14) found an overall MRSA frequency of 43.0% among confirmed S. aureus isolates, with higher prevalence observed in studies conducted after 2000. Among healthcare workers specifically, Jalali et al. (2020) (15) reported a concerning prevalence of 31.7%, with substantial regional variation ranging from 0% in Bandar Abbas to 96.6% in Qom. In intensive care units, Goudarzi et al. (2016) (16) documented an exceptionally high MRSA prevalence of 93.3% among hospitalized patients in Tehran hospitals. A broader review by Ghasemian & Mirzaee (2016) estimated the overall MRSA prevalence at approximately 56.5%, with higher rates (62.2%) observed in 2015-2016 compared to earlier years (17) .

This rate is comparable to findings from other regional studies, such as 47.56% in Fars Province (18) and 38.64% in central Iran (19) , yet lower than the 75.6% reported in high-risk populations in other regions (20) . The observed variation in MRSA prevalence may be attributed to differences in patient demographics, hospital infection control practices, antibiotic prescribing patterns, and methodological approaches.

This study demonstrates a high prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) (40.80%) among hospitalized patients in a tertiary care hospital in Tehran, reflecting the substantial burden of healthcare-associated infections in Iranian medical settings.

Research on demographic factors and MRSA acquisition presents mixed findings. Ajao et al. (2013) (21) found that MRSA colonization was significantly associated with subsequent infection but this association did not differ significantly by age group among 7,405 patients. However, Kupfer et al. (2010) identified male gender as a significant risk factor for MRSA acquisition (p<0.001) in their analysis of 798 MRSA cases, with 75% of MRSA-positive patients being over 50 years of age (22). Asensio et al. (1996) (23) demonstrated that age was independently associated with MRSA infection/colonization, with each 10-year increase in age showing an odds ratio of 1.3.

Notably, no significant association was observed between MRSA infection and age or sex, suggesting that demographic factors alone may not be primary drivers of MRSA acquisition in this setting. This finding aligns with studies indicating no significant gender-based disparity in MRSA rates (24) , although some reports suggest higher susceptibility among elderly or male patients (25-26). Instead, clinical and healthcare-related factors emerged as more influential determinants of MRSA colonization and infection.

Prolonged hospitalization (≥7 days) was significantly associated with MRSA infection (p = 0.001), reinforcing the role of healthcare exposure in the development of resistant infections. Extended hospital stays increase the risk of cross-transmission, invasive device use, and cumulative antibiotic exposure—key contributors to MRSA emergence (27). Similarly, immunocompromised status was a significant predictor of MRSA, with a prevalence of 47.50% in this subgroup compared to 37.65% in immunocompetent individuals (p = 0.001). This highlights the increased vulnerability of patients with underlying conditions such as diabetes, malignancy, or those receiving immunosuppressive therapy, who are at greater risk of acquiring multidrug-resistant pathogens (28, 29).

Research consistently demonstrates a strong association between antibiotic exposure and methicillin-resistant *Staphylococcus aureus* (MRSA) development. A comprehensive meta-analysis of 76 studies involving 24,230 patients found that antibiotic exposure increased MRSA risk by 1.8-fold, with quinolones showing the highest risk (RR=3.0), followed by glycopeptides (RR=2.9) and cephalosporins (RR=2.2) (30). This

relationship reflects antimicrobial selection pressure that maintains resistance within populations (31). Real-world evidence from Scotland demonstrated that national antibiotic stewardship programs targeting specific antibiotics (4C antibiotics and macrolides) reduced MRSA prevalence density by 54% in hospitals and 37% in communities (32). Modeling studies further confirm that antibiotic prescription patterns significantly influence MRSA spread, particularly in intensive care units where CA-MRSA prevalence can range from 3-20% depending on exposure patterns (33). These findings collectively support implementing robust antimicrobial stewardship programs to combat MRSA emergence.

Recurrent antibiotic use was strongly linked to MRSA (p = 0.001), with 63.64% of MRSA cases occurring among frequent users. This finding underscores the impact of antimicrobial pressure in selecting resistant strains, a well-documented phenomenon in both hospital and community settings (34). The high rate of prior antibiotic exposure (91.2%) in our cohort further emphasizes the urgent need for robust antimicrobial stewardship programs in Iranian healthcare facilities.

Interestingly, MRSA was less prevalent among patients with infected wounds (38.37% vs. 46.15%, p = 0.001), a counterintuitive result that may reflect differences in infection source—such as a higher proportion of methicillin-sensitive S. aureus (MSSA) in chronic wounds—or prior targeted therapy reducing MRSA burden. This contrasts with studies linking wound infections to MRSA (35). suggesting possible regional variations in strain distribution, infection control practices, or host-pathogen interactions.

The mecA gene was detected in 21.57% of isolates, which is lower than some reports, such as 78.12% in burn patients in Tunisia (36) . This discrepancy may reflect differences in SCCmec types, detection sensitivity, or gene expression levels. Real-time PCR confirmed mecA in a subset of isolates, supporting its role in molecular confirmation of resistance, although phenotypic methods like oxacillin screening remain valuable in resource-limited settings due to cost-effectiveness (37, 38).

Conclusion

This study reports a high MRSA prevalence (40.8%) among hospitalized patients in Tehran, driven by prolonged hospitalization, immunosuppression, and antibiotic use—highlighting antibiotic pressure as a key resistance driver. The mecA gene was confirmed in isolates, albeit at variable rates, and MRSA was less common in wound infections. Findings support targeted infection control integrating clinical risk assessment and microbiological surveillance.

Ethics approval and consent to participate

No formal ethical code was applicable to this retrospective analysis, as it utilized anonymized laboratory data without direct human subject involvement..

Conflict of interest

The authors declare no conflict of interest.

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